Nitrogen uptake pattern of dry direct-seeding rice and its contribution to yield in a cool temperate climate

Mari Namikawa[a,b*], *Takayuki Yabiku*[a], *Maya Matsunami*[b], *Toshinori Matsunami*[a], *and Toshihiro Hasegawa*[c]

[a] *Tohoku Agricultural Research Center, National Agriculture and Food Research Organization (NARO), Morioka, Japan;* [b] *The United Graduate School of Agricultural Sciences, Iwate University, Morioka, Japan;* [c] *Institute for Agro-environmental Sciences, NARO, Tsukuba, Japan*

E-mail for the *corresponding author: mnamikawa@affrc.go.jp

Word count: 10099 words.

Nitrogen uptake pattern of dry direct-seeding rice and its contribution to yield in a cool temperate climate

Abstract

Dry direct-seeding rice (DDSR) cultivation is expected to reduce production costs compared with transplanted rice (TPR); however, its low nitrogen (N) use efficiency (NUE) has hindered cost reduction. We conducted DDSR and TPR field experiments in a cool climate for three years using two cultivars and three N regimes to determine factors limiting NUE. We confirmed that DDSR had a lower yield and agronomic NUE (ANUE) than TPR by 11% and 30%, respectively. Our analysis revealed that both fertilizer recovery and crop NUE (CNUE, yield per unit N uptake) contributed to the lower ANUE. The difference in CNUE between DDSR and TPR resulted partly from the N uptake pattern, where the amount and proportion of N uptake until panicle initiation (PI) in DDSR were smaller than those in TPR, which had a greater contribution to spikelet density than the N uptake thereafter. We discovered that the growth and N uptake before the fifth leaf stage is key to enhancing N uptake until PI and thereby spikelet density. Split urea application was almost as productive as coated urea application; thus, suggesting that it might represent a cost-effective alternative although it needs improvement for the initial growth and N recovery rate. A high-yielding cultivar, Yumiazusa, consistently outyielded a standard cultivar, Akitakomachi, under both planting methods. DDSR had lower grain filling and single grain mass, possibly restricted by solar radiation and temperature in the grain-filling period. The results provide useful insights for improving the DDSR production efficiency in a cool climate.

Keywords: coated urea, dry direct-seeding rice, nitrogen uptake pattern, nitrogen use efficiency, spikelet density.

Introduction

In Japan, the population involved in agriculture decreased substantially from 11.7 million in 1960 to 2.1 million in 2015, a reduction of 86% (MAFF, 2018), while increasing the cultivating land per farmer. This trend is projected to continue, and labor-saving and cost-effective technologies are expected to gain prominence. Dry direct-seeded rice (*Oryza sativa L.*) (DDSR) cultivation is thus becoming an important alternative for rice farmers in Japan as it can reduce labor costs compared to conventional transplanted rice (TPR) cultivation (Farooq et al., 2011).

The Tohoku region is one of Japan's main rice cultivation areas, producing approximately 29% of the national rice production (MAFF, 2021a). This region has comparatively larger paddy fields than the rest of Japan, suitable for large-scale mechanized farming. Although both wet direct-seeded rice and DDSR technologies are available, DDSR has greater potential for saving labor than wet direct-seeded rice as larger agricultural machinery can be introduced for land preparation and planting practices (Kanmuri et al., 2017). DDSR technology, however, has some drawbacks that prevent it from being the more popular choice in the Tohoku Region. First, DDSR yields are generally lower than TPR. MAFF (2021b) reported that direct-seeded rice, including dry-seeded and wet-seeded rice, yielded 7% less than TPR when averaged over a total of 436 fields and a three-years. Second, DDSR is associated with low nitrogen (N) use efficiency and requires greater fertilizer inputs (Farooq et al., 2011); thus, increasing production costs. Third, appropriate varieties for better performance in DDSR have not been studied in a cool climate.

Many of these problems are related to the climatic conditions in the Tohoku region, located in a cool temperate zone, where rice productivity has been severely limited by low temperatures (Horai et al., 2013). DDSR seedlings are exposed to low

field temperatures from an earlier stage in contrast to the conditions characteristic of TPR seedlings grown, which are covered or protected in nurseries such as greenhouses. One of the important advantages of ponding water is to protect rice plants from cold temperatures because water is generally much warmer than air, particularly in the early stages of rice growth in the cool temperate regions, which is known as the water blanket effect (Shimono et al., 2007). Shimono et al. (2007) demonstrated that in northern parts of Japan, simulated grain yields could be halved in the absence of surface water even without water stress because low temperatures at shoot bases hampered rice growth and yield. In DDSR, seedlings are grown under upland conditions without the water blanket for more than a month before permanent flooding, possibly limiting the yield potential of DDSR in a cool climate.

Temperatures are also highly influential on rice N uptake (Shoji et al., 1976; Takahashi et al., 1976), particularly during the vegetative growth stage (Shimono et al., 2012). The effects of N management on the growth and yield of DDSR have frequently been studied in warm areas such as northwest India, Southern U.S, and central China (Ali and Thind, 2015; Griggs et al., 2007; Liu et al., 2015; Mahajan et al., 2011; Qi et al., 2012; Xu et al., 2021) in the absence of cold temperature stresses. Still, evidence exists that DDSR needs a higher rate of N application than TPR for the same yield (Farooq et al., 2011; McDonald et al., 2006). However, little is known about rice N uptake during early-stage DDSR in cool climate zones. but N uptake of DDSR limited by low temperatures could be another reason for the lower N use efficiency (NUE).

The use of control-release urea is empirically recommended for DDSR in northern regions, such as the Tohoku region, and generally, a greater amount is recommended for DDSR than that for TPR (NARO, 2021), thus hindering the efforts toward the reducing production costs. Additionally, polymer-coated urea uses

microplastic capsules, which have recently been identified as a source of environmental pollution (Katsumi et al., 2020). Consequently, attempts to reduce the use of polymer-coated urea have already started in Japan and will likely expand in the future. Although the split application of urea might represent a low-cost alternative and has been used as a basic method globally, including for DDSR the Europe and the USA, it needs to be investigated whether split application of urea can be an effective option for DDSR in the Tohoku region.

High-yielding and good-quality cultivars are an important option for higher profitability; however, a comparison of cultivars for DDSR suitability in the Tohoku region has not been performed. In a cool climate, low temperatures limit growth duration, and DDSR without protected nursery periods takes longer to mature than TPR. Cultivars for DDSR, therefore, need to be early maturing, which is often not associated with high-yielding characteristics. Recently, however, an early-maturing, high-yielding, and good-eating quality cultivar, Yumiazusa, has been bred and released by NARO and is proven to be high-yielding for transplanting practice in cool temperate areas (Ohta et al., 2017; Yabiku et al, 2021).

Here, we examined how cultivar and N management combinations affect rice growth and yield under DDSR in a cool climate using a standard cultivar (‘Akitakomachi’) and a high-yielding cultivar (‘Yumiazusa’) grown under different N regimes. We focused on the N uptake pattern of DDSR and its relevance to yield and yield components compared to those of TPR. We hypothesized that differences in N uptake at the early growth stage are a large source of variation in DDSR yields. More specifically, we attempted to answer the following questions:

(1) Can the split application of urea be an alternative to control-release fertilizer in terms of yield and NUE?

(2) Does a high-yielding cultivar under transplanting practice retain its yield advantage over a standard cultivar under dry direct-seeding practice?

(3) Which phase of N uptake is critical for yield determination?

To answer these questions, we conducted N fertilizer and cultivar DDSR experiments for three years in Northeast Japan and analyzed the temperature-driven N uptake patterns and their association with grain yield and NUE. These results were compared with TPR experiments conducted side-by-side using the same cultivars (Yabiku et al., 2021).

Material and methods

Study sites

We conducted field experiments at the Experimental Farm of Tohoku Agricultural Research Center, NARO, in Morioka city, Iwate prefecture, Japan (39°45'N 141°08'E) in 2018, 2019, and 2020 rice growing seasons. In each season, two fields were used for the experiments: one for DDSR and the other for TPR. The soils of all experimental fields were classified as Andisols, and some of their chemical properties are shown in Table S1.

Climatic data were obtained from the meteorological observation at the NARO Tohoku Agricultural Research Center located 2 km from the experimental fields. Table S1 presents mean temperatures and mean precipitations from April to October of the experimental years (2018–2020).

Experimental design and field management

Plant materials and N treatments

We used two temperate *japonica* cultivars: 'Akitakomachi' (AK) and 'Yumiazusa' (YA). AK is an old but still widely planted cultivar in northeast Japan, whereas YA is a high-yielding cultivar with high eating quality, released in 2017 (Ohta et al. 2017). For DDSR, we had three N regimes: two with N fertilizers and one without (0N). In the two fertilized plots, polymer-coated urea (CU) and normal urea (NU) were used. For the CU treatment, we applied 12 g N m^{-2} of CU fertilizer as basal, containing three types of slow-release fertilizer differing in the rate and pattern of N release (Chokuha senyou 211, Kumiai Hiryou Co. Ltd., Iwate, Japan). For the NU treatment, a total of 15g N m^{-2} of urea was split-applied four times at 40, 66, 82, and 94 days after sowing (DAS) in 2018, 45, 66, 80, 94 DAS in 2019, and 52, 67, 80, 95 DAS in 2020, at a dose of 6 g N m^{-2}, 3 g N m^{-2}, 3 g N m^{-2}, and 3 g N m^{-2}, respectively. In the 0N treatment, no N fertilizer was applied. Equal amounts of P and K were applied to all plots at the rate of 3.4g P m^{-2} and 3.1g K m^{-2} as basal, respectively, on 23 April (2018), 17 April (2019), and 15 April (2020).

All experiments in DDSR were laid out in a randomized complete block design, where a factorial combination of two cultivars (AK and YA) and three N regimes (0N, CU, and NU) was randomly assigned to each of the three blocks. The size of each plot was 192 m^2 (40 m × 4.8 m, CU treatment) or 48m^2 (20 m × 2.4 m, 0N, and NU treatment) in 2018, and 81 m^2 (15 m × 5.4 m) or 124 m^2 (23 m × 4.8 m) in 2019 and 2020.

In TPR, details are presented in Yabiku et al. (2021). Briefly, the same cultivars were grown under three different N doses: 0, 8, and 18 g m^{-2}, denoted as 0N, 8N, and

18N, respectively. Among the three N treatments, 8N was considered a standard level for TPR, comparable to the CU treatment for DDSR. The N treatments and cultivars were laid out in a split-block design for three years with three replicated each year.

Cultivation details

In DDSR, the dry soil was plowed, power-harrowed, and laser-leveled in April. Basal fertilizer was applied approximately three days before sowing. After basal fertilizer application, the soil was tilled with a vertical harrow tiller and was compacted using a Cambridge roller. Before sowing, seeds were coated with a thiram-based repellent (Kihigen R-2 flowable, Yonezawa Chemical Co., Ltd., Kyoto, Japan). We sowed the dried seeds to a depth of approximately 2 cm using the high-speed seeder (Clean seeder; NTP-6AFP and NTP-8AFP, Agritechno Search Co. Ltd, Hyogo, Japan) at a row spacing of 0.3 m on 27 April (2018), 22 April (2019), and 17 April (2020). Immediately after seeding, the lands were suppressed by land roller for good seedling establishment and impermeability (Kanmuri et al., 2017). The sowing rate was 5.1 g m^{-2} (2018), 5.7 g m^{-2} (2019), and 5.7 g m^{-2} (2020). We applied flush irrigation about once in three days to prevent the lands from drying until seedling establishment. From early June to approximately three weeks before harvest (late September), the field was kept flooded with approximately 3–10 cm ponding water.

Before permanent flooding, we applied glyphosate-potassium at a rate of 4.8 mL m^{-2} before emergence (mid-May) and cyhalofop-butyl (0.3 mL m^{-2}), and bentazone (2 ml m^{-2}) after emergence (early June), using a boom sprayer. After flooding (late June), we applied a compound herbicide containing pyriftalid, mesotrione, and metazosulfuron

at a rate of 90 mg m^{-2}, 240 mg m^{-2}, and 80 mg m^{-2}, respectively, using a backpack spreader.

For TPR, the dry soil was plowed in mid-April, and basal fertilizers were broadcast on the soil surface and incorporated into the soil with the rotary tractor during early May. During mid-May, the field was submerged and puddled with a power harrow system. Seedlings were raised in a greenhouse and at about five-leaf ages, seedlings were mechanically transplanted on May 17, 21, and 19 in 2018, 2019, and 2020, respectively with a spacing of 30 × 19 cm. The field was kept flooded from May until the end of August, after which the surface water was withdrawn for harvest in late September.

Plant measurements, sampling, and tissue N determination

Leaf age was measured on the same three plants in each replicate at one-week intervals from emergence to flag leaf development. At the same time, tillers were counted on 0.3 m^2 (one 1 m row). From approximately 45 DAS until harvest, we sampled aboveground plant organs weekly or bi-weekly, washed, oven-dried them at 80 °C for at least 72 hours, and weighed them to determine the aboveground biomass. The sampled area at each harvest was 0.3 m^2 (one 1 m row). Part of the dried samples was ground, and tissue N concentrations were measured using an elemental analyzer (Variomax, Elementar, Germany).

At harvest, plants were collected from an area of 2.4 m^2 in each plot for yield determination. We air-dried the samples, weighed the total mass, threshed and dehulled the grains, and screened them through a 1.85 mm-thick sieve. Straw and grain moisture

contents were also measured. Brown rice yield was expressed on the measured sample for yield determination. The water content of brown rice samples was corrected to 15%.

Agronomic NUE (ANUE) in the fertilized plots was calculated as:

$$ANUE(g\ g-N^{-1}) = \frac{Y_F - Y_0}{N_F} \quad (1)$$

where Y_F and Y_0 are brown rice yields from the fertilized and 0N plots, respectively, and N_F is the amount of N fertilizer applied.

Aboveground crop N uptake (N_{up}) was calculated by multiplying the biomass and tissue N concentrations. The apparent N recovery rate (ANR) in the plots was defined as:

$$ANR\ (\%) = \frac{N_{up_F} - N_{up_0}}{N_F} \times 100 \quad (2)$$

where N_{up_F} and N_{up_0} are N_{up} in the fertilized plot and 0N plot, respectively.

We also derived crop NUE (CNUE), which was defined as yield per unit of N uptake at maturity and partial CNUE fertilization (CNUEF) as:

$$CNUE(g\ g^{-1}) = \frac{Y}{N_{MT}} \quad (3)$$

$$CNUEF(g\ g^{-1}) = \frac{Y_F - Y_0}{N_{MT_F} - N_{MT_0}} \quad (4)$$

Analysis of N uptake

N uptake is closely related to temperature conditions (Shimono et al., 2012) and is known to be approximated by the function of cumulative temperature. In transplanted rice, Takahashi et al. (1976) showed that the accumulated effective thermal index

(AETI) could reproduce well the N uptake pattern under different conditions (cropping seasons or years), which is defined as

$$AETI = \sum_{i=1}^{N} \theta_i * \qquad (5)$$

where $\theta_i *$ is the daily effective thermal index, which is the product of the hourly air temperature and *effective air temperature coefficient* (Table S2), averaged for the i-th day (Hanyu and Uchijima 1962), and N is the number of days from start to end of the calculation. In this study, we set i=1 at the 5th leaf age, and the exact date when leaf age becomes 5.0 was derived from the linear interpolation of the weakly observations. The hourly temperatures (T_j) were approximated using the following cosine curve (Sun et al., 2018):

$$Tj = \frac{Tmax+Tmin}{2} + \frac{Tmax-Tmin}{2}\cos\left(\frac{j-14}{12}\pi\right) \qquad (6)$$

where T_{max} and T_{min} are the daily maximum and minimum air temperature, respectively, j is the time of day, and T_j is the hourly air temperature at time j.

Previous studies showed that the N uptake has two distinctive phases (Shoji et al., 1976; Takahashi et al., 1976). In phase 1, N_{up} exponentially increases, limited by the crop growth rate, and can be expressed as an exponential function of AETI*:*

$$N_{up} = N_{LA5} \exp(RNR \times AETI) \qquad (7)$$

where N_{LA5} is N_{up} when leaf age is 5.0 (LA5), and *RNR* is the relative N_{up} rate. In phase 2, N_{up} increases linearly, limited by soil N supply, and can be expressed as:

$$N_{up} = N_{up_b} + \alpha\ (AETI - AETI_b) \qquad (8)$$

where N_{up_b} is N_{up} at the breakpoint from phase 1 to phase 2, which occurs at $AETI_b$, and α is the N_{up} relative to AETI.

Accordingly, N_{up} for the whole growing season can be characterized by four parameters: N_{LA5}, RNR, $AETI_b$ (or N_{up_b}), and α. To derive these parameters, we first determined the duration of the exponential phase (1), by checking the linearity of the log-transformed N_{up}. We then regressed log (N_{up}) on AETI to obtain N_{LA5} and RNR, using data where the linearity is assured (AETI < 600 °C d). The rest of the data was used to estimate α by fixing N_{up_b} at $AETI_b$ = 600 °C d using equation 5. All parameters were calculated in each replicate in 2018 and 2019 and provided for the statistical analysis (next section). We calculated the root mean square error (RMSE) between observed, and predicted N uptake as:

$$RMSE = \sqrt{\frac{1}{n}\sum_{i=1}^{n}(yi - \hat{y}i)^2} \quad (9)$$

where yi and ŷi are observed and predicted N uptake on the i-th DAS, respectively, and n is the number of observations.

Statistical analysis

Statistical analyses were performed using JMP12.0.1 (SAS Institute, 2015). *P* values < 0.05 were considered significant. We conducted an analysis of variance (ANOVA) for yield components and N uptake parameters using a split-plot analysis, where the year was treated as the main plot, and cultivar and N treatment as the subplots. The model included three fixed effect variables (Year (Y), cultivar (C), and N treatments (N)) and one random effect variable (block) with the interactions (Y*C, C*N, Y*N, Y*C*N). Tukey's HSD test was performed on some effects of ANOVA, with a significant level of P<0.05.

Yield and yield components are related to plant N uptake. We computed the analysis of covariance (ANCOVA) for brown rice yield and each yield component as

objective variables, N uptake at maturity stage (N_{MT}) as explanatory variables, and planting method (DDSR or TPR, dummy variables) as covariance. 54 plots of each experiment on DDSR and TPR were used on ANCOVA.

Although N uptake before the heading stage is a critical determinant for spikelet density (Hasegawa et al., 1994; Sasaki, 2007), the N regimes may alter the relative contribution of N uptake at different stages. We, therefore, applied multiple regression analysis of spikelet density as a function of N uptake at panicle initiation (N_{PI}) and N uptake from then to heading ($N_{PI\text{-}HD}$). The interaction between *Year* and *Numerical parameters* was not considered. In the same way, a multiple regression was performed with spikelet density as an objective variable, N_{PI}, N uptake at the heading (N_{HD}), or the balance of N uptake at the heading stage from N uptake at the panicle initiation stage ($N_{PI\text{-}HD}$), and additional categorical variables (*Planting method* (DDSR or TPR), *Cultivar* ('Akitakomachi' or 'Yumiazusa'), and *Year* (2018, 2019, or 2020)).

The standard least-squares method was used as a fitting methodology. The adjusted coefficient of determination (R^{*2}), corrected Akaike's Information Criterion (AICc), and Bayesian Information Criterion (BIC) were obtained to compare the two models. The standardized partial regression coefficient (β) was also computed to determine which parameter has the higher impact on N_{PI} in each regression parameter of the selected model.

We further tested how N_{up} -related parameters influence N_{PI} using regression analysis, and the sensitivity of N_{PI} to changes in the N-related parameters using the regression results by changing *RNR* and N_{LA5} values from the 25th percentile, median to 75th percentile of all experimental data.

Results

Crop development of DDSR

Emergence occurred at 18–21 DAS, and plants reached the second leaf stage (LA2) at 21–27 DAS in all DDSR experiments (Table S3). Dates of PI and heading were similar across different environments and cultivars, ranging from 91–96 DAS and 114–117 DAS, respectively. The period from heading to maturity tended to be longer for YA than for AK. We reported the results of tiller number and biomass growth in the supplemental materials (Figure S3 and S4).

Leaf age growth was impacted by the N regimes from the early stages (approximately 50 DAS; Figure S1). Days from sowing to LA5 were significantly longer in 0N and NU than in CU by as much as 10 days (Table S3). The difference in leaf age between 0N and CU persisted until the final leaf development, resulting in 0.8–1.4 fewer total leaves on the main culm in 0N than in CU (Figure S2), of which approximately 65% were accounted for by the difference at the seedling stage (45 DAS in 2018 and 49 DAS in 2019). Leaf age increase in the NU treatment was initially slow but recovered from 50 DAS, narrowing the gap with CU to approximately 0.5 DAS at the end of the experiment.

Yield and yield components

DDSR brown rice yield under CU conditions was 388 g m^{-2} averaged for the three years and two cultivars, 89% of the average yield of transplanted rice of the same varieties (Table 1). DDSR brown rice yield differed significantly among years, with the 2019 crop showing a considerably lower yield than the 2018 and 2020 crops (P = 0.0215, Table 1). The effects of N treatments were different over three years (P = 0.0581 for Y

× N interaction, Table 1). In the 0N plot, a large year-to-year variation is shown for the brown rice yield, greater than that observed for the two fertilized plots (Table 1). The two fertilized plots (CU and NU) were not significantly different yields on Tukey's HSD test; however, the NU/CU ratio tended to be lower than 1.0. Of the two cultivars, YA had an 8% greater yield than AK, averaged over treatments and years (P = 0.0041), although the cultivar effect was not consistent among years (P = 0.0030 for Y * C interaction, Table 1). The yield advantage of YA over AK was apparent in relatively high-yielding years (16% in 2018 and 12% in 2020, Table 1), while AK had a higher yield in 2019 averaged over the three N regimes. [Table 1 near here]

Panicle density and spikelet density were comparable between CU and NU (Table 1), although leaf age (Figure S1) and tiller number (Figure S3) progress differed between the two fertilized plots. The effects of different N regimes on panicle and spikelet density were a smaller NU/CU ratio in 2018 than in the other two years. Conversely, the NU/CU ratio for 91% of ripened spikelets and single grain mass was greater in 2018, thus compensating for the limited sink size under NU conditions. The N regime significantly affected single grain mass (P = 0.0016), NU having the heaviest mass, followed by CU and 0N.

The yield advantage of YA over AK was associated mainly with the greater panicle density (8%) averaged over years and N regimes. YA had slightly fewer spikelets per panicle and ripened grain percentages than AK, but these differences did not offset the yield advantage of YA. The cultivar differences in these yield components were generally consistent across different N regimes and years.

Relationship between crop N uptake at maturity and yield components

We examined the relationships between crop N uptake at maturity (N_{MT}) and yield components and compared them between DDSR and TPR to determine whether crop N

utilization for the yield formation process differs between planting methods (Figure 1). Brown rice yield increased almost linearly with N_{MT}, but the regression slope and intercept were significantly greater for TPR than DDSR, confirming that DDSR had a lower CNUE than TPR (Figure 1a). Similarly, spikelet density had a strong positive relationship with N_{MT} in both TPR and DDSR, while spikelet density tended to be lower in DDSR than in TPR for the same N_{MT} (Figure 1b). Grain filling (%) decreased with N_{MT} in TPR, while the grain filling in DDSR was not correlated with N_{MT} and was consistently lower than that in TPR (Figure 1c). The 1000-grain weight was not modeled by N_{MT}, planting method, and their interaction ($R^2 = 0.2982$; Figure 1d).

[Figure 1 near here]

N use efficiency and crop N uptake at key growth stages

N uptake of DDSR at the maturity stage was 7.9 g N m^{-2} in 0N, 11.8 g N m^{-2} in NU, and 11.2 g N m^{-2} in CU averaged over three years and two cultivars ($P < 0.0001$, Table 2), which were similar to or even greater than crop N uptake observed in TPR plants undergoing comparable N treatments (Figure 2). Between the two fertilized plots, N uptake in NU tended to be higher than in CU in 2019, while it was similar in 2018 (Table 2), resulting in no significant differences in N regimes by the Tukey HSD test. Furthermore, there were no significant differences between cultivars.

The ANR was significantly different between years ($P = 0.0436$; Table 2), with lower values in 2018 than in the other two years. The ANR was similar between the two DDSR fertilized plots ($P = 0.328$), averaging 26% in CU and 29% in NU for the three years, 76–85% relative to the 8N conditions of TPR (Table 2). Conversely, ANUE was significantly lower in the NU than in the CU ($P = 0.0030$; 26% decrease) in both cultivars. The DDSR/TPR ratio for ANUE was 0.7 when averaged for NU and CU

(Table 2). ANUE was also significantly different among years (P = 0.0430) and similar to ANR, it was lower in 2018. CNUE was significantly different between N treatment (P = 0.0012; Table 2). CNUE of the CU treatment was significantly higher than that of the 0N and NU treatments (Tukey's HSD test). CNUEF, the criteria of NUE that excluded soil N supply, showed an interaction with the year, cultivar, and N treatment variables. However, the CNUEF of the CU was consistently higher than that of the NU. [Table 2 near here]

N_{HD} was also similar between DDSR and TPR and between cultivars at each comparable N level (Figure 2). However, a marked difference was observed in the in-season crop N before HD, where DDSR had a much lower N uptake by PI at both 0N and fertilized plots than TPR (Figure 2). This resulted in a greater N uptake between PI and HD01 in the DDSR than in TPR plants. In the two DDSR fertilized plots, CU enhanced N uptake by PI more than NU, while NU overtook CU before HD (Figure 2). [Figure 2 near here]

Analysis of the relationships between N uptake and spikelet density

Spikelet density was also significantly correlated with N_{PI} and N_{HD}, and it was observed that the regression lines were different between TPR and DDSR at both stages (Figure 3, significantly different intercepts; P = 0.002). However, there was a clear difference between the two stages, where the regression line for DDSR was higher than that for TPR at PI, and it was reversed at HD (Figures 3a, b). [Figure 3 near here]

The difference in the spikelet-N relationship between planting methods and between growth stages could be related to the different crop N uptake patterns, as shown in Figure 4. We, therefore, applied a multiple regression analysis to examine the relative contribution of N uptake on spikelet density at different growth stages (Table

3). We found that both N_{PI} and $N_{PI\text{-}HD}$ significantly contributed to spikelet density, while the *Planting method* (DDSR or TPR), *Year* (2018, 2019, or 2020), or *Cultivar* (‘Akitakomachi’ or ‘Yumiazusa’) had no significant effects. A simplified model without these categorical variables (Model 2) demonstrated that the coefficient for N_{PI} was almost twice as large as that of $N_{PI\text{-}HD}$. The differences in crop N uptake pattern and sensitivity of spikelet density to N uptake at different growth stages accounted for much of the variation in spikelet density due to N application method, *Planting method*, *Year*, and *Cultivar*. [Table 3 near here]

Crop N dynamics and model parameters

Crop N uptake increased almost exponentially until 100 DAS and linearly until maturity, with noted differences among environments (Figure 4 and S5). When crop N was plotted against AETI (Equations 5 and 6), the differences narrowed (Figure S5), and the model combining the exponential and linear functions of AETI explained the crop N pattern better than the model with DAS (Table S4), thus confirming that crop N uotake patterns for the whole growing season can be well represented by the model parameters (N_{LA5}, *RNR*, and α). [Figure 4 near here]

N_{LA5} varied greatly among years showing considerably smaller values in 2019 (Table 4). N regimes significantly affected N_{LA5} in all experiments (P = 0.0003; Table 4), whereas CU was generally associated with greater N_{LA5} than 0N or NU. *RNR* also differed considerably among experiments with greater values observed in 2019 than in 2018, a trend opposite to N_{LA5} (Table 4). *RNR* was smaller in 0N and similar between CU and NU. Cultivars did not have a significant effect on N_{LA5} or *RNR.* YA had higher α than AK while exhibiting similar N_{LA5} and *RNR* (Table 4). The α parameter was not

significantly different for all interactions. The N application either NU or CU increased α (P = 0.0007).

Multiple regression model for N_{PI} estimation (Table 5) used *RNR*, N_{LA5}, and *Year* as parameters, which were selected for N_{PI} estimation. In the model including the *Year* parameter (categorical valuable, 2018, 2019), R^{*2} (0.83) was higher, besides AICc and BIC (75 and 81) being lower than those of the model that excludes the environment parameter (0.79, 80, and 85, respectively). The *Year* had a significant effect (P = 0.0099). The standardized partial regression coefficients were 1.92 (N_{LA5}) and 1.23 (*RNR*) in the model excluding the *Year* effect (Table 5). [Tables 4 and 5 near here]

N_{PI} estimation was performed using the model that excluded the *Year* effect and used the median, 75th percentile, and 25th percentile of *RNR* and N_{LA5} (Table 6). The estimated N_{PI} by the selected model increased as *RNR* and N_{LA5} increased. If *RNR* or N_{LA5} was changed from the median to the 75th percentile of this study, N_{PI} may increase by 2.5 g N m^{-2}, whereas the change of both *RNR* and N_{LA5} from the median to the 75th percentile may increase the N_{PI} by 4.9 g N m^{-2}. [Table 6 near here]

Discussion

Our three-year field experiment confirmed that DDSR had a lower yield than TPR by an average of 11%, similar to that reported by Shinoto et al., (2021). Furthermore, the ANUE was lower in the DDSR than in the TPR by 30%, which is in line with the observations that DDSR requires a greater amount of fertilizer to be productive (Otani et al., 2013). In this section, we first explore possible reasons for the low ANUE in DDSR compared to TPR. Then we discuss how N regimes and cultivars affect production efficiency.

Why is the ANUE of DDSR low?

ANUE can be considered a product of fertilizer N recovery and the efficiency of converting crop N into grain yield. We predicted that the former, represented by ANR, was largely responsible for the lower ANUE in DDSR relative to TPR as a large fertilizer loss was expected for the DDSR, particularly in an Andosol with a high percolation rate (Kanmuri et al., 2017). Our analysis, however, indicated that both ANR and CNUE were lower in the DDSR than those in the TPR, both contributing to the low ANUE of DDSR.

The unexpectedly low CNUE of DDSR was mainly induced by two factors. One was the unfavorable climatic conditions for DDSR during the grain filling period compared with TPR; thus, limiting source capacity for grain growth. DDSR heading occurred approximately 10–17 day later than that of TPR, resulting in lower solar radiation and temperatures during the grain-filling period for DDSR plants. This is reflected in the poor grain filling (%) and single grain mass at all N levels (Figure 1). Therefore, optimum growing seasons for DDSR need to be determined, accounting for cultivar phenological properties and site-specific climatic conditions.

Secondly, spikelet production per unit of N uptake at heading or maturity was limited in the DDSR relative to the TPR. As shown in Figure 2, N_{HD} or N_{MT} were comparable between DDSR and TPR, but the N uptake pattern was considerably different. While TPR took up approximately 17–20% of the total crop N before the PI stage, DDSR only approximately 10% at PI while accelerating the N uptake toward heading and ultimately surpassing the N uptake observed in the TPR at the heading stage. The multiple regression analysis of spikelet density on N_{PI} and $N_{PI\text{-}HD}$ revealed that the contribution of N_{PI} was twice as large as that of $N_{PI\text{-}HD}$ (Table 3), suggesting that DDSR having a smaller N_{PI} and larger $N_{PI\text{-}HD}$ leads to low spikelet production efficiency

per unit N uptake. These results indicate that enhancing N_{PI} might be effective in increasing CNUE and ANUE.

Can the split application of urea be an alternative to control-release fertilizer in terms of yield and NUE?

The NU split application can be an alternative to the CU application because no significant differences in yield were observed between the two fertilized plots. The NU/CU ratio ranged between 0.93 and 1.0 since a greater amount of NU application than CU application rate may be able to the same yield of DDSR maintaining cheaper production cost. The fertilizer price of NU was approximately three times cheaper than that of CU per N unit.

The yields of DDSR in a cool climate can be further improved. The ANR observed in this study was generally lower for DDSR than TPR (Table 2), which highlights the need for improvements regarding N application methods for DDSR. No significant evidence was observed that the CU treatment had a higher ANR than that of the NU treatment. Alternatively, the ANUE associated with the NU treatment was significantly lower than that associated with the CU treatment (Table 2). This indicates that the lower yield of the NU treatment might have been caused by the imperfect application regimes rather than differences in recovery. Therefore, we can increase the yield of the NU treatment by analyzing the optimal N proportion.

We decide the first N application time of the NU treatment to be done just before flooding referencing the southern US, the area in which dry-seeded, delayed-flooded systems are commonly used (Linquist et al., 2013; Singh et al., 2017). However, the application of normal urea before flooding, such as commonly practiced in the southern US, may not be suitable for increasing N_{LA5} in a cool climate region. The

first N application time during the NU treatment was approximately 8 days before arriving at the LA5 in Morioka. At this time, it may be too late for initial N application because rice seedlings become autotrophic at the 3–4 leaf stage (Fageria, 2007). The difference in N_{LA5} among the years may suggest that the difference in soil N mineralization by the previous crop also affects N_{LA5}. The soil N mineralization has been shown to increase by drying the soil (Kautsar et al., 2020; Toriyama et al., 1988); hence, the rice seedlings in 2018 can increase N uptake by previous dryland cropping (soybean) compared to previously flooded cropping (irrigated rice) in 2019. However, there is no data on available soil N under the same condition in each experiment.

Does a high-yielding cultivar under transplanting practice retain its yield advantage over a standard cultivar under dry direct-seeding practice?

YA (high-yielding cultivar) retains its yield advantage under dry direct-seeding practice. The YA brown rice yield was significantly higher than that of the AK (Table 1). Model 1 for spikelet determination showed weak significant differences between AK and YA (Table 3, P = 0.0686). YA may exhibit slightly higher NUE than AK. Yabiku et al. (2020) reported that the radiation use efficiency of YA was not significantly higher than AK following TPR. This indicates that although the YA might have higher NUE, it exhibits the same radiation use efficiency as the conventional cultivar. YA had comparatively higher panicles and 1000-grain weight but significantly lower grain filling than AK; thus, causing a lower YA yield than AK yield in 2019. Furthermore, the DDSR in a cool climate may not provide YA with the appropriate conditions to express its high-yielding abilities as the cropping environment is restricted by solar radiation. Conversely, there were no significant differences in the cultivar effect on N_{LA5} and *RNR* (Table 2). YA did not show a higher N uptake ability than that AK.

How can we improve N_{PI}?

N uptake before the PI stage is critical for yield determination. From equation 7 (Model 2), the influence of N_{PI} on spikelet density was approximately two times higher than that of $N_{HD\text{-}PI}$. Furthermore, figure 3 indicates that N_{PI} following DDSR affects spikelet density more than it does following TPR; thus, increasing N_{PI} is more important for DDSR than that for TPR. Alternatively, Model 1 showed that the N_{PI} and $N_{HD\text{-}PI}$ relationship was not significantly influenced by the planting method (DDSR or TPR), thus allowing us to show the universality of the relationship between spikelet density and N uptake for rice grown using different planting methods. Additionally, the parameters of the N uptake pattern significantly influenced the spikelet density with N_{LA5} and *RNR* exhibiting higher impacts (Table 5 and 6), thus suggesting that N uptake during the early stage, even N uptake at LA5, affects the yield.

To generally analyze the temporal N uptake pattern in rice, the N uptake was investigated corresponding to temperature. Shoji et al. (1976) and Takahashi et al. (1976) divided the N uptake of rice into phase 1 (exponentially increase phase) and phase 2 (linearly increase phase) corresponding to air temperature. In phase 1, N uptake is limited by the growth rate of the rice plant, while in phase 2, N uptake is limited by soil N supply and N mineralization (Toriyama, 1990). N uptake is explained by temperature in phase 2, hence N uptake is limited by soil N supply, rather than the plant's ability. The phase-change point in N uptake on transplanted rice was reported at approximately 400 °C d on AETI (from transplanted), at the commencement of ear-primordia formation in field experiments on transplanted rice (Shoji et al., 1976; Takahashi et al., 1976). This borderline (the ear-primordia formation stage) is in agreement with the vegetative-reproductive phase change in rice phenology (Fageria, 2007), at least on transplanted rice. Furthermore, we must consider phase 0: before

seedling establishment because those studies of transplanted rice do not consider it before transplanting. The seeding stage may play an important role in DDSR rice growth in a cool temperate zone.

The N application before LA5 plays an important role in leaf development and contributes to biomass and yield production. We discovered that different N applications affected leaf development already at the seedling stage (LA5; Table S5), and these differences account for more than half of the differences in the final leaf number (Figure S2). Additionally, the N application for increasing booting from 7 to 11 leaf ages may increase the yield as the productive culm number increases at these stages (Table S6), which might be related to the spikelet density (Table 3). The delay in leaf development decreases the opportunity of booting, causing lower yield.

However, it is worth investigating if it is possible to increase the N_{LA5}. Sheehy et al. (1998) suggested that the critical N concentration of rice plants in the vegetative stage was generally similar in tropical and temperate environments. According to their observations, the N_{LA5} of the plants used in this study had room to reach the critical N content rate at a rate of biomass. This indicates that theoretically, we could increase N_{LA5} through N management. To increase the N_{PI} of DDSR plants to the same levels observed in TPR plants, it would be required to increase the N application by 0.5 g N m^{-2} (Figure 2). From model 4 presented in Table 5, the 0.05 g N m^{-2} N_{LA5} needs to increase by N_{PI} with 0.5 g N m^{-2} with a constant *RNR*. Such values appear achievable for DDSR high-yielding rice based on aboveground biomass data.

The cultivar and N treatments had significant effects on α, hence α was affected by plant N availability and N fertilizer application. However, topdressing (or later CU release) affects these results. The cultivar differences appeared on α, while N_{LA5} and *RNR* were not different as a function of the N treatment. In phase 2, there is no

sufficient available N per area, thus the cultivars that have large or effective root systems may get more N. Furthermore, considering the year effect, the fields with different soil types or soil fertility may have different α. Although the differences in α are suggestive, the suppression of α is likely to be more important rather than the increase of α. In Japan, N uptake after the meiosis stage is not preferred because that makes brown rice reach a high protein content and Japanese consumers prefer the taste of low protein rice. To control the N uptake and increase the yield, N_{LA5} and *RNR* are more appropriate to be manipulated. Nevertheless, α determination is important to estimate the final N uptake.

Regarding the DDSR in northern Japan, the phase change point of N uptake may be slightly different from the phenology phase change point. The turning point in the N uptake phase of rice was reported at approximately 400 °C d on AETI (from transplanted), at the commencement of ear-primordia formation in field experiments (Shoji et al., 1976; Takahashi et al., 1976). In this study, the phase change point was defined at 600 °C d. Although it may not align with the above-mentioned study due to the different AETI beginning points (from transplanted or LA5), 600 °C d in this study (2018 and 2019) is the later date for PI (Table S3). The phase change points agree with the offset point of soil ammonium N in transplanted rice (Shoji et al., 1976; Takahashi et al., 1976). This means the phase change point of phenology and N uptake is similar in transplanted rice, however, different when the DDSR method is used in northern Japan.

Conclusions

The NU treatment presented in this study can be an alternative to the more expensive CU treatment. However, the first N application during the NU treatment should be performed earlier as N_{LA5} was shown to affect spikelet number by promoting the

increase in N_{PI}. A small amount of N should be applied earlier to save ammonia volatilization and further N losses in the split application method (NU). YA (high-yielding cultivar) retains its yield advantage under dry direct-seeding practice. YA seems to have a higher NUE, but the same radiation use efficiency as the conventional cultivar. The DDSR method in a cool climate may not provide adequate conditions for YA to express its high-yielding abilities as the cropping environment is restricted by solar radiation. This study was able to show the universality of the relationship between spikelet density and rice N uptake using different planting methods. Figure 3 shows that for DDSR, N_{PI} effects spikelet density more than it does for TPR; thus, increasing the N_{PI} is more important for the DDSR than for the TPR method. The parameters characterizing the N uptake pattern significantly influence the yield (especially N_{LA5} and *RNR*). These results suggest that N uptake during the early stage affects the yield. Lastly, the differences in leaf ages at the seedling stage between the CU and 0N treatments explained 65% of the final differences, suggesting the importance of the initial stage.

Disclosure statement

No potential conflict of interest was reported by the authors.

Acknowledgements

We thank Hisaya Tanaka, Hiroshi Kawamukai, Eisaku Kumagai, Fumihiko Saito, and Kafumi Segawa for their help with in-field management and sampling. Professor Hiroyuki Shimono of Iwate University provided valuable suggestions. The colleagues of NARO Tohoku Agricultural Research Center and the United Graduate School of Agricultural Sciences-Iwate University supported our study.

Funding

This work was supported by the commissioned project (high-technology development project; “Daikibo suiden einou”) of the Ministry of Agriculture, Forestry and Fisheries, Japan.

References

Ali, A. M., Thind, H. S., Varinderpal-Singh, & Bijay-Singh. (2015). A framework for refining nitrogen management in dry direct-seeded rice using GreenSeeker™ optical sensor. *Computers and Electronics in Agriculture*, *110*, 114–120. https://doi.org/10.1016/j.compag.2014.10.021

Fageria, N. K. (2007). Yield Physiology of Rice. *Journal of Plant Nutrition*, *30*(6), 843–879. https://doi.org/10.1080/15226510701374831

Farooq, M., Siddique, K. H. M., Rehman, H., Aziz, T., Lee, D.-J., & Wahid, A. (2011). Rice direct seeding: Experiences, challenges and opportunities. *Soil and Tillage Research*, *111*(2), 87–98. https://doi.org/10.1016/j.still.2010.10.008

Griggs, B. R., Norman, R. J., Wilson, C. E., & Slaton, N. A. (2007). Ammonia Volatilization and Nitrogen Uptake for Conventional and Conservation Tilled Dry-Seeded, Delayed-Flood Rice. *Soil Science Society of America Journal*, *71*(3), 745–751. https://doi.org/10.2136/sssaj2006.0180

Hanyu, J., & Uchijima, T. (1962). Studies on the Relation of the Weather Conditions to the Growth of Crops. *Journal of Agricultural Meteorology*, *18*(3), 109–117. https://doi.org/10.2480/agrmet.18.109

Hasegawa, T., Koroda, Y., Seligman, N. G., & Horie, T. (1994). Response of Spikelet Number to Plant Nitrogen Concentration and Dry Weight in Paddy Rice. *Agronomy Journal*, *86*(4), 673–676. https://doi.org/10.2134/agronj1994.00021962008600040016x

Horai, K., Ishii, A., Mae, T., & Shimono, H. (2013). Effects of early planting on growth and yield of rice cultivars under a cool climate. *Field Crops Research*, *144*, 11–18. https://doi.org/10.1016/j.fcr.2012.12.016

Kanmuri, H., Sekiya, H., & Otani, R. (2017). Applicability of Dry Direct-seeding Rice Cultivation using Plowing and Compaction Based on Soil Conditions of Paddy Rice Fields. *Japanese Journal of Farm Work Research*, *52*(2), 63–75. https://doi.org/10.4035/jsfwr.52.63

Katsumi, N., Kusube, T., Nagao, S., & Okochi, H. (2020). The role of coated fertilizer used in paddy fields as a source of microplastics in the marine environment. *Marine Pollution Bulletin*, *161*, 111727. https://doi.org/10.1016/j.marpolbul.2020.111727

Kautsar, V., Cheng, W., Tawaraya, K., Hattori, S., Yamada, S., Kobayashi, K., & Toriyama, K. (2020). Air-drying and drying-rewetting effects in Japanese Andosols subjected to long-term organic rice farming. *Soil Science and Plant Nutrition*, *66*(5), 714–723. https://doi.org/10.1080/00380768.2020.1816123

Linquist, B. A., Liu, L., van Kessel, C., & van Groenigen, K. J. (2013). Enhanced efficiency nitrogen fertilizers for rice systems: Meta-analysis of yield and nitrogen uptake. *Field Crops Research*, *154*, 246–254. https://doi.org/10.1016/j.fcr.2013.08.014

Liu, H., Hussain, S., Zheng, M., Peng, S., Huang, J., Cui, K., & Nie, L. (2015). Dry direct-seeded rice as an alternative to transplanted-flooded rice in Central China. *Agronomy for Sustainable Development*, *35*(1), 285–294. https://doi.org/10.1007/s13593-014-0239-0

MAFF, 2018. Report on results of 2015 census of agriculture and forestry in Japan In: Ministry of Agriculture, Forestry and Fisheries, Japan [WWW Document]. URL https://www.e-stat.go.jp/en/stat-search/files?page=1&layout=datalist&toukei=00500209&tstat=000001032920&cycle=0&tclass1=000001077437&tclass2=000001097415&tclass3val=0 (accessed 6.14.22)

MAFF, 2021a. The areas of direct sowing of paddy rice in 2019 (in Japanese) In: Ministry of Agriculture, Forestry and Fisheries, Japan [WWW Document]. URL https://www.maff.go.jp/j/syouan/keikaku/soukatu/chokuha.html (accessed 10.11.21).

MAFF, 2021b. About the current direct-seeding paddy rice (in Japanese) In: Ministry of Agriculture, Forestry and Fisheries, Japan [WWW Document]. URL https://www.maff.go.jp/j/syouan/keikaku/soukatu/chokuha.html (accessed 10.11.21).

Mahajan, G., Chauhan, B. S., & Gill, M. S. (2011). Optimal Nitrogen Fertilization Timing and Rate in Dry Seeded Rice in Northwest India. *Agronomy Journal*, *103*(6), 1676–1682. https://doi.org/10.2134/agronj2011.0184

McDonald, A. J., Riha, S. J., Duxbury, J. M., Steenhuis, T. S., & Lauren, J. G. (2006). Water balance and rice growth responses to direct seeding, deep tillage, and landscape placement: Findings from a valley terrace in Nepal. *Field Crops Research*, *95*(2–3), 367–382. https://doi.org/10.1016/j.fcr.2005.04.006

NARO, T.A.R.C. (2021). Dry direct-seeding cultivation technology manual (ver.3.2) - Plow cultivation and soil compaction system- (in Japanese).

Ohta, H., Yamaguchi, M., Fukushima, A., Yokogami, N., Tsuda., N, Kaji, R., & Nakagomi, K. (2017). "Yumiazusa", a New Rice Cultivar with Blast Resistance, High Yield and High Eating Quality (in Japanese with English abstract). *Bulletin of the NARO, Agricultural Research for Tohoku Region, 120, 31-45.*

Otani, R., Sekiya, H., Kanmuri, H., Nakayama, S., & Saito, H. (2013). Crop rotation system of three-crop per two-year utilizing dry direct-seeding of rice with plowing in large scale paddy field (in Japanese). *Journal of the Japanese Society of Agricultural Machinery*, *75*(4), 220-224.

Qi, X., Nie, L., Liu, H., Peng, S., Shah, F., Huang, J., Cui, K., & Sun, L. (2012). Grain yield and apparent N recovery efficiency of dry direct-seeded rice under different N treatments aimed to reduce soil ammonia volatilization. *Field Crops Research*, *134*, 138–143. https://doi.org/10.1016/j.fcr.2012.05.010

Sasaki, J. (2007). Application to diagnosis technology of the growth information for the production of rice with high grain quality and good eating quality. *Journal of the Japanese Society of Agricultural Machinery*, *69*(5), 12-16.

Sheehy, J. E., Dionora, M. J. A., Mitchell, P. L., Peng, S., Cassman, K. G., Lemaire, G., & Williams, R. L. (1998). Critical nitrogen concentrations: implications for high-yielding rice (Oryza sativa L.) cultivars in the tropics. *Field Crops Research*, *59*(1), 31–41. https://doi.org/10.1016/S0378-4290(98)00105-1

Singh, V., Zhou, S., Ganie, Z., Valverde, B., Avila, L., Marchesan, E., Merotto, A., Zorrilla, G., Burgos, N., & Norsworthy, J. (2017). Rice Production in the Americas. In Bhagirath, S. C., Khawar, J., & Gulshan, M. (Eds.), *Rice Production worldwide* (pp.137-168). Springer Cham.

Shimono, H., Fujimura, S., Nishimura, T., & Hasegawa, T. (2012). Nitrogen Uptake by Rice (Oryza sativa L.) Exposed to Low Water Temperatures at Different Growth

Stages. *Journal of Agronomy and Crop Science*, *198*(2), 145–151. https://doi.org/10.1111/j.1439-037X.2011.00503.x

Shimono, H., Hasegawa, T., Kuwagata, T., & Iwama, K. (2007). Modeling the Effects of Water Temperature on Rice Growth and Yield under a Cool Climate: II. Model Application. *Agronomy Journal*, *99*(5), 1338–1344. https://doi.org/10.2134/agronj2007.0149

Shinoto, Y., Otani, R., & Matsunami, T. (2021). Growth and Yield of Rice in Andosol by Dry Direct Seeding Cultivation using Plowing and Compaction in a Cold Climate Region of Japan (in Japanese with English abstract). *Japanese Journal of Crop Science*, *90*(3), 343–351. https://doi.org/10.1626/jcs.90.343

Shoji, S., Nogi, T., Takahashi, J., & Wada, G. (1976). The Fate of Fertilizer Nitrogen Applied to the Paddy Field and its Absorption by Rice Plant: VIII. Comparative studies of the fate of basal nitrogen and the absorption of nitrogen by rice plant in different paddy fields. *Japanese Journal of Crop Science*, *45*(2), 226–231. https://doi.org/10.1626/jcs.45.226

Sun, T., Hasegawa, T., Tang, L., Wang, W., Zhou, J., Liu, L., Liu, B., Cao, W., & Zhu, Y. (2018). Stage-dependent temperature sensitivity function predicts seed-setting rates under short-term extreme heat stress in rice. *Agricultural and Forest Meteorology*, *256–257*, 196–206. https://doi.org/10.1016/j.agrformet.2018.03.006

Takahashi, J., Wada, G., & Shoji, S. (1976). The Fate of Fertilizer Nitrogen Applied to the Paddy Field and its Absorption by Rice Plant: VII. Relationships between the absorption of nitrogen by the rice plants and the growth of the plants. *Japanese Journal of Crop Science*, *45*(2), 220–225. https://doi.org/10.1626/jcs.45.220

Toriyama, K. (1990). The growth simulation of rice and the soil nitrogen mineralization, in: The Nitrogen Mineralization of Paddy Soil and Application. pp. 125-156 (in Japanese).

Toriyama, K., Sekiya, S., & Miyamori, Y. (1988). Quantification of effect of air-drying of paddy soil before submergence on amount of nitrogen mineralization. *Japanese Journal of Soil Science and Plant Nutrition (Japan)*.

Xu, L., Yuan, S., Wang, X., Chen, Z., Li, X., Cao, J., Wang, F., Huang, J., & Peng, S. (2021). Comparison of yield performance between direct-seeded and transplanted double-season rice using ultrashort-duration varieties in central

China. *The Crop Journal*, 10(2), 515–523. https://doi.org/10.1016/j.cj.2021.07.003

Yabiku, T. Namikawa, M. & Hasegawa, T. (2021). Development of a Simplified Growth and Yield Prediction Model for Supporting the Management of Paddy Rice Fertilization for Commercial Rice Varieties in the Tohoku Region (in Japanese with English abstract). *Japanese Journal of Crop Science*, 90(4), 430-443.

Table 1. Brown rice yield and yield components of dry-direct seeded rice under different nitrogen regimes and cultivars

Planting Method	Cultivar	N regimes	Brown rice yield[1] (g m^{-2})	Panicle density (m^{-2})	Spikelet density (10^3 m^{-2})	Grain Filling (%)	1000-grain weight (g)
DDSR Mean (2018-2020)	AK	0N	260	244	16.2	88.3	21.4
		CU	437	339	24.8	87.3	21.5
		NU	423	350	25.6	86.3	21.8
	YA	0N	301	262	16.6	81.6	21.7
		CU	467	375	27.1	83.1	22.0
		NU	443	370	25.2	81.5	22.2
	NU/CU ratio		0.96	1.01	0.98	0.99	1.01
	YA/AK ratio	0N	1.16	1.07	1.03	0.92	1.01
		CU	1.07	1.11	1.10	0.95	1.03
		NU	1.05	1.06	0.98	0.95	1.02
ANOVA	Year(Y)		0.0215	0.4615	0.1164	0.0319	0.2036
	Cultivar(C)		0.0041	0.0937	0.1104	0.0002	0.0043
	N regimes (N)		<.0001	<.0001	<.0001	0.6829	0.0163
	Y*C		0.003	0.0473	0.0797	0.2060	0.0003
	Y*N		0.0581	0.3189	0.4659	0.0695	0.0880
	C*N		0.7541	0.8453	0.3614	0.6904	0.6232
	Y*C*N		0.1291	0.6211	0.4148	0.8952	0.3340
TPR Mean (2018-2020)	AK	0N	315	251	15.7	94.2	23.1
		8N	483	350	23.2	94.3	22.8
		18N	628	411	33.6	91.6	22.6
	YA	0N	366	257	18.2	94.2	23.1
		8N	536	342	26.0	94.7	22.8
		18N	689	419	34.8	93.2	22.5
DDSR/TPR ratio	AK	0N	0.83	0.97	1.03	0.94	0.93
		CU/8N	0.90	0.97	1.07	0.93	0.94
	YA	0N	0.82	1.02	0.91	0.87	0.94
		CU/8N	0.87	1.10	1.04	0.88	0.97

[1] Grain thicker than 1.85 mm was counted to calculate brown rice yield. Brown rice yield and 1000-grain weight are presented at a water content of 15%.

The individual DDSR data for each year are shown in the supplemental material (Table S6).

DDSR, dry direct seeding rice; TPR, transplanted rice.

AK and YA mean the cultivar's names, 'Akitakomachi' and 'Yumiazusa', respectively.

0N, CU, NU, 8N, and 18N represent the N treatments, no N treatment, coated urea treatment (12 g N m^{-2}), split application of normal urea (15 g N m^{-2}), 8 g N m^{-2}, 18 g N m^{-2} treatment, respectively.

Table 2. Rice plant's nitrogen uptake during the maturity stage and apparent nitrogen recovery rate of dry-direct seeded rice under different nitrogen regimes and cultivars

Planting method	Cultivar	N regimes	N uptake (g m^{-2})	ANR (%)	ANUE (g g-N^{-1})	CNUE (g g-N^{-1})	CNUEF (g g-N^{-1})
DDSR	AK	0N	7.7	-	-	31	-
Mean		CU	10.9	29	14.9	41	51
(2018-2020)		NU	11.9	37	11.8	32	38
	YA	0N	8.1	-	-	31	-
		CU	11.5	31	14.8	41	45
		NU	11.7	32	10.3	32	39
	NU/CU ratio		1.05	1.15	0.74	0.78	0.80
	YA/AK ratio	0N	1.05	-	-	1.0	-
		CU	1.06	1.07	0.99	1.0	0.88
		NU	0.98	0.86	0.87	1.0	1.03
	ANOVA	Year(Y)	0.1972	0.0436	0.0430	0.0130	0.3416
		Cultivar(C)	0.4943	0.6990	0.4710	0.0483	0.1827
		N regimes (N)	<.0001	0.3276	0.0030	0.0012	0.0268
		Y*C	0.4484	0.1090	0.2039	0.0130	0.1864
		Y*N	0.1475	0.1598	0.9024	0.1708	0.8111
		C*N	0.5739	0.4478	0.5625	0.8470	0.0942
		Y*C*N	0.2316	0.4645	0.1391	0.8746	0.0371
TPR	AK	0N	7.1	-	-	44	-
Mean		8N	9.1	34	21.3	53	53
(2018-2020)		18N	13.9	38	17.8	45	46
	YA	0N	7.3	-	-	50	-
		8N	10.0	33	21.5	54	63
		18N	14.0	32	18.1	49	48
DDSR/TPR ratio	AK	0N	1.08	-	-	0.70	-
		CU/8N	1.20	0.85	0.70	0.77	0.96
	YA	0N	1.11	-	-	0.62	-
		CU/8N	1.15	0.94	0.69	0.76	0.71

The individual DDSR data for each year are shown in the supplemental material (Table S7).

DDSR, dry direct seeding cultivation; TPR, transplanted cultivation.

0N, CU, NU, 8N, and 18N represent the N treatments, no N treatment, coated urea treatment (12 g N m-2), split application of normal urea (15 g N m-2), 8 g N m-2, 18 g N m-2 treatment, respectively.

AK and YA mean the cultivar's names, 'Akitakomachi', and 'Yumiazusa', respectively.

Table 3. The effects of nitrogen uptake at different growth stages (PI and HD), planting methods (DDSR and TPR), cultivars (AK and YA), and years (2018, 2019, and 2020) on spikelet density

	Model 1		Model 2	
Explanatory variables	Coefficient	*P*-value	Coefficient	*P*-value
Intercept	7639	<.0001	8005	<.0001
N_{PI}	3268	<.0001	3038	<.0001
$N_{PI\text{-}HD}$	1698	<.0001	1753	<.0001
N_{HD}	-	-	-	-
Planting method (DDSR)	-51	0.9225	-	-
(TPR)	51	-	-	-
Cultivar (AK)	-713	0.0686	-	-
(YA)	713	-	-	-
Year (2018)	-1070	0.1311	-	-
(2019)	-16	-	-	-
(2020)	1086	-	-	-
Fitting criteria				
R^{*2}	0.60		0.58	
RMSE	3665		3746	
AICc	1743		1742	
BIC	1761		1751	

DDSR, dry direct seeding cultivation; TPR, transplanted cultivation.

AK and YA mean the cultivar's names, 'Akitakomachi', and 'Yumiazusa', respectively.

Table 4. Nitrogen uptake when five-leaf age (N_{LA5}), relative N uptake rate (RNR), and N uptake rate constant α of dry-direct seeded rice under different environments, nitrogen regimes, and cultivars

Year	Cultivars	N regimes	N_{LA5} (mg N m^{-2})		*RNR* (×10^{-3} (°C d)$^{-1}$)		α (mg N m^{-2} (□ d)$^{-1}$)	
2018	AK	0N	360	±12	21	±0.097	7.2	±0.16
		CU	400	±29	23	±0.11	9.3	±0.71
		NU	400	±10	23	±0.05	9.1	±0.49
	YA	0N	390	±11	22	±0.12	7.6	±0.38
		CU	480	±10	22	±0.051	9.5	±0.8
		NU	440	±11	21	±0.059	11.7	±0.01
2019	AK	0N	55	±13	57	±0.4	3.9	±0.64
		CU	81	±11	67	±0.056	5.4	±0.72
		NU	63	±6	71	±0.12	5.9	±0.37
	YA	0N	27	±1	53	±0.038	5.4	±0.69
		CU	84	±30	68	±0.41	7.0	±0.27
		NU	52	±11	76	±0.037	6.5	±1.4
ANOVA								
Year(Y)			0.0012		0.0002		0.0148	
Cultivar(C)			0.0627		0.9976		0.0125	
N regimes(N)			0.0003		<.0001		0.0007	
Y*C			0.0033		0.6538		0.8314	
Y*N			0.3836		0.0002		0.4011	
C*N			0.1786		0.5400		0.7357	
Y*C*N			0.8145		0.3371		0.2237	

Means ± standard error is shown (n = 3). AK and YA represent the cultivar's names, 'Akitakomachi', and 'Yumiazusa', respectively. 0N, CU, and NU represent the N treatments, no N treatment, coated urea treatment (12 g N m^{-2}), and split application of normal urea (15 g N m^{-2}), respectively.

Table 5. Multiple regression of N_{PI} on nitrogen uptake parameters with (Model 3) and without the Year terms (Model 4)

	Model 3			Model 4		
Explanatory variables	Coefficient	Standardized coefficient	P-value	Coefficient	Standardized coefficient	P-value
Intercept	-4.82		0.0001	-4.42		0.0001
RNR	508.18	0.82	<.0001	766.12	1.23	<.0001
N_{LA5}	21.494	2.74	<.0001	15.037	1.92	<.0001
Year (2018)	-1.738	-1.23	0.0099			
(2019)	1.738	1.23				
Fitting criteria						
R^{*2}	0.83			0.79		
AICc	75			80		
BIC	81			85		

Table 6. Estimated N uptake during the panicle initiation stage using multiple regression analyses for changes in N and environment parameters

Estimated N uptake at panicle initiation stage (g m^{-2})		*RNR* ((°C d)$^{-1}$)		
		25th percentile	median	75th percentile
N_{LA5}(g N m^{-2})	25th percentile	-1.9	-0.9	1.6
	median	0.9	1.9	4.4
	75th percentile	3.3	4.4	6.8

25th and 75th percentile as well as the median were decided following data analysis.

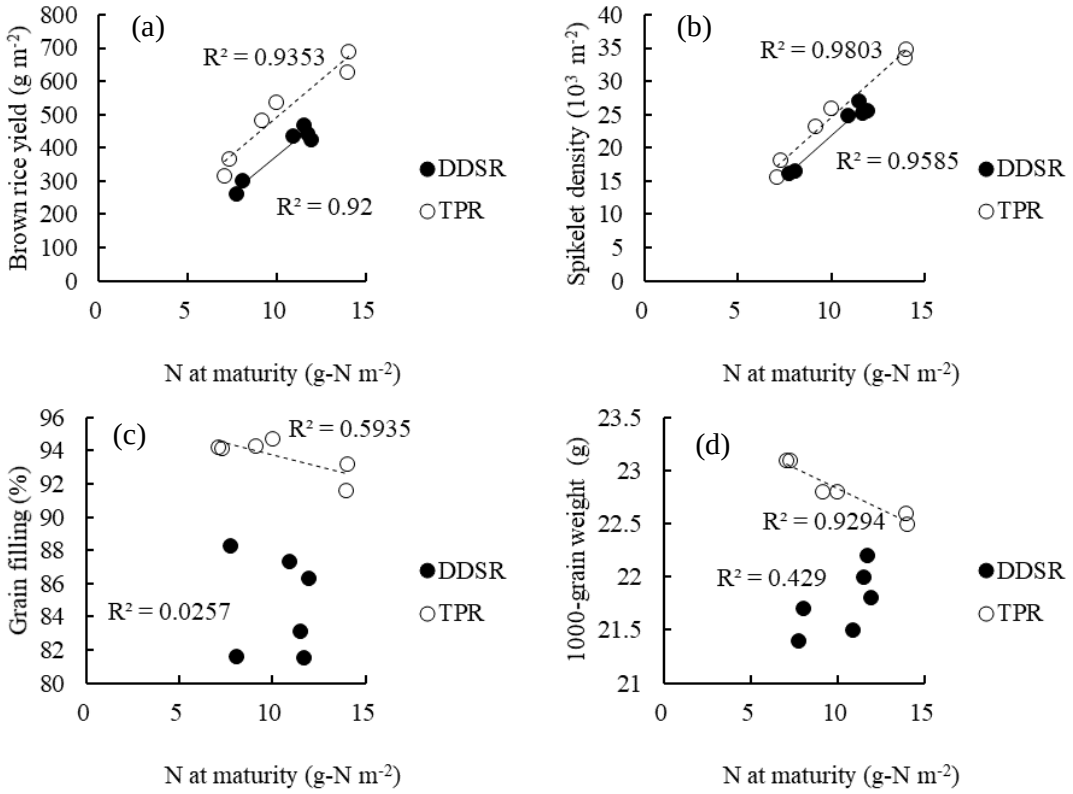

Figure 1. The relationship between N uptake at maturity (N_{MT}) and yield components. From the analysis of covariance, the regression slope and intercept for brown rice yield were different between TPR and DDSR (P = 0.019 for intercept and P < 0.0001 for slope; (a)). The spikelet density relationship with N_{MT} was not different as a function of the planting method (P = 0.8708; (b)). The regression intercept was different for grain filling (%) between TPR and DDSR (P < 0.0001; (c)). The 1000-grain weight was not influenced by N_{MT}, planting method, and that interaction (R^2 = 0.2982, (d)).

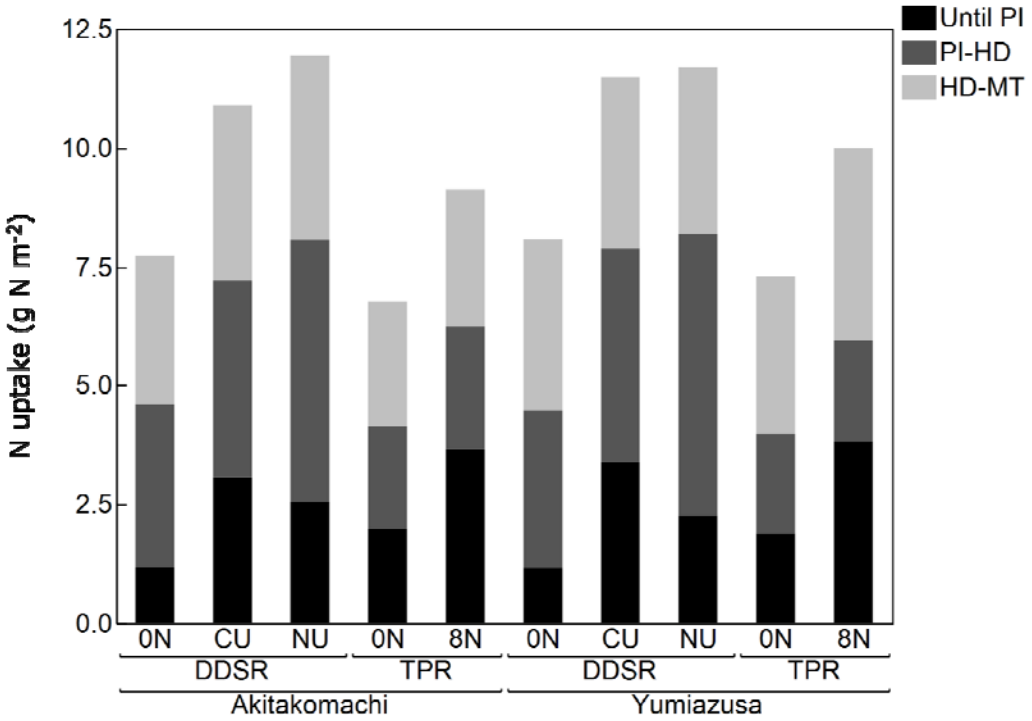

Figure 2. The N uptake for each N treatment, plantation method, and cultivar. 0N, CU, NU, and 8N represent the N treatments, no N treatment, coated urea treatment (12 g N m^{-2}), split application of normal urea treatment (15 g N m^{-2}), and local standard treatment on TPR (8 g N m^{-2}), respectively.

DDSR and TPR represent the dry direct seeding rice and transplanted rice, respectively.

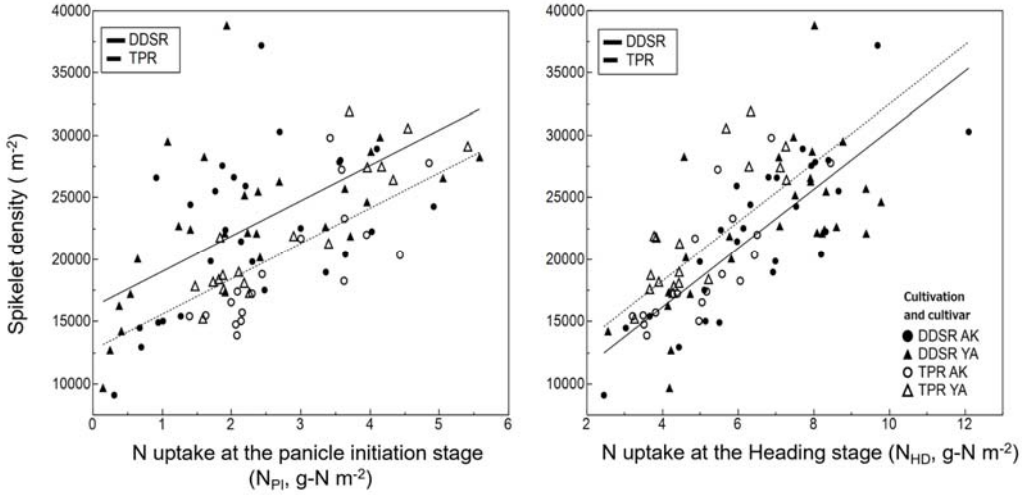

Figure 3. The relationship between spikelet density and N uptake under different planting methods N_{PI} and N_{HD} exhibit different relationships between planting method and spikelet density. The difference relationship in N_{HD} was smaller than that of N_{HD} and reversed. The interaction between N_{HD} and the *Planting method* was not significant.

DDSR and TPR represent the dry direct seeding cultivation and transplanted cultivation, respectively.

AK and YA represent the cultivar's names, 'Akitakomachi', and 'Yumiazusa', respectively.

In our analysis of covariance, the regression lines were different between TPR and DDSR at the panicle initiation stage (left side, $P = 0.0002$) and heading stage (right side, $P < 0.0001$).

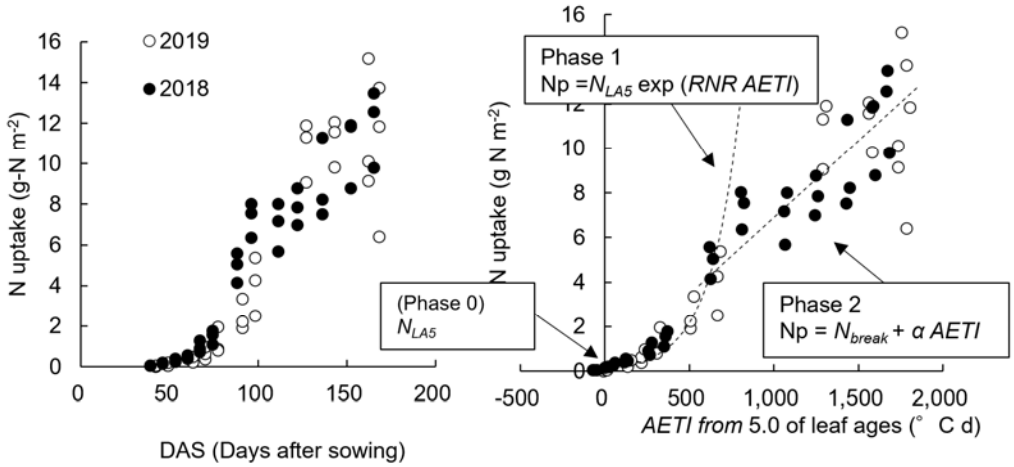

Figure 4 The relation between rice N uptake and DAS or AETI from LA5.

Graph for YA-CU (Yumiazusa and Coated urea) treatment. Supplemental material (Figure S2) shows the relation of all treatments.

Np is the N uptake of the rice plant. N_{LA5} is the N uptake at 5 leaf age. *RNR* is the relative N uptake rate and α is the rate of N uptake in phase 2.