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Research Article

Influence of Seedling Age on Grain Yield in Rice with a Low Seeding Rate Grown under Double Cropping Conditions with Machine Transplantation

Zheng H, Li B, Mo W and Tang Q*

Department of Rice Science, college of Agronomy, Hunan Agricultural University, Changsha, P.R. China

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2. Keywords

Seedling age; Seed rate; Yield

1. Abstract

Aging of rice seedlings can detrimentally impact plant growth and grain yields. We examined the effects of increased seedling age on seedling quality, tillering rate, panicle-bearing tiller rate, biomass, and seed yield attributes using four rice cultivars with a Low Seeding Rate (LSR) grown under machine-transplanted conditions. Compared to 20-day-old (15) seedlings of rice with a Conventional Seeding Rate (CSR), LSR seedlings showed a significantly increased grain yield at higher seedling ages (5–10 days). In LSR seedlings, the average number of spikelets per panicle was 18.9% higher in 25-day-old Early Rice (ER) and 15.9% higher in 20-day-old Late Rice (LR) compared to CSR seedlings; however, there was no significant difference in the number of panicles per m2 between the LSR and CSR seedlings. In addition, increased seedling age (5–10 days) in LSR plants resulted in higher seedling quality and higher tillering and panicle-bearing tiller rates. A significant difference in total biomass was observed in delayed age (5-day intervals) LSR seedlings and 20-day-old (15) CSR seedlings ($p<0.05$). Our results suggest that the prolonged aging of the LSR seedlings compensated for the negative influence of delayed transplantation under machine-transplanted conditions.

3. Introduction

To improve the operational efficiency and reduce the labor intensity of large-scale farming, mechanical transplantation has largely replaced hand transplantation in the cultivation of Chinese rice. The mechanical transplantation of young rice seedlings was first reported in Japan [1], with about 200 g of seeds per tray (60 cm x 28 cm) and 2.1 tillers per plant. Subsequently, other ecological and production conditions were developed [2], including mid-growth (3.1–3.5 tillers per plant and 60–100 g of seeds per tray) and late-growth (3.6–4.0 tillers per plant and 35–50 g of seeds per tray) transplantation. In southern China, where the amount of sunlight and temperatures are suitable for year-round rice cultivation, about 20% of the planting area is occupied by early- and late-transplantation rice; the mid-growth transplantation of seedlings is usually the result of a strained growing season. Generally, the growth period of mid-growth seedlings is 20 days in the early season and 15 days in the late season; these growth periods are about 5–10 days shorter than those used with hand transplantation. When multiple cropping is utilized, mechanical transplantation shortens the growing period and can affect rice grain yields. To overcome this, high-yielding, short-duration rice cultivars have been developed and older seedlings have been cultivated.

The aging or delayed transplantation of rice seedlings can detrimentally impact plant growth and grain yields [3] mainly due to low tiller numbers, a shortened vegetative period, decreased dry matter accumulation [4], and a decreased filling ratio [3]. Therefore, how to effectively compensate for the grain yield loss caused by older seedlings has become an important issue for some researchers. [5] found that lowering the seeding rate could improve the grain yield of older rice seedlings. [6] reported that in addition to cultivation density, enhancing the amount of pre-anthesis dry matter and the remobilization efficiency could mitigate the grain yield losses observed in 30- and 35-day-old seedlings. [7] suggested that for mechanically transplanted rice, adopting dry seedling nursery technology could enhance seedling quality and prolong the age at which seedlings are deemed suitable for transplantation.

***Corresponding Author (s):** *Huabin Zheng, Department of College of Agronomy, Hunan Agricultural University, Changsha, P.R. China, E-mail: qytang@hunau.edu.cn*

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These reports support the notion that the negative effects of older seedlings can be overcome using various agronomic methods.

We hypothesized that a Low Seeding Rate (LSR) could increase the quality of older rice seedlings (e.g., improved tillering rate, panicle-bearing tiller rate, and increased grain yield). Accordingly, we conducted field experiments to determine the effects of the delayed transplantation of four rice cultivars (two early and two late rice cultivars) on seedling quality, tillering rate, panicle-bearing tiller rate, biomass, and seed yields under machine-transplanted conditions.

4. Materials and Methods

4.1. Plant Materials and ExperimentalDesign

The field experiments were carried out in Yongan (28°14N, 113°18'E), Liuyang County, Hunan Province, between March and October 2016. The planting site is a typical double cropping rice district in China. The maximum and minimum temperatures between March 25th and November 22nd were 26.4°C and 19.3°C. The early rice cultivars were Zhongzao39 (ZZ39, inbred) and Zhuliangyou189 (ZLY189, hybrid); the late rice cultivars, Hyou518 (HY518, hybrid) and Taiyou390 (TY390, hybrid), are the most commonly planted cultivars locally.

The experiment utilized a randomized block design with three replications and a plot of 25 m2. In the early season, there were two different dry seeding rates: LSR (30g per tray for inbred cultivars and 15g per tray for hybrid cultivars) and conventional seeding rate (CSR; 100g per tray for inbred cultivars and 70g per tray for hybrid cultivars). Rice seeds were uniformly sown on the 25th of March. The LSR seedlings were grown for 20, 25, 30, and 35 days; 20-day-old CSR seedlings were used as a control. In the late season, there were two different dry seed rates: LSR (15g per tray for hybrid cultivars) and CSR (70g per tray for hybrid cultivars). The LSR seedlings were grown for 20, 25, 30, and 35 days; 20-day-old CSR seedlings were used as a control. The rice seedlings were uniformly transplanted on July 26th.

For the LSR seedlings, a new seed sowing system for single seedling machine-transplanted rice production was adopted [8]. For the CSR seedlings, seeds were manually sown in 58 cm x 28 cm seedling trays. The transplantation density was 28.5 tillers per m2 in the early season and 36.4 tillers per m2 in the late season. The level of nitrogen was 150kg N per hm in the early season and 165kg N per hm in the late season, with 70% of the total N applied at basal dressing and 30% applied at panicle initiation. The levels of phosphorus and potassium were 75.0kg P2O5 per hm and 120.0kg K2O per hm in the early season and 82.5kg P2O5 per hm and 132.0kg K2O per hm in the late season. Phosphorus was applied at basal dressing and potassium was split equally between basal dressing and panicle initiation. Insects, diseases, and weeds were intensively controlled using chemical methods to avoid yield losses.

4.2. Sampling and Determination

Three seedling samples (30 seedlings per sample) were randomly selected for each seedling age on the day of transplantation to examine the traits of the seedlings, including the leaf number per plant, plant height, stem thickness, and dry shoot weight. Excluding the three border plants, 20 hills were labeled in each plot to count basic seedlings 3 days after transplanting and to count the maximum number of tillers at 3-day intervals from 30 to 40 days after transplanting. Ten hills were sampled to determine the aboveground biomass at the flowering stage. At the mature stage, 12 hills were sampled to determine yield components, including the spikelets per panicle, filling ratio, and grain weight. At the end of the growth period, a 5m2 area was selected and sampled to determine the grain yield while 20 hills in the same area were used to determine the number of effective panicles per m2. The tillering rate was calculated as the difference in maximum tillers and basic seedlings per m2 and basic seedlings. The panicle-bearing tiller rate was calculated as the number of panicles per m2 and the maximum number of tillers per m2.

4.3. Data Analysis

Based on the parameters mentioned above, the biomass per plant height was calculated using shoot dry matter and plant height, the tillering rate was calculated as the difference between the maximum tillers and basic seedlings per m2 and basic seedlings, and the panicle-bearing tiller rate was calculated as the number of panicles per m2 and the maximum number of tillers per m2. A statistical analysis was performed using a one-way analysis of variance (ANOVA) with Statistix 8 (Analytical Software, Tallahassee, FL, USA). The means of the treatments and years were compared usingthe least significant difference test; statistical significance was set at $p < 0.05$.

5. Results

Seedling traits at different seeding rates and seedling ages. For 20-day-old Early Rice (ER) or 15-day-old Late Rice (LR) seedlings, the height of the LSR seedlings exceeded that of the CSR seedlings, and the biomass per height of the LSR seedlings was equal to or slightly lower than that of the CSR seedlings (Table 1). With increasing age, the traits of the LSR seedlings were improved over

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those of the CSR seedlings. In addition, the biomass per plant height in ER increased continually with increased seedling age, while an initial increased followed by a decrease was observed in LR. The maximum biomass per plant height was observed in 20–25-day-old seedlings.

Yield variation based on seeding rate and seedling age. For 20-day-old ER or 15-day-old LR seedlings, the average grain yield of the LSRseedlingswas 10.3% higherfor ER. Moreover, theaveragegrain yield of LSR seedlings was 3.5% lower than that of CSR seedlings, but the difference was not significant ($p > 0.05$, Table 2). As the seedling age increased in LSR rice, the maximum grain yield was recorded on day 25 in ER, day 20 in cultivar 'HY518' (30) (LR), and day 30 in cultivar 'TY390' (LR), whereas a significant difference ($p < 0.05$, Table 2) was observed between older LSR seedlings (25-day-old ER seedlings and 20-day-old LR seedlings) and 20-day-old (15) CSR seedlings. In terms of their yield attributes, the spikelet number per panicle in LSR seedlings was an average of 18.9% higher (25 days in ER) and 15.9% higher (20 days in LR) than that in CSR seedlings. No significant differences were observed in the number of panicles per m2, filling rate, or grain weight between aged LSR seedlings (ER or LR) and 20-day-old (15) CSR seedlings. Therefore, an increase in seedling age (> 5 days) for LSR seedlings, as compared to 20-day-old (15) CSR seedlings, significantly increased the grain yield from mechanically transplanted rice.

Biomass variation according to seeding rate and seedling age. The total biomass of 15-day-old LSR seedlings was slightly higher than that of CSR seedlings in ER, and slightly lower than that of CSR seedlings in LR (Table 3). For LSR seedlings, the total biomass was initially increased and then decreased as seedling age increased; the peak value was 1266 g per m2 for cultivar 'ZZ39' and 1230 g per m2 for cultivar 'ZLY189' in ER, and 1915 g per m2 for cultivar 'HY518' and 1888 gperm2 forcultivar'TY390' in LR. Asignificant difference in total biomass was observed with increased seedling age (5-day intervals) in LSR seedlings compared to 20-day-old (15) CSR seedlings ($p < 0.05$). Furthermore, there was a significant difference in biomass based on reproductive stage betweenincreased seedling age (5-day intervals) in LSR seedlings and 20-day-old (15) CSR seedlings ($p < 0.05$).

Basic seedling, tillering, and panicle-bearing tiller rates according to seeding rate and seedling age. The basic seedling rate of CSR seedlings was double that of LSR seedlings in ER, and an average of 30% greater than that of LSR seedlings in LR (Figure 1a, Figureb). For LSR seedlings, the maximum basic seedling rate was 68 seedlings per m2 for ER and 74 seedlings per m2 for LR. The tillering rate for LSR seedlings was higher than that of CSR seedlings (Figure 1c, Figure 1d). In ER, the panicle-bearing tiller rate of aged LSR seedlings (0–10-day-old) was 9.6–19.1% higher than that of CSR seedlings, and there was a significant difference in the panicle-bearing tiller rate between 25-day-old LSR seedlings and 20-day-old CSR seedlings ($p < 0.05$). Similarly, for LR, the panicle-bearing tiller rate in aged (0–10-day-old) LSR seedlings was an average of 6.2% (-2.3–14.6%) higher than that of age-matched CSR seedlings (Figure 1e, Figure 1f).

Table 1: Influence of seedling age on plant height, leaf number per plant, stem thickness and biomass per plant height of seedlings variation with different seeding rate. Values are means ± standard error of the mean.

Season	Cultivars	Dry Seeding rate (g/tray)	Seedling age (d)	Plant height (cm)	Leaf number per plant	Stem thickness	Biomass pe plant heigh (mg/ cm)
ER	ZZ39	100	20	12.9±2.3	2.9±0.2	2.1±0.1	1.2
		30	20	14.0±2.4	3.2±0.3	2.4±0.6	1.2
			25	17.0±1.8	4.8±0.3	3.2±0.1	1.5
			30	16.8±3.3	4.1±0.5	3.0±0.5	1.7
			35	17.3±1.7	4.4±0.4	3.0±0.1	2.6
		Mean#		**16.3**	**4.1**	**2.9**	**1.8**
	ZLY189	70	20	13.1±2.0	3.0±0.2	2.4±0.1	1.2
		15	20	14.4±2.6	3.0±0.1	2.2±0.1	1.1
			25	15.7±1.3	3.2±0.2	2.8±0.1	1.6
			30	19.7±2.8	3.6±0.2	3.1±0.1	2.1
			35	19.0±2.2	3.6±0.2	3.1±0.2	3.4
		Mean		**17.2**	**3.3**	**2.8**	**2.1**
LR	HY518	70	15	19.5±2.4	3.7±0.3	4.1±0.2	2.8
		15	15	20.6±1.5	3.5±0.2	4.4±0.8	2.7
			20	23.4±4.1	4.0±0.5	4.8±0.4	4.2
			25	24.6±4.7	4.5±0.6	5.3±0.9	4.2
			30	30.5±3.3	4.4±0.5	5.2±0.3	3.6
			35	34.1±3.8	4.5±0.4	5.3±0.7	3.8
		Mean		**26.6**	**4.2**	**5**	**3.7**
	TY390	70	15	16.1±2.6	3.3±0.4	3.1±0.4	2.8
		15	15	20.4±2.1	3.4±0.1	4.1±0.2	2.7
			20	22.9±2.9	3.6±0.6	4.5±0.5	3.7
			25	23.4±3.5	4.2±0.3	4.8±0.8	4.5
			30	25.4±3.5	4.3±0.4	5.0±0.6	4.1
			35	31.5±3.6	4.1±1.0	5.2±0.7	4
		Mean		**24.7**	**3.9**	**4.7**	**3.8**

ER, early rice; LR, late rice. # mean values of different seedling ages in the dry seed rate of 15 (30) $gtray^{-1}$

Table 2: Influence of seedling age on grain yield and its component changes with different seeding rate in the early- and late- season

Season	Cultivars	Dry seeding rate(g/ tray)	Seedling age (d)	Grain yield (t/ ha)	Panicle m^{-2}	Spikelet Panicle-1	Filling rate (%)	Grain weight(mg)
ER	ZZ39	100	20	6.19c	371ab	101b	78.0ab	23.2b
		30	20	6.85bc	332a	113ab	79.8ab	23.7b
			25	7.98a	338b	119a	79.6ab	24.2a
			30	7.14b	340a	124a	82.6a	24.3a
			35	6.64bc	323ab	126a	75.0b	24.2a
		Mean#		**7.15**	**333**	**120**	**79.5**	**24.1**
	ZLY189	70	20	6.79b	361a	96b	80.3a	24.9a
		15	20	7.47ab	337a	119a	80.9a	25.0a
			25	8.23a	369a	115a	79.6a	25.0a
			30	8.17a	329a	120a	79.5a	24.6a
			35	6.99b	338a	116a	75.6a	23.8b
		Mean		**7.72**	**343**	**117**	**78.9**	**24.6**
LR	HY518	70	15	7.73b	375a	135c	71.9b	25.4a
		15	15	7.31b	352ab	149ab	64.4c	25.5a
			20	8.68a	372a	158a	73.6b	25.3a
			25	7.97ab	374a	139c	79.0a	25.4a
			30	7.85b	335b	145ab	79.2a	24.0b
			35	7.34b	349ab	136c	73.3b	24.0b
		Mean		**7.83**	**357**	**145**	**73.9**	**24.8**
	TY390	70	15	6.19c	377ab	156c	63.6a	22.4a
		15	15	6.07c	339b	182a	55.5b	22.1ab

			20	7.45a	352ab	179ab	63.0a	22.4a
			25	6.56bc	364ab	170ab	64.8a	22.0ab
			30	7.73a	387a	167bc	63.0a	22.4a
			35	7.12ab	370ab	170ab	62.7a	21.7b
		Mean		**6.99**	**362**	**174**	**61.8**	**22.1**

ER, early rice; LR, late rice. Values followed by different letters represent significant difference at $p<0.05$.
mean values of different seedling ages in the dry seed rate of 15 (30) g tray^{-1}.

Table 3: Influence of seedling age on aboveground biomass and HI variation with different seeding rate and seedling age in the early- and late season

Season	Cultivars	Dry seeding rate(g/tray)	Seedling age (d)	Biomass in the vegetative stage(g m^{-2})	Biomass in the reproductive stage (g m^{-2})	Total biomass (g m^{-2})	HI%
ER	ZZ39	100	20	541a	454b	995b	50.4b
		30	20	577a	503b	1080b	50.7b
			25	591a	675a	1266a	56.1a
			30	611a	573ab	1185a	52.3b
			35	603a	436b	1039b	51.7b
		Mean#		**595**	**547**	**1142**	**52.7**
	ZLY189	70	20	691a	412c	1103c	55.8a
		15	20	640a	537abc	1178ab	56.6a
			25	646a	584ab	1230a	57.3a
			30	536b	668a	1204ab	57.5a
			35	619ab	504bc	1123bc	55.7a
		Mean		**610**	**573**	**1183**	**56.8**
LR	HY518	70	15	785ab	906c	1691c	54.7bc
		15	15	737bc	874c	1610cd	53.5c
			20	822a	1093ab	1915a	57.1a
			25	692c	1120a	1813b	57.3a
			30	686c	964bc	1650c	56.0ab
			35	680c	860c	1540d	54.1c
		Mean		**723**	**982**	**1706**	**55.6**
	TY390	70	15	959a	696c	1655b	50.7a
		15	15	846c	796bc	1642b	46.1c
			20	996a	892ab	1888a	49.7ab
			25	935ab	827bc	1762ab	48.3b
			30	856bc	923ab	1779ab	51.3a
			35	738d	1010a	1748ab	50.8a
		Mean		**874**	**890**	**1764**	**49.2**

ER, early rice; LR, late rice. Values followed by different letters represent significant difference at $p < 0.05$. # mean values of different seedling ages in the dry seed rate of 15(30) g tray^{-1}.

6. Discussion

Seedling age is an important element that affects the number of spikelets per panicle, panicle length, grain weight, and grain yield in rice [9]. Previous studies have indicated that timely transplantation and appropriate seedling age are essential for producing a higher grainyield[5]. Reportedthat 20 dayswastheidealseedling age, and that the grain yield was reduced when the seedlings were morethan 20daysold[11]. Reportedthatwhenriceseedlingswere mechanically transplanted at 20 days old, the grain yield decreased markedly by 4.66% compared to seedlings that were hand-transplanted[4]. Reported that the grain yield of 42-day-old seedlings that were mechanically transplanted was 31.3% lower thanthat of hand-transplanted seedlings. The results of this study show that older LSR seedlings (> 5 days), compared with 20-day-old (15) CSR seedlings, significantlyincreasedthegrainyieldunderconditions of mechanical transplantation. However, when the age of the LSR seedlings was more than 10 days, the rice grain yield decreased. This result is inconsistent with previous studies. This contradiction may be ascribed to the intense competition among individual seedlings under the limited growth conditions of the seed-nursery bed.

A higher seeding rate also affects seedling quality and grain yields. For example, [4] found that the sowing density was 74g per m2 for hand-transplanted rice and 412g per m2 for mechanically transplanted rice. Meanwhile, [10] found that the seeding rate was 11 g of dry seeds per tray. A lower seed rate (25g seeds per m2) could improve the grain yield of 30-day-old seedlings during the dry season. However, lowering the seeding density from 25 g to 12.5 g of seeds per m2 and increasing nitrogen topdressing has not been shown to enhance grain yields[5].

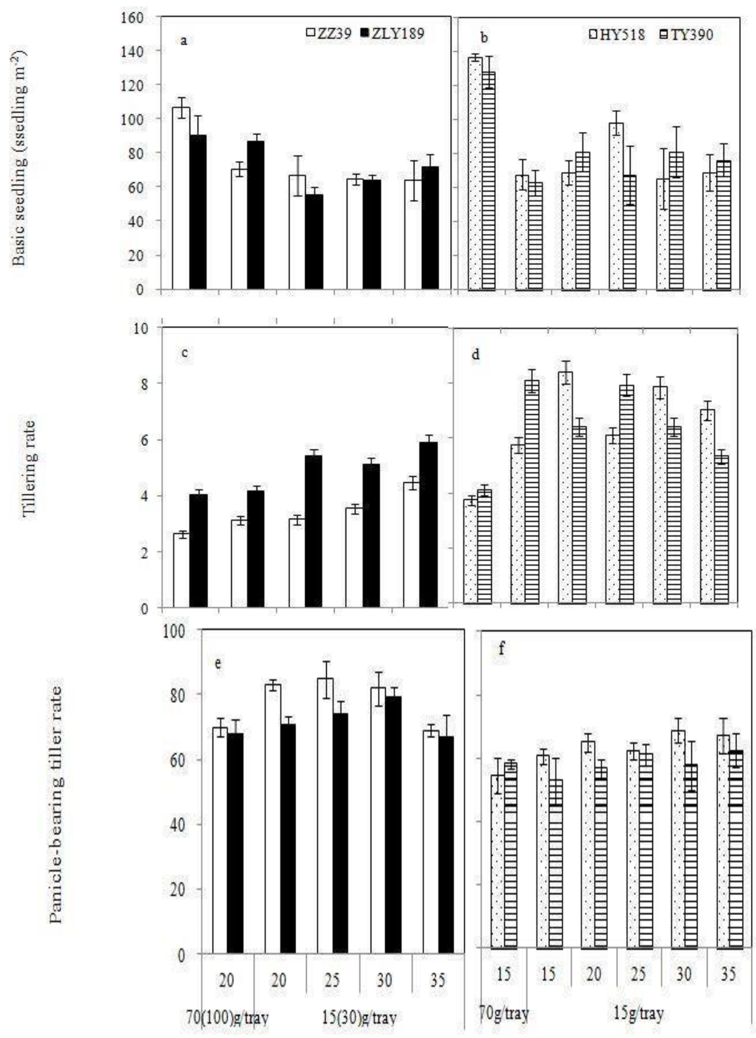

Figure 1: Influence of seedling age on basic seedling (seedling m^{-2}), tillering rate and panicle bearing tiller rate (%) variation with different seeding rate in the early- and late season. The value of the X-axis was represent to different seedling age.

A possible reason for this is that the uniformity and order of seedings can affect seedling quality. It can be difficult to ensure a uniform and well-organized distribution of seedlings using hand transplantation, whereas a uniform and well-organized distribution can be achieved using a mechanical seed-sowing system (as in the present study) [3]. Therefore, the use of older seedlings with a lower seeding rate could compensate for the negative influence of older seedlings on grain yield.

In CSR seedlings, young seedlings between 15 and 20 days ofage were characterized by a suitable plant height ranging from 12–20 cm, and stubborn individual plants with a sufficient nutrient supply from the seedling-nursery tray are suitable for mechanical transplantation [7]. Compared with the CSR seedlings, a 5–10-day increase in age in LSR seedlings could improve seedling quality while not impacting the operation of the rice transplanter. Because of the higher seedling height, the average plant height in the ER and LR was 15.8 cm and 22.6 cm, respectively, while our results also show that there was an increase in leaf number per plant, stem thickness, and biomass per plant height of LSR seedlings. Therefore, the advantages of the older LSR seedlings were: (1) a shorter period for seedling establishment, earlier tillering, and higher tillering rate (Figure 1), improved spikelet per panicle, filling rate, and grain weight (Table 2) due to stronger rice seedlings; (2) increased seedling age could prolong the growth period and alleviate seasonal conflicts brought on by mechanical transplantation; and (3) the suitable transplantion period was increased by 5–10 days (15 [20] to 25 [30] days in CSR seedlings), which is moresuitable for large-scale mechanized rice production.

Our results show that LSR seedlings generally had fewer panicles per m2 than CSR seedlings, and the greatest panicle number per m2 was observed in 5-to 10-day-old LSR seedlings. Panicle number per m2 is a function of the maximum tiller number per m2 and panicle-bearing tiller rate [11]. In this study, the higher panicle number per m2 observed in older LSR seedlings was mainly attributed to the high tillering rate in LR and the higher panicle-bearing tiller rate in ER compared with CSR seedlings, while there was no significant difference between prolonged seedling age in LSR and CSR seedlings. This is in agreement with a report by [11], who observed that there was a tight positive relationship between panicle number per m2 and the maximum tiller number per m2. In contrast to panicle number per m2, the spikelet number per panicle was greater in older LSR seedlings than in older CSR seedlings. In this regard, it is suggested that a strong compensatory mechanism exists between the panicle number per unit land area and spikelet number per panicle [12, 11]. Therefore, it is possible that a yield increase resulted from the spikelet number per panicle in older (5–10-day delay) LSR seedlings, rather than the panicle number per unit land area; further study is required to confirm this speculation.

Grain yield is determined by the aboveground biomass and harvest index [12]. In this study, the harvest index was comparable between treatments; therefore, further improvements in rice yield may be more dependent on the ability to increase biomass production than on an increase in the harvest index [13, 14, 15]. The highest total biomass was observed in 5 day-delayed LSR seedlings compared to age-matched CSR seedlings, while there was no significant difference between prolonged seedling age in LSR versus CSR seedlings. Further analysis found that a significant difference in biomass was observed in the reproductive stage, but not in the vegetative stage.

7. Conclusions

An increased seedling age in LSR cultivars can result inimproved seedling quality, an improved tillering rate, and an improved panicle-bearing tiller rate, which in turn can improve biomass accumulation and grain yields. Compared with 15-to 20-day-old CSR seedlings, a 5–10-day delay in seedling age in LSR seedlings resulted in an increase in rice growth and development and a higher and more stable grain yield. Finally, the uniform and orderly transplantation of LSR seedlings is important for mechanically transplanted seedlings.

8. Acknowledgements

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