water

Article

Crop Performance and Water Productivity of Transplanted Rice as Affected by Seedling Age and Seedling Density under Alternate Wetting and Drying Conditions in Lao PDR

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Abstract: Drought is common under rainfed lowlands in Lao People's Democratic Republic, and with the uncertain onset of rains during the wet season, delay in transplanting results in yield reduction. This study aims to explore ways to ameliorate the negative influence of delayed transplanting on rice crop. A field experiment was conducted for two wet seasons to investigate the effect of seedling age and seedling density on crop performance in terms of grain yield and water productivity. The experiment was laid out in a split–split plot design in four replicates, with seedling age as the main plot, seedling density as the subplot, and varieties as the sub-sub plot. In both years, there were significant seedling age and variety interactions on grain yield. Higher grain yields were observed with older seedlings having stronger tillering propensity. Seedling density did not affect grain yields in both years, but on grain yield components. Shorter duration variety received less supplemental irrigation than longer duration varieties. Late transplanting improved total water productivity but decreased irrigation water productivity due to harvesting delay. The total crop growth duration (from sowing to maturity) was prolonged with transplanting delay. However, the total stay of plants in the main field (from transplanting to maturity) was reduced by 3–5 d for every 10 d delay in transplanting. The results indicated that a good selection of varieties and increasing seedling density improve crop performance and water productivity with delayed transplanting.

Keywords: delayed transplanting; seedling age; seedling density; wet season

1. Introduction

Efficient water use in rice cultivation is a prerequisite to sustain food security for the rice-consuming population of the world. In recent years, increasing water scarcity has been a major threat to rice production in Asia, where by 2025, about 15–20 million of irrigated rice is estimated to suffer [1]. If today's food production and environmental trends continue, crises in many parts of the world will arise. Action should now be taken to improve water use in agriculture to address severe water challenges for the next 50 years [2].

Rice is a key staple in the Lao People's Democratic Republic (PDR) and is an important component of food security efforts in the country. Lao PDR has one of the world's highest per capita consumption of rice, with around 179 kg per capita per year recorded in 2007 [3]. Rice production in the country is the primary source of livelihood for 724,000 producers. The rainfed lowland rice system dominates with only 13% of the total area being irrigated, considering the total paddy harvested area of 830,000 ha in 2011 [4]. Availability and access to water have been identified as the major constraints to the improvement of rice-based farming systems. Lao PDR has seen a high incidence of significant floods and droughts, which severely affected agricultural production in the country [5].

Rice is the biggest user of water in agriculture and in fact, one of the biggest users of the world's fresh water resources [1]. Most rice fields are under conventional continuous flooded conditions [6], which leads to high amounts of surface runoff, seepage, and percolation losses that account for 50%–80% of total water input [7]. With decreasing water availability for agriculture and with increasing demand for rice, water input in rice production should be reduced and water productivity must be increased. Many water-saving practices, including alternate wetting and drying (AWD), have been identified and promoted for widescale dissemination in Asia to reduce water input and increase water productivity [8]. AWD has been successfully evaluated and introduced at the farmers' demonstration fields in the drought-prone southern provinces of Lao PDR in the 2011 and 2012 dry seasons, respectively [9]. Comparison between AWD and the farmers' water management practice of continuous flooding resulted to similar yields, but a 19%–25% water input reduction with AWD was observed. These results were consistent with what had been reported in other countries that tried the technology [8]. Seedling age at transplanting is an important factor to consider in attaining the uniform crop stand [10] and for regulating growth and yield [11]. When rice seedlings are transplanted at the right age, optimum tillering and growth are achieved. However, if transplanting is delayed, fewer tillers are produced during the vegetative stage resulting to poor yield [12]. "Delayed transplanted rice" or "rice with old seedling age" is the term usually used when transplanted seedling age is more than 25 d [13]. Delayed transplanted rice is common in rainfed lowland fields or in irrigated areas in Lao PDR [5]. The annual cropping cycle in Vientiane province begins either in May or June, depending on the onset of rains, with the preparation of the nursery seedbed and the sowing of seeds for the nursery. In Lao PDR, seedlings are usually transplanted about 30 d or more after sowing. However, the untimely release of irrigation in both dry and wet seasons in irrigated areas or the late onset of rainfall in rainfed areas in the wet season for land preparation and crop establishment activities result in a delay in the transplanting of seedlings. Delayed transplanting may result in yield reduction [13,14].

In this paper, we hypothesize that increasing seedling density at transplanting and use of varieties with stronger tillering propensity ameliorates the effects of delayed transplanting on crop performance. Moreover, delayed transplanting reduces irrigation requirement and increases irrigation water productivity during the rainy season. To test this hypothesis, a field experiment was conducted to evaluate the interactive effects of seedling age, seedling density, and variety on post-transplanted rice crop development, grain yield, and water productivity.

2. Materials and Methods

2.1. Site Description

A field experiment was conducted at the Agricultural Research Center (ARC) of the National Agriculture and Forestry Research Institute (NAFRI) in Vientiane, Lao PDR to investigate the effect of seedling age (SA) and seedling density (SD) on post-transplanting performance of selected waxy Lao rice varieties in terms of crop growth, grain yield, water input, and water productivity. The experimental area (16°29′ N, 104°49′ E) in the research center was characterized by loam soil (21% clay and 39% silt), which is typical for lowland rice. The soil properties are shown in Table 1. The area was previously used as a production plot and was cropped with lowland rice in both wet and dry seasons. Vientiane province has a monsoonal climate, with the southwest monsoons being associated with distinct wet (May–October) and dry (November–April) seasons. Mean annual rainfall at the experimental area (taken at a nearby agrometeorological station 200 m away) is about 1790 mm, of which 1500 mm

(89% of total rainfall) falls during the wet season, and only about 197 mm (or 11%) falls during the dry season. Annual mean solar radiation in the area is about 20 MJ m^{-2} d^{-1}, with a range of 18–24 MJ m^{-2} d^{-1}. Temperatures increase gradually from around 28–30 °C in January to about 35 °C in April. Peak monthly maximum temperature is recorded in April, immediately before the start of the wet-season rains. The temperature remains above 30 °C between April and October and starts to decline from late October. Minimum temperatures also follow a similar pattern, with 18−20 °C in January to about 26−27 °C in April, then gradually declining from late October [5].

Table 1. Soil properties of the top soil layer (0–20 cm) in the experimental site.

Soil Property	Mean
% clay	21.0
% silt	39.0
% sand	40.0
pH	6.0
Organic C (%)	2.3
Total N (%)	6.9
Available P (mg kg^{-1})	28.4
Available K (mg kg^{-1})	29.1
CEC (meq 100^{-1}g)	4.5

2.2. Experimental Setup and Treatment Details

The field experiment was implemented for two wet seasons (2014 and 2015) and was laid out in a split–split plot design with three replicates. In this experiment, seedling age at the time of transplanting (SA) was assigned as the main plot, seedling density (SD) as the subplot, and variety (V) as the sub-subplot. Four levels of SA were used: 15 (SA_{15}), 25 (SA_{25}), 35 (SA_{35}), and 45 (SA_{45}) d-old seedlings; three levels for SD: One (SD_1), three (SD_3), and five (SD_5) seedlings per hill; and three different waxy or glutinous varieties: IRUBN0300–63–5–4 (V_1), TDK10239–SSD4–303–1 (V_2), and TDK-8 (V_3). V_1 was considered a high-tillering variety, while V_2 and V_3 were relatively low and medium tillering varieties, respectively. V_3 (TDK-8) is a variety widely grown in southern Lao PDR because of its good eating quality (used as check variety in this experiment), while V_1 and V_2 were two of the most promising waxy rice varieties tested at ARC. A total of 108 plots (4 SA × 3 SD × 3 V × 3 replications) were established in each year of the field experimentation, with a sub-sub plot size of 12 m^2 (2 m × 6 m). Only the main plot (SA) was separated by bunds, but a buffer space of about 0.5 m was established in each sub-subplot.

The experimental field was the same for both years and was prepared with one dry plowing (after all crop residues were removed from the field), followed by land soaking and two wet harrowing operations to achieve thorough puddling in each year. Bunds and canals were constructed and plastic linings were installed to 40 cm depth in the sides of the bunds to reduce seepage losses around the main plot. Transplanting schedules were based on seedling age treatments. At a 10 d transplanting interval, SA_{15} was transplanted first, then followed by SA_{25}, SA_{35}, and SA_{45} treatments, respectively. Transplanting dates during 2014 were the following: 1 July (SA_{15}), 10 July (SA_{25}), 20 July (SA_{35}), and 30 July (SA_{45}); whereas during 2015, transplanting dates were 2 July (SA_{15}), 12 July (SA_{25}), 23 July (SA_{35}), and 2 August (SA_{45}). Transplanting of seedlings was carefully done at a regular spacing of 20 cm × 20 cm in all treatments to minimize seedling transplanting shock and to allow the plants to recover quickly in the experimental field. The number of seedlings planted per hill varied according to the seedling density treatments (one, three, and five seedlings per hill) of the experiment.

2.3. Seedbed Management

Seedlings used in the experiment were grown in the seedbed adjacent to the experimental field. Three well-puddled seedbed plots were prepared with one seedbed plot for each variety used.

Each plot was equally divided into four divisions for the different seedling ages. To raise healthy and vigorous seedlings in the seedbed, seeding rate and fertilizer management recommendations from [14] were used. Basal fertilizer was applied (2 g N + 3 g P + 2 g K m^{-2}) in the seedbed using complete fertilizer (14–14–14). After testing for germination rate, the pregerminated seeds of the three varieties were carefully sown in the seedbeds separately by variety at a uniform seeding rate of 25 g m^{-2}. After sowing, the seedbeds were kept wet all the time to avoid hardening of the soil, which may damage seedling roots when pulled out. The seedbeds were flooded as seedling height increased. Seedlings were carefully hand-pulled 1 d before transplanting, and the pulled seedlings with intact roots were placed in plastic trays with water. Only needed seedlings were hand-pulled for specific seedling age treatments. Seedbeds were continuously managed until the transplanting of the last seedling age treatment (SA_{45}) in the main field was completed.

2.4. Water Management in the Main Field

During the first 3 wk after transplanting, soil was kept saturated to promote better seedling establishment. Thereafter, water management based on safe AWD practice was used at the vegetative stage and after the flowering stage, where timing of irrigation was based on water depth in the field water tubes installed in each plot. This was done by irrigating the plots only when field water depths in the field were about 15 cm below the ground surface. During the flowering stage, plots were under continuous flooding (1–5 cm depth) to avoid possible yield losses. Water stress at the flowering stage may induce spikelet sterility and will thus result in yield loss. Near the end of crop maturity (10–15 d before harvesting), terminal drainage was implemented.

2.5. Fertilizer and Other Cultural Management

In each season, a total of 60 kg ha^{-1} of nitrogen (N) was applied in three splits: (1) Basal or before transplanting (30 kg ha^{-1}), (2) 21–23 d after transplanting (DAT) or mid-tillering of crop growth stage (15 kg ha^{-1}), and (3) panicle initiation (15 kg ha^{-1}). Basal application of phosphorus (P) and potassium (K) were also done at 30 kg ha^{-1} each. Complete (14–14–14) and urea (46–0–0) fertilizers were used as source of N,P and K for basal application, whereas urea was used as a source of N during the crop growth period. All fertilizers were applied during flooded soil condition in all treatments. To keep the experiment field weed-free, preemergence herbicide (butachlor) was applied a few days after transplanting, and then spot hand weeding was done during the vegetative growth stage until a full canopy cover was achieved. Plants were also protected from pests and diseases using agrochemicals.

2.6. Data Collection and Calculation

Field water tubes were used to guide the implementation of AWD [6], and to estimate the amount of irrigation input in the field experiment. Field water depths in the main plots of the experiment were regularly monitored using field water tubes that were installed at a depth of 15 cm below the ground surface. Monitoring of field water depths was done every other day, between 8:00 and 9:00 AM for all treatments from transplanting until 15 d before harvesting. Water level inside the tube was measured from the top to the level of the water inside the tube. To get the actual depth of water inside the tube, the reading will be subtracted from the height of the AWD tube that protruded above the soil surface.

Irrigation dates were all noted and during irrigation events, field water tubes were read before and after each irrigation. The irrigation water input under AWD conditions was then computed using a procedure outlined by [15] as follows:

$$I = d_f - ((\theta_s - \theta_i) \times D), \quad (1)$$

where I = irrigation (mm); d_f = final water depth above soil surface (mm); θ_s = soil water content at saturation (cc/cc); θ_i = soil water content when field water falls below the ground surface (cc cc^{-1}), which was assumed as the field capacity, especially when the perched water table is 15 cm or more from the soil surface; and D = depth of the perched water table.

Since flooded conditions were essential during flowering stage, irrigation water input was computed as:

$$I = d_f - d_s, \tag{2}$$

where d_s = initial field water depth (mm) above the soil surface.

Total water input was the sum of total irrigation and rainfall from transplanting to 15 d before harvesting. Rainfall was taken from the automatic weather station (Vantage Pro2 Weather Station, Davis Instruments Corp, USA) installed adjacent to the experimental area. Groundwater depths were also monitored using three observation wells (2-inch diameter PVC pipe, 2 m long, driven down to 1.5 m) installed along the bunds in the upper, middle, and lower portions of the experiment field. Reading of the water levels in the observation wells was also done every 2 d, from transplanting to 15 d before harvest between 8:00 AM and 9:00 AM. The water level in the observation wells was measured from the top to the level of the water inside the tube. To get the actual depth of the groundwater, the reading was subtracted from the height of the tube that protruded above the soil surface.

Plant height and number of tillers per hill were monitored every 2 wk from transplanting to flowering. Phenology dates (mid-tillering, panicle initiation, flowering, grain filling, and physiological maturity) were monitored for each sub-subplot. At harvest, crop cut samples were taken from 110 h at the center of each plot to determine grain yield. Moisture content of the grains was measured with a digital grain moisture meter (OGA Electric Co., Ltd, Japan), and grain yield was calculated at 14% moisture content. Grain yield components, number of panicles m^{-2}, % spikelet sterility, number of spikelet per panicle, 1000-grain weight, and total biomass of grains and vegetative matter were determined in five-hill samples near the harvest area. Harvest index (in percent) was also determined by dividing grain yield by total biomass and then multiplying it by 100. Irrigation water productivity (WP_I) and total water productivity (WP_{I+R}) were calculated as kg grain m^{-3} water input. For WP_I, water input was from the sum of all irrigations, while for WP_{I+R}, water input was from total rainfall and the sum of all irrigations received by the plants from transplanting up to 15 d after harvesting.

Selected weather data (rainfall, minimum, and maximum temperatures) were taken from the agrometeorological station in the ARC. Seasonal means and sums were reported based on the actual growth duration per treatment.

2.7. Statistical Analysis

The data were subjected to an analysis of variance [16], using International Rice Research Institute's (IRRI) open-access software for statistical analysis, which was implemented in the R statistical package [17]. Treatment means were separated using the least significant difference (LSD) tests and compared at the $p \leq 0.05$ level of significance. Significant interactions of factors used in this study were reported.

3. Results

3.1. Weather

The total wet season rainfall was higher in 2015 (1993.0 mm) than in 2014 (1478.0 mm). Most of the rains occurred during the wet season (May–October), with about 89.3% and 73.7% of the total rainfall in 2014 and 2015, respectively. During the crop growth period (July–October), however, the total accumulated rainfall was higher in 2014 (1078.4 mm) than in 2015 (905.7 mm) as shown in Figure 1, where the highest accumulated monthly rainfall was observed in July (451.9 mm) during 2014 and in September (322.7 mm) during 2015. Mean daily maximum and minimum temperatures during the crop growth period in 2014 and 2015 were relatively similar during the same period (July–September), ranging from 27.5 to 35.3 °C (Figure 1). However, the mean daily minimum temperature was lower in 2014 (23.4 °C) than in 2015 (24.9 °C).

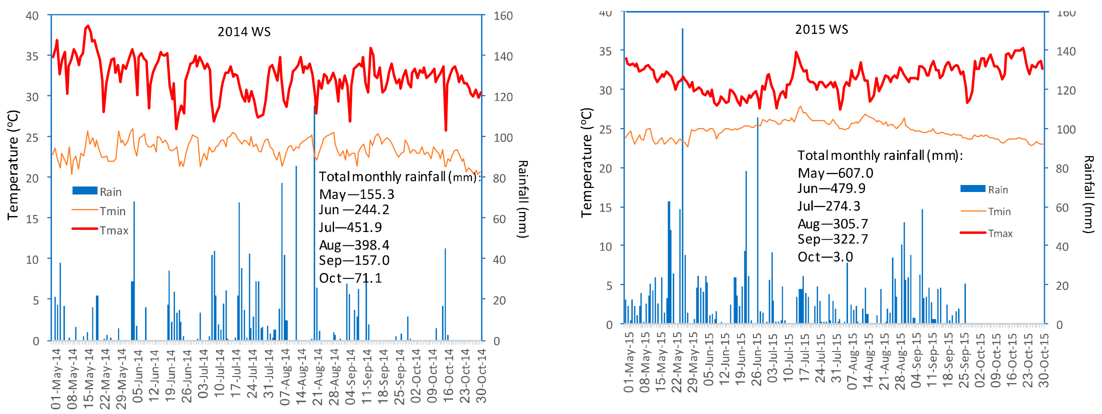

Figure 1. Daily maximum and minimum temperature, and daily rainfall in the study site, 2014 and 2015.

3.2. Hydrological Conditions and Water Level Changes

Groundwater table depths were shallow during wet seasons, especially from July to September. Similar trends of groundwater level fluctuations were observed for both years. As shown in Figure 2, groundwater depth below the ground surface fluctuated between 21 and 33 cm in July, between 18 and 57 cm in August, and between 20 and 63 cm in September, gradually declining after the second week of September.

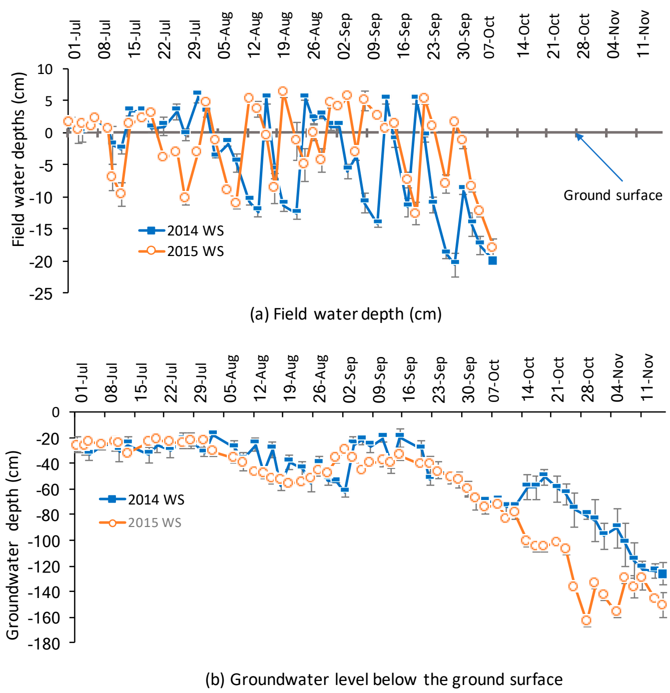

Figure 2. Average field water (**a**) and groundwater (**b**) level fluctuations in the study site during 2014 and 2015. Field water level fluctuations presented were the average of $SA_{15} \times SD_1 \times V_1$ plots, while average groundwater level was from the three groundwater observation wells installed in the site.

In October, groundwater table depths ranged from 51 to 84 cm during 2014 and 60 to 163 cm in 2015. In terms of field water depths, (using average water level data for $SA_{15} \times SD_1 \times V_1$ plots as example), it fluctuated between 5 cm (above the surface) and −15 cm (below the ground surface) during the crop growth stage in both years. Water levels were slightly above the ground surface (0–2 cm) during the first week after transplanting; after the plants fully recovered from transplanting shock, water levels fluctuated between 6 cm and –15 cm. While rice plants received ample rains in 2014 and 2015, the rains were not evenly distributed during the crop growth period. As a result, the paddy soil surface experienced five drying cycles in 2014 and about six drying cycles in 2015. This means that there was some form of AWD condition of the soil surface in the field experiment. Field water level fluctuations in September and October (Figure 2) were due to the application of supplemental irrigation as the rains became limited.

3.3. Crop Growth and Development

On average, total crop growth duration (sowing to harvest) was longer in 2015 than in 2014 by about 4 d (Table 2). SA and V significantly affected crop growth duration among the treatments. No significant interactions between treatments were revealed in the ANOVA. The number of days from sowing to panicle initiation and from sowing to harvest was highest in SA_{45} and lowest in SA_{15} between SA treatments. Mean differences in total crop growth duration between SA_{45} and SA_{15} were 17 d in 2014 and 20 d in 2015. The mean difference between SA_{35} and SA_{15} (10–13 d) was observed in both years, while a difference of 6–7 d was observed in both years between SA_{25} and SA_{15}. In terms of plant growth duration in the main field (from transplanting to harvest), the older the seedlings at transplanting, the shorter the crop duration. This means that SA_{45} had the shortest duration and SA_{15} had the longest duration in the main field in both years. A difference of 4–5 d was observed between SA_{15} and SA_{25}, 7–10 d between SA_{15} and SA_{35}, and 10–13 d between SA_{15} and SA_{45}. In terms of varieties, V_3 had the longest crop growth duration, followed by V_2 and V_1, respectively.

Table 2. Crop duration from sowing to panicle initiation and harvest, 2014 and 2015 wet seasons.

Season/ Treatment	2014			2015		
	Sowing to PI (d)	**Sowing to Harvest (d)**	**Transplanting to Harvest (d)**	**Sowing to PI (d)**	**Sowing to Harvest (d)**	**Transplanting to Harvest (d)**
Seedling age						
SA_{15}	58 d	117 d	102 a	61 d	119 d	104 a
SA_{25}	68 c	122 c	97 b	68 c	125 c	100 b
SA_{35}	73 b	127 b	92 c	79 b	132 b	97 c
SA_{45}	84 a	134 a	89 c	89 a	139 a	94 d
Seedling density						
SD_1	70 a	124 a	94 a	74 a	127 a	97 a
SD_3	72 a	126 a	96 a	74 a	129 a	99 a
SD_5	71 a	126 a	96 a	76 a	130 a	100 a
Variety						
V_1	68 b	123 b	93 b	71 c	125 c	95 c
V_2	69 b	125 b	95 b	73 b	129 b	99 b
V_3	75 a	127 a	97 a	79 a	132 a	102 a
ANOVA results						
SA	*	*	*	*	*	*
SD	ns	ns	ns	ns	ns	ns
V	*	*	*	*	*	*
SA × SD	ns	ns	ns	ns	ns	ns
SA × V	ns	ns	ns	ns	ns	ns
SD × V	ns	ns	ns	ns	ns	ns
SA × SD × V	ns	ns	ns	ns	ns	ns

[1] Within each column, season and treatment, means followed by the same letter are not significantly different at $p < 0.05$. In the ANOVA results, single (*) asterisk means that the F-value was significant at 5% level, while ns means not-significant.

In general, the average number of tillers (per hill) was higher in 2015 than in 2014 (Table 3). The tillering ability of the rice plants was significantly influenced by seedling age, seedling density, and variety. There was, however, no significant interactions among treatments on tiller count (number of tillers per hill) in both years. In 2014, across seedling age treatments, average tiller count at around panicle initiation stage (42–44 DAT) was highest in SA_{15} (11.7) and lowest in SA_{45} (6.6), while SA_{25} and

SA_{35} had the same number of tillers (10.6; Table 3). In 2015, a similar trend was observed: Highest average tiller count was seen in SA_{15} (15), and the lowest was observed in SA_{45} (9.8). SA_{25} and SA_{35} had similar tiller counts (12.7). During 2014 and at the panicle initiation stage, SD_1 provided the lowest tiller count across SD treatments, while SD_5 had the highest count, although not significantly it was different from SD_3. In 2015, differences in tiller count among seedling age treatments were significant, with average tiller counts being highest in SD_5 (14.8), followed by SD_3 (12.9). The lowest was noted in SD_1 (10.5). Across varieties, as expected, V_1 provided the significantly highest average tiller count in both years (10.9 in 2014 and 14.5 in 2015). In 2014, tiller counts of V_2 and V_3 were similar, while in 2015, V_2 had higher tiller count than V_3.

Table 3. Mean comparison of plant height and tiller count at the panicle initiation stage, 2014 and 2015 wet seasons [1].

Treatment	Plant Height (cm)		Tiller Per Hill (no.)	
	2014	**2015**	**2014**	**2015**
Seedling age				
SA_{15}	55.4 a	63.1 a	11.7 a	15.0 a
SA_{25}	53.3 a	63.0 a	10.5 b	12.9 b
SA_{35}	47.0 b	55.5 b	10.7 b	12.7 b
SA_{45}	38.3 c	54.2 b	6.6 c	9.8 c
Seedling density				
SD_1	44.7 a	55.8 a	6.5 b	10.5 c
SD_3	49.3 a	59.3 a	10.6 a	12.9 b
SD_5	51.4 a	62.0 a	11.9 a	14.8 a
Variety				
V_1	41.9 b	53.2 c	10.9 a	14.5 a
V_2	53.3 a	60.9 b	9.5 a b	13.1 b
V_3	50.2 a	63.0 a	9.2 a b	11.7 c
Interaction significance				
SA × SD	ns	ns	ns	ns
SA × V	ns	ns	ns	ns
SD × V	ns	ns	ns	ns
SA × SD × V	ns	ns	ns	ns

[1] Within each column and treatment, means followed by the same letter are not significantly different at $p < 0.05$. In the interaction significance, ns means that the F-value is not significant.

On the average, plants were taller in 2015 than in 2014 (Table 3). At the panicle initiation stage, only seedling age and variety significantly influenced plant height. No significant interactions among treatments on plant height were observed. In both years, early transplanted seedlings (SA_{15} and SA_{25}) were significantly taller than late transplanted ones (SA_{35} and SA_{45}). In 2014, the mean difference in height between SA_{15} and SA_{45} was about 15 cm, while in 2015, the difference was about 9 cm. No significant difference in plant height was found between SA_{15} and SA_{25} in both years; but between SA_{35} and SA_{45}, a difference of about 9 cm was observed in 2014. Across varieties, plant heights of V_2 and V_3 were similar in both years and were, on average, significantly taller than V_1 by 10 cm.

3.4. Grain Yield and Grain Yield Components

Comparing between years, grain yields were significantly lower in 2014 (3.9 t ha^{-1}) than in 2015 (4.5 t ha^{-1}). There was also a significant interaction between year and SA in the experiment (ANOVA results not shown). As shown in Figure 3, the highest value was observed in SA_{15} in 2014 (albeit not significantly different from the SA_{25}, SA_{35} and SA_{45} during 2014, and SA_{45} in 2015), while in 2015, the highest value was obtained in SA_{15} (although not significantly different from SA_{25} and SA_{35} in 2015).

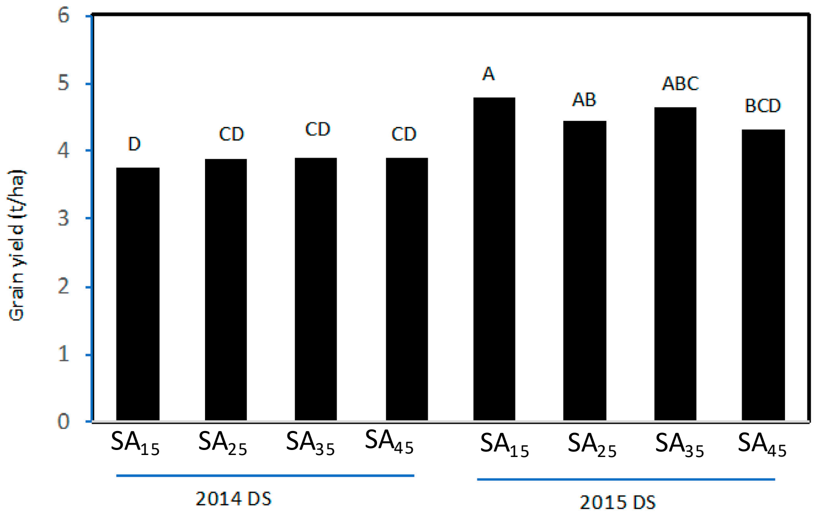

Figure 3. Comparison of yields across treatments and years (2014 and 2015). Columns with common letter are not significantly different at $p < 0.05$.

Among treatment factors, ANOVA results indicated that V significantly affected grain yield in each year (data not shown), while SA and SD did not. Higher (although not statistically different) yields were observed in V_2 and V_3, while lower grain yields were observed in V_1 in both years (Table 4). A significant SA × V interaction on grain yield was also observed in both years. Comparing SA at each level of V during 2015, a lower grain yield of SA_{45} (albeit not significantly different from SA_{25}) was observed in the V_3 level, while in the V_1 and V_2 levels, grain yields in all SA treatments were similar (Table 5). In 2014, however, there were mixed yield trends when comparing SA at each level of V: Significantly lower yields for younger seedlings (SA_{15}) in V_1 and V_2, while lower yields in late transplanted seedlings (SA_{45}) in V_3. In 2015, comparing V at each level of SA showed significantly lower yields in V_1 compared with the other two varieties (particularly under SA_{15}, SA_{25}, and SA_{35}), while grain yields in V_2 and V_3 were the same under seedling age levels. Similarly, in 2014, grain yields were lower in V_1, although not significantly different from V_3 under SA_{25} and SA_{45}.

In terms of grain yield components, no significant effect of year (2014 vs. 2015) was found in any of the yield components. However, in terms of experiment treatment effects in each year, the number of panicles (per m^2) was significantly influenced by all three factors (SA, SD, and V) during 2014 and by two factors (SD and V) in 2015. However, no significant interaction effects of treatments were found in both years. As shown in Table 4, across seedling age the significantly highest number of panicles was observed in SA_{25} particularly in 2014, and the lowest in SA_{45} (although not significantly different from SA_{15} and SA_{35} in 2014 and from SA_{15}, SA_{25}, and SA_{35} in 2015). Across seedling densities, a greater number of panicles per m^2 were observed with higher seedling densities than with lower seedling densities. In both years, the highest number of panicles was observed in SD_5 (although not significantly different from SD_3 in 2015); it was significantly lowest in SD_1. Across varieties, V_1 consistently produced the significantly highest number of panicles compared with the other varieties; no significant difference in the number of panicles was found between V_2 and V_3 varieties in both years.

Table 4. Effects of seedling age, seedling density and variety on grain yield, grain yield components, and harvest index of rice, 2014 and 2015 [1].

Season	Treatment	Panicle (no. m^{-2})	Spikelet (no. panicle^{-1})	1000-Grain Weight (g)	Filled Grain (%)	Grain Yield (t ha^{-1})	Harvest Index (%)
2014	Seedling Age						
	SA_{15}	180.6 b	106.2 c	29.1 a	79.3 c	3.7 a	39 a
	SA_{25}	192.2 a	109.0 b	29.7 a	82.2 b	3.9 a	39 a
	SA_{35}	180.8 b	107.6 b c	29.3 a	83.9 a	3.9 a	36 a
	SA_{45}	176.2 b	114.1 a	28.8 a	80.5 c	3.9 a	42 a
	Seedling Density						
	SD_1	167.3 c	120.1 a	28.9 a	80.4 a	3.9 a	40 a
	SD_3	177.0 b	107.0 b	29.2 a	82.3 a	3.8 a	40 a
	SD_5	203.0 a	100.5 c	29.5 a	81.8 a	3.9 a	36 a
	Variety						
	V_1	236.3 a	97.5 c	21.2 c	82.6 a	3.5 b	42 a
	V_2	150.1 b	108.5 b	36.9 a	79.4 a	4.1 a	38 a
	V_3	161.0 b	121.6 a	29.5 b	82.5 a	3.9 a	37 a
	Interaction Significance						
	SA × SD	ns	ns	ns	ns	ns	ns
	SA × V	ns	ns	ns	*	**	ns
	SD × V	ns	ns	ns	ns	ns	ns
	SA × SD × V	ns	*	ns	ns	ns	ns
2015	Seedling age						
	SA_{15}	182.0 a	108.8 b	33.1 a	78.9 a	4.8 a	31 a
	SA_{25}	196.0 a	114.3 a	28.9 b	79.3 a	4.4 a	32 a
	SA_{35}	179.2 a	104.24 b	28.8 b	79.0a	4.7 a	32 a
	SA_{45}	179.0 a	94.2 c	28.7 b	79.4 a	4.3 a	32 a
	Seedling density						
	SD_1	168.3 b	111.2 a	31.1 a	80.0 a	4.6 a	35 a
	SD_3	192.8 a	103.2 b	29.6 a	79.9 a	4.5 a	31 b
	SD_5	193.0 a	101.8 b	29.0 a	77.6 a	4.5 a	30 b
	Variety						
	V_1	227.2 a	100.1 a	22.8 c	78.4 b	4.3 b	35 a
	V_2	161.9 b	108.7 a	36.6 a	75.5 b	4.7 a	31 a
	V_3	165.0 b	107.6 a	30.3 b	83.7 a	4.7 a	30 a
	Interaction significance						
	SA × SD	ns	ns	**	ns	ns	ns
	SA × V	ns	ns	ns	ns	*	ns
	SD × V	ns	ns	ns	ns	ns	ns
	SA × SD × V	ns	ns	ns	ns	ns	ns

[1] In a column and within the same treatment factor, means with the same lower case letter are not significantly different at $p < 0.05$. In the interaction significance, double (**) and single (*) asterisks mean that the F-value are significant at 1% and 5% level, respectively, and ns means not significant.

Table 5. Effects of seedling age × variety (SA × V) interaction on grain yield (2014 and 2015) and percent filled spikelet (2014) and of seedling age × seedling density (SA × SD) interaction on 1000–grain weight (2015) [1].

Seedling Age	Grain Yield (t ha^{-1})			1000-Grain Weight (g)			Filled Spikelet (%)		
	V_1	V_2	V_3	SD_1	SD_3	SD_5	V_1	V_2	V_3
					2014				
SA_{15}	3.4b C	3.8b B	4.1ab A				78.6b B	77.0b B	82.4a A
SA_{25}	3.6ab B	4.2a A	3.8bc B				84.4a A	78.6b B	83.7a A
SA_{35}	3.4b B	4.1a A	4.2a A				84.0a A	83.0a A	84.9a A
SA_{45}	3.7a B	4.4a A	3.7c B				83.4a A	78.9b B	79.0b B
					2015				
SA_{15}	4.6a B	4.9a A	5.0a A	38.6a A	31.2a B	29.4a C			
SA_{25}	4.2a B	4.5a A	4.6ab A	28.2b A	29.2b A	29.4a A			
SA_{35}	4.2a B	4.9a A	4.9a A	28.7b A	29.0b A	28.8a A			
SA_{45}	4.3a A	4.4a A	4.3b A	28.7b A	28.9b A	28.5a A			

[1] In a column and within the same year, means with the same lowercase letter are not significantly different (comparison of SA at each level of V for grain yield and percent filled spikelet, and comparison of SA at each level of SD for 1000-grain weight). In a row and within the same year, means with the same uppercase letter are not significantly different (comparison of V at each level of SA for grain yield and filled spikelet, and comparison of SD at each level of SA for 1000-grain weight).

Significant effects of SA, SD, and V were observed on the number of spikelets per panicle (spikelet count) in 2014, while in 2015 only significant effects of SA and SD on spikelet count were noted. No significant interactions were found among factors on the spikelet count in both years. Across SA treatments in 2014, the spikelet count was highest in SA_{45}, followed by SA_{25}, then SA_{35} and SA_{15}, although there was no significant difference between SA_{15} and SA_{35} and between SA_{25} and SA_{35} (Table 4). In 2015, there was an opposite trend: SA_{45} provided the lowest spikelet count among the four SA treatments. Highest spikelet count was recorded in SA_{25}, followed by SA_{15} and SA_{35}, but as in 2014, the difference between SA_{15} and SA_{35} was not significant. Across SD, highest spikelet count was observed in SD_1 and the lowest was seen in SD_5 in both years. The difference in spikelet count between SD_3 and SD_5 was only significant in 2014. Across varieties, the highest spikelet count was found in V_3 and the lowest in V_1 in 2014. Spikelet counts were similar in all varieties used in the 2015 experiment.

In 2014, the 1000-grain weight (1000 GW) was significantly influenced by V, while by all three factors (SA, SD, and V) in 2015. There was a significant SA × SD interaction on 1000 GW in 2015. In both years, consistently highest 1000 GW was produced in V_2, with the lowest seen in V_1 (Table 4). In 2015, comparison of SA at each SD level (Table 5) indicated that SA_{15} was significantly highest among the four SA treatments under SD_1 and SD_3, whereas similar values were observed among SA_{25}, SA_{35}, and SA_{45}. Under SD_5, no difference in 1000 GW was found among the SA treatments. Comparing SD at each level of SA, a significant difference in 1000 GW was only found under SA_{15} level, where $SD_5 < SD_3 < SD_1$; the differences in means across SD were not significant at the SA_{25}, SA_{35}, and SA_{45} levels.

The percentage of filled spikelet was significantly influenced by SA and V in 2014 and in 2015 by V. There was also a significant SA × V interaction on percent filled spikelet during 2014. Comparing SA at each level of V (Table 5), the lowest percent filled spikelet was observed in SA_{15} under V_1 and V_2 levels, although SA_{15} was not significantly different from SA_{25} and SA_{45}, especially under V_2. However, under V_3, SA_{15}, SA_{25}, and SA_{35} were similar but were significantly higher than SA_{45}. Comparing V at each level of SA (Table 5), significant differences in percent filled spikelet between varieties were observed in SA_{15}, SA_{25}, and SA_{45}. Percent filled spikelet of V_3 was significantly higher than that of V_1 and V_2 under SA_{15}, and significantly higher than V_2 under SA_{25}. However, V_3 was statistically similar to V_2 under SA_{45}, but significantly lower than V_1. Between V_1 and V_2, values were similar under SA_{15}, but V_1 was significantly higher than V_2 under SA_{25} and SA_{45}. Mean harvest index (HI), was relatively higher in 2014 (38.7%) than in 2015 (32%), since both grain yield and dry biomass were higher in 2015 (data not shown). All treatments and their interactions did not influence HI in 2014, but, in 2015, HI was significantly influenced by seedling density. SD_I had higher HI compared with other SD treatments (Table 4), whereas SD_1, SD_3, and SD_5 were not significantly different.

3.5. Water Input and Water Productivity

Rice plants that were planted earlier received more rainfall than those planted later because more rainfall events occurred from July to early September (Table 6 and Figure 1). In 2014, total rainfall received by SA_{15} was 65 mm, 123 mm, and 363 mm more than SA_{25}, SA_{35}, and SA_{45}, respectively. Seedling age and variety significantly affected seasonal irrigation, and total water (irrigation plus rainfall) inputs in both years, while seedling density and interactions among the treatments were not significant. In 2015, the differences in total rainfall between SA_{15} and the other seedling age treatments were 96 mm (SA_{25}), 192 mm (SA_{35}), and 279 mm (SA_{45}). In general, more rainfall was received by plants in 2014 than in 2015 in all SA treatments, ranging from 4% to 20%, with the highest percent difference observed in SA_{35} and the lowest in SA_{45}.

Table 6. Irrigation and rainfall water input during 2014 and 2015 under different seedling age by a variety treatments, ARC, Vientiane, Lao PDR [1].

Treatment	2014			2015		
	Total Irrigation (mm)	Total Rainfall (mm)	Total Water Input (mm)	Total Irrigation (mm)	Total Rainfall (mm)	Total Water Input (mm)
Seedling age						
SA_{15}	439.3 d	1009.2 a	1448.5 a	317.0 d	902.8 a	1219.8 a
SA_{25}	469.5 c	944.4 b	1413.9 b	456.3 c	807.0 b	1263.3 a
SA_{35}	506.4 b	886.2 c	1392.6 b	522.4 b	711.1 c	1233.5 a
SA_{45}	596.9 a	647.2 d	1244.1 c	577.3 a	623.7 d	1201.0 a
Seedling density						
SD_1	500.2 a	876.9 a	1377.1 a	501.3 a	761.2 a	1262.5 a
SD_3	537.7 a	875.6 a	1413.3 a	488.9 a	761.2 a	1250.1 a
SD_5	505.2	862.7 a	1367.9 a	500.3 a	761.2 a	1261.5 a
Variety						
V_1	493.3 b	863.7 b	1357.0 a	465.3 b	759.4 a	1224.7 a
V_2	505.7 a	866.5 b	1372.2 a	470.3 b	759.4 a	1229.7 a
V_3	510.1 a	884.8 a	1394.9 a	524.3 a	764.7 a	1289.0 a
Interaction significance						
SA × SD	ns	ns	ns	ns	ns	ns
SA × V	ns	ns	ns	ns	ns	ns
SD × V	ns	ns	ns	ns	ns	ns
SA × SD × V	ns	ns	ns	ns	ns	ns

[1] In a column and within the same treatment, means with the same letter (lower case) are not significantly different at $p < 0.05$. In the interaction significance, ns means that the F-value is not significant.

With the abundance of rain in the wet season, total irrigation inputs received by the plants were 30%–45% of the total water input in 2014 and 26%−48% in 2015, with delayed transplanted rice receiving a higher fraction of the water input from irrigation than early transplanted seedlings. The lowest amount of irrigation input was received by SA_{15}, whereas older seedlings (SA_{35} and SA_{45}) received more irrigation input in both years (Table 6). The total irrigation input was higher by 36% in SA_{45}, 15% in SA_{35}, and 7% in SA_{25} in 2014 in comparison with SA_{15}. Higher percent differences between irrigation inputs were observed in 2015, wherein against SA_{15}, irrigation input was higher by 82%, 65%, and 44% in SA_{45}, SA_{35}, and SA_{25}, respectively. On a per season basis, the total water input was 1244–1449 mm in 2014 (while it was 1201–1333 mm in 2015). Among SA treatments, SA_{45} received the lowest and SA_{15} received the highest total water input in 2014; in 2015, total water input was similar among SA treatments. V_3 received the highest water input among the varieties tested and V_1 received the lowest, particularly from irrigation, in both years. However, we did not find any significant difference in total rainfall and total water input between varieties during the field experiment.

Irrigation water productivity (WP_I) and total water productivity (WP_{I+R}) were only significantly influenced by V and by SA × V interactions in both years. In 2014, WP_I and WP_{I+R} values ranged from 0.78 to 1.01 kg m^{-3} and from 0.23 to 0.40 kg m^{-3}, respectively; in 2015, the corresponding range were 0.80–1.01 kg m^{-3} and 0.32–0.40 kg m^{-3}. Comparing SA at each level V in 2014, the WP_I of SA_{45} was significantly higher than those of SA_{35} and SA_{15} in V_1 and significantly higher than that of SA_{15} in V_2. However, SA_{45}'s WP_I under V_3 became significantly lower than those of SA_{35} and SA_{15} (Table 7). Comparing V at each level of SA, V_1 had higher WP_I than V_2 at all levels of SA; it was also higher than V_3 at SA_{15} and SA_{35} levels, respectively, in 2014. In 2015, there were no significant differences in WP_I among SA treatments in each variety level. However, comparing WP_I of the different varieties V at each level of SA, V_1 had the lowest and V_3 (although not significantly different from V_2) had the highest WP_I values. In terms of WP_{I+R}, the significantly highest WP_{I+R} value was found in SA_{45} in all levels of varieties in 2014 (Table 7). The lowest values were observed in SA_{15} and SA_{35} under the V_1 level and in SA_{15} and SA_{25} under the V_3 levels; SA_{15}, SA_{25}, and SA_{35} were similar under the V_2 level. Comparing WP_{I+R} of the different varieties V at each level of SA in 2014, V_1 had the lowest values, particularly under SA_{15} and SA_{35} levels, respectively, while, V_2 and V_3 were similar (except in SA_{45}). In 2015, there were no significant differences in WP_{I+R} among SA treatments under V_1 and V_3 levels. Significant differences were only observed under the V_2 level, wherein the most delayed transplanted seedlings (SA_{45}) produced higher WP_{I+R} than did the earliest transplanted seedlings (SA_{15}) in 2015 (Table 7). Comparing varieties at each level of SA, lower values of WP_{I+R} were observed with V_1 under the SA_{25} and SA_{35} levels, whereas no significant differences in WP_{I+R} were found among varieties under SA_{15} and SA_{45}.

Table 7. Seedling age × variety (SA × V) interaction on irrigation water productivity (WP_I) and total water productivity (WP_{I+R}) during 2014 and 2015 wet seasons, respectively [1].

Season/Seedling Age	Irrigation Water Productivity (WP_I)			Total Water Productivity (WP_{I+R})		
	V_1	V_2	V_3	V_1	V_2	V_3
2014						
SA_{15}	0.78 b B	0.89 b A	0.94 a b A	0.23 c B	0.27 b A	0.29 c A
SA_{25}	0.84 ab B	0.96 ab A	0.89 bc AB	0.28 b A	0.30 b A	0.28 c A
SA_{35}	0.79 b B	0.96 ab A	0.97 a A	0.24 bc B	0.30 b A	0.32 ab A
SA_{45}	0.87 a B	1.01 a A	0.86 c A	0.33 a B	0.40 a A	0.33 a B
2015						
SA_{15}	0.89 a B	0.99 a A	1.01 a A	0.32 a A	0.33 b A	0.36 a A
SA_{25}	0.97 a A	1.01 a A	0.98 a A	0.32 a B	0.36 ab AB	0.37 a A
SA_{35}	0.80 a B	0.92 a A	0.97 a A	0.33 a B	0.40 a A	0.40 a A
SA_{45}	0.82 a A	0.84 a A	0.82 a A	0.37 a A	0.37 a b A	0.36 a A

[1] In a column and within the same year, means with the same lowercase letter are not significantly different (comparison of SA at each level of V); in a row and within the same year, means with the same uppercase letter are not significantly different (comparison of V at each level of SA).

4. Discussion

In our experiment, seedling age significantly affected the tillering dynamics of the rice plants. Younger seedlings (SA_{15}) had the highest tiller count, while older seedlings (SA_{45}) had the least in both years, regardless of planting density and variety used. This indicates that rice seedling age is vital in determining tiller occurrence and confirmed the findings of [18] that, with increasing seedling age, tillering was depressed, thereby resulting in reduced tiller number. Increasing seedling density ameliorated the effects of delayed transplanting on tillering performance in our experiment. Transplanting with three (SD_3) to five (SD_5) seedlings per hill produced more tillers per hill than with one seedling per hill (SD_1), and therefore the tiller number of older seedlings could be induced by increasing seedling density at transplanting. The result is in line with the findings of [13,19], but the effect may vary with seasons [20].

Seedling age also affected crop growth duration. In the present study, the phenological development of the rice plants was delayed with the transplanting of older seedlings, resulting in longer crop duration. The panicle initiation and physiological maturity were delayed by 5–11 d and 5–7 d, respectively, for every 10-d delay of transplanting in both years. Similar findings were reported by [14]: An increase of 6–9 d of vegetative phase (sowing to panicle initiation) with 10-d-old seedlings vs. 30-d-old seedlings in Central Luzon Philippines during two dry seasons, and by 8 d during one wet season of field experimentation. With increased total crop duration (sowing to physiological maturity) using older seedlings, the expected shortening of the stay of the crop in the main field did not correspond to the number of days of delay in transplanting. In our study, crop duration in the main field was only shortened by 3–5 d for every 10-d delay in transplanting. Seedling density did not show any effect on the total rice growth duration, regardless of the variety used. Seedling age could be an important factor in determining phenological stages and crop duration. Indisputably, varietal characteristics dictated the tiller number, plant height, and growth duration.

Many research studies have shown that the use of younger seedlings (not older than 25 d old) produced positive impacts on grain yield [21–23], although many authors have also contradicted this [24–26]. In our study, a significant SA $\times$ V interaction on grain yield was found in both years. This means that the influence of seedling age on grain yield depends on the variety used. The use of varieties with a stronger tillering propensity ameliorated the effects of delayed transplanting on crop yield.

Availability of water is crucial in deciding whether to transplant rice seedlings at an early or later stage of the season. When the water supply is uncertain, delay in transplanting becomes inevitable. Rainfall is abundant during the wet season in Lao PDR. However, much of these rains occur from May to September, when crops are in their vegetative growth stage. In our study site, the rains were not evenly distributed during the rainy months and the number of dry spell periods interrupted the regular monsoon rainfalls, which occurred particularly during critical crop growth stages. Therefore, supplemental irrigation is vital to avoid water stress during periods with no rains. In our study, we hypothesized that the shortening of stay in the main field had implications on total water input of the rice crop. However, our results indicated that the shortened stay of older seedlings in the main field did not translate in irrigation reduction. Higher rainfall occurred from July to August in 2014, and from July to September in 2015, which favored early transplanting. Although with relatively shorter stay in the main field, older seedlings were harvested later than younger seedlings; an additional one or two supplemental irrigations were applied (data not shown) before terminal irrigation. Compared with older seedlings (SA_{45}), younger seedling age treatment (SA_{15}) received about 56% more rainfall and about 26% less irrigation in 2014 and about 45% more rainfall and 45% less irrigation in 2015. At the end of the season, total water input (rainfall plus irrigation) was similar among the SA treatments, considering that older seedlings received less rainfall but more irrigation, while younger seedlings received more rainfall but less irrigation. A similar study conducted in the Philippines during the dry season (with comparable seasonal rainfall in Lao PDR) has indicated that the total irrigation input decreased with older seedlings but not in the wet season when rain could not be easily controlled in the field [14]. A significant difference in irrigation input was also found among the varieties used in the experiment. As expected, irrigation received by the shorter duration variety was lower than that of the longer duration variety (i.e., $V_1 > V_2 > V_3$) in both years because of one additional irrigation needed for V_3 before terminal drainage was implemented (data not shown). In terms of water productivity, a significant SA $\times$ V interaction was found in both years.

There is a common understanding among farmers that planting old seedlings (especially with TDK 8) could result in yield losses. The experiment results further confirmed their suspicion. The inference from the results can be given only to the selected varieties used in the experiment. However, as the seeding age at transplantation is more important for better crop productivity, breeders should take note on testing this effect more systematically before releasing the varieties for rainfed lowland rice cultivation.

The overall water management practice in the experimental field was safe AWD and although rainfall was abundant, the field experiments attained an average of five to six wetting and drying cycles in the main field. Safe AWD was developed to reduce irrigation water input without sacrificing rice grain yields particularly in water-short areas and seasons. In our experiment, we have demonstrated that safe AWD is possible during the wet season, although the wetting and drying cycles were fewer compared to what we expected during the dry season with better control of irrigation.

5. Conclusions

Grain yield was significantly influenced by the interaction of seedling age and variety and their interactions subjected to AWD conditions during the study. Grain yields of V_1 (IRUBN0300-63-5-4) and V_2 (TDK10239-SSD4-303-1) increased with increasing seedling age, while grain yield of V_3 (TDK-8) decreased with increasing seedling age, especially in 2014. The effect of seedling age on grain yields largely depended on the variety used particularly during the wet season in Lao PDR. Regardless of the variety, the seedling density treatments (one, three, and five seedlings per hill) had significant effects on tillering dynamics but not on grain yield. Moreover, the use of varieties with a stronger tillering propensity ameliorated the effects of delayed transplanting on crop performance. Late transplanting of seedlings improved total water productivity, but it might decrease irrigation water productivity due to the delay in harvesting. Total crop growth duration was extended with late transplanting, which required one or two supplemental irrigations until harvest when rainfall is not available. However, the total duration of stay of the rice plants in the main field was only reduced by 3–5 d for every 10 d delay in transplanting. Proper selection of varieties to use under delayed transplanting coupled with appropriate seedling density can improve crop performance, yield, and water productivity. More so, safe AWD can be implemented in rainfed areas, thus a high potential for methane emissions reduction during wet seasons.

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