## Effect of seedling age and submergence on chlorophyll content of rice cultivars

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## ABSTRACT

An experiment was conducted to investigate the impact of complete submergence of 7,10 and 15 days duration on the chlorophyll content of two flood susceptible cultivars and two flood resistant cultivars at the seedling stages(7-day old,10-day old and 15-day old). In comparison to the normal plants, total chlorophyll contents of chlorophyll a and chlorophyll b of the submerged seedlings were found to be lower at all the three growth stages of all the four cultivars. The rate of reduction of chlorophyll content during complete submergence was found to be remarkably higher in flood susceptible cultivars compared to that of resistant cultivars. However, this was more pronounced with increased duration of submergence.

Key words: Rice seedling, submergence, chlorophyll content

Different rice growing areas of India receive high rainfall from southwest monsoon and northeast during the months from June to October, resulting in flash floods during this period (Patnaik *et al.*, 1996).The submergence stress affects the crop in both shallow and semi-deep rainfed lowlands.Stagnant flooding suppresses or inhibits tillering and enhances lodging.Rainfed lowland rices are not high yielders but possess stability and adaptability to sustain vagaries of the environment.Present work was undertaken with an objective to find out the impact of seedling age and duration of submergence on the chlorophyll content of rice seedlings at a very early age which is likely to be one of the indicator trait for submergence tolerance.

The experiment was conducted at the Rice Research Station of the Orissa University of Agriculture and Technology,Bhubaneswar in green house tanks and in pot culture.The materials taken for the experiment were two semi-dwarf high-yielding varieties Jagannath and Manika and two improved varieties FR13A and FR 43B. In spite of their low yield potential and undesirable characters FR 13A and FR 43B were selected for having proven resistance for flash flood submergence.After surface sterilization with 0.1 per cent HgCl2 solution, presoaked seeds were sown in rows in enamel trays (40x25x5 cm), each containing 4 kg.of fine soil.After 5 days, the plants were thinned out leaving only ten uniform and normal healthy seedlings in each row. They were subjected to submergence in three separate tanks at three different ages, i.e.7 days, 10 days and 15days.Water level was maintained 60cm above the soil surface. Trays were taken out successively after 7 days, 10 days and 15 days of submergence after the completion of treatment. Water was drained off and plant samples were collected for the determination of chlorophyll a, chlorophyll b and total chlorophyll content.Experiment was conducted with three replications containing four sets of trays in each, where the first three were subjected to submergence and fourth was the control. Fresh samples (excluding roots) were immediately utilized for the determination of the chlorophyll content of the seedlings following the procedure of Yoshida et al, (1976). The absorbance of the plant extract was measured at 663nm and 645nm.

Chlorophyll content of the rice seedlings of three different ages (7-day old,10-day old and 15-day old) of four cultivars recorded before and after submergence have been presented in Table 1. The analysis of variance exhibited significance difference between the genotypes for chlorophyll a chlorophyll b and total chlorophyll content (Table 2).

| Varieties   | Age of<br>seedlings<br>(days) | Duration of submergence in days |         |               |                  |                  |                  |                  |                  |                  |                  |                 |                   |
|-------------|-------------------------------|---------------------------------|---------|---------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|-----------------|-------------------|
|             |                               | Befor                           | e subme | ergence       |                  | 7                |                  |                  | 10               |                  |                  | 15              |                   |
|             |                               | Chl.a                           | Chl.b   | Total<br>Chl. | Chl.a            | Chl.b            | Total<br>Chl.    | Chl.a            | Chl.b            | Total<br>Chl.    | Chl.a            | Chl.b           | Total<br>Chl.     |
| Jagannath   |                               | 1.127                           | 1.627   | 2.753         | 0.28<br>(75.15)  | 0.26<br>(84.02)  | 0.54<br>(80.38)  | 0.127<br>(88.73) | 0.153<br>(90.6)  | 0.28<br>(89.83)  | 0.11<br>(90.24)  | 0.11<br>(93.24  | 0.22<br>)(92.01)  |
| Manika      |                               | 0.93                            | 1.35    | 2.28          | 0.253<br>(72.8)  | 0.217<br>(83.93) | 0.47<br>(79.39)  | 0.12<br>(87.1)   | 0.183<br>(86.44) | 0.303<br>(86.71) | 0.12<br>(87.1)   |                 | 0.227<br>)(90.04) |
| FR 13A      | 7                             | 0.883                           | 1.24    | 2.123         | 0.553<br>(37.37) | 0.703<br>(43.31) | 1.257<br>(40.79) | 0.31<br>(64.89)  | 0.373<br>(69.92) | 0.683<br>(67.83) | 0.177<br>(79.95) | 0.243<br>(80.4) | 0.42<br>(80.22)   |
| FR 43B      |                               | 0.94                            | 1.3     | 2.24          | 0.613<br>(34.79) | 0.83<br>(36.15)  | 1.443<br>(35.58) | 0.33<br>(64.89)  | 0.443<br>(65.92) | 0.773<br>(65.49) | 0.173<br>(81.6)  | 0.277<br>(78.69 | 0.45<br>)(79.91)  |
| Susceptible |                               | 1.028                           | 1.488   | 2.516         | 0.266<br>(74.12) | 0.238<br>(84)    | 0.505<br>(79.92) | 0.123<br>(88.03) | 0.168<br>(88.71) | 0.291<br>(88.43) | 0.115<br>(88.81) |                 | 0.223<br>)(91.13) |
| Tolerant    |                               | 0.911                           | 1.27    | 2.181         | 0.583<br>(36.0)  | 0.766<br>(39.68) | 1.35<br>(38.1)   | 0.32<br>(64.87)  | 0.408<br>(67.87) | 0.728<br>(66.62) | 0.175<br>(80.79) | 0.26<br>(79.53  | 0.435<br>)(80.05) |
| Jagannath   |                               | 1.57                            | 1.52    | 3.09          | 0.277<br>(82.36) | 0.287<br>(81.12) | 0.563<br>(81.78) | 0.187<br>(88.09) | 0.203<br>(86.64) | 0.39<br>(87.38)  | 0.12<br>(92.36)  |                 | 0.277<br>)(91.04) |
| Manika      |                               | 1.367                           | 1.36    | 2.727         | 0.283<br>(79.3)  | 0.32<br>(76.47)  | 0.603<br>(77.89) | 0.18<br>(86.83)  | 0.267<br>(80.37) | 0.447<br>(83.61) | 0.117<br>(91.44) | 0.09<br>(93.38  | 0.207<br>)(92.41) |
| FR 13A      | 10                            | 1.05                            | 1.803   | 2.853         | 0.867<br>(17.42) | 0.957<br>(46.92) | 1.823<br>(36.1)  | 0.467<br>(55.52) | 0.593<br>(67.11) | 1.06<br>(62.85)  | 0.28<br>(73.33)  | 0.35<br>(80.59  | 0.63<br>)(77.92)  |
| FR 43B      |                               | 1.187                           | 1.68    | 2.867         | 0.863<br>(27.3)  | 1.083<br>(35.53) | 1.947<br>(32.09) | 0.55<br>(53.66)  | 0.743<br>(55.77) | 1.293<br>(54.9)  | 0.287<br>(75.82) | 0.383<br>(77.2) | 0.67<br>(76.63)   |
| Susceptible |                               | 1.468                           | 1.44    | 2.908         | 0.28<br>(80.92)  | 0.303<br>(78.96) | 0.583<br>(79.95) | 0.183<br>(87.53) | 0.235<br>(83.68) | 0.418<br>(85.62) | 0.118<br>(91.96) |                 | 0.242<br>)(91.68) |
| Tolerant    |                               | 1.118                           | 1.741   | 2.86          | 0.865<br>(22.63) | 1.02<br>(41.41)  | 1.885<br>(34.09) | 0.508<br>(54.56) | 0.668<br>(61.63) | 1.176<br>(58.88) | 0.283<br>(74.69) | 0.366<br>(78.98 | 0.65<br>)(77.27)  |
| Jagannath   |                               | 1.397                           | 1.8     | 3.197         | 0.743<br>(46.81) | 0.49<br>(72.77)  | 1.233<br>(61.43) | 0.293<br>(79.03) | 0.243<br>(86.5)  | 0.537<br>(83.2)  | 0.14<br>(89.98)  |                 | 0.313<br>)(90.21) |
| Manika      |                               | 1.413                           | 1.397   | 2.81          | 0.783<br>(44.59) | 0.563<br>(59.7)  | 1.347<br>(52.06) | 0.337<br>(76.15) | 0.317<br>(77.31) | 0.653<br>(76.76) | 0.12<br>(91.51)  |                 | 0.257<br>)(90.85) |
| FR 13A      | 15                            | 1.667                           | 2.613   | 4.28          | 1.127<br>(32.39) | 1.46<br>(44.13)  | 2.587<br>(39.56) | 0.67<br>59.81    | 0.83<br>(68.24)  | 1.5<br>(64.95)   | 0.367<br>(77.98) | 0.523<br>(79.98 | 0.89<br>)(79.21)  |
| FR 43B      |                               | 1.64                            | 2.677   | 4.317         | 1.153<br>(29.69) | 1.647<br>(38.48) | 2.817<br>(34.75) | 0.697<br>57.5    | 0.02<br>(61.9)   | 1.717<br>(60.23) | 0.417<br>(74.57) | 0.523<br>(80.46 | 0.94<br>)(78.23)  |
| Susceptible |                               | 1.405                           | 1.598   | 3.003         | 0.763<br>(45.69) | 0.526<br>(67.08) | 1.29<br>(57.04)  | 0.315<br>77.58   | 0.28<br>(82.48)  | 0.595<br>(80.19) | 0.13<br>(90.75)  |                 | 0.285<br>(90.51)  |

Table 1.Effect of submergence on chlorophyll content (mg./litre) of rice seedlings

Values in parentheses indicate percentage decrease in chlorophyll content.

In seven day old seedlings, with seven days and ten days of complete submergence, percentage of reduction of chlorophyll b was always found to be higher in comparison to that of chlorophyll a in both susceptible and tolerant varieties. But with fifteen days of complete submergence, susceptible varieties had higher percentage reduction of chlorophyll b (92.74%) than that of chlorophyll a (88.81%). However, tolerant cultivars had shown equal level of reduction (80.79% and 79.53%) of chlorophyll a and chlorophyll b. Ten day old seedlings had more reduction of chlorophyll a in susceptible cultivars while chlorophyll b reduction was higher in tolerant cultivars after both seven days and ten days of complete submergence. With enhanced duration of submergence to fifteen days, percentage of loss of chlorophyll was still more as the leaves were in the condition of decaying. It was found to be 91.68% in susceptible cultivars and 77.27% in tolerant

|            |           |                  | Analysis of variance |            |         |         |           |            |         |           |  |
|------------|-----------|------------------|----------------------|------------|---------|---------|-----------|------------|---------|-----------|--|
| Sources of | Degree of | Mean Square (MS) |                      |            |         |         |           |            |         |           |  |
| variation  | freedom   | 7-day old        |                      | 10-day old |         |         |           | 15-day old |         |           |  |
|            | (df)      | chl a            | chl b                | Total chl  | chl a   | chl b   | Total chl | chl a      | chl b   | Total chl |  |
| Rep.       | 2         | 0.001            | 0.004                | 0.003      | 0.001   | 0.001   | 0.002     | 0          | 0.001   | 0.001     |  |
| Var.       | 3         | 0.06**           | 0.144**              | 0.391**    | 0.143** | 0.725** | 1.499**   | 0.395**    | 2.417** | 4.772**   |  |
| Err.a      | 6         | 0                | 0.002                | 0.002      | 0.001   | 0.003   | 0.003     | 0.002      | 0.004   | 0.004     |  |
| Ds         | 3         | 1.663**          | 3.545**              | 10.049**   | 2.821** | 4.233** | 13.958**  | 3.742**    | 7.407** | 21.544**  |  |
| Var.xDs    | 9         | 0.04**           | 0.105**              | 0.271**    | 0.163** | 0.057** | 0.349**   | 0.006**    | 0.136** | 0.154**   |  |
| Err.b      | 24        | 0                | 0.001                | 0.001      | 0.001   | 0.004   | 0.003     | 0.001      | 0.003   | 0.001     |  |
| Total      | 47        |                  |                      |            |         |         |           |            |         |           |  |

Table.2 Effect of submergence on chlorophyll content(mg/litre) of rice seedlings

\*\*Significant at 0.05 and 0.01 level of probability

Rep.-Replication Err.-Error Var.xDs-Interaction of varieties and duration of submergence Var-Varieties Ds-Duration of submergence

cultivars.Loss in chlorophyll a and chlorophyll b were almost equal. In fifteen day old seedlings percentage of loss in total chlorophyll, chlorophyll a and chlorophyll b was found increasing gradually with the increased period of submergence. Averaged over the period of submergence treatments the two tolerant varieties FR13A and FR43B were found to have more chlorophyll after submergence. Reduction in chlorophyll b content was observed to be low in FR13A and FR43B irrespective of the period of submergence.

In the present study in comparison to the normal plants, total chlorophyll content as well as the contents of chlorophyll a and chlorophyll b of the submerged seedlings were found to be lower at all the three growth stages.Rate of loss of chlorophyll pigments during complete submergence was found to be more in susceptible varieties like Jagannath and Manika compared to that of tolerant varieties like FR13A and FR 43B and with the increased duration of submergence, the rate of depletion of chlorophyll content also increased. This confirms the observations of Ramakrishnayya et al. (1990) and Srivastava et al. (2007). Lower chlorophyll content in the submerged seedlings may be apparently due to the low light intensity. Deficiency of oxygen during complete submergence might also be a reason for slower synthesis of chlorophyll pigment (Kordan, 1976), which is drastic under 15 days of submergence. At early stages of growth, particularly at seedling stage, carbohydrate and other metabolites impart submergence tolerance (Adak and Das Gupta, 2001), which might be linked to the chlorophyll content.Decrease in chlorophyll content due to waterlogging has been linked with reduction in dry matter (Aleman *et al.*, 1984). Yellowing of leaves (chlorosis) was found to be a symptom of submergence of seedlings that agrees with the study of Rai *et al.* (2004). Due to low chlorophyll content, the younger seedlings might have shown higher rate of depletion.

Thus, the rate of depletion of chlorophyll pigments was maximum in flood susceptible cultivars in comparison to tolerant cultivars.Low light intensity or diffuse light and hypoxic/anoxic condition during complete submergence may be regarded as two major reasons for higher rate of depletion of chlorophyll content of submerged rice seedlings.

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