A Physiological Approach: Nitrogen Management and Sub-1 Rice Varieties Grown in Flood Prone Ecosystem

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ABSTRACT

Farmers in flood prone areas mostly use only urea without any solid recommendations. Possibilities of recurrent flooding/submergence during the season are one of reasons for avoiding nutrient application, through it has a strong bearing on regeneration growth and yield of rice varieties after floods, hence suitable nutrient management strategies are essential to enhance the productivity. However, higher dose of N (60 Kg ha⁻¹ as basal) showed positive response on plant growth during submergence but higher elongation caused plant mortality during post oxidative phase. Meanwhile, popular package and practices among flood prone farmers, addition of Zero Kg N before submergence to minimized risk was not justified. So far, higher N applied as basal showed negative effect on survival during post submergence. Plants grown without N fertilizer before submergence showed 12-23% plant mortality in both Sub-1 rice varieties during post oxidative phase even though submerged field was substituted with higher dose of N @60 kg ha⁻¹ at 5th days after desubmergence, mainly due to energy starvation during submergence. After de-submergence frequent addition of split doses of N might be helpful to meet out the demand of submerged plants for faster recovery.

Keywords
Physiology Approach; Nitrogen Management; Flood Prone; Sub-1; Rice

Introduction

Rice is semi aquatic plants. Thus, traditionally grown rice cultivars in flooded soil have a reputation for growing well under flooded conditions. About 22 million ha of rice in South Asia is prone to flash flooding. In India, about 17.4 million ha of rainfed lowland rice are grown each year, of which 5.2 million ha are submergence-prone, out of the 2.65 million ha flash-flood prone areas, about 1.6 million ha rice are frequently inundated. Even during normal years, approximately 20% of the geographical area is affected by flooding, due to serious crisis most of the rice cultivars die within days of complete submergence, often resulting in total crop loss (Mackill et al., 2012).

These losses heavily affect rice farmers where alternative livelihood and food security options are limited. Farmers of flood prone
ecosystem kept their land fallow because of severe water stagnation. The productivity of such area is also very low because of excess water inundation and flooding. Overall, the estimated annual yield loss in deep water ecosystem alone amount to 1 million t. It these losses are particularly recovered, the average productivity in rainfed lowlands and flood prone area can be easily raised to 2 t ha$^{-1}$. A wide knowledge gap still exists between researchers and farmers about the need and progress in rice technology development for flood-prone ecosystem. Even the available technologies are not adopted by farmers because of inherent risk of crop failure and runoff losses of nutrient during floods. Poor characteristics of the soil and hydrology of flood prone environments also seems to limit technology development and option on a wider scale.

One of the major constraints to rice productivity enhancement across flood prone environment is lack of suitable improved seed, nutrient efficient and responsive varieties. The recent progress in knowledge about the development of flood tolerant varieties like Swarna Sub-1 and other sub1 consisting mega rice varieties. Sub-1 gene introgressed in it showed higher yield and survival in comparison to original Swarna, IRRI showed that sub-1 varieties give an average of 1–3.8 tones higher yield than non-sub-1types under 12–17 days of complete submergence (Singh et al., 2009) and which is still grown over 5 million ha and is currently the most popular rice variety of India.

Apart from this new technology developed for flood tolerant varieties, SUB1A gene has been transferred to 8 rice varieties, including the five mega rice varieties of India and Bangladesh (Collard et al., 2013). The new versions have a small segment of the donor genome containing SUB1A, while retaining the entire genome of the original varieties (Sarkar and Bhattacharjee, 2011). SUB1A was subsequently identified as the major determinant of submergence tolerance (Singh et al., 2010). In addition, balanced nutrition (NPK and FYM) together with lower seedling density in the seedbed are also very crucial in realizing full potential of these flood tolerant varieties. Recent research has shown that leaf N concentration is negatively correlated with plant survival under flooded conditions and addition of P seemed to enhance tolerance of plants grown on P-deficient soil (Ella and Ismail, 2006) or rainfed lowland soils (Singh et al., 2006).

In Sub1 rice, during flooding leaf foliage’s are decayed and after de-submergence new leaves emerged. Therefore, rice plants needs more N for faster recovery after de-submergence. Existing recommendation is not sufficient to fulfill the requirements of submerged rice plants. Most of the N flashes out due to flooding. Experiments on nutrient management before and after flooding (“recovery”) reveal that significant increase in yield could be achieved through application of nutrients, particularly nitrogen, because of its effects on stimulating recovery and early tillering (Ram et al., 2009). The rudimentary objective of this investigation is not to replace the existing recommendations; but to provide knowledge and advice on how these recommendations need to be adjusted in flood-prone areas.

**Materials and Methods**

The field experiment was conducted in wet seasons of two consecutive year 2018 and 2019 at the Instructional Farm, Department of Crop Physiology, Narendra Dev University of Agriculture and Technology, Kumarganj, Faizabad, situated between a latitude of $26^\circ.47'$ north and longitude of $82^\circ.12'$ east, on altitude of 113 meters above sea level in the gangetic alluvium of eastern Uttar Pradesh,
India. Present study, two Sub-1 rice varieties were used (Sambha Mahsuri Sub-1: V1, BR-11 Sub-1 V2. Nursery raising, seeds of Sambha Mahsuri Sub-1 and BR-11 Sub-1 varieties were sown in 2x2m² plot size. Transplanting was done in newly constructed cemented submergence tank (size: 20x17x1.5m; ground surface was not cemented). Thirty days old seedlings were transplanted at the spacing of 20x15 cm using multiple seedlings per hill in plot size 2.5x2m² in Randomized completely block design (RCBD) with 3 replications.

The experiments were comprises three nitrogen management practices including recommended practice (@N120:P40:K40 Kg ha⁻¹) i.e. (T1) ½ N (60 Kg ha⁻¹ through urea) and full dose of P (single super phosphate) and K (muriate of potash) applied at the time of transplanting and rest N apply in two split at consecutive 5th day after de-submergence and 1 week before flowering; (T2): ¼ N (30 Kg ha⁻¹) and full dose of P and K of recommended dose was applied at the time of transplanting, rest N applied in three split(@ 30 Kg ha⁻¹ in each), at 5th day, at 20th day de-submergence (at recovery) and 1 week before flowering and farmers practices of flood prone ecosystem (T3), only P and K (@40 Kg ha⁻¹) were applied as basal at the time of transplanting (BS) and N was applied during post flood @ 60, 30 and 30 Kg N ha⁻¹ at 5th days, 20th days de-submergence and one week before flowering respectively.

Stagnant submergence treatment was given at 60 days crop age (after 30 days transplanting) in submergence tanks. 40-45cm water depth was maintained by fresh water till 18th day of complete submergence. Plant survival was recorded at 5th and 20th days (at recovery) after desubmergence respectively. Recommended agronomic cultural practices and protective measure were applied accordingly. Three plants per replicate were initially tagged for growth observations which were recorded over three replications. Growth observations viz. plant height (cm), tiller number plant⁻¹, survival (%), dry weight (mg/p), soluble sugar content (mg/dry wt.), N-content (%), N-uptake (Kg/ha⁻¹), N use efficiency, days of 50% flowering, days to physiological maturity, regenerations (new leaf emergence) were taken at three consecutive events i.e. before submergence, after de-submergence and at recovery stage.

The total regenerated plants and new leaf emergence are counted at 5th days de submergence and recovery stage (after 20 days de-submergence).

Biochemical analysis

Biochemical estimation and nutrient analysis was done at before submergence, just after submergence, at recovery and maturity stages. Traits and methodology used viz; Total chlorophyll content (Arnon 1949), total soluble sugar (Yemm and Willis 1954), nitrogen content (Linder 1944), nitrogen uptake (computed in Kg ha⁻¹), nitrogen use efficiency (Quanbao et al., 2007). The statistical analysis of treatment on the patterns of randomized completely block design (RCBD) was carried out. The data were analyzed by appropriate statistical analysis (Gomez and Gomez, 1984).

Results and Discussion

In the present investigation various parameters used for evaluation of split doses of N, time of application and its combination with P and K. In normal condition application of higher nitrogen fertilizer alone or with potassium and phosphorus provide motility or strength to the plant. Application of nitrogen in main field greatly increases vigor in terms of plant height and dry matter accumulation before submergence in Sambha Mahsuri Sub-1 and BR-11 Sub-1 rice varieties.
parameters like plant height showed higher values (45-52) for the treatment with application of 60 Kg ha\(^{-1}\) N in combination with P 40 Kg ha\(^{-1}\) and K 40 Kg ha\(^{-1}\) as basal in both sub-I rice varieties (Table 1). It seems that high nitrogen in combination with phosphorus and potassium helpful in shoot growth. Present study also indicated that chlorophyll content and nitrogen uptake in treatment comprises higher dose of nitrogen was considerably more than lower dose and Zero Kg ha\(^{-1}\) N applied as basal. The uptake of higher nitrogen was observed in T\(_1\) followed by T\(_2\) and T\(_3\) i.e., (0.87-0.59 Kg ha\(^{-1}\)), (0.67-0.53 Kg ha\(^{-1}\)), (0.19-0.17 Kg ha\(^{-1}\)) in Sambha Mahsuri Sub\(_1\) and BR-11 Sub\(_1\) respectively. It is clearly indicated that higher dose of N helps in crop establishment, the above hypothesis also supported by Cassman and Stephen (2003). Significantly Sub\(_1\) rice varieties showed more than 90% survival and higher elongation rate when 60 days old plants were subjected for 18 days complete submergence in clear water and stagnant condition. Plant mortality due to submergence was very less in all treatments, because of older plant has paid advantages to sustained plant growth during submergence. Survival percentage was recorded after 5\(^{th}\) day of de-submergence maximum survival was recorded with (N30 Kg ha\(^{-1}\)) followed by (N60Kg ha\(^{-1}\)) and (N0 Kg ha\(^{-1}\)) i.e., (100%), (98-99%), (93-94%) respectively. Recent studies also indicated that older seedling up to (40-45days) had better survival than younger seedling (21-25days). Chaturvedi et.al (1995), reported that old seedling tend to have large carbohydrate reserves, therefore good survival during submergence. Present investigation, in spite of Sub\(_1\)-mediated suppression of elongation both Sub\(_1\) rice varieties showed (1.67 to 1.75 mm/day) elongation during submergence. This study clearly indicates indicated that shoot elongation during submergence act as constitutive traits when plant vigor enhanced through proper nutrient management before flood onset or older seedling subjected to flooding. Similarly in contrast Voesenek et al., 2006 reported that rapid shoot elongation increases carbohydrate consumption which resulting less survival percentage after flooding and Ella and Ismail 2006 also suggested that plant enrichment with nitrogen before submergence adversely affected survival after submergence. The correlation study clearly indicated that negative correlation between survival and N uptake (r= -0.09). The adverse effect of submergence of observed in post submergence phase when plants experience sudden increases in O\(_2\) concentration on the re-entry of air after submergence. Visual symptoms of injury normally are not apparent immediately after submergence, but these symptoms develop gradually during the post-oxidative phase. Present study also reflected that higher dose (N60:P40:K40 Kg ha\(^{-1}\)) or imbalanced fertilizer (N0:P40:K40 Kg ha\(^{-1}\)) resulted higher seedling mortality when flood receded from field. Several studies revealed that post oxidative damage leads tissue death. Setter et.al (2010) reported that after de-submergence leaf desiccated mainly due to large reduction in hydraulic conductivity in the leaf sheath. The water deficits are an important cause in the sequence of events rather than a mere result of injury. Survival after 20 days of de-submergence was higher when (30Kg ha\(^{-1}\)) N were applied as basal followed by (60 Kg ha\(^{-1}\)) N were applied as basal before submergence. Subsequently advantages of N rich plants of Sub\(_1\) rice varieties were observed in respect to faster recovery. Initial plant grown with (0Kg ha\(^{-1}\)) N before submergence exhausted soon therefore, higher plants mortality was recorded at 20\(^{th}\) day of de-submergence. Present study showed that maximum mortality were recorded (11.6 to 23.3) followed with higher doses of N (6.03 to 14.4) and (2.3 to 3.9) of both Sub\(_1\) rice varieties. Maximum mortality was obtained with (0N Kg ha\(^{-1}\) as basal before
submergence) because of plant suddenly shifted from anaerobic to aerobic condition so, that post oxidative damage done and reason for post oxidative damage is before submergence plant vigor was poor and plant were weaker in comparison to treatments T₁ and T₂ (60 Kg ha⁻¹ and 30Kg ha⁻¹ as basal respectively). So, that very less soluble CHO was available to generate more energy for their survival as well as for growth and development under submerged condition. Unlikely in T₁ and T₂ shoot elongation is higher during submergence resulting in poor vigor’s which causes tissue damage and mortality (Table.2). Further data generated regarding regeneration at recovery indicates that post submergence nitrogen application in field might be beneficial for recovery growth. Significantly the response of nitrogen was clearly shown in T₃ (0Kg ha⁻¹N) applied as basal. The correlation study clearly indicate that strong positive correlation between survival and N content (r= 0.85). Growth parameters like the dry weight and N uptake showed significantly high values (307-300%) and (550-300%) respectively (Table.3).

**Table.1** Effect of nitrogen management on survival (%), regeneration and new leaf emergence of Sub1 rice varieties grown under submerged condition (18 days of complete submergence)

| Treatments | Plant no. before submergence/ plot | Plant no. after submergence/ plot | Survival at 5th & 20th day after de-submergence (%) | Plant no. at recovery/ plot (20th day of de-submergence) | New leaf emergence |
|------------|-----------------------------------|----------------------------------|--------------------------------------------------|--------------------------------------------------|-------------------|
| T₁V₁       | 257                               | 255                              | 99                                               | 239.6 (-6.03)                                    | 93.9              |
| T₂V₁       | 256                               | 255                              | 100                                              | 249.0 (-2.3)                                     | 97.6              |
| T₃V₁       | 261                               | 250                              | 97                                               | 221.0 (-11.6)                                    | 88.4              |
| T₁V₂       | 264                               | 261                              | 98                                               | 240.3 (-14.4)                                    | 92                |
| T₂V₂       | 280                               | 280                              | 100                                              | 269.0 (-3.9)                                     | 96                |
| T₃V₂       | 294                               | 287                              | 93                                               | 220.0 (-23.3)                                    | 76.6              |

**Interaction** | V×T | V | T |

**CD at 5%** | 6.37 | 3.68 | 4.50 |

**Table.2** Effect of nitrogen management on plant height (cm) and dry weight (g) of Sub1 rice varieties grown under submerged condition (18 days of complete submergence)

| Treatments | Before submergence | After submergence | Elongation (mm/day) | At recovery (20th day after de-submergence) |
|------------|--------------------|-------------------|---------------------|---------------------------------------------|
|            | Plant height | Dry weight | Plant height | Dry weight | Plant height | Dry weight |
| T₁         | 48.9 | 3.23       | 79.6 | 2.28      | 1.70          | 101.4 | 3.83   |
| T₂         | 44.4 | 3.11       | 72.3 | 2.02      | 1.55          | 105.8 | 4.11   |
| T₃         | 39.6 | 2.72       | 57.2 | 1.16      | 0.97          | 77.2  | 4.68   |
| CD (P=0.05)| 3.11 | 0.23       | 5.78 | 0.09      | NS            | 3.45  | 0.15   |
Table.3 Effect of nitrogen management on total chlorophyll content (mg g\(^{-1}\) fresh weight), carbohydrate content (mg/g dry wt. of leaf) and nitrogen content (%) in shoot of Sub1 rice varieties grown under submerged condition (18 days of complete submergence)

| Treatments | Before submergence | After submergence | At recovery (20\(^{th}\) day after de-submergence) |
|------------|--------------------|-------------------|--------------------------------------------------|
|            | Total Chlorophyll  | Soluble Sugar     | N content | Total Chlorophyll | Soluble Sugar | N content | Total Chlorophyll | Soluble Sugar | N content |
|            | content            | content           |           | content          | content       |           | content          | content       |           |
| T1         | 1.60               | 160               | 1.67      | 0.84             | 121           | 0.90      | 2.06             | 130           | 1.10      |
| T2         | 1.38               | 142               | 1.61      | 0.64             | 112           | 0.80      | 2.62             | 128           | 1.21      |
| T3         | 0.87               | 103               | 0.90      | 0.38             | 78            | 0.53      | 3.06             | 142           | 1.40      |
| CD (P=0.05)| 0.90               | 6.63              | 0.19      | 0.32             | 3.77          | 3.25      | 3.08             | 4.94          | 0.39      |

Fig.1 Effect of nitrogen management on N uptake (Kgha\(^{-1}\)) of Sub1 rice varieties grown under submerged condition (18 days of complete submergence)

Fig.2 Effect of nitrogen management on N use efficiency (Kgha\(^{-1}\)) of Sub1 rice varieties grown under submerged condition (18 days of complete submergence)
It is concluded that nitrogen management in main field for sub1 interrogated rice varieties is not clear yet. Recommended package (N$_{120}$:K$_{40}$:K$_{40}$ Kg ha$^{-1}$) and practices 60Kg ha$^{-1}$ (1/2 dose of N) applied as basal was found not beneficial for sub1 interrogated rice varieties. It induced higher elongation when plants were subject for 18 days complete submergence compared with 30Kg ha$^{-1}$N and zero Kg ha$^{-1}$N with 40 Kg ha$^{-1}$P and K applied as basal.

Present investigation recommended dose of N was adjusted with four split doses i.e. 30Kg ha$^{-1}$ with combination of 40Kg ha$^{-1}$P and K applied as basal, subsequently rest N was applied 5$^{th}$, 20$^{th}$ days de-submergence and one week before flowering. Further, application of N was tested according to adopted practices of farmers, avoid to loss due to heavy rainfall i.e. 60Kg ha$^{-1}$N applied as basal 5$^{th}$ day of de-submergence and consequently rest amount of N applied in two split doses (30Kg ha$^{-1}$ each) at 20$^{th}$ days de-submergence and one week before flowering.

Maximun survival was obtained i.e. 97.6 and 92.0 percent in Sambha Mahsuri sub1 and BR-11 sub1 respectively. Therefore, higher dose of N as basal induce shoot elongation during submergence. Several other studies indicated that higher dose of N is found non-significant; Ella and Ismail (2006) reported that higher ‘N’ concentration of rice leaves is not beneficial when rice is subjected to flash flooding. In case of 0Kg ha$^{-1}$N and rest N applied in three split doses i.e. (5$^{th}$, 20$^{th}$, and 60$^{th}$ days after transplanting) was found non-beneficial due to poor vigor of plant before submergence.

Thus found more mortality % at recovery (20$^{th}$ d after de-submergence) stage of plant. Higher dose of N (60Kg ha$^{-1}$) and Zero Kg N ha$^{-1}$ were found non-beneficial due low survival % at post-oxidative phase. Whereas, in case of (30Kg ha$^{-1}$) N as basal and rest N is applied in three split doses i.e. (5$^{th}$ and 20$^{th}$ day after de-submergence and one week before flowering found beneficial and effective in submergence condition due to mortality % counted very squat after 18 days of complete submergence and at post-oxidative phase.

However, application of lower dose of N (30 Kg ha$^{-1}$) as basal and rest amount of N in three split doses along with P and K (40 Kg ha$^{-1}$) in field might be exploit to improve submergence tolerance and to obtained higher yield under flood prone eco-system due to higher survival after de-submergence corresponding to less post-oxidative damage through proper N management during, before and post submergence period.

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How to cite this article:
Anand Kumar Pandey, A. K. Singh, Alok Kumar Singh and Yadav. R. K. 2020. A Physiological Approach: Nitrogen Management and Sub-1 Rice Varieties Grown in Flood Prone Ecosystem. Int.J.Curr.Microbiol.App.Sci. 9(05): 38-45. doi: https://doi.org/10.20546/ijcmas.2020.905.003