

## Biochemical Changes in Some Rice Varieties in Response to Waterlogged and Submerged Conditions

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### ABSTRACT

A field experiment was conducted during Kharif 2014 at Adaptive Research Station, Sakhigopal, Puri, Odisha to study the biochemical traits of different rice varieties grown under waterlogged and submerged conditions. Total chlorophyll content of the leaf was found to be highest in Sabita (3.26 mg/g FW). The carbohydrate content was found highest in Sabita (68.3 mg/g of D.LW) before the submergence and the percentage of reduction of carbohydrate was low during submergence which enabled the variety to sustain and regenerate after the submergence. Proline accumulation was found less and the percentage of increase due to submergence stress is less (+27.02%).

**Key words:** Biochemical, Chlophyll, Proline, Rice varities, Submergence

### INTRODUCTION

Rice a seed of grass species (*Oryza sativa*, Asian rice) or (*Oryza glaberrima*, African rice) is a monocot and normally grown in the tropical environment. It can also survive as a perennial crop. It is grown worldwide in varied ecosystems ranging from flood to drought condition and consumed by 60 percent of the world population. It is the agricultural community with the third world wide production after sugarcane and maize. It meets about 22 and 17 percent of the total calories and protein requirement respectively. Rice is

one of the world's important staple food crop, not only provides food but also influences religions, cultures and life styles since vedic period. According to the food and agricultural organization (FAO, 2009-10) rice is cultivated over an area of 161.80 million hectares with the production of 678 million tons in the world with the average productivity of 4.3 tons per ha. About 45 % of the rice area is under rain fed condition which is mainly distributed in south and southeast Asia but contributes only 25 % of the total rice production.

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As per the statistics published by International rice research institute (IRRI) estimated that 11 % of rice area in developing countries is under flood prone environment. With the advent of new technologies along with adoption of high yielding rice varieties coupled with improved agricultural management the rice production has been increased in last three decades enabling to reduce the chronic deficiency and excessive dependence of the imported food grains to period of self-sufficiency and surplus . Considering the population growth in India (2.72 percent / annum) our rice requirement ought to be increased to 25-30 million tons of milled rice in every decade. The pressure is likely to be accumulated in future and to achieve the targeted yield under reduced cultivable area, limitation of irrigation water and declined input efficiency and more over changing climate in all the major rice based cropping systems. This is a challenging task for our rice scientist to reduce the gap between the population growth rate and food production demand in forthcoming years. In India rice is grown in 43 million hectare of land more than a quarter of global rice (FAO STAT, 2007) with production of 91 million metric tons. It contributes a little less than a quarter of global production i.e. 0.1 million tons (IRRI, 2012). Rice production in India has increased during last 6 years by about 3.5 tons from 250.3 lakh tons during first five year plan period to 857.3 lakh tons during the tenth plan period. The average productivity of rice in India is 2.2 tons/ha which is far below than the global average of 2.7 tons/ha. India is expected to surpass the demand by the year

2030. According to Paroda<sup>7</sup> rice production in India is increasing very little due to several constraints. Hence, to achieve the targeted yield the genetical potential of the variety should be exploited. Oreke<sup>6</sup> reported that the genetical potential of the crop variety is limited by the environment including both biotic and abiotic stresses. Besides biotic stress, the abiotic stresses mainly drought, water logging, light, temperature, soil salinity greatly affect plant growth and yield (Dalmiya and Sawhny). The ability to design a plant ideotype for submergence prone environment depends on the available database on the physiology of crop plants, the nature of its environment and the interaction between the plant and environment.

## MATERIALS and METHODS

The field experiment was conducted during Kharif 2014 at Adaptive Research Station, Sakhigopal, Puri, Odisha to study the biochemical traits of different rice varieties grown under waterlogged and submerged conditions. The soil of experimental field of Sakhigopal farm is clay loam and texture slightly acidic in nature. The location is situated in 19°48' North latitude and 85°52' East longitude 20 km away from the Bay of Bengal with an altitude of 6 m amsl. The climate is relatively warm and humid in nature with short mild winter. The average annual rainfall of the region is 1408.8 mm which is received from southwest monsoon. Rice varieties taken as treatments in the experiment is in table-1.

**Table 1: Details of treatments**

CR DHAN 401- T <sub>1</sub>	OR-2331/14 - T <sub>12</sub>
FR-43B - T <sub>2</sub>	IR 85085 SUB17- T <sub>13</sub>
MANIKA- T <sub>3</sub>	TANMAYEE- T <sub>14</sub>
KALASIRA- T <sub>4</sub>	RAMBHA- T <sub>15</sub>
MAYURAKANTHA- T <sub>5</sub>	JALAMGNA - T <sub>16</sub>
OR-2328/05- T <sub>6</sub>	MAHALAXMI- T <sub>17</sub>
CR DHAN-500- T <sub>7</sub>	OR142/99 - T <sub>18</sub>
JALAMANI- T <sub>8</sub>	SALIBAHANA- T <sub>19</sub>
SABITA- T <sub>9</sub>	BANKOI- T <sub>20</sub>
CR-1030- T <sub>10</sub>	CR DHAN-505- T <sub>21</sub>
URBASI- T <sub>11</sub>	JAYANTI DHAN- T <sub>22</sub>

The date of sowing in the nursery bed was done from -. The nursery bed was developed for planting of 22 varieties of rice as mentioned above. Required amount of FYM and phosphatic fertilizers were well mixed with the soils of nursery for development of fertility of soil, before date of sowing. The twenty two varieties were sown by in lines with keeping appropriate spacing between the varieties. The irrigation channels were kept surrounding the speed beds. Frequent sprinkler irrigation was given for seed bed initially and after germination management was done in such a manner that the raised seed bed remained moistened without any standing water over its surface for one week. Thereafter standing water was maintained up to 3 cm. For the better growth of seedlings minimum N-fertilizer was given in seed bed. Before 7 days of rooting of seedling granular pesticide as per as recommendation was applied in seed bed in order to avoid the infection of disease and pest after the trans planting. After 21 days of sowing the seedling was up rooted for trans planting. 3.5.2 Land preparation. The main land (50mx40m) was ploughed with tractor after harvest of the previous crop. The FYM @5ton/ha was applied over the field. Again the yield was cross ploughed and leveled properly. Two days prior to transplanting for each sowing the irrigation was given to a plot size of 400 sq.m (50mlx8m) for puddling by the power tiller and a little standing water was maintained in the field. Further, main plot (50mx8m) was divided into three stripes representing three replication.

Each replication was subdivided into 22 subplots for the allocation of varieties. 3.5.3 Fertilizer application Before transplanting of seedlings and basal dose of 15 Kg N,30 Kg P2O5 and 30 Kg K2O per hectare were applied and mixed thoroughly in soil during puddling. Rest nitrogen was top dressed twice. The first top dressing of nitrogen@ 30 Kg/ha in the form of urea was applied after 15 days of trans planting. The second top dressing of nitrogen @ 15 Kg/ha in the form of urea was applied at 112 days after transplanting. ~ 25 ~ 3.5.4 Trans planting Twenty one days of old seedlings of rice varieties were transplanted

with a spacing of 20 cm x 10 cm having two seedlings per hill.

Total chlorophyll content in the leaves were determined by using the method stated by Arnon<sup>2</sup>. Proline estimation was done as per the protocol described by Sadasivam and Manickam<sup>9</sup> and Gilmour *et al*<sup>4</sup>. Carbohydrate content of plant samples was determined by following procedure<sup>12</sup>.

## RESULTS AND DISCUSSION

### Chlorophyll

Variation of chlorophyll-a content was noted among the genotypes due to the effect of submergence (**Table-2**). From the data presented in the table indicated that chlorophyll-a content before the submergence was highest in Sabita (3.26 mg/g FW) followed by OR-2331/14 (2.99 mg/g FW). But the minimum value of the same was exhibited by Bankoi (2.26 mg/g FW). After the submergence there was reduction in chlorophyll-a content irrespective of the genotypes which range from 28.22% in Sabita to 38.05% in Bankoi. The maximum chlorophyll content after the submergence was contributed by Sabita (2.34 mg/g FW), whereas the lowest value of the same was exhibited by Bankoi (1.40 mg/g FW). Comparison of chlorophyll-b content measured before and after submergence was presented in **Table-2**. Data presented in table indicated that highest value was recorded in Sabita (0.99 mg/g FW) followed by FR-43B (0.98 mg/g FW) where as lowest value of the same was shown in Bankoi (0.53 mg/g FW) before the submergence. Like chlorophyll-a content there was reduction in chlorophyll-b content under submerged condition in all the cultivar. The minimum percentage of reduction (10.00%) was recorded in or-2331/14 where as the maximum reduction (16.07%) was noted in Mayurakantha. The highest value of chlorophyll-b content (0.89 mg/g FW) was contributed by Sabita but the lowest value of the same (0.45 mg/g FW) was exhibited By Bankoi. From the correlation table-18 it was found that there was positive correlation between the chlorophyll-b and nitrogen uptake by the leaf and yield and yield attributing characters.

The total chlorophyll content recorded before the submergence was highest in Sabita (4.25 mg/g FW) followed by FR-43B (3.99 mg/g FW) where as minimum value (2.80 mg/g FW) was recorded in Bankoi. In general there was reduction in total chlorophyll content irrespective of cultivars under test due to submergence. The percentage of reduction range from 23.53% in Sabita to 32.86% in Bankoi. Highest value of total chlorophyll content was contributed by Sabita (3.25 mg/g FW) where as the lowest value was shown in Bankoi (1.88 mg/g FW) under submergence condition. The present experiment indicated that there was reduction in chlorophyll contents of all the genotypes due to submergence condition. The chlorophyll reduction is accompanied with carbohydrate content before the submergence which helps in minimum shoot elongation. It ultimately regulates the plant hormone like ethylene and GA. The chlorophyllase enzyme activity is triggered by the hormone ethylene which reduced the chlorophyll contents. There is maximum degeneration of chlorophyll content in Bankoi (32.86%) followed by Kalasira (32.30%) where as minimum value of the same was shown in Sabita (23.53%). Chlorophyll reduction was less in submergence genotypes due to reduction in ethylene production<sup>10</sup>.

#### Carbohydrate content of shoot

Comparison of carbohydrate content among the cultivars estimated before and after submergence was presented in **Table-3**. Data presented in Table indicated that the carbohydrate content of the shoot before the submergence was highest in Sabita (68.3 mg/g DW) followed by FR-43B (51.26 mg/g DW). On the contrary the lowest value (32.67 mg/g DW) was exhibited by Bankoi. The carbohydrate content estimated after the submergence indicated that the highest carbohydrate content was exhibited by Sabita (60.7 mg/g DW), whereas the minimum value (18.8 mg/g DW) of the same was shown in Bankoi. The percentage of reduction of carbohydrate content range from 11.13% in Sabita to 42.45% in Bankoi among the

cultivars due to submergence effect. Significant differences among the genotypes are noted.

Submergence tolerance in rice is not only governed by a single factor by a single factor but it is governed by a number of factors. From the present experiment it was found that high carbohydrate content (68.3 mg/g DW) was noted in Sabita followed by Jalamani (60.57 mg/g DW) where as the lowest value of the same was exhibited by Bankoi (32.67 mg/g DW). From the data it was clear that higher levels of initial carbohydrate acts as buffer stock and its continued availability during submergence is critical for the survival and growth of rice under submergence condition. Metabolic energy needed by the plant during submergence is primarily supplied from stored carbohydrate present in the tissue. Irrespective of varieties it was found that there was reduction of carbohydrate content of shoot. It is perhaps due to depletion of photosynthetic rate under submerged condition and as discussed earlier it was based on loss of chlorophyll fluorescence. It may be also lowering stomata conductance, intercellular CO<sub>2</sub> concentration as well as denaturing of the photosynthetic machineries. Moreover; inundation owing to the submergence also limits the carboxylation by low/intermediate intercellular CO<sub>2</sub> concentrations that may also subside the RUBP-Carboxylase activity, rather more favoring the oxygenation. This deviation ratio of carboxylation to oxygenation under submergence is more serious for switching over the tissues to make it more prone to photorespiration. In submergence rice cultivars when the leaves, stems and roots are completely submerged the rate of depletion of carbohydrate is very slow than the susceptible variety. Drastic reduction of carbohydrate which leads to high rate of anaerobic fermentation and production of ethanol at toxic level<sup>11</sup>. It may be also happened that the genotypes tolerant to submergence has higher Alpha amylase activity in the leaf and culm correlation study indicated that survival of rice genotypes was positively correlated with the

level of carbohydrate present in the culm and leaf before the submergence<sup>3</sup> which aggravated by the present findings.

### Proline accumulation

Proline accumulation varies significantly among the genotypes under test when recorded before the submergence. From the **Table-3** it was noted that highest proline accumulation (323.33 µg/g FW) was contributed by Kalasira followed by Bankoi (315.00 µg/g FW) whereas the lowest value of the same was exhibited by Sabita (169.00 µg/g FW). When the crop was submerged for 12 days there was increase in proline accumulation in all the genotypes but the percentage of increase differ from cultivar to cultivar. The minimum percentage of proline accumulation was found in Sabita (214.67 µg/g FW) whereas the maximum percentage of same was found in Bankoi (520.33 µg/g FW). There was a positive correlation between the

percentage of increase of proline accumulation with the tolerant capacity of the variety. From the **Table-3** it was found that the proline accumulation is less in Sabita (214.67 mg/g FW) than the susceptible variety Bankoi (520.33 mg/g FW). Accumulation of proline is maximum in susceptible cultivar under submerged condition<sup>8</sup>. So high accumulation of proline is considered to be an indication of submergence damage which occurs due to the hydrolysis of the proteins. Proline accumulation increased due to the suppression of mitochondrial nutrient transport under submerged stress condition<sup>1</sup>. The tolerant genotypes under submerged condition appear to efficiently utilize better energy due to normal mitochondria activity for which proline accumulation was apparently found lower than the susceptible variety.

**Table 2: Chlorophyll content in response to submerged condition in different rice cultivars**

Variety	Chl-a		Chl-b		Total chl.		C.S.I.
	BS (75 DAS)	AS (90 DAS)	BS (75 DAS)	AS (90 DAS)	BS (75 DAS)	AS (90 DAS)	
Sabita	3.26	2.34 (-28.22%)	0.99	0.89 (-10.10%)	4.25	3.25 (-23.53%)	0.764
FR-43B	2.94	2.09 (-28.91%)	0.98	0.88 (-10.20%)	3.99	3.03 (-24.06%)	0.759
Jalamgna	2.70	1.91 (-29.26%)	0.93	0.83 (-10.75%)	3.62	2.75 (-24.03%)	0.759
OR-2331/14	2.99	2.10 (-29.77%)	0.90	0.81 (-10.00%)	3.91	2.95 (-24.55%)	0.754
IR 85085 SUB-17	2.71	1.89 (-30.26%)	0.78	0.69 (-11.54%)	3.49	2.64 (-24.36%)	0.756
JayantiDhana	2.81	1.96 (-30.25%)	0.76	0.67 (-11.84%)	3.59	2.70 (-24.79%)	0.752
Jalamani	2.69	1.86 (-30.86%)	0.77	0.67 (-12.99%)	3.46	2.59 (-25.14%)	0.748
CR dhan-500	2.68	1.84 (-31.34%)	0.76	0.66 (-13.16%)	3.45	2.56 (-25.80%)	0.742
CR dhan-401	2.64	1.80 (-31.82%)	0.66	0.60 (-9.09%)	3.35	2.47 (-26.27%)	0.737
CR dhan-505	2.59	1.76 (-32.05%)	0.70	0.61 (-12.86%)	3.29	2.42 (-26.44%)	0.735
Mahalaxmi	2.36	1.59 (-32.63%)	0.67	0.59 (-11.94%)	3.06	2.23 (-27.12%)	0.728
Manika	2.45	1.64 (-33.06%)	0.69	0.59 (-14.49%)	3.14	2.28 (-27.39%)	0.726
CR dhan-1030	2.46	1.64 (-33.33%)	0.67	0.58 (-13.43%)	3.14	2.27 (-27.71%)	0.723
OR-142/99	2.38	1.58 (-33.61%)	0.66	0.57 (-13.64%)	3.06	2.20 (-28.10%)	0.718
Tanmayee	2.38	1.56 (-34.45%)	0.61	0.52 (-14.75%)	2.99	2.15 (-28.09%)	0.719
Urbashi	2.32	1.51 (-34.91%)	0.61	0.52 (-14.75%)	3.06	2.10 (-31.37%)	0.686
Salibahana	2.31	1.49 (-35.50%)	0.65	0.55 (-15.38%)	2.96	2.10 (-29.05%)	0.709
Rambha	2.26	1.45 (-35.84%)	0.64	0.54 (-15.63%)	2.91	2.04 (-29.90%)	0.701
OR/2328/05	2.40	1.52 (-36.67%)	0.59	0.50 (-15.25%)	2.99	2.01 (-32.78%)	0.672
Mayurakantha	2.39	1.50 (-37.49%)	0.56	0.47 (-16.07%)	2.98	2.04 (-31.54%)	0.684
Kalasira	2.33	1.45 (-37.77%)	0.57	0.48 (-15.79%)	2.91	1.97 (-32.30%)	0.676
Bankoi	2.26	1.40 (-38.05%)	0.53	0.45 (-15.09%)	2.80	1.88 (-32.86%)	0.671
SEM	0.05	0.02	0.02	0.02	0.05	0.07	
C.D 5%	0.14	0.06	0.05	0.06	0.14	0.19	
C.V	4.04	1.90	3.93	6.40	4.04	4.71	

**Table 3: Variation in carbohydrate and proline content of plant in response to submerged condition of rice cultivars**

VARIETY	CARBOHYDRATE (mg/g DW)		PROLINE ( $\mu$ g/g FW)	
	BS	AS	BS	AS
SABITA	68.3	60.7 (-11.13%)	169.00	214.67 (+27.02%)
FR-43B	51.26	45.6 (-11.04%)	177.67	235.00 (+32.27%)
JALAMGNA	48.13	42.33 (-12.05%)	181.33	247.33 (+36.40%)
OR-2331/14	54.27	46.06 (-14.13%)	186.67	258.67 (+38.57%)
IR 85085 SUB-17	48.75	40.43 (-17.07%)	192.33	267.67 (+39.17%)
JAYANTI DHANA	48.43	39.67 (-18.09%)	199.33	281.33 (+41.14%)
JALAMANI	60.57	49.03 (-19.05%)	201.67	286.00 (+41.82%)
CR DHAN-500	58.36	47.23 (-19.07%)	199.00	305.00 (+53.27%)
CR DHAN-401	51.5	41.67 (-19.09%)	222.33	322.00 (+44.83%)
CR DHAN-505	47.05	37.95 (-19.34%)	227.33	325.00 (+42.96%)
MAHALAXMI	40.67	32.73 (-19.52%)	213.33	335.00 (+57.03%)
MANIKA	39.56	31.17 (-21.21%)	218.00	346.00 (+58.72%)
CR DHANA-1030	47.8	37.5 (-21.55%)	232.67	345.67 (+48.57%)
OR-142/99	36.64	28.57 (-22.03%)	229.67	349.67 (+52.25%)
TANMAYEE	44.07	34.3 (-22.17%)	224.00	360.33 (+60.86%)
URBASHI	48.4	37.23 (-23.08%)	224.67	399.67 (+77.89%)
SALIBAHANA	56.3	42.77 (-24.03%)	250.33	402.67 (60.86%)
RAMBHA	52.36	39.77 (-24.05%)	303.67	409.00 (34.69%)
OR/2328/05	45.21	34.3 (-24.13%)	290.67	445.33 (+53.21%)
MAYURAKANTHA	32.86	24.87 (-24.32%)	297.67	474.33 (+59.35%)
KALASIRA	34.57	23.47 (-32.11%)	323.33	519.33 (+60.62%)
BANKOI	32.67	18.8 (-42.45%)	315.00	520.33 (+65.18%)
SEM	0.53	1.56	1.27	1.09
C.D 5%	1.51	4.45	3.62	3.10
C.V	1.92	7.10	0.95	0.54

## CONCLUSIONS

It can be concluded that among the varieties sabita, FR-43B, Jalamgna contributed higher carbohydrate reserves, low proline accumulation and less decrease in chlorophyll content with submergence tolerance. The tolerance capacity was mainly due slow reduction of buffer stock of carbohydrate before the submergence for energy supply during flash flood. Therefore sabita, FR-43B, Jalamgna can be recommended to the farmers for cultivation under waterlogged and submerged conditions.

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