

1 RESEARCH ARTICLE

2 **SPAD Chlorophyll Meter-Based Real-Time Nitrogen Management in**
3 **Manure-Amended Lowland Rice**

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19 **ABSTRACT**

20 Precise adjustment of nitrogen (N) application time and dose under real-time nitrogen
21 management (RTNM) is crucial for optimal benefits. The study aimed to assess and recommend
22 best combination of threshold SPAD-502 chlorophyll-meter reading (SCMR) and topdressing N
23 rate for lowland flooded-rice (manure amended at 5 t ha⁻¹) to maximize yield, N use efficiency,

24 and economic gains. Four SCMR-based N scheduling (SCMR₃₄, SCMR₃₆, SCMR₃₈, SCMR₄₀),
25 each with three topdressing N rates (15, 20, 25 kg N ha⁻¹) as RTNM were evaluated and
26 compared with fixed-time N management (FTNM) (100 kg N ha⁻¹, 4 splits). Topdressing of 20
27 kg N ha⁻¹ at ≤ SCMR 38 (SCMR₃₈N₂₀) led to a higher yield (+8%), N use efficiency (+43%), net
28 return (+11%) and reduced N input (-18%) compared to FTNM. The yield advantage with
29 SCMR₃₈N₂₀ was attributed to higher internal N use efficiency and optimal source-sink balance
30 (higher harvest index). The best-fit treatment SCMR₃₈N₂₀ along with a low-yielding treatment
31 (SCMR₃₆N₂₀) was tested across seven rice cultivars for validation. Treatment SCMR₃₈N₂₀ led to
32 yield advantage (+7%), but significant only for four cultivars. In lowland rice, RTNM can
33 therefore increase yield and N use efficiency. However, cultivar-specific adjustments to the
34 SCMR threshold and N rate are necessary.

35

36 **KEYWORDS:** Real-time N management, Lowland flooded-rice, Lower Indo-Gangetic plain,
37 Agronomic N use efficiency, N recovery efficiency, Multivariate regression analysis.

38 **Introduction**

39 Rice (*Oryza sativa* L.) is predominantly grown in lowland soils that are mostly deficient in
40 available nitrogen (N) (Choudhury and Kennedy 2005; Hou et al. 2019). The current practice of
41 N management [fixed-time N management (FTNM)] in rice has several drawbacks including
42 lower N use efficiency, groundwater and atmospheric pollutions, and suboptimal productivity.
43 Now, there is a major concern over the decline in partial N factor productivity in flooded-rice
44 production primarily attributed to discrepancy in crop N demand and supply and higher
45 magnitude of losses (Fagodiya et al. 2020). This has led to an interest in crop demand-driven

46 real-time N management options to maximize nutrient use efficiency, yield, and profitability in
47 rice production systems ([Bandaogo et al. 2015](#)).

48 Crop season temporal N dynamics in anoxic-flooded rice soils depends on time and rate
49 of fertilizer/manure, crop nutrient removal, and N losses, and thus, ensuring optimal N
50 bioavailability throughout the crop season as per the crop demand is a potential challenge ([Qin et](#)
51 [al. 2020](#)). According to [Fageria et al. \(2011\)](#), the non-synchrony between crop N demand and
52 supply largely constrains rice production. In this context, sensor-based real-time N management
53 (RTNM) could be a potential approach to site-specific and demand-driven N management to
54 improve N use efficiency and reduce N losses ([Huda et al. 2016](#)). Several researchers have
55 demonstrated strong positive associations between rice plant N status and leaf chlorophyll
56 content ([Wood et al. 1993](#); [Lin et al., 2010](#)). Crop N nutrition as monitored by chlorophyll-meter
57 reading at different growth stages differs with cultivar, soil fertility, environmental conditions,
58 and locally adopted crop management practices ([Mohanty et al. 2021](#)). The precise adjustment of
59 chlorophyll-meter reading thresholds for fertilization scheduling with the optimal topdressing N
60 rate is therefore necessary in order to achieve maximum benefits of RTNM. Pre-sowing
61 application of manure in tropical soils can substantially contribute towards crop N nutrition,
62 depending on quantity, nutrient content, and rate of mineralization ([Venkatesh et al. 2017](#)).
63 Chlorophyll-meter based N scheduling would be a more realistic approach to N management in
64 integrated nutrient management module(s). However, the efficacy RTNM under integrated
65 nutrient management for yield advantage, nutrient use efficiency, and N economy module has
66 not been adequately studied. Further, variable N rates and timing as alter with different RTNM
67 treatments would provide valuable insights into N yield functions and relative weightage of N
68 application at different growth stages of rice crops.

69 In view of this, two years of calibration and one year of validation trials were conducted
70 to determine the best chlorophyll meter threshold for application scheduling and top-dressing N
71 rate in lowland rice crop that is amended with farmyard manure at 5 t ha⁻¹ to maximize
72 productivity, nutrient use efficiency, and N economy and validate the best-fit RTNM treatment
73 across predominant rice cultivars. The major hypotheses of the study were (i). RTNM with
74 optimal combination(s) of N scheduling based on threshold chlorophyll-meter reading and top-
75 dress N rate lead to yield advantage and higher nutrient use efficiency over FTNM in lowland
76 flooded rice, (ii) temporal scheduling of fertilizer-N application (time of application), total
77 applied N rate, and frequency of N topdressing as differ within the RTNM treatments would
78 have a direct influence on rice productivity, (iii) the response scale of rice to SPAD chlorophyll-
79 meter based RTNM would differ with cultivars having variable yield potential and crop duration,
80 and (iv) chlorophyll-meter based RTNM can be effectively integrated with manure amended
81 lowland rice production system for yield advantage and N economy.

82

83 **Materials and Methods**

84 *Site characteristics*

85 The experiments were conducted at the research farm of Bihar Agricultural University, Sabour,
86 Bhagalpur (25°15' N; 87°02' E, and 37 m a.s.l.). The site is located in the lower Indo–Gangetic
87 plain (IGP) region (subtropical humid climate). The weather conditions during the crop seasons
88 are presented in [Supplementary Figure 1](#). The experimental soil is a silty–clay loam and
89 classified as *Fluvisol* ([IUSS Working Group WRB, 2015](#)). The soil (0–0.15 m) had neutral pH
90 7.1, electrical conductivity of 0.47 dS m⁻¹, 4.9 g kg⁻¹ soil organic carbon, 12.2 mg kg⁻¹ of Olsen-

91 P and 66.9 mg kg⁻¹ of NH₄OAc-K (Jackson 1973). The soil available-N (98.2 mg kg⁻¹ KMnO₄-
92 N) of the field was lower than the critical soil N level (125 mg kg⁻¹ KMnO₄-N).

93

94 ***Experimental design and treatment details***

95 ***Experiment 1 (Optimization trial)***

96 In the optimization trial (rainy season 2013 and 2014), fourteen different N treatments were
97 evaluated in rice [cultivar (*cv.*) Rajendra Sweta (130–135 days duration)]. The treatments
98 comprised of one N control (no fertilizer-N application), one conventional practice of fixed-time
99 N management (FTNM) treatment, and twelve different SPAD 502 chlorophyll-meter (SCMR)
100 based real-time N management (RTNM) treatments using different combinations of SPAD 502
101 chlorophyll-meter reading (SCMR) based fertilization scheduling in which N was applied when
102 the SCMR of the youngest fully-extended leaf was less than or equaled 34 (SCMR₃₄), 36
103 (SCMR₃₆), 38 (SCMR₃₈), and 40 (SCMR₄₀), each with three top-dressing N rates (15, 20, and 25
104 kg N ha⁻¹). SCMR were recorded ($n = 15$) from each plot starting from the tiller initiation (20
105 DAT) to the fully heading stage (90 DAT) at a 10-day interval using SPAD-502 chlorophyll-
106 meter (Minolta Camera Co., Osaka, Japan). The FTNM treatment represents the current
107 recommended dose of fertilizer-N (100 kg N ha⁻¹) application in four equal splits *i.e.* basal, active
108 tillering [40 days after transplanting (DAT)], panicle initiation (60 DAT), and heading (90 DAT)
109 stages. The basal dose of fertilizer-N (*i.e.* 25 kg N ha⁻¹) was applied to both the FTNM and
110 RTNM treatments and subsequent N was top-dressed as per treatments. The source of N was
111 urea [CO(NH₂)₂]. Before the rice crop establishment, farmyard manure (FYM) at 5 t ha⁻¹ (dry
112 weight basis) was applied to each plot. The quantity of N added with the FYM was 25.6 ± 0.7
113 kg ha⁻¹ and 27.7 ± 1.1 kg ha⁻¹ (sample analyzed $n = 3$) in the year 2013 and 2014, respectively. The

114 treatments were allocated in a randomized complete block design (RCBD) with three
115 replications. The dimension of each plot was 6 m × 4 m. A filler space of 2.5 m was kept on all
116 sides of each plot.

117

118 ***Experiment 2 (validation trial)***

119 In the validation trial (year 2015), the best-fit RTNM treatment along with a relatively low-
120 yielding RTNM treatment (significant lower yield compared to the best-fit RTNM treatment)
121 were identified/selected based on the results of the optimization trial (Experiment 1). These
122 treatments were evaluated in seven high yielding rice cultivars viz. 27P31 (hybrid), RAU 724,
123 MTU 7029, Rajendra Mahsuri, Rajendra Bhagawati, Rajendra Sweta and Arize 6444 (hybrid).
124 Similar methods and management practices were employed as in the optimization trial.

125

126 ***Crop management***

127 Rice seedlings were raised on a 20 m × 5 m nursery bed (wet nursery). During the final field
128 preparation, well-decomposed FYM (5 t ha⁻¹) was added to each plot and thoroughly mixed.
129 Wet-tillage (puddling) was performed one day prior to transplanting. Twenty-two-day old
130 seedlings were transplanted on July 10 in 2013, July 15 in 2014 (optimization trial), and July 13
131 in 2015 (validation trial) at a spacing of 10 cm × 15 cm. In both the experiments, 17.2 kg P ha⁻¹
132 and 32.8 kg K were applied as a basal dose to all the treatments plots. From transplanting to
133 heading stage, continuous flooding/ponding conditions were maintained with irrigation. Weeds
134 were manually controlled by hand weeding 1-2 times. Necessary plant protection measures were
135 taken to prevent insect pests and diseases.

136

137 ***Yield and yield attributes***

138 Yield attributing parameters like panicles m⁻², grains panicle⁻¹ and 1000-grain weight were
139 recorded from five rice hills randomly selected from each plot at the maturity stage. Grain and
140 straw yields were estimated from a net-plot area of 5.0 m × 2.0 m. The harvested crop was
141 threshed using a plot thresher and the produces was sun-dried. The total biomass of the crop was
142 estimated on an oven dry weight basis, while grain yield was adjusted at 12% moisture content
143 (w/w). Harvest index (HI) was calculated by the ratio of grain yield to total biological yield
144 (aboveground dry matter) and expressed as a percentage.

145

146 ***Plant nutrient analysis and calculation of N use efficiency***

147 At maturity, plant and grain samples were collected, oven dried, ground and analyzed for total N
148 content was determined by micro-Kjeldahl method (Yoshida et al. 1976). Agronomic N use
149 efficiency (AE_N), N recovery efficiency (RE_N), internal N use efficiency (IE_N), partial factor
150 productivity of applied N (PFP_N), nitrogen harvest index (NHI) and physiological N use
151 efficiency (PE_N) were calculated by the following equations 1-6

152
$$AE_N (\text{kg kg}^{-1}) = \frac{GY_T - GY_C}{FN} \quad \text{eq. 1}$$

153
$$RE_N (\text{kg kg}^{-1}) = \frac{NU_T - NU_C}{FN} \quad \text{eq. 2}$$

154
$$IUE_N (\text{kg kg}^{-1}) = \frac{GY}{NU} \quad \text{eq. 3}$$

155
$$PFP_N (\text{kg kg}^{-1}) = \frac{GY}{FN} \quad \text{eq. 4}$$

156
$$NHI (\%) = \frac{NU_{\text{Grain}}}{NU_{\text{Total}}} \times 100 \quad \text{eq. 5}$$

157
$$PE_N (\text{kg kg}^{-1}) = \frac{AGB}{NU} \quad \text{eq. 6}$$

158 where, GY_T = grain yield in fertilizer-N treated plot ($kg\ ha^{-1}$), GY_C = grain yield in N
159 control plot ($kg\ ha^{-1}$), FN = quantity of fertilizer-N applied ($kg\ N\ ha^{-1}$), NU_T = total uptake of N
160 in fertilizer-N treated plot ($kg\ ha^{-1}$), NU_C = total uptake of N in N control plot ($kg\ ha^{-1}$), GY =
161 grain yield ($kg\ ha^{-1}$), NU = total uptake of N ($kg\ ha^{-1}$), NU_{Grain} = grain N uptake ($kg\ ha^{-1}$),
162 NU_{Total} = Total uptake of N ($kg\ ha^{-1}$), AGB = total above ground biomass ($kg\ ha^{-1}$).

163

164 ***Calculation nitrous oxide emission***

165 Total N_2O emission from each treatment plot was calculated indirectly following the
166 mathematical model (eq. 7) suggested by Tubiello et al. (2015) with the necessary modifications
167 at the regional scale and cropping period. A global warming potential equivalent factor 265 was
168 multiplied with the N_2O emission value to express the total emission as $kg\ CO_2$ equivalent ha^{-1} .

$$169 \quad N_2O \text{ emissions } (kg\ ha^{-1}day^{-1}) = N \times EF_1 \times \frac{44}{28} \quad \text{eq. 7}$$

170 where N_2O emissions from the fertilizer-N, manures, and residues added to the managed
171 soil; N = N consumption from fertilizers and crop residues ($kg\ N\ input\ year^{-1}$); EF_1 = emitting
172 factors of 0.01 for N_2O emissions from N input ($kg\ N_2O-N\ kg^{-1}\ N\ input$).

173 ***Economic analysis***

174 Costs and prices of all consumed inputs and outputs were considered in the economic analysis.
175 All input cost components were summed up and denoted as working cost of cultivation, on
176 which 3.5% interest, transportation and miscellaneous costs were added to calculate total
177 variable cost of cultivation (TVCC). The total economic return from the grain and straw output
178 was calculated as the gross return. The year-wise minimum support price of rice (paddy)
179 (Government of India) and local market price of rice straw was used to convert the grain and
180 straw yield to their economic value. Net return (NR) was calculated as the difference between

181 gross return and TVCC. The ratio of net return to TVCC was denoted as the benefit-cost ratio
182 (BCR). Marginal return (MR) from N management treatments was computed using eq. 9.

183
$$\text{MR } (\$ \text{ ha}^{-1}) = \frac{\text{NR}_T - \text{NR}_C}{\text{TVCC}_T - \text{TVCC}_C} \quad \text{eq. 10}$$

184 NR_T = net return with fertilizer-N treatment, NR_C = net return from N control treatment, TVCC_T
185 = total variable cost of cultivation in fertilizer-N treatment, TVCC_C = total variable cost of
186 cultivation without fertilizer-N treatment (N control).

187

188 *Statistical analysis*

189 The data of optimization trial was analyzed following analysis of variance (ANOVA) of RCBD,
190 while validation trial data were subjected to split-plot ANOVA (Cochran and Cox 1957).

191 Principal component analysis was performed using statistical software PAST (version 3.26). The
192 multivariate regression analysis was performed using ‘Data Analysis Toolpak’ Add-In of
193 Microsoft Excel to explain the independent variables (X_1 - X_n) as yield function and their
194 corresponding coefficient weightage (C_1 - C_n) [$Y = C + C_1X_1 + C_2X_2 + \dots + C_nX_n$].

195

196 **Results**

197 **Optimization trial (Experiment 1)**

198 *Fertilizer-N scheduling*

199 The total fertilizer-N rate ranged between 55 kg (SCMR₃₄N₁₅) to 92 kg (SCMR₄₀N₂₅) N ha⁻¹
200 within the RTNM treatments (Table 1). The fertilizer-N rate was consistent over replications and
201 cropping years, except for the treatment SCMR₄₀N₂₀ (Supplementary Table 1). The frequency of
202 N splits ranged from 2 to 4 (SCMR₄₀N₁₅) (Table 1).

203

204 ***Crop response to N management treatments***

205 The treatment SCMR₃₈N₂₀ had 8% ($p < 0.05$) higher yield than the FTNM treatment (Table 1).
206 The RTNM treatments SCMR₃₄N₁₅, SCMR₃₄N₂₀, SCMR₃₆N₁₅, and SCMR₃₆N₂₀ resulted in a
207 yield reduction (11-24%, $p < 0.05$) as compared to the FTNM, whereas the remaining RTNM
208 treatments had a similar yield to FTNM. The treatment SCMR₃₈N₂₀ had a lower panicle m⁻² but
209 equivalent grains panicle⁻¹, stover yield and HI to the FTNM. Under fertilizer-N deficient
210 conditions (control), yield traits decreased (compared with FTNM) in the order of straw yield (-
211 55%), grain yield (-45%), panicle m⁻² (-43%), grains panicle⁻¹ (-17%) and 1000-grain weight (-
212 10%), but increased HI (+11%) ($p < 0.05$). The highest HI was recorded in the treatment
213 SCMR₃₆N₂₅ (48.8%) followed by the N control treatment (47.1%). Grain yield showed higher
214 positive correlation with panicle density ($r = 0.86$) and straw yield ($r = 0.80$) ($p < 0.05$) (data not
215 presented). Under RTNM, a higher N rate increased yield in SCMR 34, 36, and 38, but not with
216 SCMR 40. Similarly, increasing SCMR threshold increased yield under N rate (kg ha⁻¹) of 15 kg
217 ($r = 0.996$), 20 kg ($r = 0.888$), but not 25 kg N ($r = 0.221$) (data not presented).

218

219 ***N uptake and N use efficiency***

220 The highest N uptake was recorded in the FTNM treatment (113 kg N ha⁻¹), while N uptake
221 within the RTNM treatments ranged between 69 (SCMR₃₄N₁₅) to 112 (SCMR₃₈N₂₀) (Table 2). In
222 the N control treatment, a greater magnitude of reduction (compared to FTNM treatment) was
223 observed in the N uptake (62%) than in the scale of grain yield loss (45%). Strong positive
224 correlations were observed between the N rate and N uptake in both the cropping years ($r =$
225 0.911-0.938, $p < 0.01$) (Figure 1). Treatments SCMR₃₈N₂₀, SCMR₃₈N₂₅ led to the highest
226 increase in AE_N over FTNM, while, except SCMR₃₈N₂₀, all the RTNM treatments had either

227 equal or higher AE_N over FTNM (Table 2). The RE_N ($kg\ kg^{-1}$) was higher in treatments
228 SCMR₃₈N₂₅ (0.87), SCMR₃₆N₂₅ (0.86), SCMR₃₈N₂₀ (0.84), which were significantly higher than
229 the FTNM (0.70). The parameters IUE_N , PFP_N , NHI, and PE_N were higher in the RTNM
230 treatments than the FTNM treatment. The highest IUE_N , NHI, and PE_N were recorded in the N
231 control treatment (Table 2).

232 *Multivariate analysis, economic return, and N₂O emission*

233 Variable N rates, particularly at 50, 70, 80, and 90 DAT had a significant and higher positive
234 influence on yield ($p < 0.05$). Likewise, among the variables N rate, N splits and NTI (an index
235 that defines the earliness or delay in the application of fertilizer–N), only the N rate exhibited a
236 significant positive influence on rice yield. There are marked year-to-year variations in the
237 regression coefficient for the NTI (Figure 1). According to the PCA, treatment-induced variations
238 in N splits, AE_N , RE_N , net return, and N rate. The strong positive association between total N rate
239 and straw yield was evident from PCA results (Supplementary Figure 2).

240 The RTNM treatment SCMR₃₈N₂₀ led to the highest net return and benefit–cost ratio,
241 which were +11% and +4% higher than the FTNM treatment ($p < 0.01$) (Table 3). With an
242 increase in N top-dress rate, the marginal return was increased in SCMR₃₄ (up to 25 $kg\ N\ ha^{-1}$)
243 SCMR₃₈ only (up to 20 $kg\ N\ ha^{-1}$). A reduction in seasonal N₂O emission (simulated) was
244 estimated in all the RTNM treatments (2.4–12.9 $kg\ CO_2\ equivalent\ ha^{-1}$) ($p < 0.001$) as compared
245 to the FTNM treatment ($N_2O\ GWP = 37.8\ kg\ CO_2\ equivalent\ ha^{-1}$) (Figure 2)

246

247 **Experiment 2 (Validation trial)**

248 Treatment SCMR₃₈N₂₀ exhibited higher precision for N rate within replications over the
249 treatment SCMR₃₆N₂₀ (Supplementary Table 2). Notably, the yield difference between
250 treatments (SCMR₃₆N₂₀, SCMR₃₈N₂₀) for *cv.* Rajendra Sweta was 15.6% ($p < 0.05$) that was
251 almost similar to the optimization trial (13.9-28.7%, $p < 0.05$) (Table 4, Table 1). The yield
252 increment with the treatment SCMR₃₈N₂₀ over SCMR₃₆N₂₀ was recorded as $> +10\%$ for the *cvs.*
253 RAU724, Rajendra Sweta, and Rajendra Mahsuri, but remained comparable for *cvs.* MTU 7029,
254 Rajendra Bhagawati, and Arize 6444. The treatment SCMR₃₈N₂₀ had a higher straw yield and N
255 uptake than the treatment SCMR₃₆N₂₀ (Table 4, Table 5).

256

257 **Discussion**

258 **Experiment 1 (Optimization trial)**

259 The results show that SPAD-meter-based RTNM could be an efficient N management approach
260 to lowland flooded-rice production. Meanwhile, our results show that precise adjustment of
261 SCMR threshold for N scheduling along with optimal N top-dress rate are essential for yield
262 advantage and higher N use efficiency over conventional FTNM. As, FYM (5 t ha⁻¹) was applied
263 to each treatment plot prior to crop establishment, therefore the organically bound N in manure
264 could have complemented the crop N requirement. In tropical climates, the faster mineralization
265 of organically bound N is readily accessible to the immediate crop; however, this may be
266 partially limited under flooded-rice soils due to anoxic soil conditions (Borase et al. 2020). The
267 release kinetics of organically bound nitrogen can differ with crop stage, which is hard to
268 monitor. Therefore, RTNM could be a realistic approach to synchronize the demand and supply
269 of N based on relative chlorophyll meter readings and thereafter optimum scheduling of N
270 fertilization.

271 According to the results, not all RTNM treatments are effective in producing higher yield
272 but fertilizer savings is common in all treatments. Results indicate that RTNM following a lower
273 SCMR threshold (SCMR₃₄, SCMR₃₆) in combination with a low or medium top-dressed N rate
274 (15 and 20 kg N ha⁻¹) could lead to a yield loss (compared to the FTNM treatment), while, with a
275 higher top-dress N rate (25 kg N ha⁻¹) had attained a similar yield to conventional FTNM
276 treatment with a saving of 25 kg N ha⁻¹ (Table 1). This result suggests that a higher N
277 topdressing rate is essential to have an extended effect of top-dress N in RTNM following a
278 SCMR threshold below 38. A topdressing N rate of 20 kg N ha⁻¹ in combination with fertilizer-N
279 scheduling with SCMR₃₈ could produce higher yields, fertilizer-N use efficiency, and higher
280 economic return compared to FTNM (Tables 1, 2, and 3), and was therefore recommended as a
281 best-fit RTNM treatment for the medium duration rice *cv.* Rajendra Sweta. The positive impact
282 of the treatment SCMR₃₈N₂₀ was possibly attributed to synchronous and balance N application as
283 per crop demand. The N fertilization schedules in the SCMR₃₈N₂₀ treatment are basal, 50, 70, 90
284 DAT, while in the FTNM treatment they are basal, 40, 60, 90 DAT (Supplementary Table 1,
285 Supplementary Figure 3), and thus, the delayed application of topdressing fertilization may have
286 been the cause for balanced growth, whereas the higher N rates applied in the FTNM treatment
287 resulted in higher vegetative growth, but no apparent increase in yield attributing traits. The
288 multivariate regression analysis results indicate that the variable N application at 50, 70, 80, and
289 90 DAT had a direct positive influence on yield, which closely matched with the best-fit RTNM
290 treatment SCMR₃₈N₂₀ (Figure 1). The consistency in the results within the cropping year also
291 validates the regression model. The treatment SCMR₃₈N₂₀ had a lower panicle density than the
292 FTNM treatment, whereas the number of grains panicle⁻¹ and seed weight was higher in the
293 treatment SCMR₃₈N₂₀ over the FTNM treatment (Table 1), and this finding specifically hints a

294 higher intra-plant competition in the FTNM treatment possibly attributed to increased tiller
295 production that might have reduced number of grains panicle⁻¹ and their weights. The reduction
296 in yield determining traits like grains panicle⁻¹ and seed weight are mostly genetically
297 configured, but an imbalance in growth (higher biomass growth, ([Supplementary Figure 4](#)) may
298 lead to affect source-sink balancing, which is further confirmed with the results of HI. [Wei et al.](#)
299 ([2018](#)) also reasoned that the source-sink relationship during grain filling is not merely a matter
300 of carbon supply versus demand, but highly coupled with grain N demand.

301 Results indicate that the RTNM following a higher threshold SCMR (> 38) could
302 increase topdressing frequency but not necessarily increase yield over the FTNM. This is
303 especially true when N splits are not uniformly distributed (temporal) during the crop season.
304 Additionally, faster growth may result in the dilution of chlorophyll in leaf tissue, misleading the
305 SCMR-based RTNM.

306 Besides, a faster growth may cause dilution of chlorophyll in leaf tissue and thereby
307 mislead the SCMR-based RTNM. Concerning the increased yield benefits from N application at
308 the later growth stages (50-90 DAT) a dynamic approach of N management (lower N topdressing
309 rate at the initial growth stages and higher rate at the later growth stages) would result in further
310 up-scaling of N use efficiency. [Peng et al. \(2006\)](#) also suggested that a quantum leap in AE_N is
311 possible in the intensive rice-growing areas in China by simply reducing the current N rate and
312 by allocating less N at the early vegetative stage. PCA reveals that the frequency of N split
313 application is not significantly correlated with higher N use efficiency (especially AE_N) as a
314 right-angle relationship between variables (or traits) represents a non-significant correlation
315 ([Supplementary Figure 2](#)). A trait vector length represents the sensitivity scale of a variable to N
316 management treatments, and the results suggest that the benefits of RTNM are much more

317 prominent on N use efficiency parameters (AE_N and RE_N) than yield and its attributes. The
318 prominent year-wise deviations in the regression coefficient of NTI could be attributed to the
319 seasonal variation in weather conditions (particularly rainfall) and mineralization rate (Figure 1).

320 The RTNM treatment $SCMR_{38}N_{20}$ exhibited higher AE_N and RE_N over the FTNM
321 treatment, which is due to the yield gain over the FTNM treatment as well as reduced use of
322 fertilizer-N. This study corroborates the findings of [Khurana et al. \(2008\)](#) and [Singh et al. \(2002\)](#)
323 in western parts of India with a SPAD critical value of 37.5. The increased IUE_N and PE_N with
324 higher yield certainly indicate improved intra-plant nutrient use efficiency and N remobilization
325 from source to sink in the treatment $SCMR_{38}N_{20}$ over the FTNM. The study also indicates the
326 advantage of RTNM in reducing the NO_2 emission from a lowland flooded rice soil primarily by
327 reducing the fertilizer-N input. Lowland flooded rice ecosystems contribute largely toward total
328 agricultural greenhouse gas emissions. Fertilizer-N applied to flooded rice crops is much prone
329 to several losses such as N leaching, denitrification etc., which ultimately downscale the N use
330 efficiency. The imbalance in N fertilization scheduling and a higher rate of N topdressing might
331 have resulted in higher N losses for FTNM treatment, whereas higher N application at early
332 stages might have led to higher N losses.

333 A higher topdressing frequency with a low top-dress N rate is considered an ideal
334 approach to upscale yield and N use efficiency; however, the economic feasibility must be taken
335 into consideration to determine the optimum N splits. For instance, chlorophyll-meter-based
336 RTNM following a higher SCMR as a threshold could increase the frequency of N topdressing
337 particularly when the N topdressing rate is low as observed for the treatment $SCMR_{40}N_{15}$ ([Table](#)
338 [1](#)). The treatment $SCMR_{40}N_{15}$ recorded a comparable grain yield (5204 kg ha^{-1}) to the treatment

339 SCMR₃₈N₂₀, however, the net return was higher in the SCMR₃₈N₂₀ treatment over the treatment
340 SCMR₄₀N₁₅ (Table 5) and therefore SCMR₃₈N₂₀ was selected as the best treatment.

341 The savings of fertilizer-N and a higher yield in best-fit RTNM treatment (SCMR₃₈N₂₀)
342 led to favourable production economics. The researchers noted the exorbitant cost of rice
343 cultivation was due to the overuse of fertilizer-N (Koch et al. 2004). The economics were
344 calculated based on the government-subsidised price of urea (Government of India). Therefore,
345 in reality, the cost involved in N management would be higher than the estimated cost of
346 fertilizer N in the study. Thus, the margin of economic return between the best-fit RTNM
347 treatment and FTNM with unsubsidised fertilizer-N (urea) price would be more significant.

348

349 **Experiment 2 (validation trial)**

350 The results demonstrate a need for cultivar-specific calibration of optimal SCMR for fertilization
351 scheduling in lowland flooded rice. According to the validation trial result, the test *cv.* Rajendra
352 Sweta showed a similar response to that of the optimization trial, and thus, the result verifies the
353 field scale efficacy/precision of RTNM treatments for a particular rice cultivar in lowland
354 ecologies. Nevertheless, the yield advantage with best-fit RTNM treatment varied greatly across
355 cultivars. Differences in yield potential, N demand, and crop duration might explain the
356 differential response of cultivars to RTNM. Given the differential response of rice cultivars to
357 the selected RTNM treatments, a cultivar-specific RTNM recommendation would be essential to
358 have the best results. Additionally, the results indicate that the precision of RTNM treatment for
359 the applied N rate and timing within replicates may be equally considered for precise adjustment
360 and validation of cultivar and location-specific N management in rice. Shukla et al. (2004) also

361 reasoned that site-specific nutrient management must consider the wider variability in leaf colour
362 traits within the cultivars.

363

364 **Conclusions**

365 The study concluded that SCMR-based RTNM could be a sustainable approach of N
366 management in lowland flooded-rice systems over the conventional FTNM. However, precise
367 adjustment of the SCMR threshold in combination with optimal N rate is vital to realize the
368 optimal benefits of RTNM. The results suggested that SCMR based RTNM in lowland flooded
369 rice could be effectively applicable under integrated nutrient management module(s). The
370 optimization trail results revealed that basal application of 25 kg N ha⁻¹ followed by top-dressing
371 of fertilizer-N at 20 kg N ha⁻¹ when the SCMR ≤ 38 could improve yield, N use efficiency, net
372 return and reduced fertilizer-N rate over FTNM. The yield gain in the treatment SCMR₃₈N₂₀
373 over the FTNM was primarily attributed to improved source-sink balance with a timely (delayed)
374 supply of N during crop season. The regression analysis results explained that N application at
375 later growth stages (panicle initiation to full heading stage) had a higher scale of impact on rice
376 yield over the early applications. Therefore, a variable N top-dress rate [a lower dose at early
377 growth stage(s) and a higher in the later growth stages] would further improve the N economy in
378 RTNM in lowland rice, which is to be ascertained with systematic studies. The study suggests
379 that there is a need for cultivar-specific calibration of SCMR threshold limit in combination with
380 optimal top-dress N rate for maximum benefits.

381

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385

386 **Disclosure statement**

387 No potential conflict of interest was reported by the authors.

388

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392

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473 **Table 1.** Effect of different N management practices on yield attributes of rice (pooled data of
 474 2013 and 2014) [Experiment 1].
 475

Treatment	Fertilizer-N applied (kg ha ⁻¹)	N split (nos.)	Panicles m ⁻² (nos.)	Grains panicle ⁻¹ (nos.)	Test weight (g)	Grain yield (kg ha ⁻¹)	Straw yield (kg ha ⁻¹)	Harvest index (%)
SCMR ₃₄ N ₁₅	55	2	235 ^e	73.3 ^{cd}	22.9 ^{cd}	3753 ^e	4403 ^h	45.9 ^{bcd}
SCMR ₃₄ N ₂₀	65	2	260 ^d	83.1 ^a	23.7 ^{abc}	4164 ^d	4992 ^{fg}	45.5 ^{cde}
SCMR ₃₄ N ₂₅	75	2	281 ^{bcd}	79.7 ^{abc}	24.1 ^{ab}	4893 ^{bc}	5365 ^{ef}	47.7 ^{ab}
SCMR ₃₆ N ₁₅	63	3	256 ^d	74.5 ^{bcd}	22.9 ^{cd}	4132 ^d	4867 ^{gh}	46.1 ^{bc}
SCMR ₃₆ N ₂₀	65	2	263 ^d	78.7 ^{abc}	24.1 ^{ab}	4412 ^c	4983 ^{fg}	46.9 ^{abc}
SCMR ₃₆ N ₂₅	71	2	271 ^{cd}	80.8 ^{ab}	24.5 ^a	4767 ^c	5008 ^{fg}	48.8 ^a
SCMR ₃₈ N ₁₅	70	3	280 ^{bcd}	75.8 ^{bc}	23.3 ^{bc}	4764 ^c	5496 ^{def}	46.4 ^{bc}
SCMR ₃₈ N ₂₀	82	3	280 ^{bcd}	84.9 ^a	24.6 ^a	5323 ^a	6277 ^{bc}	46.0 ^{bc}
SCMR ₃₈ N ₂₅	75	2	284 ^{bc}	75.9 ^{bc}	24.6 ^a	5106 ^{abc}	5745 ^{cde}	46.9 ^{abc}
SCMR ₄₀ N ₁₅	85	4	282 ^{bc}	82.8 ^{ab}	24.5 ^a	5204 ^{ab}	5927 ^{bcd}	46.8 ^{abc}
SCMR ₄₀ N ₂₀	85	3	286 ^{abc}	79.3 ^{abc}	23.7 ^{abc}	5005 ^{abc}	6545 ^a	43.2 ^e
SCMR ₄₀ N ₂₅	92	3	290 ^{ab}	79.6 ^{abc}	24.0 ^{abc}	4862 ^c	6340 ^{ab}	43.6 ^{de}
FTNM	100	3	303 ^a	81.2 ^{ab}	24.2 ^{ab}	4952 ^{bc}	6800 ^a	42.3 ^e
Control	-	-	173 ^f	67.5 ^d	21.7 ^d	2700 ^f	3045 ⁱ	47.1 ^{abc}
LSD (<i>p</i> = 0.05)	7.4	-	17.5	7.2	1.2	346	563	2.4
Pooled ANOVA	<i>(p value)</i>							
Year	1.00		0.065	0.052	0.147	0.013	0.056	0.004
Treatment	< 0.001		< 0.001	0.002	< 0.001	< 0.001	< 0.001	< 0.001
Year × Treatment	0.124		0.150	0.564	0.309	0.002	0.027	< 0.001

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 477 *SCMR*, SPAD 502 chlorophyll meter reading; *N*, fertilizer-N topdressing rate (kg N ha⁻¹);
 478 *FTNM*, Fixed-time N management. *a-i*, treatments with different lowercase superscript letters
 479 are significantly different at *p* ≤ 0.05.
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488 **Table 2.** Effect of different N management practices on agronomic N use efficiency (AE_N),
 489 nitrogen recovery efficiency (RE_N), internal N use efficiency (IUE_N), partial factor productivity of
 490 applied N (PFP_N), nitrogen harvest index (NHI) and physiological N use efficiency (PE_N) of rice
 491 (pooled data of years 2013 and 2014) [Experiment 1].
 492

Treatment	Total N	AE _N	RE _N	IUE _N	PFP _N	NHI	PE _N
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	uptake (kg ha ⁻¹)	(kg kg ⁻¹)	(%)	(kg kg ⁻¹)			
SCMR ₃₈ N ₂₀	69.0 ^f	19.1 ^d	0.47 ^h	54.3 ^b	68.2 ^a	0.64 ^{ab}	125 ^b
SCMR ₃₈ N ₂₅	80.7 ^e	22.5 ^{cd}	0.57 ^g	51.6 ^{bc}	64.1 ^{abc}	0.62 ^{bc}	120 ^{bc}
SCMR ₃₄ N ₂₅	103.7 ^{bc}	29.2 ^{ab}	0.80 ^{bc}	47.2 ^{efg}	65.2 ^{ab}	0.59 ^{de}	115 ^{cdef}
SCMR ₃₆ N ₁₅	81.4 ^e	22.5 ^{cd}	0.61 ^g	50.7 ^{cd}	66.6 ^a	0.62 ^{bc}	121 ^{bc}
SCMR ₃₆ N ₂₀	89.9 ^d	26.3 ^{bc}	0.72 ^{ed}	49.0 ^{cde}	67.9 ^a	0.61 ^{cd}	115 ^{cdef}
SCMR ₃₆ N ₂₅	104.0 ^{bc}	28.9 ^{ab}	0.86 ^{ab}	45.8 ^{fgh}	67.6 ^a	0.58 ^{ef}	112 ^{def}
SCMR ₃₈ N ₁₅	97.6 ^c	29.5 ^{ab}	0.78 ^{cde}	48.8 ^{cdefg}	68.1 ^a	0.61 ^{cd}	119 ^{bc}
SCMR ₃₈ N ₂₀	111.6 ^a	32.2 ^a	0.84 ^{abc}	47.8 ^{defg}	65.6 ^{ab}	0.60 ^{cde}	112 ^{def}
SCMR ₃₈ N ₂₅	108.7 ^{ab}	32.1 ^a	0.87 ^a	46.8 ^{efg}	68.1 ^a	0.60 ^{cde}	110 ^f
SCMR ₄₀ N ₁₅	110.0 ^{ab}	29.4 ^{ab}	0.78 ^{cde}	47.4 ^{efg}	61.2 ^{bc}	0.59 ^{de}	118 ^{cd}
SCMR ₄₀ N ₂₀	107.0 ^{ab}	26.7 ^{bc}	0.74 ^{def}	47.0 ^{efg}	59.2 ^{cd}	0.59 ^{de}	119 ^{bc}
SCMR ₄₀ N ₂₅	107.5 ^{ab}	23.6 ^c	0.70 ^f	45.5 ^{gh}	54.0 ^{de}	0.56 ^{fg}	117 ^{cde}
FTNM	113.3 ^a	22.5 ^{cd}	0.70 ^f	43.7 ^h	49.5 ^e	0.55 ^g	111 ^{ef}
Control	43.4 ^g	-	-	62.2 ^a	-	0.67 ^a	142 ^a
LSD ($p = 0.05$)	7.08	4.4	0.07	3.1	5.3	0.03	6.8
Pooled ANOVA (p value)							
Year	1.00	0.019	0.034	0.054	0.006	0.119	< 0.001
Treatment	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Year × Treatment	0.124	0.039	0.003	0.309	0.563	0.659	0.320

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494 SCMR, SPAD 502 chlorophyll-meter reading; N, topdressing N rate (kg N ha⁻¹); FTNM, fixed-
495 time N management; DAT, days after transplanting. *a-h*, treatments with different lowercase
496 superscript letters are significantly different at $p \leq 0.05$.

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502 **Table 3.** Effect of different N management practices on economics of rice production (pooled
503 data of years 2013 and 2014) [Experiment 1].

Treatment	Total variable cost of cultivation (\$ ha ⁻¹)	Gross return (\$ ha ⁻¹)	Net return (\$ ha ⁻¹)	Benefit cost ratio	Marginal return (\$ ⁻¹ ha ⁻¹)
SCMR ₃₄ N ₁₅	454 ^d	697 ^f	243 ^f	1.53 ^e	2.19 ^d
SCMR ₃₄ N ₂₀	465 ^c	774 ^e	309 ^e	1.66 ^d	2.77 ^c
SCMR ₃₄ N ₂₅	483 ^b	903 ^{bcd}	420 ^{cd}	1.87 ^{bc}	3.44 ^{ab}
SCMR ₃₆ N ₁₅	465 ^c	767 ^e	302 ^e	1.65 ^d	2.67 ^c
SCMR ₃₆ N ₂₀	470 ^c	816 ^e	346 ^e	1.73 ^d	3.06 ^{bc}
SCMR ₃₆ N ₂₅	480 ^b	876 ^{de}	397 ^d	1.82 ^c	3.30 ^{bc}

SCMR ₃₈ N ₁₅	481 ^b	883 ^{bcd}	402 ^d	1.83 ^{bc}	3.31 ^{bc}
SCMR ₃₈ N ₂₀	495 ^a	989 ^a	494 ^a	2.00 ^a	3.76 ^a
SCMR ₃₈ N ₂₅	487 ^b	944 ^{abc}	457 ^{abc}	1.93 ^{ab}	3.68 ^{ab}
SCMR ₄₀ N ₁₅	496 ^a	964 ^{ab}	468 ^{ab}	1.94 ^{ab}	3.47 ^{ab}
SCMR ₄₀ N ₂₀	484 ^b	938 ^{abcd}	455 ^{abc}	1.93 ^{ab}	3.78 ^a
SCMR ₄₀ N ₂₅	482 ^b	911 ^{bcd}	430 ^{bcd}	1.89 ^{bc}	3.59 ^{ab}
FTNM	486 ^b	933 ^{abcd}	447 ^{bc}	1.92 ^b	3.62 ^{ab}
Control	392 ^e	499 ^g	107 ^g	1.27 ^f	-
LSD ($p = 0.05$)	7.7	62.2	44.8	0.08	0.45
Pooled ANOVA (p value)					
Year	0.015	0.018	0.018	0.020	0.147
Treatment	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Year × Treatment	< 0.001	0.003	0.005	0.013	0.029

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506 SCMR, SPAD 502 chlorophyll-meter reading; N, topdressing N rate (kg N ha⁻¹); FTNM, fixed-
507 time N management. *a-g*, treatments with different lowercase superscript letters are
508 significantly different at $p \leq 0.05$.

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515 **Table 4.** Crop yields, N uptake, and partial factor productivity of applied N (PFP_N) of different
516 rice cultivars to RTNM treatments [Experiment 2].
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Cultivar	Crop duration (days)	Grain yield (kg ha ⁻¹)			Straw yield (t ha ⁻¹)
		RTNM ₁	RTNM ₂	Mean	RTNM ₁
27P31	128-132	5920 ^{B#}	6467 ^A	6194 ^d	13854 ^B
RAU724	~150	5630 ^B	6480 ^A	6055 ^d	13595 ^B
MTU7029	~150	6487 ^A	6770 ^A	6629 ^c	14297 ^B
Rajendra Sweta	130-135	4127 ^B	4753 ^A	4440 ^e	10418 ^A
Rajendra Bhagawati	120-125	4416 ^A	4260 ^A	4338 ^e	10246 ^A
Rajendra Mahsuri	~150	6612 ^B	7330 ^A	6971 ^a	14647 ^B
Arize 6444	135-140	6727 ^A	6683 ^A	6705 ^b	14231 ^A
Mean		5702 ^B	6106 ^A		12755 ^B
LSD ($p = 0.05$)					
Cultivar (C)	-	194			319
N treatment (N)	-	351			796
C × N interaction (p value)	-	< 0.001			0.001

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519 *A-B*, different uppercase superscript letters indicates significant difference within the RTNM
520 treatments at $p \leq 0.05$. RTNM₁ = SCMR₃₆N₂₀, RTNM₂ = SCMR₃₈N₂₀. *a-e*, treatments (cultivars)

521 with different lowercase superscript letters indicates significant difference at $p \leq 0.05$ based on
522 the LSD value of split plot ANOVA. # two-sample t -test at $p \leq 0.05$ was employed to compare
523 the significant difference within the N management treatments for individual cultivar.

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527 **Table 5.** Crop yields, N uptake, and partial factor productivity of applied N (PFP_N) of different
 528 rice cultivars to RTNM treatments [Experiment 2].
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Cultivar	Fertilizer N applied (kg ha ⁻¹)			N uptake (kg ha ⁻¹)			PFP _N
	RTNM ₁	RTNM ₂	Mean	RTNM ₁	RTNM ₂	Mean	RTN
27P31	78.3 ^A	85.0 ^A	81.7 ^b	117.5 ^A	124.8 ^A	121.2 ^c	75.6
RAU724	71.7 ^A	85.0 ^A	78.4 ^b	98.1 ^B	128.7 ^A	113.4 ^d	78.5
MTU7029	78.3 ^A	85.0 ^A	81.7 ^b	128.2 ^A	131.2 ^A	129.7 ^b	82.8
Rajendra Sweta	71.7 ^A	85.0 ^A	78.4 ^b	89.3 ^A	95.3 ^A	92.3 ^e	57.6
Rajendra Bhagawati	78.3 ^A	85.0 ^A	81.7 ^b	88.9 ^A	86.3 ^A	87.6 ^e	56.4
Rajendra Mahsuri	105.0 ^A	91.7 ^A	98.4 ^a	127.0 ^B	142.6 ^A	139.8 ^a	63.0
Arize 6444	85.0 ^A	78.3 ^A	81.7 ^b	128.2 ^A	123.0 ^A	125.6 ^{bc}	79.1
Mean	81.2 ^A	85.0 ^A		111.0 ^B	118.9 ^A		70.2
LSD (<i>p</i> = 0.05)							
Cultivar (C)	9.9			6.08			9.1
N treatment (N)	<i>ns</i>			11.4			<i>ns</i>
C × N interaction (<i>p</i> value)	<i>ns</i>			<i>ns</i>			<i>ns</i>

530
 531 *A-B*, different uppercase superscript letters indicates significant difference within the RTNM
 532 treatments at *p* ≤ 0.05. RTNM₁ = SCMR₃₆N₂₀, RTNM₂ = SCMR₃₈N₂₀. *a-e*, different lowercase
 533 superscript letters indicates significant difference within the cultivars at *p* ≤ 0.05 based on the
 534 LSD value of split plot ANOVA. # two-sample *t*-test at *p* ≤ 0.05 was employed to compare the
 535 significant difference within the N management treatments for individual cultivar.
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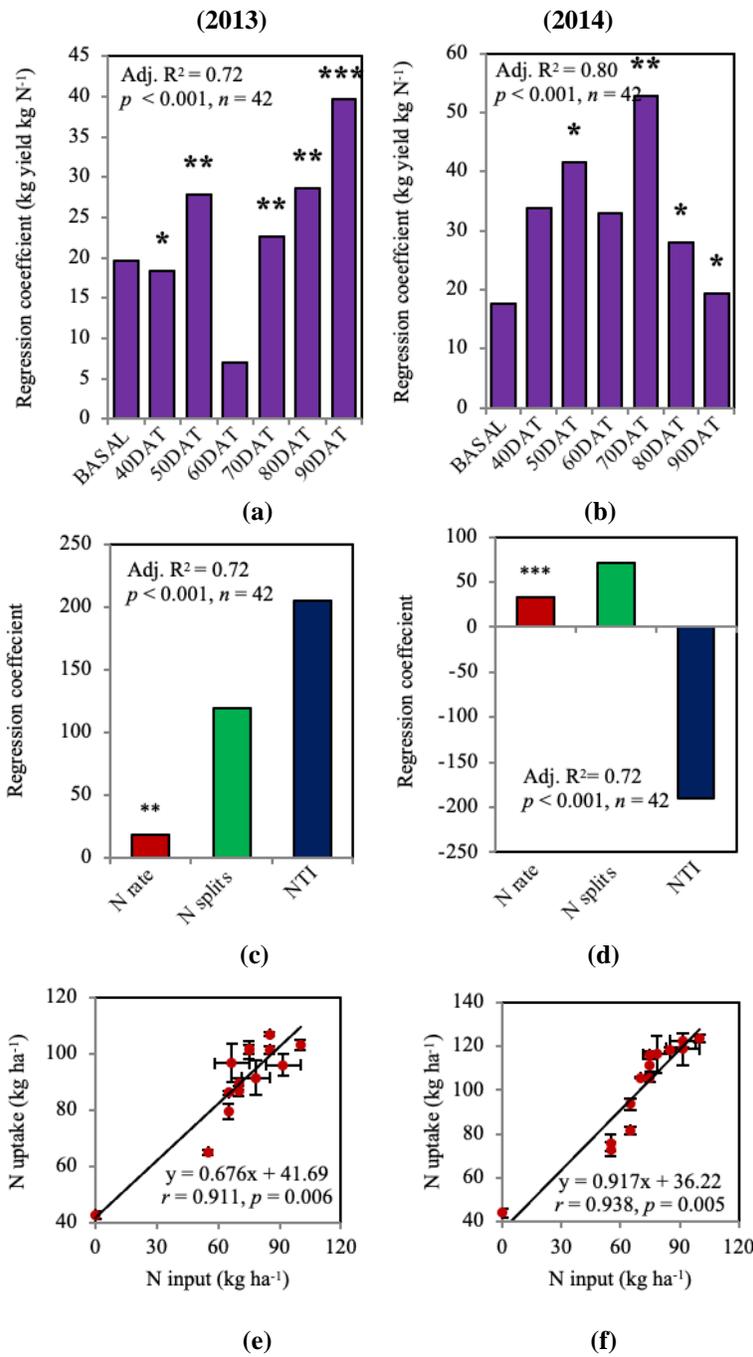
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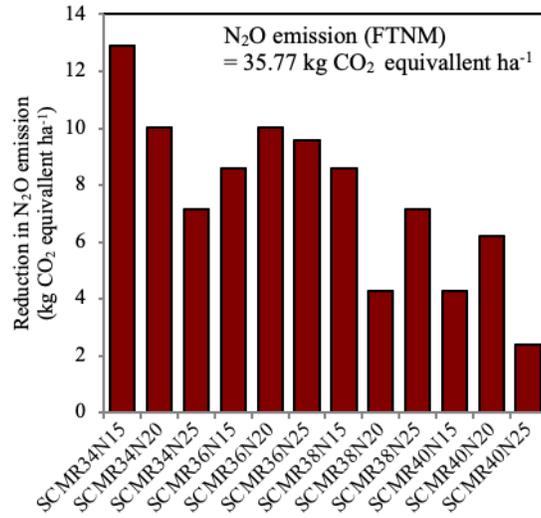


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Figure 1. Weightage of regression coefficient of N scheduling at different growth stages (independent variables) to explain their relative influence on rice grain yield (dependent variable) based on linear mixed regression model (a, b). Weightage of regression coefficient of N rate, N splits and NTI variables (independent variables) to explain their relative influence on rice yield (c, d). Linear relationship between fertilizer-N input and N uptake in rice in the year 2013 and 2014 (Experiment 1, Optimization trial) (e, f). NTI, N timing index defines the earliness or delay in the application of fertilizer-N. DAT, days after transplanting. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.



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Figure 2. Simulated estimates of the reduction in N₂O emission (projected) in different RTNM treatments compared to the FTNM treatment (Experiment 1).