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Grain yield and dry matter accumulation response to enhanced panicle nitrogen application under different planting methods (*Oryza sativa* L.)

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Abstract

A four-year field experiment (2006-2009) was conducted to assess the effect of enhanced panicle nitrogen application on rice yield and its dry matter accumulation characteristics in direct-seeded (DSR) and transplanted rice (TPR). The field experiment included two planting patterns (TPR and DSR) and four nitrogen treatments: (F1) low panicle nitrogen fertilizer (PNF) with high total nitrogen amount (TNA; 225kg N ha^{-1}); (F2) low PNF with low TNA (150kg N ha^{-1}); (F3) high PNF with high TNA; (F4) high PNF with low TNA. There was little difference in mean yield of planting patterns between F2 and F4. However, the mean yield in F1 was 10.15% greater than in F3. The mean total dry matter in F3 was 12.0% greater than in F1. These results indicated that enhanced PNF might benefit dry-matter accumulation but lead to yield decline. We found that rice yield was linearly correlated with grain filling percentage ($R^2 = 0.47$, $p < 0.01$) as well as grains m^{-2} ($R^2 = 0.55$, $p < 0.01$). Grains m^{-2} was quadratically correlated with stem dry matter at the heading stage, which suggests that over-accumulated dry matter in the stem before grain filling might play a negative role in grain yield. Grain filling percentage and grain m^{-2} were found to be linearly correlated with leaf dry matter at the heading stage, which suggests that leaf growth at panicle initiation and heading stage might be critical characteristics in yield performance.

Keywords: Rice; Grain yield; Plant dry matter; Direct seeding; Transplanting, Panicle Nitrogen fertilizer.
Abbreviations: TPR: Transplanted rice; DSR: Direct-seeded rice; PNF: Panicle nitrogen fertilizer; TNA: Total nitrogen amount; MT: Mid-tillering stage; HS: Heading stage; PI: Panicle initiation stage; GFP: Grain filling percentage

1. Introduction

The amount of N absorbed during panicle initiation makes the most effective contribution to spikelet production as well as grain filling. A larger amount absorbed increases specific leaf weight and N content in leaves, which leads to enhancement of photosynthetic capacity and promotion of carbohydrate accommodation in culms and leaf sheaths (Mae, 1997). Therefore, heavy nitrogen fertilization during panicle development, so called panicle nitrogen fertilizer (PNF), has been popular in China to improve population dynamics, make fertilizer use more efficient and enhance grain yield in recent years (Jiang et al., 2004; Lin, 2000). In addition, a switch in planting methods from transplanting to direct seeding has occurred in China (and many other countries) as labor costs have risen and the need to intensify rice production through double and triple cropping has provided economic incentives (Dawe, 2005; De Datta, 1986; Naklang, 1996; Pandey et al., 2002). Meanwhile, most high-yielding rice varieties released in China in the last decade have been characterized as having high nitrogen tolerance with high dry matter production (Lin et al., 2009; Ottis et al., 2008). Moreover, farmers have discovered that applying enhanced fertilizer at mid-tillering (MT) encourages early population development of rice and saves labor by reducing the time needed for topdressing, especially in Southeast China. As a result, the total nitrogen fertilizer amount has increased to a very high level to maintain yield performance, which has led to a huge waste of nitrogen fertilizer followed by a series of environmental problems. Whether PNF could help to maintain yield or minimize yield loss in rice paddies when the total nitrogen is reduced is still unknown. DSR cultivation provides a completely different growth environment for rice, particularly at the seedling stage, compared with that under the transplanting system. The proportion of plant N derived from applied fertilizers is about 40-60% in young seedlings (Mae, 1997). Nitrogen absorbed during the vegetative period mainly promotes the early growth of the plant and increases the number of tillers (Makino et al., 1984). In addition, DSR has a shorter time from seeding to emergence (Pandey et al., 2002), stronger root activity (De Datta and Nantasomsaran, 1991), higher grain-setting percentage and greater biomass production in the early period (Naklang, 1996) compared to TPR. These differences affect nitrogen absorption and physiological utilization. In addition, higher PNF results in reduced MT nitrogen fertilizer topdressing, which might depress the early development of DSR. However, literature about the effects of PNF on the early growth and enhanced reproductive stage of DSR is limited. Until now, most rice varieties have been specifically bred for transplanted conditions. Nitrogen fertilization strategies have also been developed based on the agronomy practice of transplanting and have been widely adopted to improve rice yield with a high total nitrogen level. In fact, grain yields of most rice varieties are achieved by high nitrogen input in China. With increasing use of panicle fertilization, there has been less fertilization applied at basal or topdressing in MT, but responses of rice yield and dry matter accumulation in DSR and TPR have been little studied. Thus, it is of urgent importance to understand the relationship between planting pattern and nitrogen management under the Chinese system. The objective of this study was to determine the effects of nitrogen fertilization strategies on DSR yield in terms of dry matter accumulation and redistribution and to test the PNF effect on DSR when the total nitrogen level was reduced. This would help high-yielding rice varieties to establish the proper N fertilization strategy for DSR with the aim of improving nitrogen use efficiency and maintaining high yield level.

2. Results

2.1 Effect of N treatments on grain yield

In general, the mean yield across the fertilization of DSR was slightly decreased compared with that of TPR. The difference among the mean yields of N treatments was significant: F1 > F3, F2, F4 in DS and F1 > F3 > F2, F4 in TPR. However, rice yield was variable across years. The mean yields of F1 across the years were 11%-18% and 11%-19% higher than that of other N treatments in TPR and DSR, respectively. The highest yield of TPR was 11.10 t/ha obtained by F1 management in 2009, and the highest yield of DSR was 10.43 t/ha obtained by F1 in 2009 (Table 2).

Rice yield across years was also influenced by the total nitrogen amount and PNF levels. The interaction between planting patterns (PP) and nitrogen fertilization treatments (NF) was significant (Table 2). Apparently, grain yield depended on the total nitrogen amount, especially in TPR, where rice yield at the rate of 225 kg N ha^{-1} was greater than that of 150 kg N ha^{-1} in both PNF levels except in 2007. However, in DSR, the effect of total nitrogen amount was only found in low PNF. The PNF effect on grain yield varied within different total nitrogen amounts. In comparison with low PNF, rice yield in high PNF was reduced when the amount was 225 kg N ha^{-1} and stayed consistent when the amount was 150 kg N ha^{-1}. However, the PNF effect on yield difference between DSR and TPR was not significant. The year's effect on yield difference between DSR and TPR was significant, so it is difficult to draw conclusions about the PNF effect on yield difference between TPR and DSR across the years. Furthermore, rice yield was significantly decreased due to unusual climatic conditions during 2007 (extremely high temperature during reproductive phase).

Effects of N management on total dry matter accumulation and harvest index

The N management effect on the total dry matter accumulation and harvest index (HI) under DSR and TPR are shown in Table 2. In general, total dry matter was greater in DSR than in TPR, while the opposite was found for HI. Significant differences in total dry matter accumulation and HI were found among N treatments, with F3 greater than the other methods regardless of planting patterns. The PNF effect on total dry matter varied depending on the total nitrogen amount. Total dry matter in high PNF was greater than that in low PNF when the amount was 225 kg N ha^{-1}. However, little difference was found between low and high PNF when the amount was 150 kg N ha^{-1}. The PNF effect on HI varied with different N managements, being F1 > F4 > F2, F3. Compared with low PNF, high PNF in HI was increased when the amount was 150 kg N ha^{-1}, but decreased when the amount was 225 kg N ha^{-1}. Because the interactions of PP and NF on total dry matter and HI were not significant, little difference between TPR and DSR in N management effect was found in total dry matter and HI.

Effects of N management on dynamic change of plant dry matter accumulation

Dynamic total dry matter accumulations in different N managements were determined for DSR and TPR methods (Figure 1). Before MT, dry matter accumulation was generally greater in DSR than in TPR, but the opposite was found from MT to HS in all years except for F1 in 2007 and F2 in 2009. On average, dry matter accumulated in DSR was 43% greater than in TPR before MT, and 23% less from MT to HS. Little difference in dry matter accumulation was found between total nitrogen amount of 150 and 225 kg N ha^{-1} in both DSR and TPR regardless of growth periods. The PNF effect on dry matter accumulation varied with planting patterns and growth periods. TPR produced more dry matter during PI to HS with high PNF (F3 and F4) than with low PNF (F1 and F2) in all years and planting patterns except for 2006 in TPR. DSR produced more dry matter in the vegetative phase, while TPR produced more in the reproductive phase. Self-regulation of plant group might partly explain the different dry matter accumulation rates.

Effects of N management on net photosynthetic and SPAD value

Flag leaf net photosynthetic rate and leaf SPAD value were determined to evaluate the capacity of leaf CO_2 assimilation and nitrogen status (Figure 2). In general, flag leaf net photosynthetic rate was greater in TPR than in DSR when high PNF was applied in all the years except 2007 (Figure 2a). However, the difference between TPR and DSR was not significant when low PNF was applied. Furthermore, flag leaf net photosynthetic rate in high PNF was greater than in low PNF when the same TNI was applied in both DSR and TPR in all years except for 2006 and 2008 in DSR and 2007 in TPR. The effect of N management on leaf SPAD value varied by year (Figure 2b). The difference in SPAD value between DS and TP was significant when high PNF was applied, with TPR significantly greater than DSR; however, little difference was found when low PNF was applied, except in 2009. Meanwhile, the effect of TNI on the difference in SPAD value between DSR and TPR was not significant.

Relationships of rice yield and dry matter accumulation

Relations between yield components and grain yield were analyzed to determine the component factors that might influence the final grain yield. We found that grain filling percentage (GFP) and grains m^{-2} were linearly related with grain yield in both TPR and DSR. The relationship between yield and GFP is shown in Figure 3b; no substantial difference between DSR and TPR was found. However, there was a clear difference between DSR and TPR in the relationship between grains m^{-2} and grain yield (Figure 3a). Grains m^{-2} for a given grain yield was 10% larger in DSR than in TPR on average. Further analysis was carried out to evaluate the relationships between dry matter accumulation characteristics and grains m^{-2}/GFP. For grains m^{-2}, characteristics of total plant dry matter at MT, leaf dry matter at PI and HS was linearly related with grains m^{-2} (Figures 4a, b, c). Moreover, a quadratic curve relationship was found between stem dry matter at HS and grains m^{-2} (Figure 4d). There was a clear difference between DSR and TPR in the relationship between GFP and leaf dry matter, with GFP about 14% larger in DSR than in TPR.

Table 1. Fertilizer application treatment (kg N ha^{-1}) for DSR and TPR.

Fertilizer treatment			[a]Rate	Total N	[b]Basal dressing	[c]Topdressing	
	TNA	PNF				MT	PI
F1	225	10% of TNA	3:6:1	225	67.5	135	22.5
F2	150	10% of TNA	3:6:1	150	45	90	15
F3	225	30% of TNA	2:5:3	225	45	112.5	67.5
F4	150	30% of TNA	2:5:3	150	30	75	45

a: The proportion of N fertilizer application rate at basal, MT and PI. b: Compound Fertilizer (N:P:K=15%:15%:15%), c: Urea. Note: DSR: direct seeding rice; TPR: transplanting rice; TNI: total nitrogen amount; PNF: panicle nitrogen fertilzer;

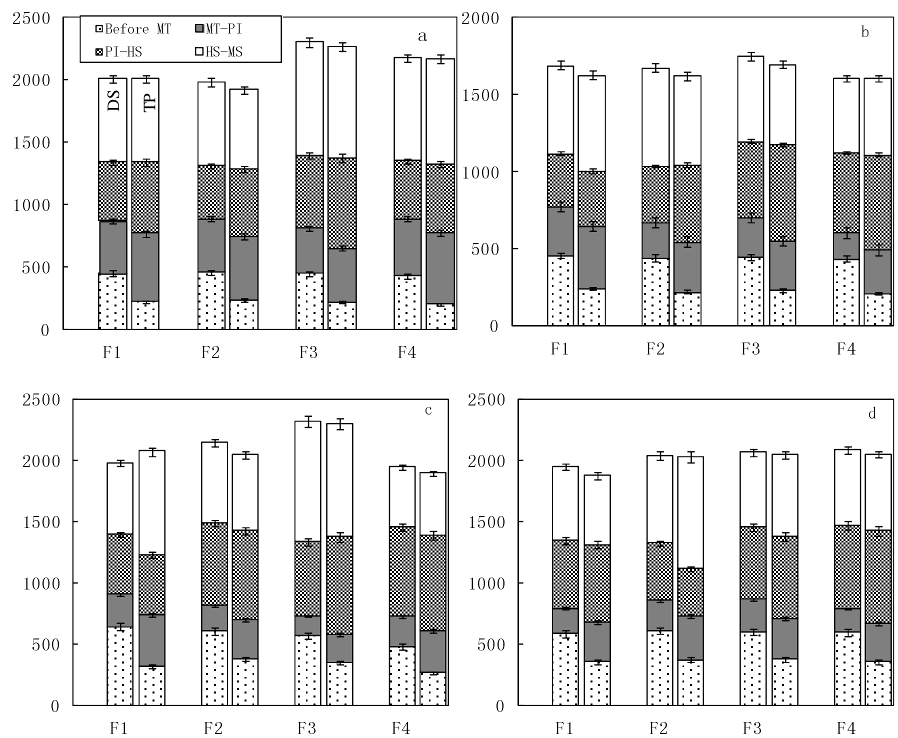

Fig 1. Plant total dry matter accumulation before MT, from MT to PI, from PI to HS and from HS to MS of rice under TP and DS with different nitrogen fertilization in Jiaxing China from 2006 to 2009 (a - d).

Discussion

PNF effect on grain yield and total dry matter with different planting patterns

To improve nitrogen use efficiency (NUE) in order to increase farmers' profitability and reduce negative environmental externalities, studies have previously been performed on aspects of rice yield and N utilization under different N application rates and patterns. However, the conclusions have been inconsistent (Jing et al., 2007; Li and Zhang, 1981; Shi et al., 2008; Tan et al., 1981). Recently, panicle N fertilization was recommended for use in rice production in China to increase grain yield and reduce nitrogen loss in the field (Ling, 2000). Belder et al. (2004), Jiang et al. (2004), Jing et al. (2007), and Shi et al. (2008) all used differential responses to N management of rice cultivars (inbred rice, two-line hybrid, and three-line hybrid, etc.), assuming that a higher panicle nitrogen fraction would be a sound strategy for improving NUE and yield. However, our results showed that enhanced panicle nitrogen fertilizer had a negative effect on the yield production of rice variety Xiushui 09 in both TPR and DSR, especially when the total nitrogen amount was high (225kg N ha^{-1}). Furthermore, the grain yield depended on the nitrogen amount: rice yield was greater at the rate of 225kg N ha^{-1} than at the rate of 150kg N ha^{-1}. Cultivar's differential responses to nitrogen management in rice yield, dry matter and nitrogen accumulation and redistribution have been reported by previous researchers (Bufogle et al., 1997; Jiang et al., 1995; Souza et al., 1998). Jiang et al. (2004) also pointed out that rice varieties with long growth duration could increase grain yield by appropriately increasing panicle nitrogen rate. The dominant cultivar in South China, Xiushui 09, has a long growth duration of 150-155 days. Improving the panicle nitrogen rate increased the plant dry matter significantly; however, the yield was dramatically decreased. These results imply that growth duration might not be a good indicator for choosing the appropriate nitrogen strategy. Most previous studies of panicle nitrogen fertilizer were conducted based on the proper total nitrogen input for traditionally transplanted rice but ignored the fact that high rice yield was obtained by overuse of early nitrogen topdressing and increased total nitrogen input (Cheng, 2012). It is still not clear whether nitrogen management options developed for traditionally transplanted rice are unsuitable for direct-seeded rice because direct-seeded rice is subject to more diverse and heterogeneous soil environments than transplanted rice (De Datta and Nantasomsaran, 1991). The current study showed that the mean yield across fertilization methods in DSR was slightly decreased compared with that in TPR (Table 2), and little difference in the effect of nitrogen management between

Table 2. Grain yield, above dry matter and HI of rice under TP and DS with different N treatments.

PP	DS				TP			
NF	F1	F2	F3	F4	F1	F2	F3	F4
Yield (t ha^{-1})								
2006	[a]10.05a	9.64b	9.85ab	9.23b	10.77a	9.67b	10.00a	9.34b
2007	7.91a	6.03b	6.23b	6.49b	7.98a	6.08c	6.33b	6.54b
2008	10.21a	8.49b	9.85ab	8.77ab	10.98a	8.85c	9.90b	9.10c
2009	10.43a	9.38b	9.90a	9.45ab	11.10a	9.54c	10.07b	9.50c
Average	9.65a	8.38c	8.95bc	8.48c	10.21a	8.53c	9.07b	8.62c
		8.86b				9.11a		
[b]*ANOVA*	*NF*	*147.8***	*Y*	*839.9***	*NF*Y*	*5.02**	*PP*Y*NF*	*0.55ns*
	PP	*32.99***	*PP*NF*	*5.44**	*PP*Y*	*4.12**		
Total dry matter (t ha^{-1})								
2006	20.8b	19.7b	22.9a	21.6ab	20.0b	19.1b	22.6a	21.6a
2007	16.8b	16.7b	17.4a	16.1c	16.2b	16.2b	16.9a	16.0c
2008	20.7b	21.4b	24.1a	17.4c	20.7b	20.4b	22.9a	17.2c
2009	19.4b	19.3b	20.8a	19.8b	18.7b	19.2b	20.4a	19.4b
Average	19.4b	19.3bc	21.3a	18.7c	18.9b	18.7bc	20.7a	18.5c
		19.7a				19.2b		
ANOVA	*NF*	*100.22***	*Y*	*320.63***	*NF*Y*	*2.20ns*	*PP*Y*NF*	*1.56ns*
	PP	*15.2**	*PP*NF*	*0.35ns*	*PP*Y*	*0.39ns*		
HI								
2006	0.50a	0.49a	0.42b	0.43b	0.54a	0.50a	0.44b	0.44b
2007	0.47a	0.36b	0.36b	0.40a	0.49a	0.37b	0.37b	0.41a
2008	0.52a	0.40b	0.41b	0.50a	0.53a	0.43b	0.43b	0.54a
2009	0.55a	0.46b	0.48b	0.48b	0.59a	0.47b	0.49b	0.50b
Average	0.51a	0.43c	0.42c	0.45b	0.54a	0.44c	0.43c	0.47b
		0.45b				0.47a		
ANOVA	*NF*	*306.4***	*Y*	*245.25***	*NF*Y*	*4.60**	*PP*Y*NF*	*1.08ns*
	PP	*63.78***	*PP*NF*	*1.15ns*	*PP*Y*	*1.69ns*		

Notes: [a] Means of N treatments followed by the same letter for each planting pattern are not significantly different at the 5% level by Tukey test, n=4; [b] The effect planting pattern (PP), nitrogen fertilizer (NF) , Year (Y) effects as well as their interaction effects was tested by Tukey test; ns=not significant; *=P<0.05; **=P<0.01.

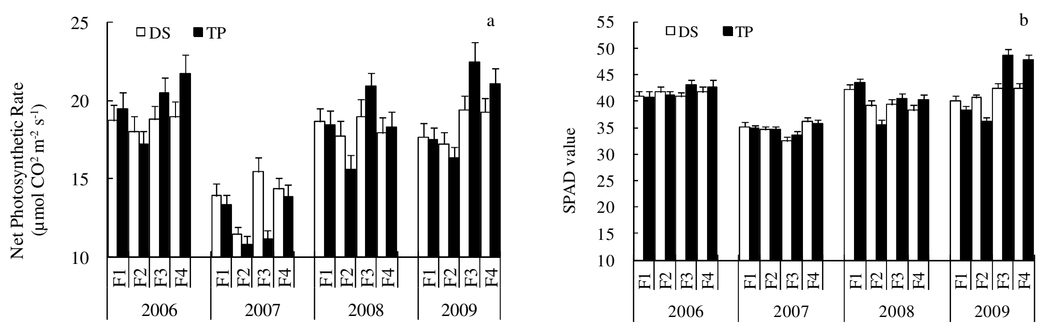

Fig 2. Net photosynthetic rate (a) and SPAD value (b) of rice at heading stage at Jiaxing from 2006 to 2009. Error bar represent SE, n=4.

DSR and TPR was found. We also found that DSR had greater dry matter accumulation in the early stage, consistent with previous reports (Dawe 2005; De Datta 1986; Naklang 1996; Pandey et al. 2002). In general, the nitrogen applied at basal and mid-tillering was 22% less in high PNF than in low PNF (Table 1). Our results showed that there was little difference in dry matter accumulation between low and high PNF at the mid-tillering stage in both TPR and DSR regardless of nitrogen amount, but significant differences were found at PI between low and high PNF in TPR (except in 2009), with low PNF greater than high PNF and differences ranging from 8.5-16.8% (Figure 1). These results indicated that appropriate reduction in nitrogen topdressing in the early stage might not affect the plant dry matter accumulation in both TPR and DSR. A slightly improved effect of high PNF on grain yield was also observed at 225kg N ha^{-1} compared with that at 150kg N ha^{-1} in TPR. However, there was little difference between the rates of 150 and 225 kg N ha^{-1} under high PNF in DSR. The extra nitrogen input had little effect on the yield improvement but significantly improved the total dry matter. These results imply that increased PNF might lead to an excessive plant growth at the panicle growth stage, which ultimately affects the yield performance

Relationships between yield components and dry matter accumulation characteristics

The yield improvement of recently released varieties comes from two sources: improved dry matter accumulation and greater HI (Dingkuhn et al., 1991; Horie et al., 2001; Peng et al., 1996; Yoshida, 1981). Generally, DSR produced more dry

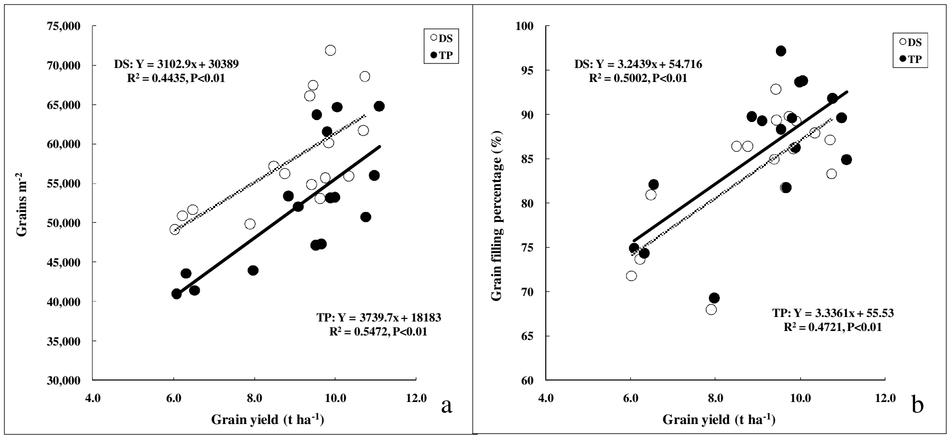

Fig 3. Relationship between grains m^{-2} (a), grain filling percentage (b) and yield of DS and TP rice grown from 2006 to 2009 at Jiaxing.

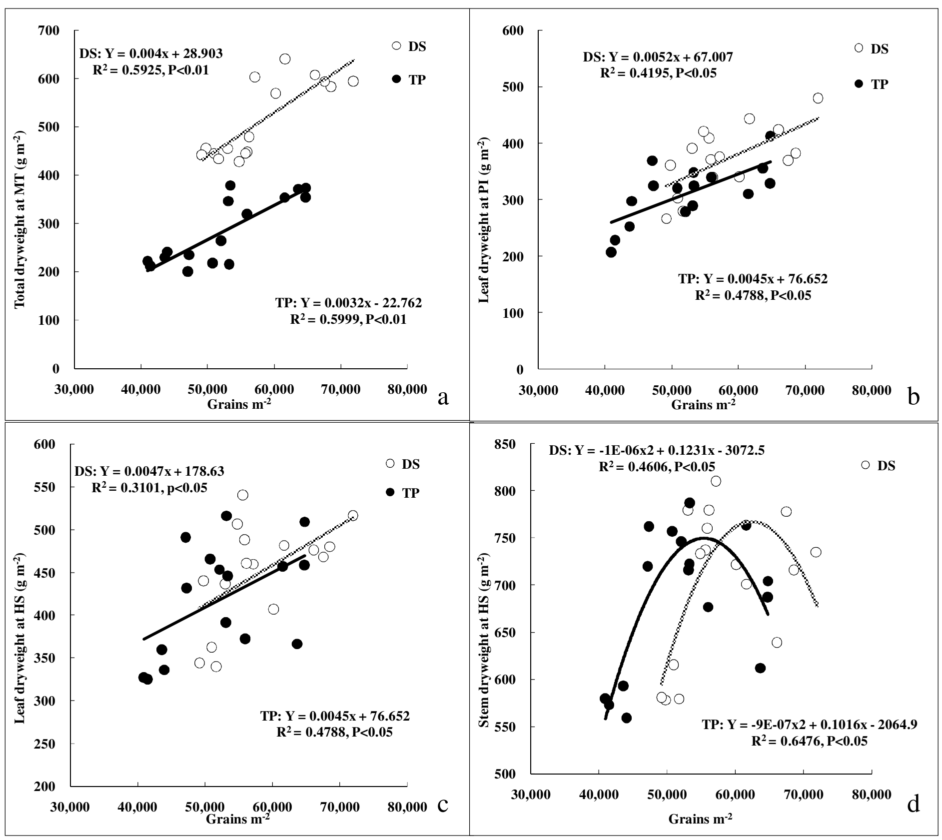

Fig 4. Relationships between total dry weight at MT (a), leaf dry weight at PI (b), leaf dry weight at HS (c), stem dry weight at HS (d) and grains m^{-2} of DS and TP rice grown from 2006 and 2009 at Jiaxing.

matter accumulation but lower HI compared to TPR (De Datta, 1986; De Datta and Nantasomsaran, 1991; Naklang, 1996; Pandey et al., 2002), which was consistent with our results. Analyzing the relationships between grain yield and yield components, we found that the yield was linearly correlated with grain filling percentage and grains m^{-2} (Figure 3). In sink-source theory, the grain filling percentage is determined by the sink size (grains m^{-2}), source capacity (leaf photosynthetic rate, photosynthesis duration, solar radiation, air temperature and so on.) and carbohydrate flow between sink and source (dry matter transportation from straw to spikelets during the grain filling period; Cassman et al., 1998; Makino et al., 1984; Peng et al., 1996; Ying et al., 1998). In the current study, we found that high PNF significantly improved the flag leaf photosynthetic rate compared with low PNF in the heading stage regardless of planting pattern and year (Figure 2), which is consistent with reports of the physiological effect of nitrogen application on the leaf photosynthesis at panicle initiation (Mae, 1997). However, the result that grain m^{-2} was quadratically correlated with

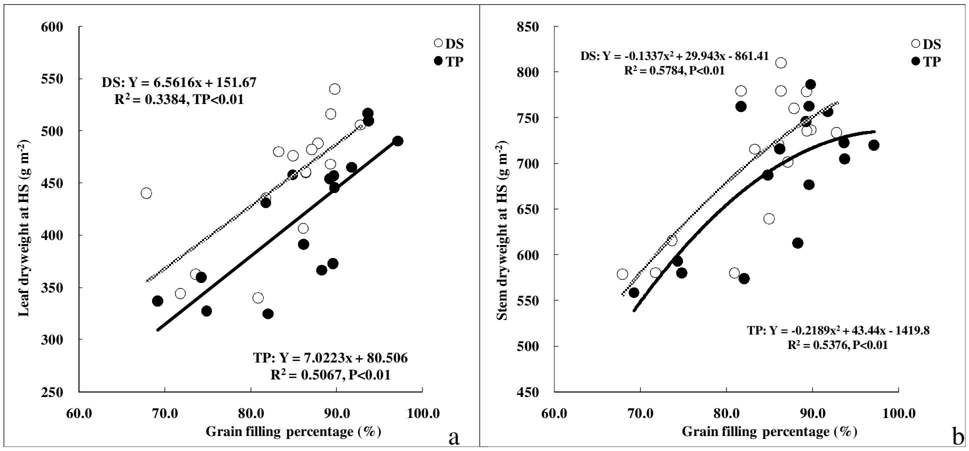

Fig 5. Relationships between leaf dry weight at HS (a), stem dry weight at HS (b) and grains filling percentage of DS and TP rice grown from 2006 and 2009 at Jiaxing.

stem and sheath dry matter accumulation at the heading stage (Figure 4d) provides evidence that over-accumulation of dry matter in the stem and sheath might have a negative effect on sink size production and result in yield decline. Dry matter accumulated in straw before heading is thought to be carbohydrate storage for the dry matter redistribution during the grain filling period (Ying et al., 1998). However, excess nitrogen applied to the crop could result in problems such as over-stimulation and excessive growth which attracts pests, delayed maturity, reduction in the quality of the crop, or lodging of rice plants (Yoon et al. 2001). In addition, the concentration of nitrogen required varies depending on the crop as well as its growth stage; high nitrogen levels may be beneficial during early growth stages but may cause yield losses during the later flowering and fruiting stages (Ayres and Westcot, 1994). These reasons might partly explain the lower yield in PNF.

Material and Method

Site description

Field experiments were conducted at the experimental farm of the Jiaxing Agriculture Research Institute, China (30°17′N, 120°45′E, 6 m altitude) from 2006 to 2009. Prior to planting, the soil at the experimental site was alluvial sandy clay loam, with pH 6.8, organic C 30.3 g/kg, total N 2.4 g/kg, available P 32.5 mg/kg and available K 58.6 mg/kg. Soil properties were determined according to standards set by the Laboratory Manual for Agriculture and Soil Analysis (Pao, 2005).

Experimental treatment

The rice (*Oryza sativa* L.) variety used was Xiuyou 5 (Xiushui 110a × Xiuhui 69), a japonica hybrid released by Jiaxing Crop Research Institute. Two planting patterns were used: direct sowing (DSR) and transplanting (TPR). For DSR, seeds were manually broadcast on the plowed plots at a seeding rate of 15 kg ha^{-1} in early June. For TPR, seedlings with three expanded leaves (about 25-30 d after sowing) were transplanted into the field at a density of 25 hills m^{-2} (20 cm × 20 cm) with two plants per hill. Sowing of TPR was 10-15 days earlier than DSR to meet the date for the heading period. Four treatments of nitrogen fertilizer application were evaluated as follows:

F1: TNA of 225 kg N ha^{-1} combined low PNF;
F2: TNA of 150 kg N ha^{-1} combined low PNF;
F3: TNA of 225 kg N ha^{-1} combined high PNF;
F4: TNA of 150 kg N ha^{-1} combined high PNF (Table 1).

Basal applications were made a week before transplantation/sowing. Total Potassium of 135 kg ha^{-1} was applied to all plots, with 50% being basal dressing in the form of compound fertilizer and 50% as topdressing at PI in the form of Potassium Chloride. Phosphorous was not supplied because of its sufficient level in the soil. A split plot design was used with the planting pattern as main plot and fertilizer application as sub-plot. The size of each sub-plot was 5.5 × 4.5 m, with three replications for each treatment. Weeds, insects and diseases were controlled as required to avoid yield loss.

Sampling and measurement

In this paper, the full-heading period was defined as the time when 80% of the panicles had emerged. After measurement of the SPAD value of individual leaves (SPAD-502, Minolta, Tokyo, Japan), net photosynthetic rates were measured using a LI-6400 portable photosynthesis system (LI-COR, Lincoln, NE, USA) on the uppermost fully expanded leaves. The crop was considered to have reached maturity when 95% of the spikelets had turned yellow. Twelve representative hills (hills with mean value of tillers per hill from each plot were collected at each sampling for measurement of dry weight of constituent organs (the hills were separated into leaf blades, culms plus leaf sheaths and panicles). All samples were oven-dried at 80-105°C for several days, weighed and powdered. At the time of harvest, twelve representative hills with the mean value of panicles per hill from each plot were collected and hand-threshed for measurement of the number of filled and unfilled spikelets. The filled spikelets were separated by submerging the hand-threshed spikelets in a NaCl solution with a specific gravity (g m^{-3}) of 1.06. The filled spikelets were then hulled and oven-dried at 105°C to a constant weight in order to determine grain dry weight. A survey of yield was carried out as follows: hills were collected from the center part of each plot of 5 m^2. Unhulled (rough) rice was obtained after reaping, threshing and wind selection. The weight of rough rice was adjusted to a moisture content of 14%.

Statistical analysis

All data were subjected to ANOVA using statistical software

SAS 8.0 for Windows. Comparisons were made between the treatments using Tukey's test; p values < 0.05 and 0.01 were considered significantly different.

Acknowledgments

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