Yield Component Differences between Direct-Seeded and Transplanted Super Hybrid Rice

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Abstract: Super hybrid rice Liangyoupeijiu was grown by transplanting (TP) at a spacing of 20 cm × 20 cm with one seedling per hill and by direct-seeding (DS) at a seeding rate of 22.5 kg ha⁻¹ (about 120 seeds m⁻²) in Changsha, Hunan Province, China in 2004–2010. Grain yield and yield components were measured each year, and some physiological factors were determined in 2009. There was no significant difference in mean grain yield across years between DS and TP. DS produced more panicles per m² but less spikelets per panicle than TP. The differences in number of spikelets per m², spikelet filling percentage and grain weight between DS and TP were not significant. A large number of panicles per m² in DS was derived from the increased number of tillers per m² rather than increased rate of panicle-bearing tillers, and the number of tillers per m² was mainly determined by the number of hills per m² because the number of tillers per hill was small in DS plants. Tiller rate, tillering duration as well as carbohydrate and nitrogen metabolism were critical to the reduced number of tillers per hill in DS. In DS plants with fewer spikelets per panicle, the numbers of primary branches and secondary branches per panicle, spikelets per primary and secondary branches, and also panicle length and spikelets per unit panicle length were smaller. These differences were attributed to the smaller source leaf size as well as lower root activity and the nutritional status at panicle initiation.

Key words: Direct seeding, Panicle number, Panicle size, Physiological factor, Super hybrid rice.

During the four decades from the 1960s to the 1990s, the yield of rice, the staple food crop in China, has rapidly increased by reducing plant height using the semi-dwarf gene and utilizing heterosis of hybrids (Zhang, 2007). Over the last 10 years, however, rice yield has remained unchanged or has even declined in most rice production provinces of China and the average annual growth rate was reduced by 0.3% from 1998 to 2006 (Fan et al., 2009). A rice yield increase of more than 1.2% per year will be required to meet the growing demand for food that will result from population growth and economic development in the next decade (Normile, 2008).

China established a nationwide mega-project to develop super rice in 1996 (Yuan, 1996). As of 2007, 61 varieties with great yield potential have been approved as super rice by the Ministry of Agriculture of China (Huang and Zou, 2009). However, rice yield depends not only on the genetic characteristics but also on the agronomic practices (Zou et al., 2003). Transplanting has been a major traditional method of rice establishment (Chen et al., 2007). However, this method requires a large amount of labor (Bhushan et al., 2007). In China, labor availability is limited because the younger generation is leaving the rural districts for jobs in the cities resulting in the aging of the farmers (Derpsch and Friedrich, 2009). The simple and labor-saving method of direct seeding is attractive for efficient agriculture in China (Wu et al., 2005).

Rice yield is determined by sink size (number of spikelets per unit land area), spikelet filling percentage and grain weight. Sink size is considered as the primary determinant of the rice yield (Kropff et al., 1994). It can be increased either by increasing panicle number per unit land area or spikelet number per panicle or both (Ying et al., 1998).

Tillering is an important agronomic trait in rice because it affects the number of panicles per unit land area (Moldenhauer and Gibbons, 2003). Many studies have been carried out to identify the genes involved in the control of rice tillering (Li et al., 2005; Miyamoto et al., 2004; Zou et al., 2005). However, the tillering characteristics are altered by the environment and by agronomic practices. Yoshida (1973) suggested that tillering of rice plants should be considered in relation to light intensity, temperature and carbohydrate metabolism. Zhong et al. (2003) reported that tillering rate increased linearly as leaf nitrogen concentration increased in rice plants under different nitrogen treatments and suggested that the leaf...
nitrogen concentration should be considered in predicting the number of tillers in the rice crop.

Panicle traits, as important determinants of sink capacity, have been studied by many genetic researchers (Cai et al., 2002; Hong and Leng, 2004; Xu et al., 2004; Mei et al., 2006; Xing et al., 2008). Many of these studies focused on panicle size (number of spikelets per panicle), which shows a large range of variation and is the major objective of improvement in rice breeding (Tian et al., 2006). The number of spikelets per panicle can be divided into four sub-components: the number of primary branches per panicle, secondary branches per panicle, number of spikelets per primary branch, and number of spikelets per secondary branch (Cai et al., 2002). Kato (1997) stated that primary branches per panicle was positively and strongly correlated with the number of spikelets per panicle, and suggested that indirect estimation of the number of spikelets per panicle via the number of primary branches per panicle was slightly more effective than direct counting of the spikelets per panicle, whereas Cui et al. (2002) and Mei et al. (2006) reported that the number of spikelets per panicle was more closely correlated with the number of secondary branches per panicle. In another approach, the number of spikelets per panicle was the function of panicle length and number of spikelets per unit panicle length (Wang et al., 2007). It is generally considered that the number of spikelets per panicle is more closely correlated with the number of spikelets per unit panicle length (Cui et al., 2002). In the last two decades, newly released rice varieties in southeastern China have been mainly characterized by a large number of spikelets per unit panicle length, i.e. a compact panicle (Wang et al., 2008).

There are some reports describing the difference in yield components between direct-seeded (DS) and transplanted (TP) rice (Yoshida, 1981; Naklang et al., 1996; Cho et al., 2001). DS rice usually produced a larger number of panicles per m\(^2\) but smaller number of spikelets per panicle than TP rice. However, limited information is available on the critical physiological factors that cause the difference in yield components between DS and TP rice, especially in the super hybrid variety. This study aimed to (1) compare grain yield and yield components of DS rice with those of TP rice and (2) identify the physiological factors that cause the difference in the yield component between DS and TP super hybrid rice.

**Materials and Methods**

Field experiments were conducted in Changsha (28°11’ N, 113°04’ E, 32 m asl), Hunan Province, China in 2004–2010. The location is situated in the East-Asian monsoon climatic zone and has a moist subtropical monsoon climate with a mean annual temperature of about 17.0°C, mean annual rainfall of about 1355 mm and mean annual sunshine hours of about 1677 hr. The soil of the experimental field was clay loam with pH=6.04, organic matter=14.96 g kg\(^{-1}\), total N=1.40 g kg\(^{-1}\), total P=1.18 g kg\(^{-1}\), total K=18.13 g kg\(^{-1}\), NaOH hydrolysable N=137.0 mg kg\(^{-1}\), Olsen P=38.35 mg kg\(^{-1}\), NH\(_4\)OAc extractable K=113.3 mg kg\(^{-1}\).

Liangyoupeijiu, the first super hybrid rice variety in China, was used in the experiment. This variety is an indica-japonica hybrid (Peiai64S ×9311) released by Jiangsu Academy of Agricultural Sciences of China in 1999. In the past few years, Liangyoupeijiu has been widely commercialized, being cultivated in about 2.5 million hectares from 12°N to 35°N in southern China and southeastern Asia, e.g., Vietnam and Philippines (Li and Zou, 2003).

In each year, Liangyoupeijiu was grown by transplanting and direct seeding in the single rice-growing season (from May to October). Plots were laid out in a randomized complete block design with four replicates using a plot size of 30 m\(^2\). TP seedlings were raised in nursery beds, and 25-day-old seedlings were manually transplanted at a spacing of 20 cm×20 cm with one seedling per hill between May 31st and June 24th. For direct seeding, pre-germinated seeds were manually broadcasted onto the soil surface at a seed rate of 22.5 kg ha\(^{-1}\) (about 120 seeds m\(^2\)) between May 11 and June 1. Fertilizers used were urea for N, single superphosphate for P and potassium chloride for K at doses of 150 kg N ha\(^{-1}\), 90 kg P\(_2\)O\(_5\) ha\(^{-1}\) and 180 kg K\(_2\)O ha\(^{-1}\). N was split-applied: 90 kg ha\(^{-1}\) at basal, 45 kg ha\(^{-1}\) at mid-tillering, and 15 kg ha\(^{-1}\) at panicle initiation. P was applied at basal. K was split equally as basal and top dressing at the panicle initiation stage. The regimen for water management was in the sequence of flooding, midseason drainage, reflooding and moist intermittent irrigation. Weeds, insects and diseases were controlled as required to avoid yield loss. However, the yield was decreased by lodging caused by a typhoon in the growing season of 2005. Hence, the data of 2005 were excluded from the analysis.

In 2004–2010, plants were sampled from a 0.48-m\(^2\) area in each plot at maturity. Plant samples were first hand-threshed and then filled spikelets were separated from unfilled spikelets by submerging in tap water. Three subsamples of 30 g filled spikelets and all unfilled spikelets were counted to calculate the number of spikelets per panicle, spikelet filling percentage and grain weight. The number of panicles in a 0.96-m\(^2\) area at maturity to determine the number of panicles per m\(^2\). Grain yield was determined from the number of harvested plants in a 5-m\(^2\) area in each plot and adjusted to the standard moisture content of 14 g H\(_2\)O g\(^{-1}\).

In 2009, plants in a 0.48-m\(^2\) area in each plot were marked to count the tillers (including main stem) starting at 35 days after sowing at a 3-day interval until the number
diminished. Tillering duration was defined as the number of days from sowing to final tillering. Tillering rate was calculated as: tillering rate = number of final tillers per hill/tillering duration. The number of spikelets per panicle, the number of spikelets per secondary branch, panicle length and the number of spikelets per cm panicle were measured in the uppermost fully expanded leaves. At panicle initiation, plants were sampled from a 0.48 m² area in each plot and were separated into leaves and stems. Area of each leaf was determined by measuring leaf length and maximum leaf width and calculated as: leaf area = leaf length x maximum leaf width x 0.75 (Umashankar et al., 2005), and then the leaves and stems were oven-dried at 70°C to determine the nitrogen, phosphorus and potassium contents. Net photosynthetic rate and chlorophyll, soluble sugar and soluble protein contents were measured on the uppermost fully expanded leaves. Bleeding sap was collected from 6 rice plants with similar tiller numbers (about 15 and 7 tillers for transplanting and direct seeding, respectively) for each plot. Main stems of these rice plants were cut off at about 20 cm above the soil surface, and were then covered with cotton balls, which were enclosed in plastic bags for 12 hours (1830-0630). Bleeding sap rate was expressed as the gain in weight of the cotton ball per hour. Soluble sugar and amino acid concentrations were determined in the bleeding sap. At maturity, panicles from a 0.48 m² area were collected to determine the numbers of primary branches and secondary branches per panicle, and the number of spikelets per primary branch, the number of spikelets per secondary branch, panicle length and the number of spikelets per cm panicle.

The net photosynthetic rate was determined with a portable photosynthesis system (LI-6400, LI-Cor, Lincoln, NE, USA) at 0900-1030. It was measured at a light intensity of 1200 μmol m⁻² s⁻¹, a leaf temperature of 30°C, a constant CO₂ concentration of 380±5 μmol mol⁻¹, and a relative humidity of 75±5% in the sample chamber. Nitrate reductase activity was measured by the in vivo method described by Harper and Hageman (1972) and expressed as μg NO₂ formed per g fresh weight (FW) per hour. Glutamine synthetase activity was assayed by the method of Chien et al. (2000) and one unit (U) of the activity was defined as the increase in absorbance at 540 nm per g FW per hour. Chlorophyll content was measured by extracting with a mixture of ethanol: acetone: distilled water = 4.5:4.5:1 (v/v/v) for 24 hr (Yang et al., 2007) according to the procedure of Arnon (1949). Soluble sugar content was determined by the anthrone color reaction (Yemm and Willis, 1954) using sucrose as the standard. Soluble protein content was determined by the

Table 1. Grain yield and yield components of super hybrid rice Liangyoupeijiu grown under transplanting (TP) and direct seeding (DS) in Changsha, Hunan Province, China in 2004–2010.

| Cultivation method | 2004  | 2006  | 2007  | 2008  | 2009  | 2010  | Mean† | LSD (0.05)‡ |
|--------------------|-------|-------|-------|-------|-------|-------|-------|------------|
| Grain yield (t ha⁻¹) |       |       |       |       |       |       |       |            |
| TP                 | 8.70  | 10.13 | 10.68 | 10.26 | 9.68  | 8.88  | 9.62 a | 0.63       |
| DS                 | 9.35  | 11.12 | 10.80 | 10.28 | 9.50  | 8.82  | 9.98 a | 1.24       |
| Panicles per m²    |       |       |       |       |       |       |       |            |
| TP                 | 214   | 195   | 215   | 201   | 199   | 209   | 205 b  | 20         |
| DS                 | 261   | 262   | 233   | 378   | 267   | 287   | 281 a  | 57         |
| Number of spikelets per panicle |       |       |       |       |       |       |       |            |
| TP                 | 171   | 259   | 261   | 223   | 222   | 228   | 227 a  | 27         |
| DS                 | 162   | 207   | 240   | 158   | 185   | 164   | 186 b  | 27         |
| Number of spikelets per m² |       |       |       |       |       |       |       |            |
| TP                 | 36.6  | 50.4  | 56.1  | 44.7  | 44.2  | 47.6  | 46.6 a | 7.4        |
| DS                 | 42.2  | 54.0  | 55.8  | 59.8  | 49.3  | 46.3  | 51.2 a | 10.2       |
| Spikelet filling (%) |       |       |       |       |       |       |       |            |
| TP                 | 77.6  | 69.4  | 72.7  | 81.4  | 71.9  | 82.1  | 75.8 a | 3.9        |
| DS                 | 77.5  | 73.1  | 76.6  | 77.0  | 72.9  | 80.7  | 76.3 a | 3.2        |
| Grain weight (mg)  |       |       |       |       |       |       |       |            |
| TP                 | 25.5  | 25.7  | 26.1  | 26.8  | 23.9  | 23.1  | 25.2 a | 0.3        |
| DS                 | 24.9  | 26.4  | 26.5  | 27.1  | 23.3  | 23.2  | 25.2 a | 0.4        |

† Means of cultivation methods for each parameter with the same letters are not significantly different according to LSD at P=0.05.
‡ LSD values are for the comparison of years for each parameter under each cultivation method.
protein-dye binding method introduced by Bradford (1976) using bovine serum albumin as the standard. Amino acid concentration in the bleeding sap was determined by the ninhydrin method (Moore and Stein, 1954) using L-leucine as the standard. Nitrogen content was determined in an autoanalyzer (Integral Futura, Alliance Instruments, Frépillon, France). Phosphorus content was determined by the ascorbic acid-molybdate method (Murphy and Riley, 1962). Potassium content was determined with a flame photometer (FP640, Shanghai Precision & Scientific Instrument Inc., Shanghai, China). Statistical analyses were performed using analysis of variance (Statistix 8, Analytical software, Tallahassee, Florida, USA). Means of values were subjected to the least significant difference test (LSD) at the 0.05 probability level.

**Results and Discussion**

There was no significant difference in mean grain yield across years between DS and TP plants (Table 1). DS plants had an average of 281 panicles per m², which was 37% higher than in TP plants. On the contrary, the mean number of spikelets per panicle in DS plants was 22% less than that in TP plants. There were no significant differences in number of spikelets per m², spikelet filling percentage and grain weight between DS and TP plants.

The number of tillers per m² in DS plants was 55% higher than that in TP plants, while the productive tiller percentage was 7% lower in DS than in TP plants (Table 2). This revealed that more panicles per m² in DS plants were derived from the increased number of tillers per m² rather than increased rate of panicle-bearing tiller. The number of tillers per m² was determined by the number of tillers per hill and the number of hills per m². In the present study, the number of tillers per hill in DS plants was less than in TP plants by 42%, whereas the number of hills per m² in DS plants was 2.68 times higher than in TP plants. This indicated that the number of tillers per m² in DS plants was mainly determined by the number of hills per unit land area and suggested that keeping a full standing of seedlings was very important for rice production by direct seeding.

The number of tillers per hill could be regarded as the time integration for tillering rate, and that was to say, the number of tillers per hill could be increased by accelerating the tillering rate or prolonging tillering duration or both. In this study, the tillering rate in DS plants was significantly lower than that in TP plants by 10% (Table 2), while tillering duration in DS plants was 24 d shorter than that in TP plants. This indicated that fewer tillers per hill in DS plants was attributed to both lower tillering rate and the shorter tillering duration. However, growth process without the setback caused by uprooting and transplanting should be partly responsible for the shorter tillering duration of DP rice (Nabheerong, 1993; Kotera et al., 2004).

At mid-tillering, although there was no significant difference in chlorophyll content between DS and TP plants, soluble protein content was 32% lower in DS plants than in TP plants (Table 3). It is well known that a considerable proportion of the soluble protein is Rubisco (EC 4.1.1.39) (Stitt and Schulze, 1994; Sarker et al., 2002), which is the most important enzyme involved in CO₂ fixation and its content is thought to be a rate-limiting factor for the light-saturated photosynthetic rate at atmospheric CO₂ concentration (Makino et al., 1985).

Many studies have demonstrated that the decrease in soluble protein content was always accompanied by a reduced photosynthesis in rice (Chen et al., 2005; Sarker et al., 2002; Weng and Chen, 1987). Also, in the present study, net photosynthetic rate in DS plants was 10% lower than that in TP plants, and soluble sugar content in DS plants was 20% lower in DS than in TP plants. These results indicated that the capacity of photosynthetic carbon fixation in DS plants was not as great as that in TP plants, which was consistent with the smaller number of tillers per hill. This was in agreement with the hypothesis that energy supply regulates tillering of plants (Mitchell, 1953) and that tiller appearance depends on carbon supply (Bos and Neuteboom, 1998; Gautier et al., 1999). On the other
hand, there was no significant difference in nitrate reductase activity between DS and TP plants, whereas glutamine synthetase activity was 9% lower in DS than in TP plants (Table 3). Glutamine synthetase is a central enzyme in the nitrogen metabolism of higher plant (Lightfoot et al., 1988) and plays a role in the primary assimilation of ammonia (Hirel and Gadal, 1980; Tsai et al., 2003), the re-assimilation of ammonia released from the photosynthetic nitrogen cycle, and the catabolism of nitrogenous storage and transport of nitrogenous compounds (Lea and Miflin, 2003; McNally et al., 1983). Moreover, rice plants in paddy fields utilize ammonium as a major nitrogen source (bhiyama et al., 2004). Therefore, nitrogen content of rice plants is greatly related to glutamine synthetase activity. In the present study, consistent with the glutamine synthetase activity, the nitrogen content of the leaf in DS plants was lower than in TP plants by 21%. Previous studies showed that the nitrogen content of the leaf is positively related to photosynthetic capacity in rice because the proteins involved in the Calvin cycle and thylakoids account for the majority of leaf nitrogen (Evans, 1989; Ohsumi et al., 2007). Thus, the lower leaf nitrogen content might be partially responsible for the smaller number of tillers per hill in DS plants, which was caused by the smaller capacity of photosynthetic carbon metabolism. This was in agreement with the report by Zhong et al. (2003) that there was a strong positive linear relationship between the relative tillering rate and nitrogen content of the leaf in rice.

At panicle initiation, chlorophyll content, soluble protein content, net photosynthetic rate and soluble sugar content in leaves were lower in DS plants than in TP plants, although the differences were not significant (Table 3). Li et al. (1998) reported that a large proportion of variation in sink capacity (number of spikelets per panicle) was explained by source leaf size. Sheehy et al. (2001) observed that there was significant relationship between the number of juvenile spikelets and leaf area per stem. Therefore, in the present study, the smaller leaf area per stem in DS plants was a critical factor that explained its smaller capacity of photosynthetic carbon metabolism.

### Table 3. Physiological factors in the uppermost fully expanded leaf at mid-tillering of super hybrid rice Liangyoupeijiu grown under transplanting (TP) and direct-seeding (DS) in Changsha, Hunan Province, China in 2009.

| Physiological factor                           | TP        | DS        |
|-----------------------------------------------|-----------|-----------|
| Total chlorophyll content (mg g⁻¹ FW)         | 2.62 ± 0.02 a | 2.46 ± 0.06 a |
| Soluble protein content (mg g⁻¹ FW)           | 23.8 ± 0.3 a  | 16.1 ± 0.5 b |
| Net photosynthetic rate (μmol CO₂ m⁻² s⁻¹)    | 22.4 ± 0.3 a  | 20.2 ± 0.6 b |
| Soluble sugar content (mg g⁻¹ FW)             | 17.0 ± 0.5 a  | 13.6 ± 0.2 b |
| Nitrate reductase activity (μg NO₂ g⁻¹ FW hr⁻¹) | 17.1 ± 0.6 a  | 16.6 ± 0.7 a |
| Glutamine synthetase activity (U g⁻¹ FW hr⁻¹) | 14.1 ± 0.1 a  | 12.9 ± 0.2 b |
| Leaf nitrogen content (%)                      | 5.93 ± 0.13 a | 4.71 ± 0.29 b |

Data are mean ± SE (n=4), means of cultivation methods for each parameter with the same letters are not significantly different according to LSD at P=0.05.

### Table 4. Panicle traits of super hybrid rice Liangyoupeijiu grown under transplanting (TP) and direct-seeding (DS) in Changsha, Hunan Province, China in 2009.

| Panicle trait                           | TP        | DS        |
|-----------------------------------------|-----------|-----------|
| Primary branches per panicle            | 11.6 ± 0.3 a  | 10.3 ± 0.2 b |
| Secondary branches per panicle          | 50.4 ± 1.2 a  | 43.2 ± 0.8 b |
| Number of spikelets per primary branch  | 4.93 ± 0.11 a | 4.75 ± 0.09 b |
| Number of spikelets per secondary branch| 3.26 ± 0.08 a | 3.13 ± 0.06 b |
| Panicle length (cm)                     | 25.2 ± 0.4 a  | 23.8 ± 0.1 b |
| Number of spikelets per cm panicle      | 8.81 ± 0.34 a | 7.31 ± 0.26 b |

Data are mean ± SE (n=4), means of cultivation methods for each parameter with the same letters are not significantly different according to LSD at P=0.05.
smaller panicle size. On the other hand, Zhang et al. (2009) reported that an improved root growth, as shown by the larger root biomass, higher root length density during the whole growing season and higher root oxidation activity and root zeatin and zeatin riboside contents at early and mid-growth stages, contributed to the large panicle size in super rice varieties. Bleeding sap rate is considered to be a useful index of root activity in rice plant, and a higher bleeding sap rate might be related to the larger root biomass (Kato et al., 2004). In this study, bleeding sap rate per stem in DS plants was 26% lower than that in TP plants (Table 5). Furthermore, sugar and amino acid concentrations in bleeding sap were 17% and 23% lower, respectively, in DS plants than in TP plants. Subasinghe (2007) stated that the chemical composition of bleeding sap could reveal useful information on the storage, mobilization, and movement of nutrients in plants and thus it could be used as an indicator of nutritional status in a number of plant species. In the present study, nitrogen, phosphorus and potassium contents of both leaves and stems were lower in DS plants than in TP plants, and the differences were significant in the nitrogen content in leaves as well as the potassium contents in both stems and leaves (Table 6). A good nutritional status is essential for increasing the number of spikelets per panicle, and this is why panicle fertilizer is widely used in rice production (Murata, 1969). Hence, lower root activity and its effects on nutritional status was another critical factor for the smaller panicle size in DS.

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Table 5. Leaf area per stem, and physiological factors in the uppermost fully expanded leaf and root at panicle initiation of super hybrid rice Liangyoupeijiu grown under transplanting (TP) and direct-seeding (DS) in Changsha, Hunan Province, China in 2009.

| Physiological factor                  | TP            | DS            |
|--------------------------------------|--------------|--------------|
| Leaf                                 |              |              |
| Leaf area per stem (cm$^2$)          | 124.4 ± 5.2 a | 82.9 ± 2.1 b |
| Total chlorophyll content (mg g$^{-1}$ FW) | 2.69 ± 0.13 a | 2.33 ± 0.15 a |
| Soluble protein content (mg g$^{-1}$ FW) | 14.1 ± 0.6 a | 13.7 ± 0.5 a |
| Net photosynthetic rate (μmol CO$_2$ m$^{-2}$ s$^{-1}$) | 17.0 ± 1.0 a | 15.3 ± 0.6 a |
| Soluble sugar content (mg g$^{-1}$ FW) | 19.6 ± 1.6 a | 17.5 ± 0.3 a |
| Root                                 |              |              |
| Bleeding sap flow rate (mg stem$^{-1}$ hr$^{-1}$) | 292 ± 15 a | 215 ± 5 b |
| Soluble sugar concentration in bleeding sap (μg mL$^{-1}$) | 122 ± 2 a | 101 ± 6 b |
| Amino acid concentration in bleeding sap (μg mL$^{-1}$) | 27.0 ± 2.0 a | 20.9 ± 3.3 b |

Data are mean ± SE (n=4), means of cultivation methods for each parameter with the same letters are not significantly different according to LSD at P=0.05.

Table 6. Nitrogen, phosphorus and potassium contents in stems and leaves at panicle initiation of super hybrid rice Liangyoupeijiu grown under transplanting (TP) and direct-seeding (DS) in Changsha, Hunan Province, China in 2009.

| Nutrient content                  | TP            | DS            |
|-----------------------------------|--------------|--------------|
| Stem                              |              |              |
| Nitrogen content (mg g$^{-1}$ DW) | 15.9 ± 0.9 a | 12.5 ± 0.5 a |
| Phosphorus content (mg g$^{-1}$ DW) | 4.41 ± 0.11 a | 4.06 ± 0.05 a |
| Potassium content (mg g$^{-1}$ DW) | 25.4 ± 0.5 a | 18.7 ± 1.0 b |
| Leaf                              |              |              |
| Nitrogen content (mg g$^{-1}$ DW) | 32.9 ± 0.2 a | 30.2 ± 0.6 b |
| Phosphorus content (mg g$^{-1}$ DW) | 3.03 ± 0.07 a | 2.95 ± 0.05 a |
| Potassium content (mg g$^{-1}$ DW) | 15.2 ± 0.5 a | 12.3 ± 0.3 b |

Data are mean ± SE (n=4), means of cultivation methods for each parameter with the same letters are not significantly different according to LSD at P=0.05.
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