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Development Concept and Early Demonstration of Nitrogen-Use-Efficient Rice Varieties

Mitsuhiro Obara, Kazuhiro Sasaki



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Preface

The Asia-Monsoon region accounts for approximately 90% of the world's paddy rice cultivation, and is critical for food security. Improving rice productivity has become an urgent challenge owing to population growth and rising food demand. Despite high-yield strategies that rely on the heavy application of chemical nitrogen fertilizers being pursued for several years, their environmental consequences are becoming increasingly apparent. The consequences include groundwater pollution, greenhouse gas emissions, pests, and diseases.

Crops absorb approximately half of the applied nitrogen. The remainder leaches into the soil or is released into the atmosphere. Particularly, nitrous oxide has a greenhouse effect approximately 300 times greater than that of carbon dioxide; therefore, its impact on climate change as an agricultural emission source is of concern. Furthermore, methane emissions from paddy fields tend to increase during the vegetative growth period. Thus, revising fertilization systems and growth periods is crucial to reduce greenhouse gas emissions.

This report introduces the concept of nitrogen-use-efficient rice varieties and presents preliminary findings. These methods are based on the current status and challenges of fertilizer application in the Asia-Monsoon region. The proposed technology improves the nitrogen use efficiency by utilizing photosynthesis during the reproductive stage. This facilitates decreased environmental impact and increased productivity.

The goal of decreasing fertilizer application while maintaining and enhancing yields is rooted in a commitment to the future of regional agriculture and the environment.

Additionally, this report addresses the challenges and prospects of adopting this technology, and we hope that it will contribute to the transition to environmentally harmonious agriculture.

Abstract

For several years, excessive chemical nitrogen fertilizers have been applied to the Asia-Monsoon region. Consequently, environmental challenges, such as groundwater pollution and greenhouse gas emissions, which sometimes lead to pest and disease proliferation, have emerged, making a shift toward sustainable rice cultivation imperative. One key technological challenge in achieving this is enhancing the nitrogen use efficiency (NUE). This report introduces technological developments that aim to achieve high yields with minimal fertilizer application, presents preliminary demonstration findings, and discusses future prospects.

Fertilizer management and breeding are two major methods for enhancing NUE in rice cultivation. The latter can have substantial potential for social acceptance and widespread adoption if the development of varieties considers regionally specific environmental conditions.

For instance, dry matter accumulation tends to stagnate during the heading stage in environments such as the Asia-Monsoon region, which is characterized by high temperature, high humidity, and frequent overcast conditions. Consequently, assimilation via photosynthesis during the reproductive stage plays a critical role in the yield. Therefore, developing varieties that maintain dry matter production during the reproductive stage is an effective strategy for enhancing NUE.

While early heading has traditionally been considered detrimental to yield potential in breeding programs, recent breeding strategies have actively used early heading to avoid weather stresses such as high temperatures and drought during the reproductive stage. Furthermore, a shorter cultivation period resulting from early heading can decrease methane emissions and conserve irrigation water.

Based on the hypothesis that efficient photosynthesis to maintain dry matter production during the reproductive stage results in enhanced NUE, we developed lines introducing early heading genes as part of the JIRCAS project (FY2016–FY2021). We developed NR160E, using the high-yielding Philippine variety NSIC Rc 160 as a background, by introducing an early heading QTL. Field trials in Japan indicated that NR160E maintained yield even at half the fertilizer application rate.

Enhanced physiological NUE (PNUE) can be achieved by promoting photosynthesis during the reproductive stage and improving dry matter accumulation and yield components. However, the effect of the early heading gene in NR160E may be limited in low-latitude regions. Thus, using high-yielding varieties suited to each region's cultivation conditions as a genetic background is crucial when disseminating early heading technology in the Asia-Monsoon regions.

In response to these difficulties, JIRCAS developed EHD10 and EHD16, which introduced various genes that produce early heading in low-latitude regions into the major variety Ciherang in Indonesia. Furthermore, preliminary verification tests in farmers' fields was conducted as part of the Green Asia Project. Under the recommended fertilization conditions, both lines showed yields 22%–25% higher than that of Ciherang and decreased nitrogen use by approximately 20%. Additionally, the yields of EHD10 and EHD16 were maintained even under no-fertilization conditions, overcoming the conventional problem of reduced yields owing to early heading.

These preliminary findings indicate the potential for improved NUE by developing early heading varieties tailored to regional environmental conditions. Nevertheless, further evidence is necessary to promote the revision of fertilizer application regimes and the transformation of fertilization systems in the Asia-Monsoon region.

1. Current Status and Challenges of Nitrogen Fertilizer Use in Rice Cultivation in the Asia-Monsoon Region

1.1: High-Yield Strategies and Environmental Impacts in the Asia-Monsoon Region

Intensive agriculture centered on rice cultivation is widely practiced in the Asia-Monsoon region. The region accounted for nearly 85% of the world's paddy harvested area and nearly 90% of global rice production in 2022 (FAO, 2025). The major producing countries include China, India, Bangladesh, Indonesia, Vietnam, Thailand, Myanmar, and the Philippines. These countries account for the majority of the world's population and comprise cultural spheres in which rice is the staple food.

Rice cultivation in this region plays an indispensable role in global food security. It is crucial that rice production in this region be maintained and strengthened to meet the global food demand and continuous increase in global population. Therefore, agricultural technologies and fertilization systems developed in this region should be viewed as global challenges.

For a long time, rice cultivation in this region has relied on a high-yield strategy involving heavy chemical fertilizer use. Although this technique increased yields in the short term, it has caused serious long-term issues, such as increased environmental impact and decreased resource utilization efficiency.

Crops absorb approximately 50% of the applied nitrogen, and the remainder either leaches into groundwater as nitrate in the soil or is released into the atmosphere as nitrous oxide (IPCC 2021), which is a potent greenhouse gas. It has a global warming potential approximately 300 times that of carbon dioxide. Moreover, emissions from agriculture substantially contribute to climate change.

Furthermore, excessive nitrogen application disrupts nutrient balance in crops and promotes pest and disease outbreaks. These phenomena have been reported globally and studies have indicated that they can be mitigated by enhancing nitrogen use efficiency (NUE) and introducing new crop varieties (Tyagi et al., 2022).

In light of this, ensuring the sustainability of rice cultivation in the Asia-Monsoon region requires re-assessing conventional high-yield strategies, developing technologies that enhance the efficiency of nitrogen fertilizer use, and shifting fertilization systems.

1.2: Synthesis of Chemical Nitrogen Fertilizers and Environmental Impact

Ammonia, the main ingredient in nitrogen fertilizers, is produced using the Haber-Bosch process. This technology, established in the early 20th century, produces

ammonia by reacting atmospheric nitrogen (N_2) and hydrogen (H_2) at high temperatures and pressures. Nearly all nitrogen fertilizers globally are still produced this way, requiring temperatures of approximately $350^\circ C$ and pressures of between 250 and 350 atmospheres. Producing one ton of ammonia requires at least 27–32 gigajoules (GJ) of energy, and the manufacturing process is only 65% efficient. The high energy consumption of this process results in dependence on fossil fuels and greenhouse gas emissions, which substantially affect the environment.

Recently, the electrochemical nitrogen reduction reaction (NRR) has gained traction as a technology for synthesizing ammonia, which uses less energy than the Haber–Bosch process. NRR uses renewable energy to supply protons from water and decreases nitrogen to ammonia. The NRR is expected to markedly decrease environmental impacts because it does not emit greenhouse gases and can proceed at ambient temperature and pressure (Jesudass et al., 2023).

However, this technology remains in the research and demonstration phases and has not yet reached implementation at the industrial scale. The reaction rate of the NRR is slow, and the catalytic performance required to cleave the triple bond of nitrogen molecules has not been sufficiently established. Additionally, the competing hydrogen evolution reaction (HER) tends to be prioritized, leading to an extremely low ammonia production efficiency (Faraday efficiency). Overcoming these challenges needs several technological breakthroughs including catalyst design, electrolyte improvement, and reaction system optimization. Several years to a decade of development is estimated to be required before practical applications become feasible (Jesudass et al., 2023).

Even if low-energy manufacturing technologies become practical, they will not solve environmental problems unless fertilizer usage changes. Thus, technological development, including cultivar development that enhances NUE and the transformation of fertilization systems, will remain a critical challenge even if nitrogen fertilizer manufacturing technology advances to the subsequent stage.

1.3: Improving NUE

In the Asia-Monsoon region, heavy application of chemical nitrogen fertilizers has long been utilized to increase crop yields. Although this has achieved short-term increases in crop yields, as discussed in Section 1.1, it has additionally brought serious challenges, such as increased environmental impacts and decreased resource use efficiency. Furthermore, nitrogen fertilizer production is highly energy-intensive and contributes to greenhouse gas emissions, as mentioned in Section 1.2.

In light of this situation, solely relying on advances in nitrogen fertilizer manufacturing technology is insufficient to ensure the sustainability of rice cultivation in the Asia-Monsoon region. The immediate challenge requiring on-site action is the development of technologies to enhance NUE.

Multiple techniques for improving NUE have been found. One approach is to optimize the fertilizer management. Optimizing the amount, timing, and method of fertilizer application improves the crop nitrogen uptake efficiency and suppresses runoff and emissions. Another approach is to improve physiological- and breeding-based NUE via crop breeding. Recently, several physiological and breeding approaches have been reported to enhance NUE. Genetic modifications can improve photosynthetic capacity, maintain dry matter production during the reproductive stage, increase yield, and improve NUE (Yoon et al., 2020). Furthermore, increasing the thousand grain weight via conventional breeding enhances the harvest index (HI) and increases the yield relative to nitrogen uptake (Obara et al., 2022; Yoon et al., 2022).

Although these technologies have been shown to improve NUE effectively, they cannot be widely adopted in all regions. Genetically modified crops may experience cultivation restrictions owing to national regulations, and the increase in thousand-grain weight depends on consumer preferences and market acceptance. Thus, developing approaches that can be widely adopted across the entire Asia-Monsoon region needs breeding efforts that consider the regional cultivation environments and social acceptability.

In environments such as the Asia-Monsoon region, which is characterized by high temperature, high humidity, and frequent overcasting, dry matter accumulation tends to stagnate during the heading stage. Consequently, assimilation via photosynthesis during the reproductive stage is critical for yield. Therefore, developing varieties that can maintain dry matter productivity during this stage is an effective strategy for improving NUE.

This technological development is key to achieving decreased environmental impact and stable food production. The next section discusses policy trends that support this development and the shift toward environmentally harmonious agriculture in the Asia-Monsoon region.

1.4: Policies for Environmentally Harmonious Agriculture

In the Asia-Monsoon region, the increasing environmental impact of excessive nitrogen fertilizer application has become a serious problem. In response, countries have shifted their policies toward environmentally harmonized agriculture in recent years. In Japan, the “MIDORI Strategy for Sustainable Food Systems” was formulated in 2021, setting a target to decrease chemical fertilizer use by 30% by 2050 (MAFF, 2021). Additionally, the “Japan-ASEAN Green Cooperation Plan” was formulated, incorporating collaborative projects that leverage new technologies and innovations developed in Japan in cooperation with ASEAN countries. Cooperation and collaboration between ASEAN countries and Japan are advancing toward “Building resilient and sustainable agriculture and food systems through innovation.”

For instance, in Indonesia, the Ministry of Agriculture is promoting the “Smart Farming 4.0” concept and advancing the modernization of agriculture by utilizing advanced technologies such as artificial intelligence, IoT, drones, and satellite imagery. The government is formulating a roadmap for smart agriculture and implementing pilot projects as part of its national strategic project for 2020-2024, with demonstrations conducted in several regions (Ministry of Agriculture, Indonesia, 2020).

Indonesia has diverse climates, soils, and agricultural structures and holds a globally crucial position as a demonstration site for environmentally friendly agricultural technologies. The findings obtained in Indonesia via the Green Asia Project are expected to spread throughout the Asia-Monsoon region, accelerating the transition to sustainable agriculture through technology optimization according to regional characteristics and policy support.

Based on this policy background and regional challenges, this report introduces technologies for improving NUE via rice breeding as part of technological development to enhance NUE. This is a crucial initiative that should be pursued along with the transformation of fertilization systems, and is positioned as a practical solution to support the transition to environmentally friendly agriculture. Chapter 2 details specific technologies. Chapter 3 presents the verification findings and analysis of NUE improvement technologies in Indonesia. Chapter 4 outlines future research topics, and Chapter 5 provides an outlook.

2. Developed Technologies for Improving NUE in Rice

2.1: Background and Overview of Technological Development

This chapter introduces the NUE improvement technologies via variety breeding using early heading and addresses the challenges outlined in Chapter 1. In particular, we developed an isogenic line, NR160E (Figure 1), by introducing early heading QTLs into the genetic background of the high-yielding variety, NSIC Rc 160 (NR160), which is widely cultivated in the Philippines (Sasaki et al., 2026). The Philippine Rice Research Institute developed NR160, a high-yielding variety, and it is a representative variety with high domestic adoption rates.

NR160E aimed to simultaneously enhance dry weight, harvest index (HI), and NUE by promoting early heading and enabling efficient photosynthesis during the reproductive stage. Cultivation trials were conducted at the JIRCAS experimental station in Tsukuba, Japan, as described in Section 2.3. NR160E achieved yields equivalent to or higher than those of high-fertilizer-applied NR160 (9.6 g N m^{-2}) even with half the fertilizer rate (4.8 g N m^{-2}). This fertilizer-reduction effect was consistently noted at several planting densities.

Thus, NR160E is a technology capable of maintaining high yields with decreased fertilizer inputs, representing an important policy-relevant achievement that simultaneously reduces the environmental impact and enhances resource efficiency.

The NR160E used in this report was developed under Project (FY2016-FY2021) at JIRCAS. The findings presented in Sections 2.2 and 2.5 were obtained under the Green Asia Project.

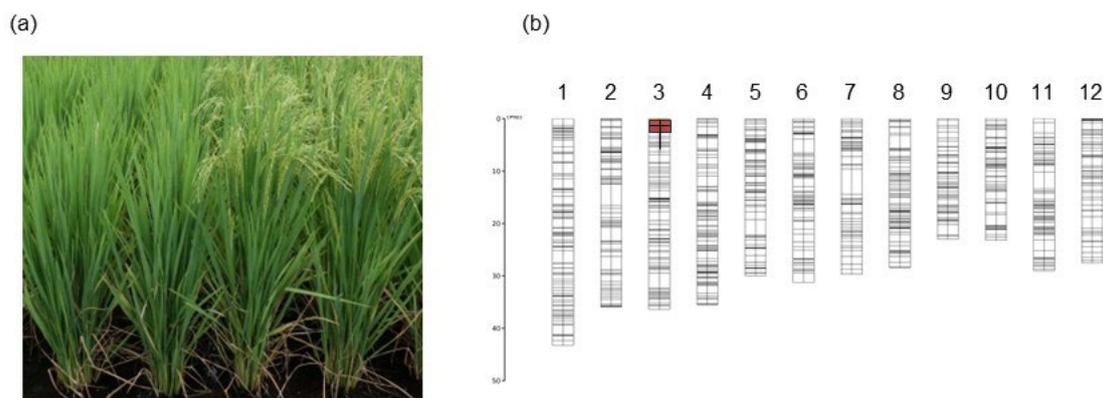


Figure 1. Characteristics of the early heading isogenic line NR160E

a: Plant appearance at heading stage NSIC Rc 160 (NR160, left) and NR160E (right). b: NR160E genotype. Rice's 12 chromosomes are indicated as vertical columns, with positions analyzed by DNA marker indicated as horizontal lines within the columns. Regions shared with the NR160 genetic background are shown in white and differing regions in orange.

2.2: Effect of Early Heading on Dry Matter Accumulation

In rice, the photosynthetic capacity temporarily stagnates around the heading stage, tending to suppress dry matter accumulation. This is owing to the worsened light reception caused by the canopy closure and physiological alterations linked to the transition from vegetative to reproductive growth. Yoshida (1981) pointed out that photosynthetic stagnation during heading is the rate-limiting factor in yield formation.

Traditionally, early heading has been considered detrimental to yield potential and has been avoided in breeding programs (Yoshida et al., 1972). However, recent breeding strategies actively use early heading to avoid weather stresses, such as high temperatures and drought, during the reproductive stage. For instance, reports have described cases in which the introduction of the *Ef-cd* gene shortened the maturation period while maintaining or increasing yield (Fang et al., 2019). However, this report provided limited descriptions of the physiological basis for yield enhancement or fertilizer reduction effects, and sufficient evidence for its use as a fertilizer-reduction technology has not been shown.

The NR160E developed in this study showed heading approximately 8–10 days earlier than the NR160. It effectively accumulated dry matter by maintaining photosynthesis during the reproductive stage. Although NR160E demonstrated a lower crop growth rate (CGR) than NR160 during the early growth stage, its CGR from just before heading to the reproductive stage exceeded that of NR160, sustaining dry matter production during grain filling (Figure 2). The final above-ground dry weight showed minimal difference between the two lines, confirming a growth pattern that compensates for the disadvantages of dry matter accumulation caused by early heading.

These findings support the hypothesis that early heading prevents photosynthetic stagnation around the heading stage and maintains dry matter production during the reproductive stage. This suggests that it contributes to improved NUE by enhancing dry matter weight.

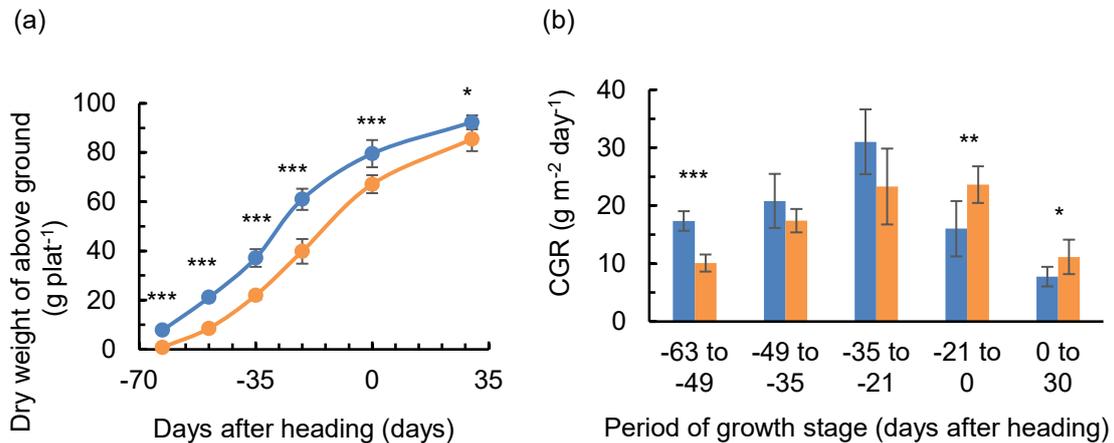


Figure 2. Changes in the above-ground dry weight (a) and crop growth rate (CGR) (b) throughout the life cycle

Evaluated at six time points relative to heading: 63 days before heading, 49 days before heading, 35 days before heading, 21 days before heading, heading day, and 30 days after heading. NR160 is indicated as blue plots or columns, and NR160E as orange plots or columns. Statistical comparisons between NR160 and NR160E were conducted at each time point using one-way analysis of variance (ANOVA) ($n = 6$). Asterisks indicate significant differences (*: $P < 0.05$, **: $P < 0.01$, ***: $P < 0.001$).

2.3: Impact of Early Heading on Fertilizer Application

As indicated in Section 2.2, early heading facilitates the efficient use of photosynthesis during the reproductive stage, thereby maintaining dry matter accumulation and optimizing the formation of yield components. This section presents the test findings from the JIRCAS experimental station in Tsukuba, Japan. Here, the effects of these physiological improvements on actual yield and HI under various fertilization conditions were investigated.

First, when comparing yields under the same fertilization conditions, NR160E consistently outperformed NR160 (Figure 3). Especially under the 9.6 g N m^{-2} condition (equivalent to 96 kg N ha^{-1}), NR160E yielded $922.9 \pm 57.7 \text{ g m}^{-2}$, whereas NR160 yielded $692.3 \pm 49.0 \text{ g m}^{-2}$, revealing a significant difference. These results show the technical basis for enhancing yields without increasing fertilizer

application rates, representing a substantial empirical achievement in the implementation of fertilizer-reduction policies. This 9.6 g N m⁻² fertilization condition aligns with the fertilization levels (approximately 90–140 kg N ha⁻¹) in intensive rice cultivation in the Asia-Monsoon region, facilitating comparisons based on the regional realities (Coggins et al., 2025).

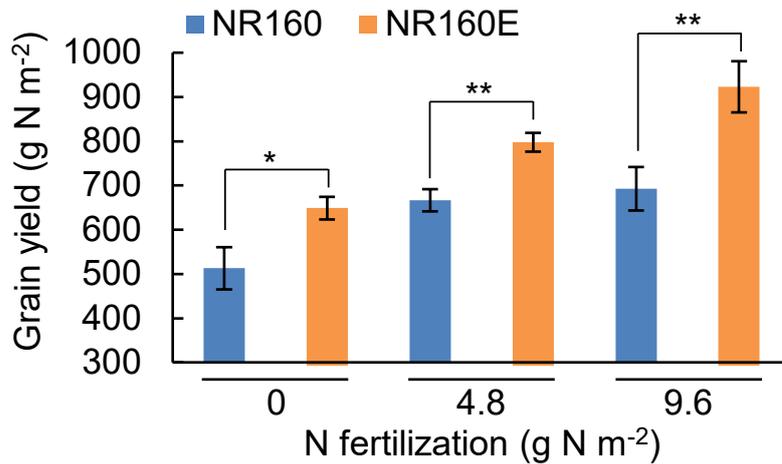


Figure 3. Comparison of the grain yield between NR160 and NR160E cultivated under various fertilizer application rates

Column values indicate the mean \pm standard deviation ($n = 4$). Comparisons between NR160 and NR160E were performed within each fertilization treatment (0, 4.8, and 9.6 g N m⁻²) and field stratum using Welch's t-test, followed by FDR adjustment (family size = 12) with the Benjamini-Hochberg procedure (* $q < 0.05$; ** $q < 0.01$). In the figures, NR160 is shown in blue and NR160E in orange. Plants were grown in Tsukuba at a JIRCAS field under a planting density of 18.5 plants m⁻².

Comparing the HI under the same fertilization conditions, NR160E revealed 0.513 ± 0.004 at 4.8 g N m^{-2} , whereas NR160 revealed 0.423 ± 0.012 , confirming a significant difference (Table 1). Rather than an increase in the dry weight of the aboveground parts, an enhancement in HI was achieved via comprehensive enhancement of the yield components.

Table 1. Comparison of the agronomic traits and yield components between NR160 and NR160E cultivated under various fertilizer application rates

Trait	N fertilization (g N m^{-2})	NR160 ^a	NR160E ^a	Significance ^b
Days to heading	0	120.5 ± 0.6	112.5 ± 1.3	***
	4.8	117.0 ± 0.8	107.8 ± 0.5	***
	9.6	118.5 ± 1.0	109.0 ± 0.8	***
Aboveground dry weight (g m^{-2})	0	$1,146 \pm 56$	$1,154 \pm 37$	
	4.8	$1,443 \pm 51$	$1,394 \pm 27$	
	9.6	$1,739 \pm 65$	$1,693 \pm 74$	
Agronomical HI	0	0.423 ± 0.021	0.511 ± 0.015	***
	4.8	0.423 ± 0.012	0.513 ± 0.004	***
	9.6	0.397 ± 0.020	0.501 ± 0.012	***

^a Values indicate mean \pm standard deviation ($n = 4$).

^b Asterisks indicate significant differences between lines within each nitrogen-field stratum (4 planting densities \times 3 nitrogen levels), evaluated using Welch's t-test with false discovery rate (FDR) correction (Benjamini-Hochberg, family size = 12): * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$.

2.4: Mechanism Behind Equivalent Yields with Lower Fertilizer

The ability of NR160E to achieve high yields even with low fertilizer application was attributed to the differences in the responsiveness of the yield components to absorbed nitrogen. The findings suggest that NR160E achieves higher yields mainly via improved grain filling and thousand-grain weight, rather than the grain number.

When we compared the relationship between grain number and absorbed nitrogen (data not shown), the NR160 and NR160E lines showed an increase in grain number corresponding to nitrogen absorption, with no significant difference noted in the slope of the regression line (covariance analysis, difference in slope $P = 0.288$). Thus, both lines showed similar nitrogen responsiveness with respect to grain formation.

Comparing the relationship between yield and absorbed nitrogen, NR160 and

NR160E lines revealed increased yields with increasing nitrogen absorption (Figure 4). Unlike the relationship between grain number and absorbed nitrogen, NR160E demonstrated higher yields than NR160, with a significant difference in the regression line slope (covariance analysis, slope difference $P = 0.006$). This indicates that NR160E could produce higher yields with the same nitrogen uptake, demonstrating an improvement in physiological NUE (PNUE).

The improvement in PNUE for NR160E is thought to be achieved via an increased yield resulting from the maintenance of photosynthesis during the reproductive stage owing to early heading, which allows absorbed nitrogen to be efficiently converted into grain formation.

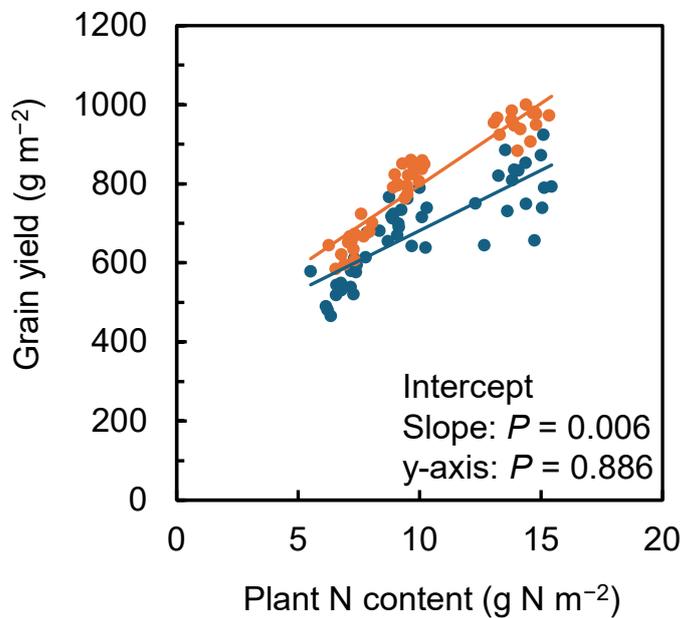


Figure 4. Relationship between absorbed nitrogen and yield in NR160 and NR160E

For each line, 48 datasets were plotted revealing the relationship between absorbed nitrogen in the aboveground portion at maturity and yield. Rice was grown under 12-plot conditions combining various nitrogen fertilizer rates (0, 4.8, and 9.6 g N m⁻²) and planting densities (18.5, 22.2, 37.0, and 44.4 plants m⁻²). NR160 is indicated as blue plots, and NR160E as orange plots. The linear regression lines for each line are shown. Covariance analysis was conducted to compare the slopes and intercepts of the regression lines between NR160 and NR160E.

2.5: Adaptability, Versatility, and Limitations of Technology for Policy Use

NR160E maintained early heading under long-day conditions, suggesting its potential adaptability to high-latitude regions (Figure 5).

Conversely, under short-day conditions, the heading difference between NR160 and NR160E decreased, indicating that the effect of the early heading gene in NR160E may be limited to low-latitude regions. Such constraints on regional deployment can be overcome by selecting and combining the introduced early heading genes.

Additionally, a key consideration for the widespread adoption of early heading technology is the use of high-yield varieties suited to local cultivation conditions as the genetic background. Introducing early heading genes into varieties that are already widely cultivated improves technology implementability and acceptability. Additionally, this makes it easier to overcome social factors such as consumer preferences and market acceptance via technological means.

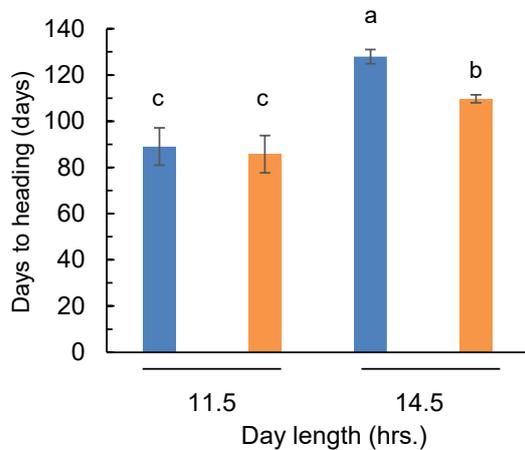


Figure 5. Days to heading of the NR160 and NR160E lines under varying photoperiods

Each line was grown under two different photoperiods (11.5 and 14.5 hours) in a climate chamber, and the heading date was recorded ($n = 10$). NR160 is indicated as blue columns, and NR160E as orange columns. Statistical analysis was conducted using Tukey's test. Different letters (a–c) indicate significant differences at $P < 0.05$.

3. Pilot Evaluation of NUE-Enhanced Rice Developed for Indonesia

3.1: Development of NUE-Enhanced Lines Derived from Ciherang

Sustainable rice production in the Asia-Monsoon region requires the development of enhanced technologies based on locally adopted high-yielding varieties. Section 2.5 emphasized the criticality of optimizing and verifying technologies tailored to regional cultivation environments and policies. Therefore, the genetic background used for verification and the early heading genes introduced were found to be key to their widespread adoption.

Ciherang is a widely adopted high-yield variety in Indonesia. Ciherang boasts a higher adoption rate than IR64 and combines traits such as a short culm, good grain quality, and several panicles, making it highly acceptable to farmers. Thus, this study developed EHD10 and EHD16 using the widely adopted major variety Ciherang as the genetic background and introduced various early heading gene donors. Both lines incorporated distinct genes for early heading in low-latitude regions.

After the crosses between Ciherang and early heading gene donors, four backcrosses were conducted using Ciherang as the pollen parent for EHD10 development and three backcrosses for EHD16 development. Through DNA marker selection, EHD10 and EHD16 were developed with genotypes that were > 99% identical to those of Ciherang.

The EHD10 and EHD16 used in this report were developed under the Project (FY2016-FY2021) in JIRCAS. All findings presented in this chapter were achieved under the Green Asia Project.

3.2: NUE-Improving Lines with Ciherang as the Background

EHD10 is an earlier heading variety, and cultivation trials at the JIRCAS experimental station in Tsukuba have confirmed its early heading and high harvest index (HI). Similarly, EHD16 plants showed early heading and high HI. Both lines were expected to enable efficient nitrogen use and maintain or increase yield by maximizing photosynthetic use during the reproductive stage.

These lines provide a technological foundation for simultaneously achieving decreased environmental impacts and increased productivity by introducing early heading traits into regional mainstay varieties and revising fertilization systems.

3.3: Methods and Trait Measurement Methods for Verification Trials

Verification trials were performed in the 2024 rainy season and the 2025 dry season in the farming fields in Parawad Village, Cikarawa District, Subang Regency, West Java Province, Indonesia. The trials included three rice varieties: Ciherang, EHD10,

and EHD16. Two fertilization treatments were applied: a no-fertilizer control and a fertilizer application following the Indonesian government's recommended rate (138 kg N ha⁻¹). Each treatment was replicated four times. Cultivation followed the farmers' customary practices throughout the period from sowing to harvest, and the key traits such as heading date, maturity date, and yield were assessed.

Heading and maturity dates were recorded as the number of days after sowing. The yield was calculated by measuring the weight of the harvested paddy from each plot, determining the paddy moisture content at harvest, and converting it to 14% moisture. The nitrogen requirement per unit grain yield (NGY) was calculated by dividing the amount of nitrogen applied by grain yield (N kg/ton GY).

Statistical analysis was performed using the Mann–Whitney U test for grain yield and NGY and Tukey's multiple comparison test for grain yield components, with $P < 0.05$ as the significance level.

3.4: Heading and Maturity Performance of NUE-Enhanced Lines in a Pilot Trial

This section presents the findings of comparing the days to heading and maturity between the major regional variety Ciherang and the NUE-improved lines EHD10 and EHD16.

During the 2024 rainy season, rice growth was delayed owing to infection with the tungro virus complex, and the subsequent appearance of tungro disease symptoms was noted approximately 30 d following transplanting. Although symptomatic plants were removed and cultivation continued, symptoms spread throughout the cultivation plot. Consequently, heading was delayed, and the number of twisted grains was extremely low, resulting in the abandonment of trait measurements.

During the 2025 dry season, rice grew healthily with no noted pests or diseases. EHD10 headed and matured the earliest under both fertilization treatments, followed by EHD16, and finally, Ciherang (Table 2). Particularly, the number of days to heading without fertilization was 66.3 days for EHD10, 72.0 days for EHD16, and 76.0 days for Ciherang. A similar trend was noted under the recommended fertilization treatments. Regarding the days to maturity, EHD10 matured approximately 13–14 days earlier than Ciherang, whereas EHD16 matured approximately 5–7 days earlier than Ciherang. These findings indicate that early heading of EHD lines shortens the growing period, maximizes photosynthetic use during the reproductive stage, and potentially helps avoid weather stress (high temperatures and drought).

Table 2. Days from sowing to flowering and harvest for the major Indonesian rice variety Ciherang and its NUE-improved line, cultivated under no fertilization and recommended fertilization conditions

Trait	Nitrogen fertilization (kg Urea ha ⁻¹)	Ciherang ^a	Letter ^b	EHD10 ^a	Letter ^b	EHD16 ^a	Letter ^b
Days to flowering	0	76.0 ± 0.0	a	66.3 ± 4.0	b	72.0 ± 0.0	b
	300	76.0 ± 0.0	a	67.3 ± 3.3	b	72.0 ± 0.0	b
Days to maturity	0	108.5 ± 1.0	a	95.5 ± 5.2	c	103.0 ± 0.0	b
	300	110.0 ± 0.0	a	96.3 ± 4.7	b	103.0 ± 0.0	b

^a Values indicate mean ± standard deviation (n = 4).

^b Different letters within each trait indicate statistically significant differences between treatments based on the Mann–Whitney U test ($P < 0.05$). Rice was cultivated in a farmer's field in Subang Regency, West Java Province, Indonesia, during the 2025 dry season. 300 kg Urea ha⁻¹ is equivalent to 138 kg N ha⁻¹.

Thus, the early heading genes introduced into EHD10 and EHD16 express early heading in the Ciherang genetic background, even in low-latitude regions.

Additionally, EHD10 and EHD16 can be widely used as maternal parents in future development of Indonesian rice breeding. Ciherang is widely used in Indonesian rice variety development, and several Inpari varieties with Ciherang as the parent have been released. For instance, the cultivation area for Inpari32 has recently expanded. Since the molecular mechanisms controlling heading (flowering) in Ciherang and Inpari32 are thought to be similar, it is highly plausible that the early heading genes introduced into EHD10 and EHD16 will also confer early heading in modern Indonesian varieties.

3.5: Yield Performance under Different Fertilization Conditions

The yields of Ciherang and the NUE-improved lines EHD10 and EHD16 were compared in the 2025 dry season cultivation trial. Under no-fertilizer conditions, yields were 5.55 ± 0.39 tons ha⁻¹ for Ciherang, 5.86 ± 0.71 tons ha⁻¹ for EHD10, and 5.91 ± 0.89 tons ha⁻¹ for EHD16, with no significant differences noted among the three varieties (Figure 6). Under the recommended fertilization conditions, however, Ciherang yielded 6.51 ± 0.56 tons ha⁻¹, whereas EHD10 yielded 7.97 ± 0.54 tons ha⁻¹ and EHD16 yielded 8.17 ± 0.32 tons ha⁻¹, both significantly higher than Ciherang. EHD10 revealed an approximately 22% increase in yield compared to Ciherang, whereas EHD16 achieved an approximately 25% increase.

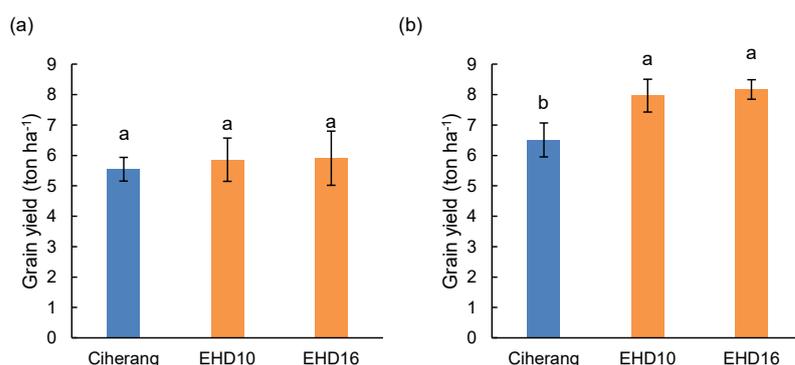


Figure 6. Yields of the major Indonesian rice variety Ciherang and its NUE-improved line cultivated under no fertilizer conditions (a) and the recommended fertilizer rate of 300 kg urea ha⁻¹ (b).

Values represent the mean \pm standard deviation ($n = 4$). Different letters on the same column indicate statistically significant differences between treatments based on the Mann–Whitney U test ($P < 0.05$). Rice was cultivated in the dry season of 2025 at a farmer's field in Subang Regency, West Java Province, Indonesia. 300 kg Urea ha⁻¹ corresponds to 138 kg N ha⁻¹.

These findings indicate that EHD10 and EHD16 can efficiently use nitrogen under fertilized conditions and consistently achieve yields exceeding those of Ciherang. Additionally, the maintenance of yield, even under no-fertilizer conditions, suggests that early heading varieties have overcome the traditional challenge of low yield. These results indicate that NUE improvement technology through early heading represents a significant achievement in Indonesia's cultivation environment, showing enhanced NUE and production stability under low-input conditions.

Increases in yield under fertilized conditions improve farmer profitability and contribute to decreased environmental effect via optimized fertilization systems. In particular, introducing high-yielding, high-NUE lines such as the EHD line represents a key technological option for future breeding and extension strategies, enabling fertilizer cost reduction and productivity gains.

3.6: Verification of the Effect of Reducing Chemical Nitrogen Fertilizer Use

This section compares the nitrogen requirement per unit grain yield (NGY: N kg/ton GY) required to produce 1 ton of paddy rice for the main regional variety Ciherang and the NUE-improved lines EHD10 and EHD16 in fields in Subang District, West Java Province, Indonesia, and evaluates their nitrogen fertilizer reduction effects.

Under the recommended fertilization conditions, NGY was 21.3 ± 1.8 N kg/ton GY for Ciherang, 17.4 ± 1.2 N kg/ton GY for EHD10, and 16.9 ± 0.7 N kg/ton GY for EHD16 (Table 3). Thus, EHD10 achieved an approximately 18% reduction in nitrogen use compared to Ciherang, whereas EHD16 achieved an approximately 21% reduction.

Table 3. Comparison of the chemical nitrogen fertilizer requirements and reduction rates

Trait	Ciherang ^a	Letter ^b	EHD10 ^a	Letter ^b	EHD16 ^a	Letter ^b
NGY (N kg / ton GY)	21.3 ± 1.8	a	17.4 ± 1.2	b	16.90 ± 0.7	b
Reduction ratio of NGY for Ciherang	0.00 ± 0.09	b	0.18 ± 0.06	a	0.21 ± 0.03	a

^a Values indicate mean ± standard deviation (n = 4).

^b Different letters within each trait indicate statistically significant differences between treatments based on the Mann–Whitney U test (P < 0.05). Rice was cultivated in 2025 during the dry season at a farmer's field in Subang Regency, West Java Province, Indonesia. 300 kg Urea ha⁻¹ is equivalent to 138 kg N ha⁻¹. NGY: Nitrogen requirement per unit grain yield.

This reduction shows that increasing the yield while decreasing nitrogen application rates is possible, confirming the effectiveness of NUE-enhancing technologies. Notably, EHD16 achieved markedly higher yields than Ciherang while substantially decreasing nitrogen requirements and successfully combining high yields with fertilizer reduction.

While these trial findings are based on the data from a limited region and only during the dry season, the benefits of NUE-improved rice can be expected, at least during the dry season, in neighboring areas with similar cultivation environments. Thus, implementation by farmers in neighboring regions is considered feasible. Implementation, that is, cultivation by farmers, requires variety registration for EHD10, EHD16, etc., and preparations for this are currently underway.

4. Future Research Topics

4.1: Multi-site validation and tuning

This chapter presents research topics for the further development of NUE improvement technologies based on the verification results to date. Key research topics include the requirement of multilocation trials with an eye toward adoption within Indonesia, decision-making techniques to estimate suitable rice fields for technological applications, and elucidation of physiological mechanisms.

EHD10 and EHD16 showed characteristics of early heading, high harvest index, and high NUE in trials in the Tsukuba field and in the Subang Regency, West Java, Indonesia. Although these findings were verified under limited environmental conditions, implementation by farmers in neighboring areas was considered feasible. For widespread adoption, multilocation demonstration trials under various climatic and soil conditions are warranted.

Furthermore, verification tailored to local conditions is required regarding the impact of the differences in fertilization systems and planting density on yield components. For instance, in Indonesia, the government-recommended fertilizer rate is 138 kg N ha^{-1} ($300 \text{ kg Urea ha}^{-1}$). However, actual farmer application rates significantly vary by region and economic circumstances. In intensive paddy fields on Java, fertilization often exceeds the recommended rate. In high-fertilizer areas, decreasing conventional fertilizer application while maintaining yields has been shown, suggesting a significant potential for fertilizer reduction using NUE improvement technologies.

Conversely, in arid regions, such as the East Nusa Tenggara Province, fertilizer application rates can be considerably lower owing to cost constraints and limited availability. In low-fertilizer regions, nitrogen is a limiting factor. Although technology adoption may show yield maintenance or improvement effects, assessing the benefits of fertilizer reduction is difficult.

Furthermore, variations in early heading effects owing to different photoperiodic conditions may affect technological stability. NR160E, introduced in Chapter 2, maintains early heading even under long-day conditions, suggesting its potential adaptability to high-altitude areas. Conversely, EHD10 and EHD16 retained their early heading traits, even under short-day conditions. Thus, tuning based on regional photoperiod environments is necessary.

Through these multilocation trials, clarifying the versatility and limitations of the technology and developing strategies for its dissemination are crucial. Particularly, verifying the adaptability of the technology to environmental factors such as fertilization systems, photoperiod conditions, and soil characteristics can enhance the implementation and policy value of NUE improvement technologies.

4.2: Development of Application Area Determination Technology

The dissemination of NUE improvement technologies requires the development of simple and practical evaluation methods to determine which paddy fields should adopt them. Conventional methods that comprehensively assess diverse factors such as soil properties, weather conditions, and fertilization history are unsuitable for immediate on-site decisions. Thus, this study explored the development of an assessment method using image analysis to assess plant growth patterns under conventional cultivation practices.

This assessment technique envisions a future operation in which farmers simply photograph their paddies at three key periods (approximately two weeks before flowering, flowering, and maturity) using a smartphone. The AI subsequently analyzes the images and automatically determines the suitability of applying the NUE technology. Realizing such a system would allow farmers to evaluate the requirement of introducing technology in their own fields, promising more efficient extension activities and improved technology acceptance.

Furthermore, image-analysis-based assessments are well-suited for large-scale deployment. Integration with drones or satellite imagery facilitates mapping of applicable paddy fields at the regional level. This allows administrative bodies and extension agencies to visualize areas where technology adoption should be prioritized, aiding in target selection for policy support.

4.3. Deepening of Physiological Mechanisms

The high NUE showed by the NR160E and EHD lines is thought to involve several physiological factors, such as maintaining photosynthetic activity following heading and improving grain-filling efficiency. Elucidating these factors directly contributes to technological improvements and development of new breeding strategies.

Particularly noteworthy mechanisms contributing to PNUE improvement include maintaining post-heading CGR, enhancing panicle number, thousand grain weight, grain filling rate, differences in photosynthetic rate and translocation efficiency, and the timing and amount of nitrogen uptake by the root system.

For instance, NR160E, introduced in Chapter 2, showed slower growth rates than NR160 before heading but demonstrated higher post-heading CGR, leading to no significant difference in the final dry weight. This is thought to stem from enhanced light use during the reproductive stage and enhanced translocation of photosynthetic products achieved by early heading before canopy closure.

Furthermore, since grain number and thousand grain weight improved, it is highly likely that the yield increase resulted not merely from a higher grain number but also from an enhanced grain filling rate and thousand grain weight. These traits

serve as crucial indicators of increased yield relative to nitrogen uptake.

Moving forward, quantitatively assessing these physiological traits and comparing them with other early heading genes or evaluating their combined effects will enable the development of higher-performance NUE-improved lines. Additionally, molecular-level expression and metabolite dynamics analyses will contribute to the elucidation of the underlying mechanisms. In particular, the analysis of the expression dynamics of photosynthesis-related genes and translocation-related enzymes following heading is expected to clarify the molecular basis of PNUE improvement.

5. Future Prospects for Technology Dissemination

5.1: Advanced Breeding Techniques

Developing rice varieties that improve NUE is a critical technological innovation for achieving sustainable agriculture. The results from verification studies in Indonesia suggest the potential for international technology deployment as a solution to the challenges common across the Asia-Monsoon region. This section outlines prospects for future technology dissemination, including advancing breeding methodologies, showing multifaceted benefits, coordinating policies, and exploring international expansion.

Conventional breeding techniques often require over five years to develop NUE-improved lines, limiting the speed of technology adoption. Contrastingly, using genome editing technologies and rapid breeding enables a significant decrease in breeding time.

Specifically, its rapid introduction into major regional varieties enhances its acceptance by local farmers and contributes to accelerated dissemination. Establishing breeding systems using molecular breeding techniques and the international sharing of breeding materials will be important in the future.

5.2: Demonstration of Multifaceted Benefits

Traditionally, early heading has been considered detrimental to yield potential and has been avoided in breeding programs. However, breeding strategies that actively utilize early heading to avoid weather stresses such as high temperatures and drought during the reproductive stage have recently gained traction.

Decreasing fertilizer application by improving NUE directly contributes to the suppression of greenhouse gas emissions. Specifically, significant decreases in methane emissions were anticipated with the introduction of early heading lines. Methane emissions in paddy fields tend to be the highest during the vegetative growth period before heading (Zhang et al., 2020). We hypothesized that shortening this period would decrease methane emissions.

Furthermore, several environmental and production benefits were achieved simultaneously, including suppressing nitrous oxide emissions via reduced fertilizer application, decreasing irrigation water use by shortening the cultivation period via early heading, and enhancing pest and disease resistance via robust early growth.

We can clarify the societal value of technology by quantitatively verifying and presenting these benefits for climate-change adaptation and mitigation. Furthermore, this would strengthen the basis for promoting its adoption by policymakers and agricultural stakeholders.

5.3: Policy Coordination

Coordination with national agricultural policies is necessary to accelerate technology adoption. For instance, linking smart agriculture policies such as Indonesia's Smart Farming 4.0, enables access to institutional support.

In countries where policy goals include decreasing environmental impacts and contributing to sustainable agriculture, introducing NUE improvement technologies aligns with policy objectives. Moving forward, the simultaneous deployment of policy advocacy and technology packages is required.

5.4: Expansion into the Asia-Monsoon Region and the Global South

The verification results in Indonesia could serve as a model for technology deployment across the Asia-Monsoon region. The similar climate, soil conditions, and cropping systems in this region enable horizontal technology transfer.

Developing technology packages tailored to the characteristics of each region and conducting joint demonstrations in collaboration with local research institutions are crucial in the future. For example, establishing a network centered on JIRCAS and developing a framework for technology transfer are expected to contribute to the international promotion of sustainable agriculture.

Authors contribution statement

M.O. provided the principal methodological innovation for the Indonesian field trials by designing the evaluation framework that enabled nitrogen-use efficiency (NUE) to be quantitatively assessed under contrasting fertilization regimes using early-heading lines introduced into the Ciherang background. This design established how the local fertilization system and growth period interact in practice, allowing NUE-related effects to be interpreted under real cultivation conditions. K.S. contributed original analytical development by integrating physiological measurements, yield data, and nitrogen-response indicators, ensuring that evidence from diverse trial components formed a coherent basis for evaluating the NUE advantages of early-heading lines. All authors reviewed the literature, synthesized the findings, and contributed to writing and revising the manuscript.

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