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# Impact of Nitrogen Application Rates on Upland Rice Performance, Planted under Varying Sowing Times

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**Abstract:** Application of suitable nitrogen (N) fertilizer application rate (NR) with respect to sowing time (ST) could help to maximize the performance and productivity of upland rice in Southern Thailand. The 2-year experiments were conducted in the sheds to evaluate the agronomic responses of the upland rice genotype, Dawk Pa–yawm, under various combinations of NR and ST between 2018–2019 and 2019–2020 aimed at obtaining sufficient research evidence for the improved design of long-term field trials in Southern Thailand. As with the initial research, four NR were applied as N0 with no applied N, 1.6 g N pot$^{-1}$, 3.2 g N pot$^{-1}$ and 4.8 g N pot$^{-1}$, and experiments were grown under three ST including early (ST1), medium (ST2) and late sowing (ST3). Results from the experiments indicate that the application of 4.8 g N pot$^{-1}$ resulted in maximum grain yield under all ST in both years. However, a maximum increase in grain yield was observed under ST2 by 54–101% in 2018–2019 and by 276–339% in 2019–2020. Maximum grain N uptake of 0.57 and 0.82 g pot$^{-1}$ was also observed at NR 4.8 g N pot$^{-1}$ under ST2 in both years, respectively. Application of NR 4.8 g N pot$^{-1}$ resulted in the highest N agronomic efficiency (NAE), nitrogen use efficiency (NUE) and water use efficiency (WUE). However, the performance of yield and yield attributes, N uptake, N use efficiencies and WUE were declined in late sowing (ST3). Significant positive association among yield, yield attributes, N uptake and WUE indicated that an increase in NR up to 4.8 g N pot$^{-1}$ improved the performance of Dawk Pa–yawm. The results suggest that the application of 4.8 g N pot$^{-1}$ (90 kg N ha$^{-1}$) for upland rice being grown during September (ST2) would enhance N use efficiencies, WUE and ultimately improve the yield of upland rice. However, field investigations for current study should be considered prior to general recommendations. Moreover, based on the findings of this study, the importance of variable climatic conditions in the field, and the variability in genotypic response to utilize available N and soil moisture, authors suggest considering more levels of NR and intervals for ST with a greater number of upland rice genotypes to observe variations in field experiments for the precise optimization of NR according to ST.

**Keywords:** upland rice; nitrogen application rate; sowing time; yield; nitrogen use efficiencies

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## 1. Introduction

Rice (*Oryza sativa* L.) contributes half of the world's staple food [1,2]. Rice production is also increasing continuously [2,3]. According to FAO [3], a 25% increase was observed only during 2000–2016. Rice is grown under various ecosystems including irrigated, lowlands and uplands. However, lowland rainfed and lowland irrigated systems are major rice production systems [4] representing 6.2 and 4.1 million hectares of production area, respectively [5]. Upland rice acreage contributes 9% in Asia [6]. Thailand is the sixth-largest producer of rice worldwide and the second-largest in Southeast Asia [3]. Rice plays a key role in Thailand's economy and food security [7]. According to USDA [5], major rice production in Thailand is in northern, central, and north-eastern regions, whereas Southern Thailand contributes 6% of the cultivated rice area [8,9]. Like other regions, lowland rice contributes to major rice production in Southern Thailand, but the cultivated area is limited due to geographic limitations. Upland rice is grown in rainfed conditions [10] and it is cultivated by small land holders during rainy seasons in Southern Thailand [11]. Rice supply in Southern Thailand is insufficient for local consumption, and it is imported from other parts of the country. To meet the rice demand and enhance local rice production, upland rice is a good alternative because it does not require additional irrigation, slopy and non-flat area can be utilized, and it can be intercropped with other crops such as young rubber and oil palm. However, efficient upland rice productivity in the southern region has not yet been achieved due to the lack of significant research evidence on agronomic management of upland rice and prevailing traditional management practices. To establish sustainable productivity and enhance upland rice yield, locally adjusted agronomic practices such as optimum nitrogen application rate (NR) with respect to adopted sowing times (ST) should be investigated and recommended.

Nitrogen (N) is a crucial nutrient that has a significant impact on upland rice growth and productivity. According to Kichey et al. [12], N, among all other nutrients, is the most critical element for plant growth, development and quality. N is used extensively to increase rice crop yield by farmers. This is because N improves crop performance, promotes leaf area, plant biomass and ultimately the crop yield [13]. Application of N fertilizer causes N deficiency in rice plants which increases yellowing in color and reduction in leaf size. Reduced N supply at tillering and panicle initiation stages ultimately lead to a reduction in grain yield. Therefore, it is recommended that a suitable N doze should be applied at critical crop stages so that crops can achieve maximum growth and produce better yield potential. Considering N fertilization in rainfed upland rice production in Thailand, various nitrogen application rates (NR) are practiced. Application of 9.8 kg N rai$^{-1}$ or 61.25 kg N ha$^{-1}$ in a yield trial of 43 upland rice genotypes in Songkhla province of Thailand [14], application of 25 kg N ha$^{-1}$ in a simulation of drought stress study on upland rice genotype, Dawk Pa–yawm [15], application of 75 kg N ha$^{-1}$ in a performance evaluation study of 16 upland rice genotypes [16] and application of 15 kg N ha$^{-1}$ as basal doze with an unknown amount of additional urea application during the crop growth period in a correlation and a path analysis study of 10 upland rice genotypes [17] have been reported. A study interviewing the farmers north of Thailand conducted by the Center for Agricultural Resource System Research, Faculty of Agriculture, Chiang Mai University, Thailand, indicated that farmers in the Chiang Mai province usually applied 1.6–12 kg N rai$^{-1}$ or 10–75 kg N ha$^{-1}$ mainly by using N–P–K (16–20–0) as fertilizer source [18]. A general application rate range of 48.75–82.5 kg N ha$^{-1}$ based on soil analysis, soil nutrient status for rainfed and irrigated rice production was recommended by the Division of Rice Research and Development, Thailand [19,20]. According to the Division of Rice Research and Development, Thailand [21], 40–45 kg N ha$^{-1}$ chemical N fertilizer should be applied in two splits at 20–25 kg N ha$^{-1}$ as basal dose and 20 kg N ha$^{-1}$ should be applied 30 days prior to flowering for the upland rice grown in foothill plains. Considering the location, specific to the experimental site (Songkhla Province) and photosensitivity of genotypes, application of 34–39 kg N ha$^{-1}$ and 59–69 kg N ha$^{-1}$ was recommended to be applied for photosensitive and photoperiod insensitive genotypes, respectively [21]. However, according to the authors, no specific study or recommendation regarding a suitable or optimum NR solely or N application according to ST for upland rice production in Thailand has been reported, indicating a research gap. Therefore, a wide range of NR (10–75 kg N ha$^{-1}$) by farmers has been observed for upland rice production under sole or intercropping systems in Thailand. Urea is commonly used as N fertilizer source to meet N requirements which is highly volatile and result in higher N losses. Due to improper N management, variations in genotypic response, fertilizer types and prevailing climatic conditions i.e., temperature and moisture availability, efficient fertilizer utilization and plant N uptake per unit area are also affected. According to Choudhury and Khanif [22], the utilization efficiency of urea–N is lower in rice systems, which is approximately 30–40%, and N recovery seldom surpasses 50% of the total N applied. This happens due to the N loss by denitrification and leaching. Qiao et al. [23] reported a positive correlation between N loss and NR applied. N uptake and upland rice growth may increase with an increase in NR, though it may result in increased N leaching losses due to a high level of N available in the plant root zone [24]. N leaching loss is also positively correlated with N input and a decrease in NR may decrease N leaching [25]. A decline in N leaching with decreased NR was observed when the NR was decreased from 300 kg N ha$^{-1}$ to 200 kg N ha$^{-1}$ without a decline in yield [26]. To avoid under or excessive application of N which results in a decline in grain yield or agronomic and economic losses, respectively, proper nutrient management is necessary [27]. In this regard, estimating plant N concentrations and uptake could help to identify optimum NR for maximized nitrogen use efficiency (NUE). The agronomic efficiency of applied N (NAE) can be used to determine the impact of N fertilizer applied to the grain yield produced. An increase in NAE can increase in N uptake by the plant resulting in reduced N losses and higher NUE in soil and plant systems. Enhanced NUE is a useful indicator for N utilization by crop plants. Higher NAE and NUE at certain levels of N application could give the indication for optimal NR for upland rice. Hence, for optimized upland rice production, increase in grain yield and higher N use efficiencies, researching the identification of suitable NR is essential.

Optimum sowing time (ST) is an important agronomic management factor that becomes more critical in the case of upland rice, as the moisture availability and prevailing climatic conditions significantly influence the nutrient use efficiency of upland rice. Optimal ST ensures that vegetative growth receives a high level of photosynthetic radiation, and grain filling occurs during favorable temperatures [28]. According to Nazir [29], too early and too delayed sowing time resulted in increased plant sterility and reduced the number of productive tillers, respectively. Significant responses of yield and yield components including the number of effective tillers per area, number of grains per panicle and grain weight under different ST have been reported. Therefore, determination of the suitable sowing period relative to rice growth and development stages is necessary. Photoperiod-sensitive genotypes are affected greatly as compared to photoperiod-insensitive genotypes. According to Watcharin et al. [30], farmers in Southern Thailand usually grow photoperiod-sensitive genotypes during the rainy season which is a critical issue in the current scenario of climate change where high variability in rainfall occurs. Variations in rainfall and moisture availability influence the nutrient availability to upland rice. It was suggested that the cultivation of photoperiod insensitive cultivars could be one of the possible solutions to stabilize the upland rice yields [31] in Southern Thailand. However, photoperiod insensitive genotypes may also suffer at different crop developmental stages due to lower or higher rainfall events which can cause drought stress or flooding leading to reduced nutrient availability for rice plants. The rainy season in Thailand prevails during May–October [7,31,32], in which most rice plantation is performed. However, in Southern Thailand, especially in the eastern part of Southern Thailand, most of the rain is received from November to the February of the next year [32]. Hot and dry intervals at the start of the rainy season and variability in rainfall thus pose potential threats to upland rice production. Water use efficiency (WUE), which is the ratio between yield produced and water consumed or evapotranspiration, is significantly affected. A significant interaction prevails between WUE and NUE. According to Gajri et al. [33], NR influenced the WUE, whereas NUE was also dependent upon water input. Adjustment in crop growth period [7] with modifications in ST results in shifting of critical crop stages to favorable parts of the season. Variations in moisture availability affect the plant nutrient uptake. Thus, the adjustment in ST could benefit with higher WUE as well as enhanced NUE. Therefore, in the current scenario, synchronization of ST with optimum NR could fulfil the rice crop requirements. Adaptation strategies to adjust ST could also help to significantly reduce the extent of climatic impact on upland rice production. Studies conducted in north-eastern Thailand also suggest that adjustment in ST according to local conditions and proper nutrient management could help mitigate the impact of climate change on rice production [34–36].

Based on the significance of NR, ST, limited research evidence availability and the wide range of NR and ST management practices in Southern Thailand, we understood that adjustments in NR with modification in ST and synchronizing their interactions could result in improved NUE, WUE and yield. Therefore, the initial objective of this research was to obtain sufficient information about the impact of NR under varying ST on upland rice performance. The results of the current research will help to adjust the appropriate gradients for NR and intervals for ST for better designing of further long-term, multilocational field trials and propose best and optimized N and ST management practices for enhanced upland rice production, especially in Southern Thailand.

## 2. Materials and Methods

### *2.1. Experimental Setup and Crop Management*

Experiments were conducted in the sheds located (7°00′16.57″ N, 100°30′01.93″ E) at the Faculty of Natural Resources, Prince of Songkla University, Hat Yai, Thailand (Figure S1) during 2018–2019 and 2019–2020. Topsoil was sieved, straw and plant roots were removed, and a composite soil sample was obtained before filling the soil in the planting pots for soil nutrient status during both years. The soil was sandy clay loam in the texture with pH, 4.77 and 5.29, organic matter of 4.73 and 4.60 g kg$^{-1}$, total N of 0.34 and 0.30 g kg$^{-1}$, available P of 13.03 and 35.58 mg kg$^{-1}$ and available $K^+$, of 41.19 and 58.67 mg kg$^{-1}$ for 2018–2019 and 2019–2020, respectively. Dawk Pa–yawm, an upland rice genotype, famous due to its aroma and commonly grown in Southern Thailand, was used in this study. Experiments were laid out using a completely randomized design with three replications during both years. Planting pots used in the experiments were conical shaped with 30 cm top diameter, 19 cm bottom diameter and 24 cm in height. Each pot was filled with 12 kg of homogenous soil. Seeds were sown at 5 cm soil depth in the pots by a direct seeding method maintaining 3 plants in each pot at the seedling stage. There were 3 pots used for each treatment in each replication, and a total of 27 plants were maintained for each treatment in each experiment. Experiments were subjected to two treatments including NR and ST. Each treatment was designated in a separate block of pots arranged at different coordinates in the shed. As a wide range of NR is practiced at the farmer’s scale for rainfed upland rice production under sole or intercropping systems, and keeping in view the current practices, and various recommendations of research institutes in Thailand, initially NR were chosen as a control, N0 with no applied N, 30 kg N ha$^{-1}$, 60 kg N ha$^{-1}$ and 90 kg N ha$^{-1}$. NR for pots were calculated on field basis using Equation (1) [37] and were applied as N0 with no applied N, 1.6 g N pot$^{-1}$, 3.2 g N pot$^{-1}$ and 4.8 g N pot$^{-1}$ as an initial study. Urea was used as the fertilizer source containing 46% N and NR were applied in two equal splits at the start of tillering and panicle initiation stages. Upland rice is grown in the rainy season and most rain in Southern Thailand prevails in May–October. A wide range sowing window prevails in Southern Thailand and farmers perform early or late planting depending upon cultivars sensitivity and moisture availability. However, major rice planting has been in practice by farmers during September–November, while minor rice planting has been in practice during April–June in Southern Thailand [8]. As of initial research, ST were selected as early sowing-ST1 on 05 September 2018, medium-ST2 on 26 September 2018 and late sowing-ST3 on 31 October 2018 for 2018–2019 and early sowing-ST1 on 01 September 2019, medium-ST2 on 06 October 2019 and late sowing-ST3 on 03 November 2019 for 2019–2020. Plants were irrigated with an automatic drip irrigation system and irrigation was applied for a specific time duration for each treatment block frequently to avoid water stress. The amount of irrigation water was then calculated by the irrigation time, dripper head water discharge capacity of 8 tiller per hour and area of pots. Each planting pot in ST1, ST2 and ST3 received 57 liters (L), 68 L and 74 L as an average total amount of irrigation water during both years. Manual weeding and insect, pest and disease management practices were performed as standard practices to reduce yield losses in both years.

$$\text{Fertilizer amount for pot} = \frac{\text{Recommended doze of fertilizer}}{1\ \text{hectare}} \times \text{Weight of pot soil} \quad (1)$$

*2.2. Sampling, Measurements and Computations*

At harvest, plant height and biomass were recorded from 3 out of 9 randomly selected plants and data collection was repeated for each treatment in three replications. Plant height was measured from the base of the stem to the topmost leaf or panicle. The number of days to 50% flowering and 50% maturity were recorded by counting the number of days from respective sowing time. The number of tillers was counted at the maximum tillering stage and tillers with at least one visible leaf were included. Rice plants were manually harvested at maturity, and the number of panicles were counted. Plant and grain samples were dried in an oven at 70 °C for different time durations until a constant weight was achieved to get grain yield and biomass on a dry weight basis. Soil sampling for N analysis was performed for each treatment and each replication at harvest to observe N concentrations. Soil samples collected from the pots from three replicates were first mixed and passed through a 1mm sieve to remove impurities for obtaining a respective composite soil sample for each N treatment for N analysis. Oven-dried plant biomass and grain samples were finely ground and passed through a 1mm sieve as well. Straw, grain and soil samples were then sent to the Central Analytical Laboratory of Faculty of Natural Resources, Prince of Songkla University, Thailand for N analysis to obtain N concentrations and calculate N uptake by plant and grains. Straw and grain N uptake in relation to applied NR [38] were calculated by multiplying straw biomass and grain yield with respective N concentrations. N efficiencies including agronomic efficiency (NAE) (2), which is the number of extra grains harvested per kg of N applied to a grain crop that drives both the agronomic and economic efficiency of fertilizer use, and N use efficiency (NUE) (3), which is the fraction of applied N that is absorbed and used by the plant, were calculated using equations mentioned by Abbasi et al. [39]. WUE was calculated as the ratio between grain yield harvested and total amount of irrigation water per pot using Equation (4) [40].

$$\text{NAE} = \frac{\text{Grain yield}_{\text{N added}} - \text{Grain yield}_{\text{control}}}{\text{Total N fertilizer applied}} \quad (2)$$

$$\text{NUE} = \frac{\text{N uptake}_{\text{N added}} - \text{N uptake}_{\text{control}}}{\text{Total N fertilizer applied}} \times 100 \quad (3)$$

$$\text{WUE} = \frac{\text{Grain yield per pot (g)}}{\text{Amount of irrigation water per pot (L)}} \quad (4)$$

*2.3. Statistical Analysis*

Data obtained from both year experiments was used in statistical software Statistix (8.1 package, analytical software, Tallahassee, FL, USA) [41] to test the significance of results and mean comparisons for the effects of applied NR and ST. A two-way analysis of variance (ANOVA) was performed for yield and yield attributes of Dawk Pa–yawm, straw and grain N uptake and WUE from three replications with effect to NR, ST and the interactions of NR and ST. Mean comparisons were made using the least significant difference (LSD), and $p$-value < 0.05 was considered significantly different [42]. Combined Pearson's correlation coefficients were computed for yield and yield attributes of Dawk Pa–yawm, computed straw, grain and total N uptake and WUE to observe associations among various parameters. The "Corrplot" [43] package of R software was used in computing correlation coefficients and graphics.

# 3. Results

## 3.1. Upland Rice Growth and Productivity

Results from the analysis of variance (ANOVA) for observed traits and computed parameters for Dawk Pa–yawm using the LSD-test ($p < 0.05$) indicated highly significant ($p < 0.001$) differences for days to flowering and days to maturity with respect to the ST, whereas there were no significant differences observed with respect to NR and NR × ST for both years (Tables S1 and S2). Flowering days and maturity duration were not significantly affected by an increase in NR for both years and the difference ranged 1–3 days for flowering (Figure 1A,B) and similarly 1–3 days for maturity (Figure 1C,D). ST influenced days to flowering, thus, days to flowering and days to maturity were increased under ST3 by 7 days for both years (Figure 1A,B). Days to flowering were decreased only under ST2 for 2019–2020 (Figure 1B). Maturity duration was increased under ST2 and ST3 for year 2018–2019 by 6–9 days (Figure 1C) while increased for 6–8 days under ST3 for 2019–2020 (Figure 1D). There were highly significant differences ($p < 0.001$) for plant height, number of tillers, number of panicles, grain yield and biomass with respect to NR during both years except moderate significant differences ($p < 0.01$) for the number of tillers and number of panicles during 2019–2020 (Tables S1 and S2). Highly significant differences ($p < 0.001$) were observed for days to flowering, days to maturity, plant height and grain yield with respect to ST in both years, whereas moderately significant differences ($p < 0.01$) were observed for the number of tillers, number of panicles and biomass in 2018–2019 and for biomass in 2019–2020 with respect to the ST (Tables S1 and S2). The number of tillers and number of panicles were significantly different ($p < 0.05$) during 2019–2020. ANOVA for the interactions of the NR and ST indicated non-significant differences for days to flowering, days to maturity, plant height, number of tillers, number of panicles, grain yield and biomass in both years except a moderate significant ($p < 0.01$) difference for plant height during 2019–2020 under the interaction of NR and ST. Plant height was gradually increased for both years with an increase in NR (Figure 2A,B) under all ST. Increase in plant height ranged 13–27% for ST1, 2–19% for ST2 and 3–19% for ST3 for 2018–2019 (Figure 2A) and 4–10% for ST1, 18–38% for ST2 and 1–8% for ST3 for 2019–2020 (Figure 2B). The number of tillers (Figure 3A,B) and the number of panicles (Figure 3C,D) were influenced by NR and ST. In total, 1–4 tillers, as well as panicles, per plant were increased under increasing NR up to 4.8 g N pot$^{-1}$. However, the number of tillers and number of panicles were decreased by 1–3 tillers as well as panicles per plant under ST3 for both years (Figure 3A–D). Grain yield (Figure 4A,B) and biomass (Figure 4C,D) were increased with increasing NR under all ST for both years. Grain yield increased by 19–64% under ST1, 54–101% for ST2 and 32–78% for ST3 in 2018–2019 (Figure 4A) while it increased by 53–121% for ST1, 276–339% for ST2 and 64–94% for ST3 in 2019–2020 (Figure 4B). Biomass increased by 52–111% under ST1, 77–127% for ST2 and 65–127% for ST3 in 2018–2019 (Figure 4C) while it increased by 43–86% for ST1, 98–153% for ST2 and 32–75% for ST3 in 2019–2020 (Figure 4D).

## 3.2. Nitrogen Uptake

Highly significant differences ($p < 0.001$) were observed for straw N uptake, grain N uptake and total N uptake with respect to NR and ST during both years, except a non-significant difference for total N uptake under ST during 2018–2019 (Tables S1 and S2). Interactions of the NR and ST indicate highly significant ($p < 0.001$) differences for straw N uptake and grain N uptake and a significant difference ($p < 0.05$) for total N uptake during 2018–2019. Straw N uptake was significantly different ($p < 0.05$), whereas non-significant differences for grain N uptake and total N uptake were observed with respect to NR × ST in 2019–2020. Straw (stem + leaves) N uptake (g pot$^{-1}$) was increased with increasing NR up to N 4.8 g N pot$^{-1}$ for both years (Figure 5A,B) when compared to pots with no applied N. However, ST affected straw N uptake resulting in variations in N uptake under all ST (Figure 5A,B). Grain N uptake was also increased with increasing NR up to 4.8 g N pot$^{-1}$ under all ST in both years (Figure 5C,D). However, maximum grain N uptake was observed under ST2 and it was then decreased under ST3 (Figure 5C,D), indicating the negative impact of delayed sowing on grain N uptake. Maximum grain N uptake valued 0.57 g pot$^{-1}$ at NR 4.8 g N pot$^{-1}$ under ST2 in 2018–2019 (Figure 5C) and 0.82 g pot$^{-1}$ at NR 4.8 g N pot$^{-1}$ under ST2 in 2019–2020 (Figure 5D). These results indicate that ST2 was the favorable ST for increased grain N uptake, and early (ST1) or delayed (ST3) sowing resulted in less translocation of N from the rice straw to grain. Total N uptake including straw and grain-N was also increased with increasing NR up to 4.8 g N pot$^{-1}$ (Figure 5E,F). However, total N uptake was decreased at NR 4.8 g N pot$^{-1}$ under ST3 in 2018–2019 (Figure 5E) and decreased at 3.2 g N pot$^{-1}$ under ST1 in 2019–2020 (Figure 5F), indicating the significant negative impact of ST on total plant N uptake. Total plant N uptake was in an increasing trend under ST2 and maximum total N uptake was observed under ST2 with a value of 2.09 g pot$^{-1}$ in 2019–2020 (Figure 5F). The results exhibit that medium ST (ST2) was the most favorable ST for maximum N extraction from soil and increase in total plant N uptake as well as for maximum mobility of N from plant parts to grains.

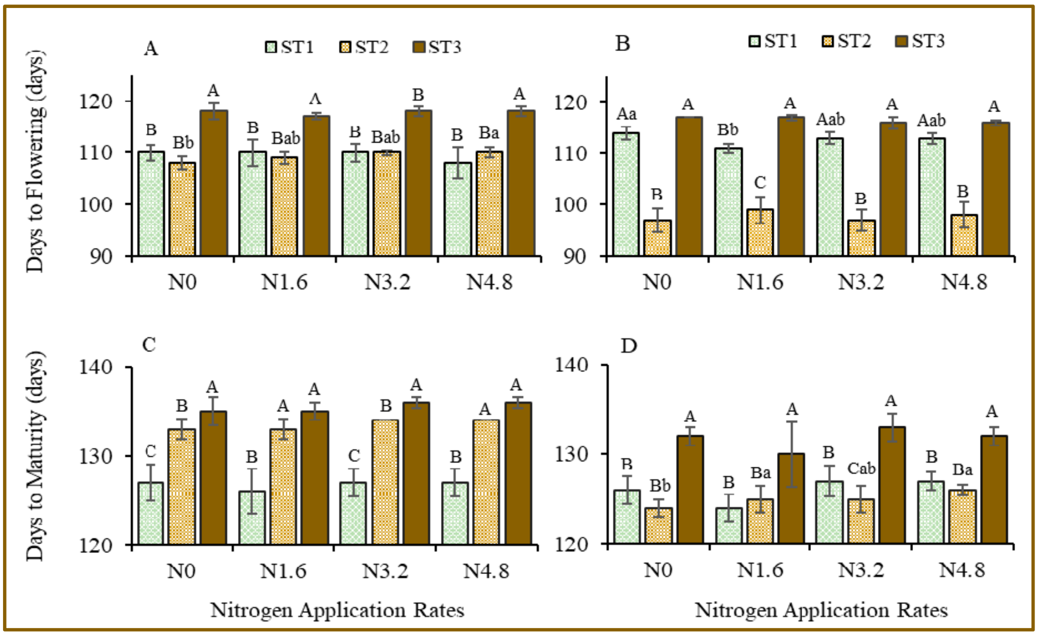


**Figure 1.** Effect of nitrogen application rates and sowing times on days to flowering (**A**,**B**) and days to maturity (**C**,**D**) during 2018–2019 (**A**,**C**) and 2019–2020 (**B**,**D**). Mean values are presented and vertical bars indicate ± standard errors of means ($n$ = 3). Uppercase letters indicate significant differences ($p$-value < 0.05) of days to flowering and days to maturity under different sowing times within each nitrogen application rate. Lowercase letters indicate significant differences ($p$-value < 0.05) of days to flowering and days to maturity at different nitrogen application rates within each sowing time. Due to the non-significant differences for sowing times within the same nitrogen application rate, no lowercase letters are presented in Figure 1C. ST1: sowing time 1 (early), ST2: sowing time 2 (medium), ST3: sowing time 3 (late). N0: no applied N, N1.6: 1.6 g N pot$^{-1}$, N3.2: 3.2 g N pot$^{-1}$, N4.8: 4.8 g N pot$^{-1}$.

### *3.3. Nitrogen Use Efficiencies*

NR significantly affected N efficiencies including agronomic efficiency (NAE) (Figure 6A,B) and nitrogen use efficiency (NUE) (Figure 6C,D) in both years. NAE was increased with applied N and increasing NR up to 4.8 g N pot$^{-1}$ under all ST in both years (Figure 6A,B). Maximum NAE was observed at N 4.8 g N pot$^{-1}$ under all ST with values 5.35, 10.18 and 8.26 kg kg$^{-1}$ for ST1, ST2 and ST3, respectively, in years 2018–2019 (Figure 6A) and 14.95, 34.16 and 10.14 kg kg$^{-1}$ for ST1, ST2 and ST3, respectively, in years 2019–2020 (Figure 6B). However, ST influenced the NAE and resulted in a decline and variations in both years (Figure 6A,B). The highest NAE was observed under ST2 in both years (Figure 6A,B) and NAE was decreased under delayed sowing ST3, indicating that ST2 was the most favorable ST for improved NAE. NUE was also increased with an increase in NR up to 4.8 g N pot$^{-1}$ under all ST in both years (Figure 6C,D). Maximum NUE was observed at NR 4.8 g N pot$^{-1}$ under all ST up to 119%, 137% and 133% for ST1, ST2 and ST3, respectively, in years 2018–2019 (Figure 6C) and 155%, 171% and 102% for ST1, ST2 and ST3, respectively, in years 2019–2020 (Figure 6D). ST influenced the NUE and resulted in differences in both years (Figure 6C,D). However, the highest NUE was observed under ST2 in both years (Figure 6C,D) and NUE was decreased under delayed sowing, ST3. NUE under ST3 was more affected in 2019–2020 (Figure 6D) as compared to 2018–2019 (Figure 6C).

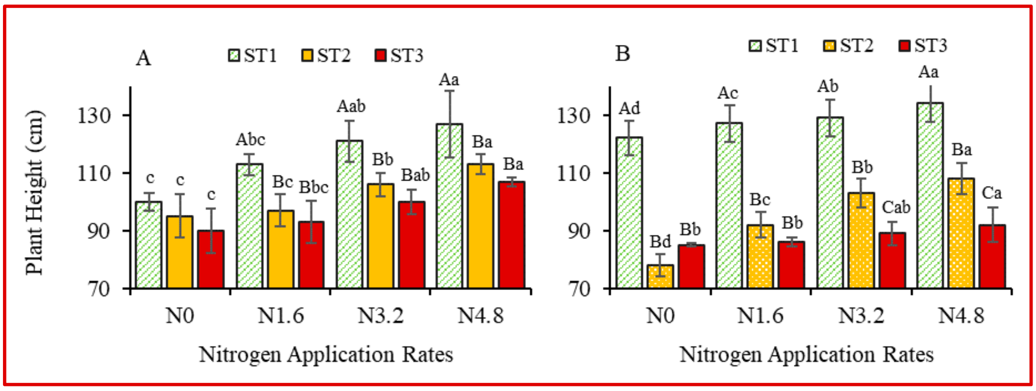


**Figure 2.** Effect of nitrogen application rates and sowing times on plant height during 2018–2019 (**A**) and 2019–2020 (**B**). Mean values are presented and vertical bars indicate ± standard errors of means ($n$ = 3). Uppercase letters indicate significant differences ($p$-value < 0.05) of plant height under different sowing times within each nitrogen application rate. Lowercase letters indicate significant differences ($p$-value < 0.05) of plant height at different nitrogen application rates within each sowing time. ST1: sowing time 1 (early), ST2: sowing time 2 (medium), ST3: sowing time 3 (late), N0: no applied N, N1.6: 1.6 g N pot$^{-1}$, N3.2: 3.2 g N pot$^{-1}$, N4.8: 4.8 g N pot$^{-1}$.

### 3.4. Water Use Efficiency

Water use efficiency (WUE) was estimated for each treatment for both years (Figure 7). There was a highly significant ($p < 0.001$) difference for WUE with respect to NR and a moderate significant difference ($p < 0.01$) with respect to ST and a non-significant difference for the interactions of NR and ST in 2018–2019 (Table S1). During 2019–2020, highly significant ($p < 0.001$) differences for WUE with respect to NR and ST and a significant difference ($p < 0.05$) with respect to the interactions of NR and ST were observed (Table S2). An increase in NR up to 4.8 g N pot$^{-1}$ significantly increased WUE in both years (Figure 7A,B). Maximum WUE was observed at N 4.8 g N pot$^{-1}$ under all ST with values 0.25, 0.31 and 0.26 g L$^{-1}$ for ST1, ST2 and ST3, respectively, in years 2018–2019 (Figure 7A) and 0.44, 0.59 and 0.26 g L$^{-1}$ for ST1, ST2 and ST3, respectively, in years 2019–2020 (Figure 7B). WUE increased up to 40% under ST1, 59% under ST2 and 42% under ST3 at NR up to N 4.8 g N pot$^{-1}$ during 2018–2019 and increased up to 50% under ST1, 92% under ST2 and 67% under ST3 at NR up to N 4.8 g N pot$^{-1}$ during 2019–2020 (Figure 7A,B). However, ST influenced the WUE and resulted in a decline in both years under delayed sowing (Figure 7A,B). The highest WUE was observed under ST2 in both years by 59% and 92%, respectively (Figure 7A,B), and it was decreased under delayed sowing, ST3 by 24% in 2018–2019 and by 84% in 2019–2020. The results indicate that ST2 was the optimal ST for better performance for WUE.

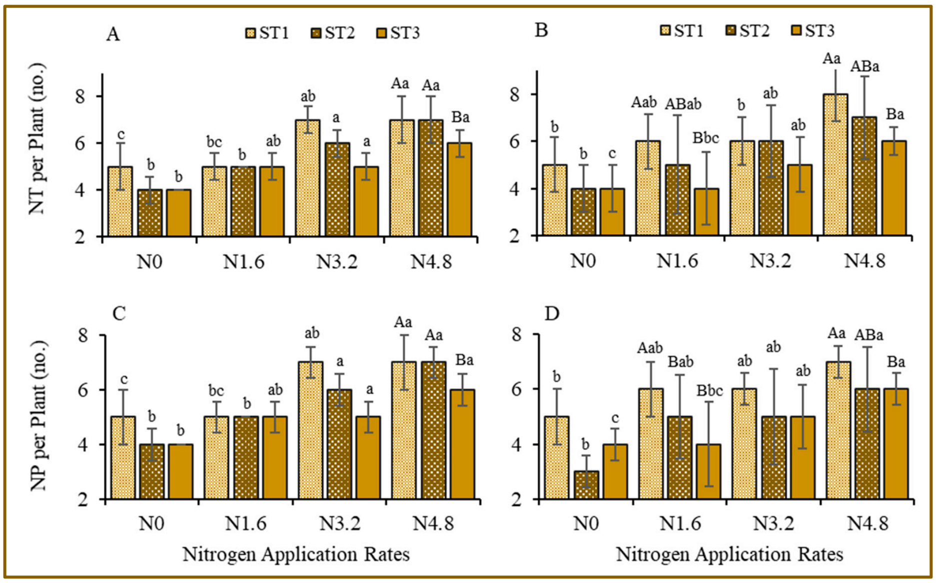


**Figure 3.** Effect of nitrogen application rates and sowing times on the number of tillers (NT) (**A**,**B**) and the number of panicles (NP) (**C**,**D**) during 2018–2019 (**A**,**C**) and 2019–2020 (**B**,**D**). Mean values are presented and vertical bars indicate ± standard errors of means ($n$ = 3). Uppercase letters indicate significant differences ($p$-value < 0.05) in the number of tillers and the number of panicles under different sowing times within each nitrogen application rate. Lowercase letters indicate significant differences ($p$-value < 0.05) in the number of tillers and the number of panicles at different nitrogen application rates within each sowing time. ST1: sowing time 1 (early), ST2: sowing time 2 (medium), ST3: sowing time 3 (late). N0: no applied N, N1.6: 1.6 g N pot$^{-1}$, N3.2: 3.2 g N pot$^{-1}$, N4.8: 4.8 g N pot$^{-1}$.

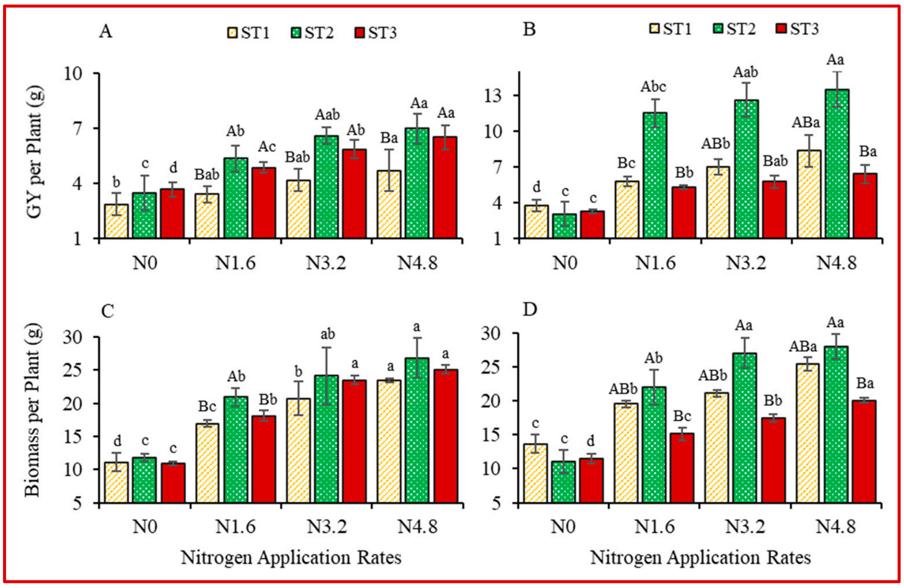


**Figure 4.** Effect of nitrogen application rates and sowing times on grain yield (GY) (**A**,**B**) and biomass (**C**,**D**) during 2018–2019 (**A**,**C**) and 2019–2020 (**B**,**D**). Vertical bars indicate ± standard errors of means ($n$ = 3). Mean values are presented and vertical bars indicate ± standard errors of means ($n$ = 3). Uppercase letters indicate significant differences ($p$-value < 0.05) of grain yield and biomass under different sowing times within each nitrogen application rate. Lowercase letters indicate significant differences ($p$-value < 0.05) of grain yield and biomass at different nitrogen application rates within each sowing time. ST1: sowing time 1 (early), ST2: sowing time 2 (medium), ST3: sowing time 3 (late). N0: no applied N, N1.6: 1.6 g N pot$^{-1}$, N3.2: 3.2 g N pot$^{-1}$, N4.8: 4.8 g N pot$^{-1}$.

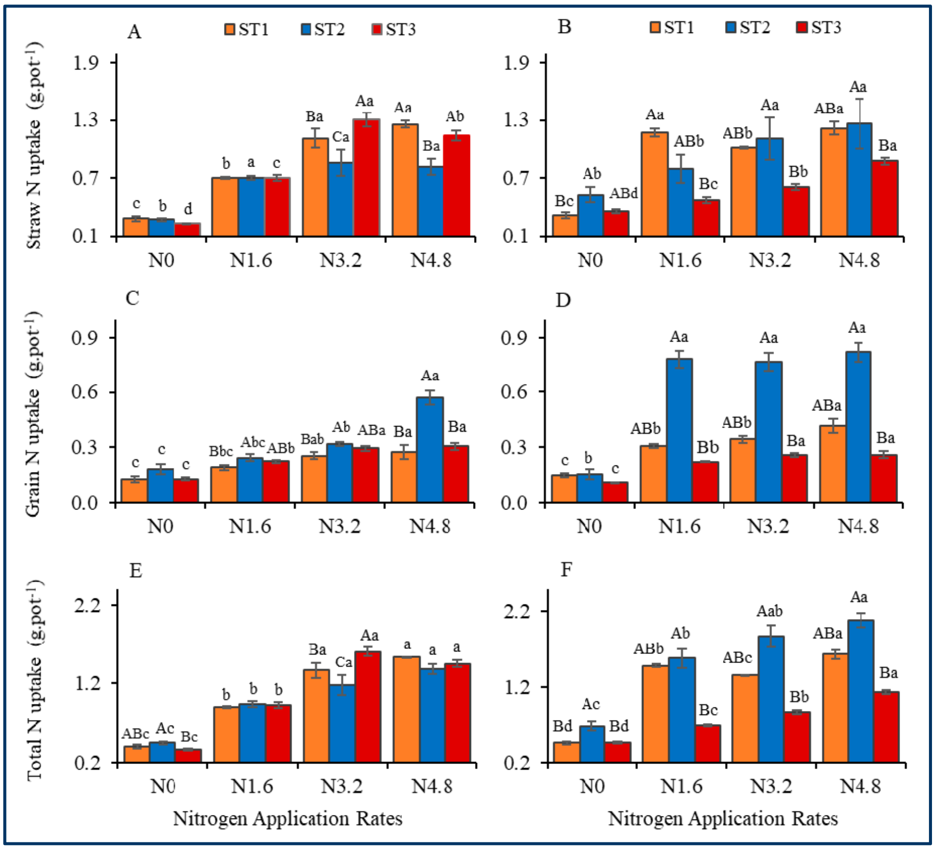


**Figure 5.** Effect of nitrogen application rates and sowing times on straw N uptake (**A**,**B**), grain N uptake (**C**,**D**) and total N uptake (**E**,**F**) during 2018–2019 (**A**,**C**,**E**) and 2019–2020 (**B**,**D**,**F**). Vertical bars indicate ± standard errors of means ($n$ = 3). Uppercase letters indicate significant differences ($p$-value < 0.05) of straw N uptake, grain N uptake and total N uptake under different sowing times within each nitrogen application rate. Lowercase letters indicate significant differences ($p$-value < 0.05) of straw N uptake, grain N uptake and total N uptake at different nitrogen application rates within each sowing time. ST1: sowing time 1 (early), ST2: sowing time 2 (medium), ST3: sowing time 3 (late). N0: no applied N, N1.6: 1.6 g N pot$^{-1}$, N3.2: 3.2 g N pot$^{-1}$, N4.8: 4.8 g N pot$^{-1}$.

### *3.5. Correlation Analysis*

Pearson's correlation analysis (Figure 8) indicates that there was a highly significant positive correlation between days to flowering and days to maturity. There was a significant positive correlation between days to flowering and grain yield. Plant height was highly significant and positively correlated with the number of tillers and the number of panicles, whereas a moderately significant and positive association was observed between plant height and biomass. Plant height was also significant and positively correlated with grain yield. The number of tillers were highly significant and positively associated with the number of panicles and biomass, whereas significant and positively correlated with grain yield and straw N uptake. There was a highly significant positive correlation among the number of panicles and biomass whereas a significant correlation was observed among the number of panicles and grain yield. Grain yield was highly associated with biomass, whereas it was significantly associated with straw N uptake. Straw N uptake was highly significant, whereas total N uptake was significantly associated with the biomass. Straw N uptake was also highly associated with the total N uptake. Grain N uptake was moderately associated with the straw N uptake, whereas it was highly associated with total N uptake. Straw N uptake, grain N uptake and total N uptake were highly significant and positively correlated with the WUE. Computed coefficient values are presented in Figure 8.

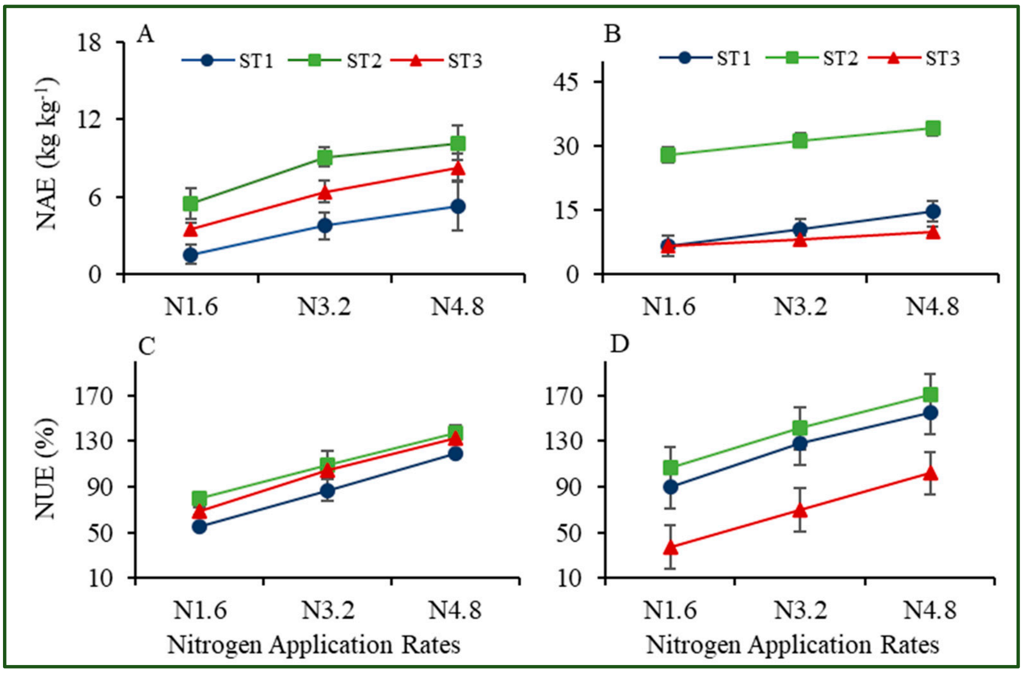


**Figure 6.** Effect of nitrogen application rates and sowing times on N agronomic efficiency (NAE) (**A**,**B**) and N use efficiency (NUE) (**C**,**D**) during 2018–2019 (**A**,**C**) and 2019–2020 (**B**,**D**). Vertical bars indicate ± standard errors of means ($n$ = 3). ST1: sowing time 1 (early), ST2: sowing time 2 (medium), ST3: sowing time 3 (late). N1.6: 1.6 g N pot$^{-1}$, N3.2: 3.2 g N pot$^{-1}$, N4.8: 4.8 g N pot$^{-1}$.

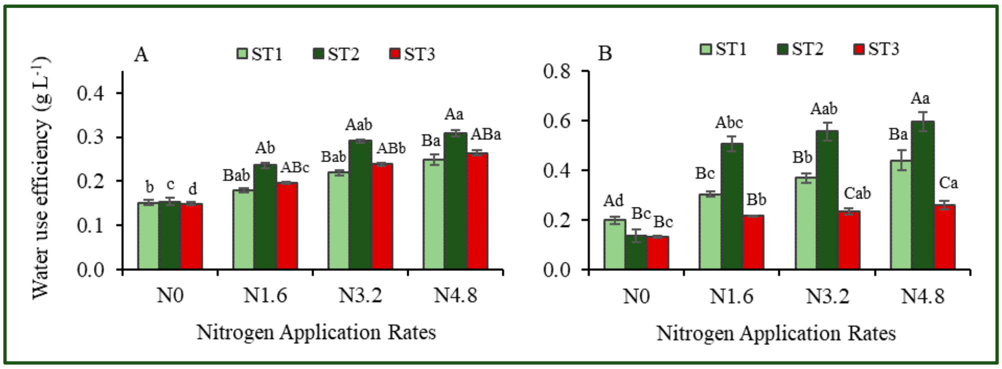


**Figure 7.** Effect of nitrogen application rates and sowing times on water use efficiency during 2018–2019 (**A**) and 2019–2020 (**B**). Mean values are presented and vertical bars indicate ± standard errors of means ($n$ = 3). Uppercase letters indicate significant differences ($p$-value < 0.05) of water use efficiency under different sowing times within each nitrogen application rate. Lowercase letters indicate significant differences ($p$-value < 0.05) of water use efficiency at different nitrogen application rates within each sowing time. ST1: sowing time 1 (early), ST2: sowing time 2 (medium), ST3: sowing time 3 (late). N0: no applied N, N1.6: 1.6 g N pot$^{-1}$, N3.2: 3.2 g N pot$^{-1}$, N4.8: 4.8 g N pot$^{-1}$.

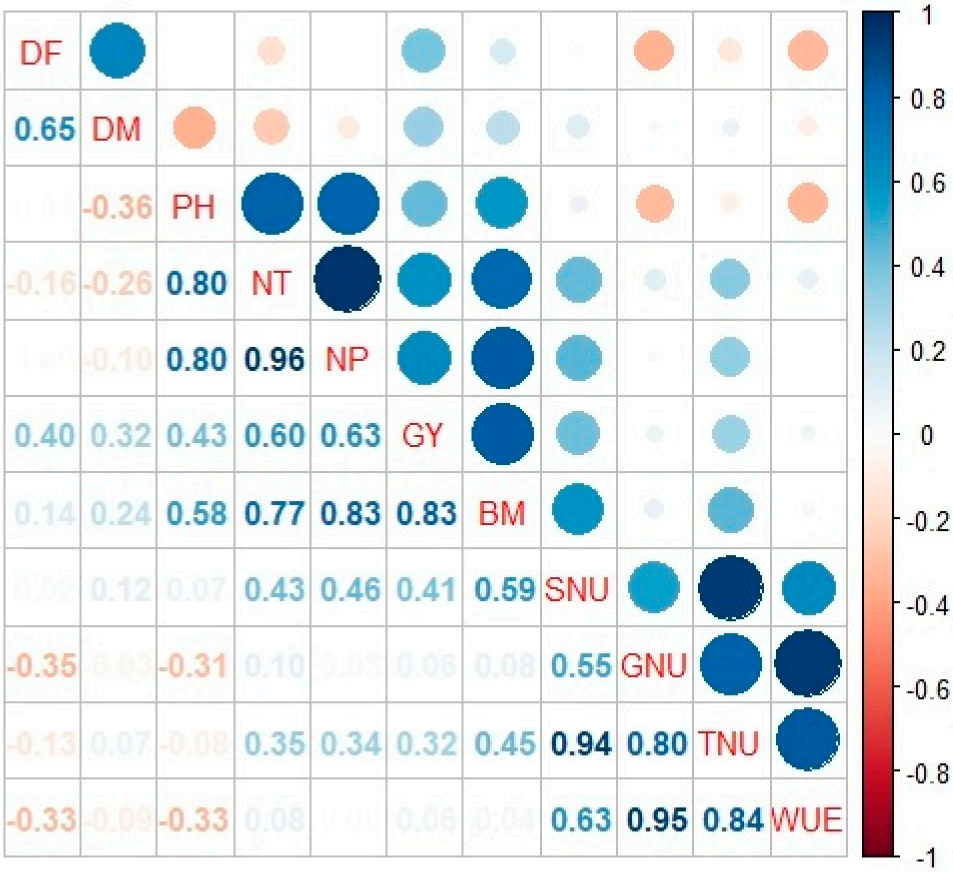


**Figure 8.** Corrplot of combined Pearson's correlation coefficients among agronomic attributes of Dawk Pa–yawm, nitrogen uptake and water use efficiency. Positive and negative associations are presented in blue and red colored circles, respectively, at the top-right diagonal and squares with an absence of colored circles represent no significant association at $p$-value < 0.005 among respective parameters. Correlation coefficient numbers are presented at the bottom-left diagonal. The intensity of colors of circles and numbers, and the size of the circles indicate the proportion of Pearson's coefficients. DF, days to flowering; DM, days to maturity; PH, plant height; NT, number of tillers; NP, number of panicles; GY, grain yield; BM, biomass; SNU, straw nitrogen uptake; GNU, grain nitrogen uptake; TNU, total nitrogen uptake; WUE, water use efficiency.

## 4. Discussion

Nitrogen (N) is an important element and the application of nitrogenous fertilizers in upland rice systems is crucial as N significantly impacts rice performance and productivity. Rice yield is significantly influenced by reduced or no N fertilizer application and the overuse of N results in increased agronomic and economic losses, as well as affects soil health. The efficiency of applied N fertilizer is influenced by various rice crop management practices, and sowing time (ST) is one of them. Improper N management and wide sowing windows adopted by small land holders and upland rice growers are major problems affecting the upland rice production in Southern Thailand. Early or late sowing alters the nutrient availability to rice plants due to variations in prevailing climatic conditions and moisture availability. To achieve viable rice productivity, optimal management of nitrogen application rate (NR) with respect to ST is necessary as upland rice performance and yield are significantly influenced by N input under various ST.

The quantity of applied N significantly influences the physiological processes and photosynthesis of plants [44], which ultimately impacts the performance of yield attributes and defines the rice yield potential. Our results indicate that the performance of yield attributes and the yield of upland rice varied significantly under varying NR and N nutrition remarkably improved the overall performance. Additional N supply resulted in increased plant height in both years. An increase in plant height occurred possibly due to the contribution of added N which improved the growth, internode length and overall metabolism. Enhanced N application is well documented in encouraging cell expansion, and it subsequently stimulated stem elongation [45,46]. Results for plant height were supported by the findings of Abbasi et al. [39] and Zhang et al. [44] who reported remarkable improvements in plant height following increased N application rate. Similar results have also been demonstrated by Jahan et al. [47] who described that an increase in N supply to rice genotypes caused a significant increase in the height of rice plants. In the present study, higher nitrogen application resulted in an additional 1–4 tillers as well as panicles per plant. Previous studies have also observed that panicle numbers were increased with an increase in NR [44]. The increase in tillering due to increased NR might be linked to more N availability at the tillering stage which plays a role in cell division. Wang et al. [48] demonstrated that N availability controls rice tiller numbers through the regulation of the nitrate transporter. An elevated nitrogen level in rice plants leads to increased tiller numbers and tiller bud outgrowth [49]. The number of panicles is one of the major contributing factors in rice yield. Cell division triggered by N supply increases the panicle formation at reproductive stages of rice crop. Jahan et al. [47] observed that N fertilization increased the number of tillers m$^{-2}$, which resulted due to the increased N availability for cell division. In our study, higher N concentration in plants resulted in a higher number of panicles, and similar findings were observed by Manzoor et al. [27] Yield attributes and yield were significantly associated with applied NR. Approximately 19–339% increase in grain yield was observed in our study with increasing NR under different ST. An increase in yield possibly occurred due to the increased performance of yield attributes. Zhang et al. [43] observed that an increase in NR significantly increased grain yield; however, this increase in grain yield was in the limited range of NR. Chen et al. [49] also observed that grain yield and biomass of rice were positively affected by increased NR. An increase in plant biomass ranging from 26 to 127% with increasing NR under different ST indicated a higher performance of biomass contributing traits including plant height and the number of tillers. An increase in plant biomass with N fertilization has also been reported in a rice experimental study by Jahan et al. [47] In our experimental results, it was noticed that grain yield was in an increasing trend up to NR 4.8 g N pot$^{-1}$, indicating the need for an increase in further levels of NR in future experimentation to observe the curve for better optimization of the N application rate.

Nitrogen application and N uptake by plants significantly influence the physiological processes of rice. Synchronization of crop N requirement and N supply is an important step to enhance N use in rice plants. The ratio between N uptake and N loss regulates plant growth and development, and higher plant biomass is produced if more N is absorbed [7]. However, there are various factors that may influence N utilization and N uptake in rice plants as N is highly susceptible to denitrification, volatilization and leaching losses in rice environments. Higher plant N uptake is desired through efficient N management. In our study, plant and grain N concentrations and N uptake varied among NR. Straw and grain N uptake was increased up to NR 4.8 g N pot$^{-1}$ during both years. Maximum grain N uptake was observed at 4.8 g N pot$^{-1}$ under ST2, and it was decreased under ST3. Variations in the increase in straw and grain N concentrations and N uptake were observed at varying NR under different ST which indicates the impact of ST. Jahan et al. [47] also reported that rice's response to applied NR was associated with growing seasons. An increase in rice straw N, grain N concentration and N uptake was also observed by Chen et al. [49]. Higher N uptake is an indication of the achievement of crop N requirement under ideal NR availability and optimal conditions. It was indicated that an increase in NR under delayed sowing could not increase grain N uptake. Total N uptake was also observed at its maximum under ST2 at 4.8 g N pot$^{-1}$, indicating that increasing NR under ST2 increased total N uptake. N uptake was also decreased in late sowing as reported by Pal et al. [50] Agronomic efficiency of applied N (NAE) is an important index to record the response of grain productivity in relation to NR. In our study, NAE was increased with increasing NR and it was observed that maximum NAE was achieved at 4.8 g N pot$^{-1}$. Nitrogen use efficiency (NUE) was also increased with increasing NR, and maximum NUE was achieved at 4.8 g N pot$^{-1}$ under ST2 as well. Enhancing N use efficiencies in upland rice systems is one of the main objectives of N fertilization. We observed that increased NR enhanced the N use efficiencies. Furthermore, water use efficiency (WUE) was also associated with NR, and higher NR 4.8 g N pot$^{-1}$ resulted in higher WUE. The association of NUE and WUE has also been well reported [7,51].

Sowing time critically impacts the utilization of environmental sources including moisture availability during crop growth, and it can influence crop yields [4]. It was observed that ST influenced the performance of Dawk Pa–yawm with respect to applied NR. Maximum grain yield was observed under ST2 and an increase in NR to 4.8 g N pot$^{-1}$ could not cause a significant increase in grain yield under delayed sowing, ST3. This indicated that NR 4.8 g N pot$^{-1}$ was suitable for ST2 while NR 3.2 g N pot$^{-1}$ was suitable for ST3 with respect to N use. Days to flowering and days to maturity were increased under delayed sowing, ST3. Crop yield and biomass [52] are highly correlated with the life cycle. Gomez–Macpherson and Richards [53] stated that phenology is one of the critical aspects of adaptation and enhancement of yield as it regulates the length of critical crop growth stages and change in crop phenology is considered one of the major indicators of climate change impact. Maximum plant height, number of tillers, number of panicles and grain yield were observed under ST2, while biomass was recorded at its maximum under ST3 during 2019–2020. It indicated that conversion of photo-assimilates to grain was decreased under ST3 as an increase in biomass under delayed sowing could not result in increased grain yield. Babel et al. [34], in a climate change impact study, predicted that the delay in ST of Thai rice genotype KDML–105 at Roi Et province (Thailand) with 30 days delay in initial sowing would increase yield by 23% during the 2050s. The predictions of Babel et al. [34] are supporting evidence for this research as it was observed that ST2, which was slightly delayed ST for upland rice, resulted in improved grain yield, N uptake, N efficiencies as well as WUE. The maximum of N use efficiencies including NAE and NUE was achieved at NR 4.8 g N pot$^{-1}$ under ST2 possibly due to the level of N matched with optimal ST and crop N requirement that was attained. Our results are in line with the findings of Yousaf et al. [54] who observed that maximum N efficiencies were observed in rice and oilseed crop rotations when the N level matched the N requirements of crops. Higher N uptake and enhanced N efficiencies under ST2 were favored in improved WUE and resulted in higher WUE under ST2. Results for enhancement in WUE and NUE in adjustment to ST were also supported by previous studies [7].

The findings of the present study indicate the importance of and are the supporting evidence for, the need for proper N management according to various ST for upland rice production in Thailand. Our study indicates that N fertilization and various NR applied under different ST produced significantly improved results for upland rice productivity. Therefore, N application practices [14–18], as well as N fertilizer recommendations based on soil analysis and soil nutrient status [19,20] and location-specific recommendations [21], are needed to be modified and improved according to various ST. However, further investigations in this field are needed to achieve more precise optimization of NR and ST for upland rice in Southern Thailand as, in the present study, soil moisture was constantly and sufficiently supplied whereas climatic conditions and rainfall variability differs under various ST in the field conditions. In addition, N uptake and utilization is not only influenced by prevailing climatic conditions, soil moisture status and NR or N availability but also varies among various genotypes of the same plants [55–58]. It was observed that high genetic variability and variation among agronomic traits prevailed amongst numerous Thai upland rice genotypes [14–17] including the studied genotype Dawk Pa–yawm. Therefore, the authors suggest that it becomes necessary to include other major upland rice genotypes being cultivated in Southern Thailand for future field investigations.

## 5. Conclusions

Ideal agronomic management of upland rice is an important strategy to enhance productivity and enhance resource input efficiency. The identification and application of optimal NR synchronized with ST are some of the principal elements of this strategy. Results obtained from the current study exhibit the significance of the optimal NR and its synchronization with ST as it was indicated that NR and ST influenced growth, productivity, nitrogen use efficiencies and WUE of upland rice. An increase in NR indicated an increased performance of yield and yield attributes. In addition to grain yield, NR and ST significantly influenced N uptake, NAE, NUE and WUE. Maximum performance for yield, yield attributes and WUE was achieved at 4.8 g N pot$^{-1}$. However, the highest plant and grain N uptake and N use efficiencies were achieved at 3.2 g N pot$^{-1}$. Considering the impact of ST, the maximum performance for yield, grain N uptake, N use efficiencies and WUE were achieved under ST2. Based on the findings of this study, and from a practical point of view, the application of 4.8 g N pot$^{-1}$ (90 kg N ha$^{-1}$) and sowing in the month of September (ST2) would enhance upland rice production. Though field investigations for current study should be considered prior to general recommendations. Furthermore, it is recommended that future experiments should investigate more upland rice genotypes, more NR gradients and ST intervals under field conditions for improved and precise NR optimization according to ST and recommendations.

**Supplementary Materials:** The following are available online at https://www.mdpi.com/article/10.3390/su14041997/s1, Table S1. Mean squares of ANOVA of yield and yield attributes of Dawk Pa–yawm, straw N uptake, grain N uptake, total N uptake and water use efficiency during 2018–2019. Table S2. Mean squares of ANOVA of yield and yield attributes of Dawk Pa–yawm, straw N uptake, grain N uptake, total N uptake and water use efficiency during 2019–2020. Figure S1: Study area at Prince of Songkla University, Songkhla in Southern Thailand (Source: adapted from ArcGIS: *v*10.5).

**Author Contributions:** Author T.H. and S.D. conceived the idea and T.H. conducted experiments. N.H. helped in experiments and data collection. T.H. analyzed the data and prepared the first draft. C.N., M.A. and S.D. edited the manuscript. S.D. supervised and contributed in finalized version and M.A. proofread the manuscript. All authors have read and agreed to the published version of the manuscript.

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