Physiological and Biochemical Perspectives of Rice: Activities, Achievements and Aspirations

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SUMMARY

Since its inception in 1960, the Division of Crop Physiology and Biochemistry at NRRI has made significant contribution in the area of basic and applied physiology and biochemistry and improvement of nutritional quality of rice. In crop physiology, over the years major thrust was given on understanding the physiological, biochemical and molecular mechanism of tolerance to different abiotic stresses *viz*., drought, salinity, submergence, waterlogging, heat, low light, etc. occurring simultaneously or in tandem during the cropping season. Screening of large numbers genotypes for multiple seasons resulted in identification of unique genotypes having novel sources of tolerance to one or more abiotic stresses, improved photosynthetic efficiency and improved plant types, which were used further for varietal development or for identification of novel QTLs. Besides this, significant progress has been made for improving photosynthetic efficiency for yield maximization in rice. These studies not only identified highly photosynthetically active rice genotypes, but also analysed the unique plant type features of these genotypes which can be transferred to other rice cultivars for enhancing their photosynthetic efficiency. Since, last few years, the work on developing C4 rice was initiated in the division with a vision to further enhance the photosynthetic efficiency and cutting the photorespiratory losses of currently grown rice cultivars. In biochemistry, since long efforts were made to identify rice genotypes with high grain protein content and understand their underlying mechanisms. Recently, this institute has released country's first high protein rice cultivar CR Dhan 310 having average grain protein content of more than 10%, using the genotypes identified at our division. Later one more variety, CR Dhan 311 was released for high grain protein and moderate Zn content, which was also developed using the genotypes identified at our division. The Division is also working on development of low Glycaemic Index (GI) rice suitable for diabetic people and multiple nutrient rich rice with improved bioavailability of essential minerals to counter wide spreading malnourishment in our country. The achievements of the division were documented in form of peer reviewed research articles in journals of national and international repute. Besides the core research activities, the Division has produced 22 and 9 PhD and MSc scholars, respectively. In this compilation major activities, achievements and aspirations of the Division are presented.

1. INTRODUCTION

Over the past three decades, the advent of modern high yielding rice varieties and associated 'Green Revolution' technologies have almost doubled the rice production globally. Several countries in South and Southeast Asia have not only achieved self-sufficiency in rice production, but also a few of them, including India, have become net exporters of rice. However, with the growing population, the demand for rice will also continue to grow. A challenges in the future would be to produce more rice from less land, with less water, less labour and lesser use of pesticides. This would require innovative research and technologies and policies that promote increased rice production. In the Division of Crop Physiology and Biochemistry, based on analysis of current status of rice production and review of the constraints, constant research efforts are undergoing for improvement of rice in terms of physiological and biochemical traits to meet the targeted production of rice. We are focussing our research on three aspects: 1) rice production under different abiotic stress environment (individual or multiple), 2) improvement of photosynthetic efficiency of rice and 3) improvement of nutritional and grain quality aspects of rice.

Abiotic stress can impose limitations on crop productivity and also limit land available for farming, often in regions that can ill afford such constraints, thus highlighting a greater need for understanding how plants respond to adverse conditions with the hope of improving tolerance of plants to environmental stresses. Most rice varieties are severely injured by abiotic stresses, with strong climatic impacts. Water deficit is a widespread challenge to sustainable agriculture. Evidence suggests that various pathways, mechanisms, and morphological modifications are among the most important factors in plant responses to dehydration. It is true that plants require water for growth but excess water that occurs during submergence or waterlogging is harmful or even lethal. A submerged plant is defined as a plant standing in water with at least part of the terminal above the water or completely covered with water, whereas waterlogging is defined as, a condition of the soil in which excess water limits gas diffusion in rhizosphere. Rice is also a salt-susceptible crop. Currently, one third of the world in agricultural land is affected by salinity. Salinity, both soil and water, has negative effect on rice production. Elevated levels of sodium in agricultural lands are becoming a serious threat to the world agriculture. This demand of increased food supply can be fulfilled only if we utilize all available land resources to their full potential. Understanding rice physiology to stress may help breeding for more tolerant varieties.

Photosynthesis is the process where plants capture energy from sunlight and convert it into biochemical energy, which is subsequently used to support nearly all life on Earth. Recent technological developments now provide us with the means to engineer changes to photosynthesis that would not have been possible previously. The concept is to switch rice from using the C_3

photosynthetic pathway, which is not very efficient, to using the more efficient C4 photosynthetic pathway. In most plants, including rice, CO_2 is first fixed into a compound with three carbon atoms, hence it is known as 'C3 photosynthesis'. The enzyme that fixes CO_2 into sugar reacts with oxygen in an energy-wasting process known as photorespiration, which occurs especially at higher temperate tropics where most rice is grown and causes a dramatic reduction in the amount of CO_2 to convert to sugar. C4 plants, on the other hand, have a naturally evolved way to minimise this energy wasting process by using an oxygen insensitive enzyme first to fix CO_2 and then efficiently convert CO_2 into sugar. This increased efficiency is accompanied by increased water and nitrogen use efficiency and improved adaptation to hotter and dryer environments. Several international consortia are currently working toward improving photosynthesis. Grafting the more efficient C4 photosynthetic pathway onto cereals that produce the most desirable grains in rice and wheat is a bold venture. A new bioengineering approach for boosting photosynthesis in rice plants could increase grain yield by an approach, called photorespiratory bypass. The genetically engineered plants would be greener and larger and showed increased photosynthetic efficiency and productivity under field conditions, with particular advantages in bright light.

As countries reach self-sufficiency in rice production, the demand by the consumer for better quality rice has increased. In our division, we are working on rice grain quality which includes combination of physical and chemical characteristics that are required for a specific use by the consumers. Thus, grain quality can be a combination of many factors such as smell (aroma), size, cooking characteristics, colour, nutritional value, percent whole grains, etc. Consumer preferences driven research on grain quality are underway in the division. Another research focus of our division is on rice grain protein, which is nutritionally superior over most of the other food cereals in terms of amino acid composition. The enrichment of rice grains with protein would have a positive effect on the health of billions of people around the globe particularly the poor and the malnourished. The Chapter provides a glimpse of rice physiology under different abiotic stress environments, the way of improving photosynthetic efficiency and grain and nutritional quality of rice.

2. GENESIS OF THE DIVISION

The Division of Plant Physiology was established at ICAR-National Rice Research Institute, Cuttack in June, 1960 as a separate section. The mandate of the Section was to study the physiology of rice under various abiotic stresses such as drought, salinity, submergence, water-logging and other environmental stresses. The Section envisaged identification and characterisation of tolerant genotypes, development of appropriate screening techniques and analysis of constraints for productivity especially under stress situations. The Section was upgraded to a Division during the 4th Five Year Plan in 1972. During this period financial help was received in terms of different national and international projects, which strengthened and intensified the research work of the division. The Rockefeller Grant (prior to 1960), Colombo Plan (aid from Japan in 1974), PL-480 Project for Photosynthesis (1980-90), Canadian line of credit (1978), IRRI-Collaborative Program under IRWYN (1982-84), National Fellow Award (1984-88), Emeritus Scientist Award (1989-92) were few of them which immensely helped in developing divisional infrastructure, establishment of good research laboratories with modern equipment for achieving research goals. A Government of India certified 'Radio Tracer Laboratory' was also established in 1972 to support various work related to radio biology in the division.

Initially, physiological research on salt-tolerance in rice was carried out at the saline sub-station located at Canning, West Bengal. With the transfer of this sub-station to CSSRI, Karnal in 1972, work on salt tolerance was shifted to the main institute. Similarly, the physiological research on drought tolerance was initially started in at Cuttack, but after the establishment of Central Rainfed Upland Rice Research Station (a sub-staion of NRRI) at Hazaribagh, Jharkhand, majority of the drought research was shifted to that center from this Institute. Currently major focus of the Division is on understanding the mechanism of tolerance to individual and multiple abiotic stresses along with improvement of photosynthetic efficiency in rice and improving grain and nutritional quality in rice. Besides, investigations on physiological analysis of chemical regulation of growth, heterosis in hybrid rice derived from cytoplasmic male sterility (CMS) lines and simulation models for low light monsoon rice also progressed well in due course of time.

Work on rice biochemistry was initiated in 1970s. The objective was to study all aspects of rice quality with particular emphasis on consumers' preference. This covered varietal differences in hulling, cooking quality and nutrient content, loss of proteins and vitamins in milling, influence of various fertiliser and cultural practices on the protein content, influence of storage on the physical and chemical changes that take place in stored grains, effect of moisture content, aging, time and method of harvesting, drying etc. on the milling quality and the genetics of the various quality attributes. Subsequently during 1980-1996 the activity of Biochemistry division was concentrated on biochemical, technological, cooking and eating quality of rice, aromatic rice, protein quality, quantity and biosynthesis, biochemical mechanism associated with biotic and abiotic stresses, rice processing and by-product utilization etc. As per the suggestion of QRT (1997-2004) the Physiology and Biochemistry divisions were merged as Biochemistry, Physiology and Environmental Science Division. Subsequently on the basis of next QRT (2005-2012) recommendation the division of Biochemistry, Physiology and Environmental Sciences was renamed as Crop Physiology and Biochemistry Division to undertake research on the following aspects: i) study and development of health rice, ii) designing and tailoring new plant type of higher potential yield and iii) stress physiology.

3. OBJECTIVES OF THE DIVISION

Since beginning, the major goal of division was to conduct basic and strategic research on rice to enhance productivity and profitability by improving the production per unit area and per unit time under different environmental conditions. On this note, the research programmes of Physiology and Biochemistry of rice has three major thrust areas *viz.* rice grain and nutritional quality, abiotic stress physiology with special emphasis on mechanistic understanding and enhancing photosynthetic efficiency. The major research programmes of the division have the following objectives.

- Study rice grain quality in relation to GI, mineral (Fe/Zn) bioavailability and protein content.
- Identification of donor sources of relevant physiological traits and understanding the physiological/biochemical/molecular mechanisms underlying the plant responses to abiotic stresses
- Enhancement of photosynthetic efficiency by introduction of C4 pathway and minimizing photorespiration.

4. MAJOR ACHIEVEMENTS

4.1. Abiotic Stress Tolerance in Rice

As 60-70% of monsoon rice (*Kharif* season) is subjected to stresses like drought, salinity or water-logging, both laboratory and field investigations were conducted under defined conditions to isolate and characterise elite lines to develop simple screening techniques.

4.1.1. Drought Stress

Since beginning, drought tolerance studies were one of the thrust areas of the Division. More than 10,000 rice germplasm accessions comprising of upland rice, lowland rice, deep water rice, wild rice, advanced breeding lines and fixed lines were screened under field condition during the dry seasons. Among all the tested genotypes, around 250 lines were identified as vegetative stage drought tolerant characterized by SES score '1' following the standard international method (IRRI SES method on 1-9 scale) under forced drought stress with an available soil moisture content of 5–8%, soil moisture tension -40 to -50 kPa and ground water table down below 100 cm. Three rice germplasm lines named Mahulata (AC No. 35186), Brahman Nakhi (AC- 35678) and Sal-kaiin (AC- 34992) were registered as new sources of vegetative stage drought tolerance through Plant Germplasm Registration Committee (PGRC), ICAR-NBPGR, New Delhi. Another unique breeding line CR 143-2-2 developed at ICAR-NRRI was identified as both vegetative as well as reproductive stage drought tolerant rice variety having a yield potential of more than 1.2 t ha^{-1} under drought stress condition, was registered in ICAR-NBPGR, New Delhi.

Besides the cultivated rice genotypes, two wild rice accessions of *Oryza nivara* (IC -330470 and IC -330611) collected from West Bengal, were also identified as vegetative stage drought tolerant lines with a SES score of '0' & '1'. For reproductive stage drought stress, BVD-109 (2.15 t ha^{-1}), Kalakeri (2.08 t ha^{-1}), IC 416249 (2.02 t ha^{-1}) and CR 143-2-2 (1.90) are identified as promising genotypes with grain yield of 1.90 to 2.15 t ha^{-1}, while AC 27675, IC 516130, Udayagiri, Indira, Vandana and CR 143-2-2 showed better tolerance in terms of other morpho-physiological traits like relative water content (RWC), drought score, chlorophyll florescence traits etc.

In shallow rain-fed rice agro-ecosystems, drought stress can occur at any growth stage and can cause a significant yield reduction. During recent years, some rice varieties possessing reproductive-stage drought tolerance have recently been developed. Besides, tolerance to vegetative-stage drought stress is also required to improve rice productivity in drought-prone regions. We have evaluated a set of rice breeding lines for their response to a range of different types of vegetative-stage drought stress in order to propose standardized phenotyping protocols for conducting vegetative-stage drought stress screening. A soil water potential threshold of 20 kPa during the vegetative stage was identified as the target for effective selection under vegetative stage with grain yield reduction of about 50% compared to irrigated control trials. Genotypes identified as showing high yield under reproductive-stage drought stress were not necessarily the genotypes showing best performance under vegetative-stage drought stress (Swain et al. 2017). Alternatively, tolerance of both vegetative-stage and reproductive-stage drought stress could be accomplished by crossing donor lines, one of which is tolerant of vegetative-stage drought stress and one of which is tolerant of reproductive-stage drought stress. The development of improved varieties with combined tolerance of drought stress at multiple growth stages will help farmers in rain-fed rice-growing regions maintain stable yields across increasingly unpredictable climatic conditions

Maintaining high turgidity (RWC>70%) during severe stress with higher photosynthetic rate and faster recovering efficiency on re-irrigation were found to be the key traits for drought tolerance. Significant negative correlation between drought score vs. RWC (r= -0.78*), drought score vs. Fv/Fm (r= -0.610*) and significant positive correlation between RWC and Fv/Fm (r=0.710*) showed that plants having higher leaf water content were able to harvest most of the photon falling on the canopy and radiated back less amount of energy leading to overall lowered canopy temperature. The genotypes *viz*. AC-43025, AC-43037 and AC-42997, AC-43012 and CR 143-2-2 were identified to have highest water use efficiency coupled with slow transpiration rate above VPD 5 kPa. Low stomatal density and lower canopy temperature (34.02°C - 41.18 °C) in these genotypes resulted in higher biomass production using less water compared to susceptible check IR 64 (ICAR-NRRI Annual Report 2015-16).

Along with higher water use efficiency, lesser number of stomata and slower transpiration rate, improved root traits also play vital role in drought tolerance in rice. We identified twenty superior genotypes *viz*. Kalakeri, Mahulata, Zhu-11-26, CR 143-2-2, Lalnakanda-41, Annapurna, RR 433-2, Sambha-Mahsuri, EC306321, CR 2430-4, RMP-1, RR 366-5, AC- 26774, Sasyashree, Salumpikit, Lektimachi, AC 26773, Khitish, IET 18817, SGS-1 for higher values for more than one root trait. This was reflected in their better ability to tolerate vegetative stage drought stress. The genotypes AC-26685, AC-35679 and EC-205334 were found to have important root markers (MRL, RV, R/S, RW) responsible for drought tolerance. The root: shoot ratio, maximum root length to shoot length ratio and root volume were observed to be the most crucial morphological markers in determining drought tolerance in rice genotypes analyzed through biplot analysis (Dash et al. 2017). AC-42994, AC- 42997, AC-43020, CR-143-2-2, Ronga Bora and Bora were found to possess desirable root traits and these genotypes can be used in the breeding programme for enhancing drought tolerance in rice (Fig. 1).

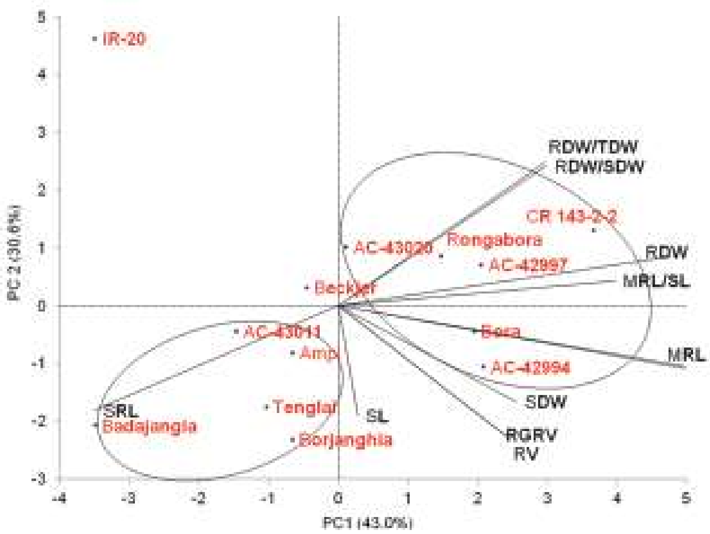

Fig. 1. Factorial plans with two principal components representing root traits averaged over three replications for selected rice genotypes under water limited condition sampled at 20 days stress period (severe stress). Variance explained by each dimension is shown as a percentage of total variance (indicated in axis legend). Coordinates correspond to the correlation coefficients between variables and principal components 1 and 2. *Black colour letters* indicate trait names and *red colour letters* indicate genotype names.

Source: Dash et al. (2017)

Similarly, studies showed significant negative correlation obtained between DSI with grain yield under stress (S) and positive correlation between DSI with yield under irrigated control (I) conditions indicated selection for this character under stress environment might result in decreasing susceptibility to stress. In general, the drought tolerant varieties were characterized by high germination in PEG (20,000) and coleoptile growth in D-mannitol (10 atm), stability to high temperature (40 °C) as apparent from less statolith starch disintegration in root. Besides, low destruction of chlorophyll, tillering in quick succession before onset of drought, fast recovery on re-watering, profuse root growth with high ramification of roots at greater depths (25 cm below), high leaf moisture content (RWC), high stem sugars and protein nitrogen in the plant tissue was also found in drought tolerant rice genotypes. Tillers produced till the end of drought were mostly productive whereas those developed during recovery phase had no influence on yield. The root length was high in ~~DR~~ drought tolerant varieties which was characterized by high peroxidase activity, high soluble sugar and lipid P in root enabling rapid rejuvenation of roots on relief of drought. Drought at panicle emergence stage was found to be more detrimental than at vegetative stage. The critical periods of injury being flowering >PI>tillering>milk stage. Certain varieties were consistently tolerant to drought especially at vegetative stage with very good recovery in terms of plant vigour after drought spell and also give satisfactory yield of ~2 to 2.5 t ha^{-1}. Among the high yielding varieties, Rasi, C 22, CR 113-84-2, CR 143-2-2, TNAU x T 65, Annada (MW 10) were identified to have considerable drought tolerance (Ramakrishnayya and Murty 1993).

4.1.1.1. Identified drought tolerant donors: Over the years, evaluation led to identification of few drought tolerant rice genotypes. Some of the noteworthy ones are AC-254, AC-263, AC-304, AC-511, AC-2298, AC-3035, AC-3111, AC-3577,AC-9066, AC-9387, ARC-7063, AC-45 (CH-45), AC-40083 (MTU-17), W-691, AC-467 (Lalnakanda-41), AC-35207 (Dular), AC-37077 (Dhan gora), AC-37127 (Black gora), AC-37291 (Kalakeri), AC-8205 (Surjamukhi), AC-34440 (Salumpikit), AC- 34256 (Kabiraj Sal), AC- 34296 (Bombay murgi), AC-35021 (Kalon dani) AC-35038 (Godhi akhi), AC-35046 (Nadi tikar), AC-35059, (Phutki bari), AC-35060 (Bhuska), AC- 35143 (Baihunda), AC-35452 (Karama), Mahulata, Brahman-nakhi, Sal-kain, CR 143-2-2, etc.

4.1.1.2. Water saving approaches for rice cultivation: Aerobic rice cultivation using alternate wetting and drying (AWD) method was found to be a potential method of rice cultivation with significant water saving during the crop growth period. Higher yield potential in rice hybrids under AWD and Aerobic conditions (more than 5.5 t ha^{-1}) with least yield loss (1.87 - 6.2%) indicated their adaptability to water shortage conditions. It might have been contributed by high biomass, high LAI (>4.22) higher moisture retention capacity (>70%) during the dry cycle with high chlorophyll content (> 3.0 mg g^{-1} FW) and high photosynthetic rate (30.2-35.6 μ mole CO_2 m^{-2} s^{-1}). Highest grain yield in CRHR

7, PA 6444, PHB 71, PA 6201 and (5.4 - 6.8 t ha^{-1}) under both AWD and normal irrigated conditions with least yield loss (1.87 - 6.2%) under AWD indicated their adaptability to water limited situations. Yield loss under AWD treatment was non-significant and total biomass reduced by 6-8 % without much change in harvest index. Thus aA better adaptable water saving technique was understood by alternate cyclic wetting and drying practice which enabled decreased requirement of irrigation water (about 33%) at the cost of 14 % yield loss in dry seasons and 3.2% yield loss in wet season.

4.1.2. Submergence and Waterlogging Stress

Erratic rainfall leading to excess water usually creates havoc for rice cultivation especially in the rain-fed shallow lowland ecology. For this we need genotypes that are capable to withstand stresses arising due to excess water. In this regard, Elite varieties (traditional and high yielding) for different water depths have been identified. These are AC 2624 (T 1237), MTU 16, Monoharsali, NC 1281 (traditional), IR 5, CR 70-91, TN1 x 65, CRM 10-4622 (HY) for shallow submergence (15-30 cm) and Intan, CN-540 (Suresh), IET 5638, IET 6206, C 4045 for intermediate depths (about 50 cm) (Sarkar 2016). Among the stages, flowering period was more sensitive for flood or submergence injury. However, CRM 10-4622, NC 1281 were relatively tolerant at all stages. It was found that waterlogging reduced chlorophyll and total sugar content in the plant with reduction in NR-A and RUBP Carboxylase activities. Under water-logging, the panicle number per unit area was reduced and the yield reduction is partially compensated by greater grain number per panicle. The adverse effect was more apparent on dry matter at harvest than on HI. Obviously, the panicle weight types were more efficient under water logged situations. Tillers above 40 cm in height at time of submergence survived better and hence varieties with greater number of primary tillers with optimum height were more efficient under submerged conditions eg. CN 540 (Suresh), IET 6205 (Reddy and Mitra 1985).

Significant progress has been made in submergence studies in rice at our institute. The genotype, FR13A (vernacular name 'Dhalaputia', Orissa, India) was identified from this institute as a true submergence tolerant genotype. FR13A is the source of submergence tolerance gene / QTL (*SUB1*) which imparts submergence tolerance. Extensive work was done with this genotype and mechanism of submergence tolerance is now fairly understood (Sarkar and Bhattacharya 2011). In other words, it can be said that NRRI (then CRRI) and Odisha presented the *SUB1* gene to the rest of the world. Genotypes with *SUB1* can withstand complete submergence depending on flood-water characteristics up to 1-2 weeks. One of the key finding for submergence tolerance of rice identified from here is that non-structural carbohydrates content before and after submergence is important for providing energy for maintenance of key metabolic processes during submergence and for regeneration and recovery of seedlings after submergence. Studies have shown that the differences in tolerance level were not necessarily associated with the initial carbohydrate status before submergence but were rather associated with the plant's ability to maintain high levels of stored energy through either slower utilization during submergence and/or greater underwater photosynthesis. The initial carbohydrate level before submergence was almost equal in rice cultivars Gangasiuli and Raghukunwar, but Gangasiuli showed better survival percentage (51%) than Raghukunwar (36%), with retention of higher chlorophyll and non-structural carbohydrate contents during submergence (Das et al. 2009).

Under submergence, varieties with high O_2 liberating power are more efficient and the tall tillers have such efficiency to withstand water-logging. Elongation of stem was associated with high pectin methyl esterase activity and air space in culm. However, in productive non lodging varieties moderate elongation with less activity of enzyme and lower air space was apparent. From stress management perspective, our studies showed foliar spray of IAA + GA, 10 days after planting (100 ppm) helped in leaf sheath elongation, better survival of the plants with 5-fold increase in grain yield at harvest. Close spacing and aged seedlings when transplanted could yield better under water logging. Direct seeded crop was better than transplanted crop because of the advantage in initial height of the tillers at submergence (Sharma 1994). Application of N- and P-fertilizer at seedling stage helped in greater elongation growth of tillers and tolerance to submergence. It was found that P enhanced acid phosphatase activity and submergence tolerance. Minimum elongation during submergence aided in survival due to greater preservation of non-structural carbohydrates (NSC) and application of Gibberellic acid (GA) showed to increase susceptibility, whereas GA biosynthesis inhibitor paclobutrazol increased the tolerance (Das et al. 2005). Seed priming with simple tap water and 2% Jamun (*Syzygium cumini* L.) leaf extract improved the grain yield both under anaerobic seeding and stress-free environment in Swarna and Swarna-Sub1.

The activities of antioxidant enzymes viz. superoxide dismutase, catalase and peroxidase were found protective in ensuring better regeneration/ rejuvenation capacity of rice seedlings during shift from anaerobic (submergence) to aerobic (normal) situations. A higher catalase activity seems to be important to withstand de-submergence injury (Sarkar et al. 2001). Evaluation of starch phosphorylase enzyme activity was indicative of its strong association with submergence tolerance. Submergence tolerant cultivars maintained greater non-structural carbohydrate (NSC) before and especially after submergence. Maintenance of underwater photosynthesis due to greater preservation of chloroplast functional and structural integrity also aided in survival. Cultivars with *Sub1A* QTL maintained greater quantities of non-structural carbohydrate contents especially after submergence and pursued the energy saving mode of carbohydrate utilization. Structural and functional integrity of photosystem II was better in cv. with *Sub1A*. Introgression of *Sub1A* had no adverse effects on yield (Panda et al. 2006). The tolerant tall varieties, Suresh and Utkal Prabha (CR 1030) are characterised by optimum air space in stem, high peroxidase and catalase activity and O_2 liberation by roots, greater chlorophyll especially Chl 'b' and high photosynthetic rate of the top leaves above water level. A novel genotype CRK-2-6, was identified as equally submergence tolerant to that of FR 13A but possesses better yield potential as well as superior grain quality. Dhulia, PD-27 and PD-33 were also identified as submergence tolerant rice genotypes. Besides a few germplasm lines IC-299929 and IC-300131 found to be tolerant to complete submergence for 14 days comparable to FR 13-A and better than Swarna-*Sub1* (Sarkar and Bhattacharya 2011).

Here are some of the most significant achievements of the division with respect to submergence and waterlogging tolerance studies.

- Data on submergence tolerance of more than six thousand germplasms were compiled and a core set and CRRI germplasm data base is prepared.
- Eight cultivars namely, Khoda (INGR 04001), Khadara (INGR 08108), Atiranga (INGR 08109), Kalaputia (INGR 08110), Gangasiuli (INGR 08111), Kusuma (INGR 08113), Bhundi (INGR 14025) and Kalaketi (INGR 14026) have been registered as submergence tolerant donors. Bhundi and Kalaketki tolerate three weeks of submergence.
- A chlorophyll fluorescence based non-destructive technique was developed and standardized to screen submergence tolerant varieties.
- Important germplasm lines tolerant to stagnant flooding are AC 42103, AC 42220, AC 42243 and AC 42254.
- AC39416 is tolerant to anaerobic germination, stagnant water flooding, drought and salinity.
- Kalaputia is tolerant to submergence, stagnant water flooding, and drought.

4.1.3. Anaerobic Germination and Germination Stage Oxygen Deficiency (GSOD) Tolerance

Besides submergence and waterlogging, excess water at the time germination is also detrimental for rice. Flash flood just after sowing imposes submergence stress by creating hypoxic condition (3% Oxygen) during germination as well as during vegetative stage. Interestingly, mode of overcoming hypoxic stress by rice plants seems to be different during germination and vegetative stages. The genes and QTLs reported for vegetative stage submergence tolerance are of no use to tolerate germination stage submergence and *vice-versa*. In general, rice coleoptile under water has been found to elongate about 1 mm h^{-1} to reach the atmosphere by rapid elongation of basal cells (up to 200 m in 12 h) immediately after emerging from embryo. However, anaerobic germination potential (AGP) varies greatly among different rice genotypes, which ultimately provide an edge to a few genotypes to perform better under oxygen deficient conditions over others. Screening over the years resulted in identification a few rice genotypes *viz*. Panikekoa, Dhulia, AC1631, AC 40413, AC 40602, AC 41658 and AC 41620 having superior anaerobic germination potential. Detailed mechanism and possible candidate gene(s) for anaerobic germination was identified in rice recently. Our studies showed effective operation of anaerobic respiration and nitrogen metabolism in tolerant rice genotypes led to more energy efficient metabolic system under oxygen limiting GSOD condition resulted in better ROS handling and cellular pH maintenance. Very recently we have registered AC41620 as a unique rice germplasm having high anaerobic germination potential (AGP) with a robust gene regulatory mechanism. We believe that it would be used as a donor for rice improvement programme to increase the AGP of direct seeded rice varieties (Vijayan et al. 2018).

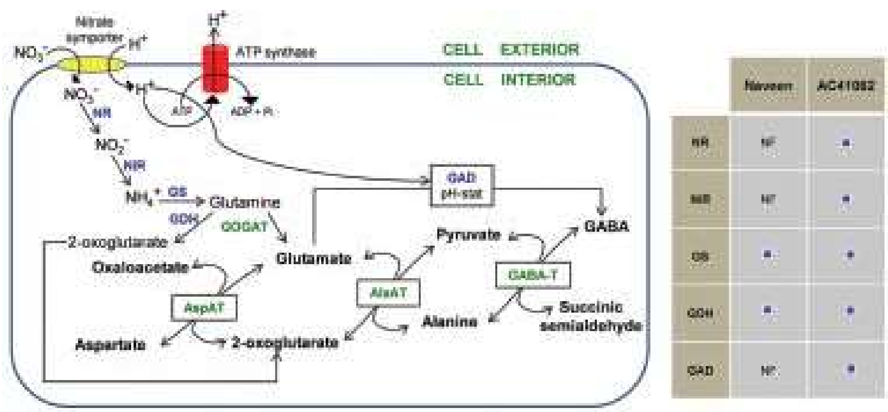

Fig. 2. A summarized network for nitrogen metabolism operative inside the cell also maintains the intracellular pH. In the network, the enzymes encoded by up-regulated genes are mentioned in blue colour and the enzymes encoded by genes which did not show any differential expression are mentioned in green.

Source: Vijayan et al. (2018)

4.1.4. Salinity stress

Work done for past few decades in the area of salt stress physiology indicated that rice is quite sensitive to salinity stress. In fact, it is the most sensitive among the cereals, having a threshold salinity level of only 3 dS m^{-1}. But, rice show considerable variability across different genotypes. Although it had been reported to be relatively tolerant during germination, active tillering and towards maturity, but shows sensitivity during early seedling and reproductive stages, which can reduce rice yield signicantly. It was found that the critical salt limit for 50% reduction in yield was 0.2% NaCl solution for salt susceptible genotypes like Co 13 and T 90 and 0.4% NaCl solution for salt tolerant lines J

349 and SR 26B. The adverse effects of salinity were more apparent in dry than in wet season. Under salt stress reduction in grain number per panicle due to increased spikelet sterility, was found to be the most affected yield component in rice. Varieties showing salt tolerance at seedling stage were not as productive under saline conditions at later stages (Janardhan and Murty 1970).

Under salinity Na, Ca, Mg, ash and Cl percent increases while that of K and P decreases, resulting in lower K/Na ratio. In salt tolerant varieties higher K/Na ratio was maintained. The N concentration increases with salinity, especially soluble N, and P content decreases in salt susceptible varieties. The uptake of P and N is also high in saline tolerant varieties (Kalarata, Damodar, Dasal, Getu, etc.). Forty-five rice landraces collected from coastal saline areas of Orissa and West Bengal were evaluated for salinity tolerance at seedling stage at an EC of 12 dS/m. Kamini, Talmugra, Paloi, Marisal, Rupsal, Ravana and Rahspanjar were found tolerant with per cent survival of 42.2 (Kamini) to 78.3 (Paloi), as against 87.9 for Pokkali (tolerant check). Nagalmutha was moderately tolerant with 22.2% survival. Studies showed that rice plants counteracted the deleterious effects of salinity and maintained greater photosynthetic activities and chloroplast structural and functional integrity by maintaining the appropriate levels of K and ascorbate in metabolically active tissues. Among the 55 salt-tolerant Pokkali collection present in the institute, it was found that accession no. AC39416 was tolerant to both salinity and waterlogging and accession no. AC41585 was tolerant to both vegetative and reproductive salt stress (Singh and Sarkar 2014).

Studies on combined stresses of salinity and partial flooding indicated that maintenance of sufficient levels of tissue K^+ and ascorbate content helps to counteract deleterious effect of salt stress. Our studies showed both photosynthetic rate and non-structural carbohydrate contents decreased due to saline treatment especially in susceptible cultivars. The significant reduction in photosynthetic rate after seven days of salt treatment was noticed. However, in Pokkali (AC No. 39416), photosynthetic rate did not decrease and the differences were non-significant compared to the controlled conditions. The reduction in variable fluorescence (Fv) and maximal photochemical efficiency (Fv/Fm) of light reaction were greater in IR 42 and FR 13A as compared to the tolerant genotypes. Other physiological characteristics changed greatly with imposition of salinity stress. Accumulation of greater quantities of H_2O_2 contents increased lipid peroxidation as MDA content showed highly significant position association with H_2O_2 content. Greater quantities of Na^+ in shoot increased H_2O_2 production whereas greater quantities of K^+ had inverse reaction which resulted lower production of MDA. Highly significant negative association was noticed between net photosynthetic rate (Pn) and H_2O_2 content and Pn and Na:K ratio. However, the association between Pn and K^+ content was positive. The data showed that under saline conditions accumulation of greater quantities of K^+ is beneficial (Sarkar et al. 2013).

One of our very recent study showed reproductive stage salt tolerance in rice is primarily governed by selective Na^+ and K^+ transport from root to upper plant part. Ionic discrimination at flag leaf governed by differential expression of Na^+ and K^+ specific transporters/ ion pumps was found to be associated with reduced panicle degeneration, spikelet sterility and reproductive stage salt-tolerance in rice. It was found that that higher expression of *HKT1;5*, *HAK5*, *HAK1* and *AKT1* in roots, and *NHX1* and *HKT1;1* in leaves governed selective ion transport in rice. It resulted in minimal Na^+ transport to flag leaf and developing panicle maintaining greater K^+/Na^+ ratio for reproductive stage salt tolerance. Increased expression of *AHA1* in leaf and *AHA7* in roots not only met the greater energy demand for selective salt ion exclusion, but also contributed to overall energy balance required for reproductive stage salt tolerance in rice.

The procedure for screening rice genotypes for salt tolerance at seedling stage has been well established and validated through number of experiments. But, unfortunately only a few protocols for screening of salt tolerance at reproductive stage are currently in use, which are based on both in-situ field evaluation and ex-situ evaluation under controlled condition. For this we have developed and standardized a novel screening protocol for precise phenotyping of salt-tolerance at reproductive stage in rice. In the new method, the setup was established with a piezometer placed in a perforated pot for continuous monitoring of soil EC and pH throughout the period of study. Further, fertilizer enriched soil was partially substituted by gravels for better stabilization and maintaining the uniformity of soil EC in pots without hindering the buffering capacity of the soil. The protocol having modified medium (soil: stone, 4:1) at 8 dS m^{-1} salinity level was validated using different rice genotypes having differential salt sensitivity. The method was found significantly efficient for easy maintenance of desired level of soil salinity and identification of genotypes tolerant to salinity at reproductive stage.

Similarly, another high precision screening protocol based on chlorophyll fluorescence imaging system was standardized for efficient phenotyping of rice genotypes for combined stresses of salinity and partial flooding. Among different fluorescence parameters such as maximal fluorescence (F_M), variable fluorescence ($F_V = F_M - F_0$), maximal photochemical efficiency of PS II (F_V/F_M) and quantum yield of non-regulated energy dissipation of PS II ($Y_{(NO)}$) were able to precisely distinguish genotypes based on their sensitivity to stress. Overall, we found suitability of chlorophyll fluorescence imaging technique for precise phenotyping of rice based on their sensitivity to combined effect of salt and partial submergence (Pradhan et al. 2018).

4.1.5. Low Light Stress

Light intensity is one of the most important environmental factors that determine the basic characteristics of rice production and productivity. However, continuous cloudy weather during the rainy season especially if it coincides with the grain-filling stage, induces a significant loss in rice yield and results in poor grain quality (Murty and Sahu, 1987). Our studies showed that low light at flowering was more detrimental. The adverse effect being in the order of reproductive stage > ripening stage > vegetative stage. Under low light, chlorophyll (Chl) content especially Chl 'b' increases with reduction in Chl a/b ratio. Among the varieties tested, Swarnaprabha (early), Vijaya (medium), Mahsuri (late) were found to be consistently better adapted to low light condition. The lowlight adapted varieties are characterised by high net photosynthetic rate (Pn) under low light, increased Chl b content and N uptake, slow senescence, low respiration and better shoot contribution to grain development. Vijaya is relatively more adapted to low light at ripening stage than at reproductive stage and its adaptability is associated with the above characters (Nayak and Murty 1979).

Foliar spray of 2, 4-D or kinetin, delayed senescence, enhanced apparent contribution rate from stem to panicle and reduced spikelet sterility under low light. The adverse effect of low light is more apparent in *Kharif* season than in *Rabi* season. Under low light the decline in photosynthetic rate is partly due to reduction in stomatal frequency and partly due to reduced stomatal conductance to CO_2. The simple characters like specific leaf weight (SLW) or leaf thickness at flowering under normal light condition is significantly associated with biomass or grain yield at harvest under low light suggesting the usefulness of this character as a preliminary selection parameter for low light adapted varieties even under normal conditions. Specific leaf weight (SLW) at flowering under normal light condition is significantly associated with biomass and grain yield at harvest under low light suggesting the usefulness of this character as selection criteria for low light adapted varieties even under normal condition (Nayak and Murty, 1980). Since expansion of leaf blade appears to be characteristic feature in increasing leaf area ratio under low light, least increase in this parameter (within 80 $cm^2 g^{-1}$) found to be an index for identifying cultivars adaptable to low light stress (Swain and Nayak 1996). Stomatal frequency and RUBISCO activity were reduced under low light. However, low light adapted varieties like Vijaya and Swarnaprabha were less affected in these traits. It was found that cultivars adapted to low light also had high Chl 'b' and high Pn under blue light. Light saturation for photosynthesis varied from 50 to 80 klx and the low light adapted varieties invariably showed low light saturation for Pn (30-40 klx). Some of the better adapted low light varieties identified are Ptb 10, Hamsa, T 90, Mahsuri, Pallavi, Swarna-prabha, Vijaya, NC 1281, Vajram and Hybrid, IR 54752A/Vajram. This hybrid has the potential for use in the low light monsoon areas (Murty et al. 1992). Among the three hybrids PHB-71, Rajalaxmi and Ajay tested under low light, Ajay had lowest yield loss over control.

The efficiency of some wild rice species was also tested to now their adaptability potential under low light stress. *O. rufipogon, O. punctata, O. barthii, O. eichingeri* and *O. nivara* were identified to be tolerant to low light stress

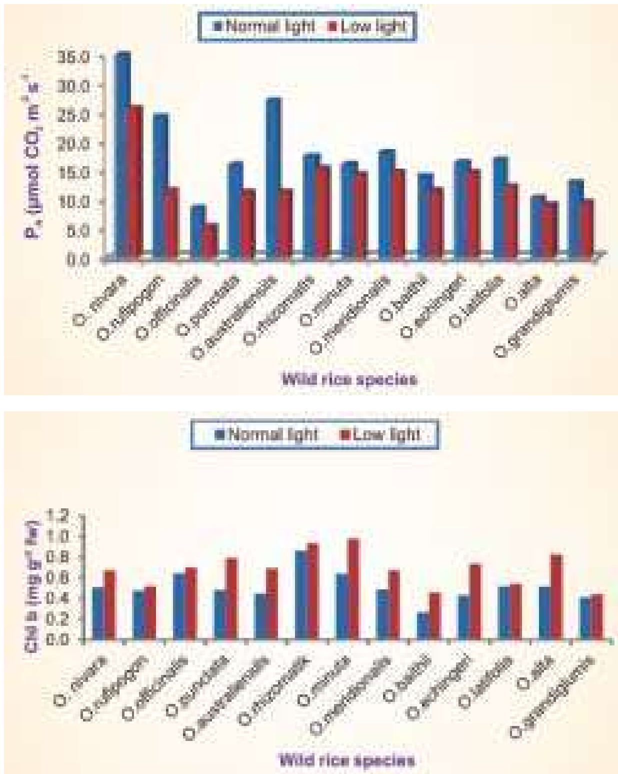

Fig. 3. Photosynthesis and chlorophyll 'b' content of different wild rice species under low light stress condition.

Source: NRRI Annual Report (2014-15)

among different species of rice (NRRI Annual Report 2014-15). Considering accumulation of more chlorophyll *b* and consequently low *Chl a/b* ratio under low light environment as the selection criteria for selecting the varieties for low light tolerance, the species *O. rufipogon (AC-100266), O. punctata (AC-100289), O. barthii (AC-100277) and O. echingeri (AC-100210) and O. nivara (AC-100298)* showed their tolerance to low light environment (Fig 3). Longer duration rice genotypes showed better photosynthesis and more chlorophyll b accumulation under low light environment. Besides, the wild rice species, some identified low light tolerant genotypes from cultivated rice are Satyam, Govinda, Vandana, Naveen, CRHR-32, Sahabhagidhan, Phalguni, Anjali, ADT-36, NC-0087, Udayagiri, Lalitgiri, Suphala, Kalinga-III, ASD-16, Saket, PB-1, Satyakrishna, Tapaswini, Chandrama, Pusa-33, Daya, Srabani, etc.

4.1.6. Heat stress

Work on heat tolerance in rice is at nascent stage in our division. Some preliminary work suggested pollen response can be used as a screening tool for heat tolerance. Among different genotypes tested, IR 8 was identified as most sensitive to temperature with a cumulative temperature response index (CTRI) of 6.8, while Annapurna as most tolerant with a CTRI of 7.5. High temperature stress resulted in reduction in chlorophyll, protein, seedling dry weight, membrane stability, but increase in relative injury (%), sugar content, peroxidase and catalase enzyme activities with significant differences in the changes in these parameters under increased duration of exposure to high temperature stress conditions. Annapurna and N-22 showed minimum yield reduction (19.3 to 21.5%) with low relative injury under high temperature stress with better partitioning of carbohydrate reserve from source to sink tissues. Accumulation of total sugar in the panicles of heat tolerant varieties (N-22) were much more pronounced than the heat susceptible Naveen. In Satabdi and Naveen, the carbohydrate concentration in grains was significantly lowered suggesting the impaired carbohydrate mobilization process existing in these.

4.2. Photosynthetic Enhancement of Rice

Utilization of light energy via photosynthetic processes plays key role and has considerable impact on biomass production and yield in rice. Important components of photosynthetic efficiency are considered to be canopy structure, nitrogen utilization, photosynthetic capacity and CO_2 diffusion rate. Modification of plant types or architecture is thought to be an important strategy to enhance the photosynthetic efficiency and potential yield of crops. Our studies showed photosynthetic rate increases with moderate N rate (90N) due to enhanced nitrogen per unit leaf area (N_{LA}) and decreases with population density because of reduced leaf thickness. Under field conditions, the canopy photosynthesis ~~Pn~~ increased up to 12 noon, remained stable till 2 P.M. (100-120 klx) and declined later indicating 100 klx as threshold light for canopy photosynthesis ~~Pn~~. However, a mid-day depression in individual leaf net photosynthetic rate (Pn) of top leaves was apparent which is associated with reduction in leaf hydration and stomatal conductance to CO_2 and increase in photorespiration and substrate accumulation (Swain et. al, 2006). Canopy photosynthesis (LA x Pn) also varied considerably (1.5-2 fold) and had association with dry matter and yield (r =0.66**). In general, leaf area and Pn, contributed 70% and 30% to grain yield indicating the importance of LAI for yield.

Net assimilation rate (NAR), which indicates the integrated field photosynthesis showed high association with specific leaf weight (SLW) suggesting the usefulness of the latter as a primary selection index for high NAR varieties. However, Ratna showed high Pn with relatively low photorespiration (PR). The tall Indica, Ptb 10 though efficient in Pn recorded high PR and dark respiration. Cultivars identified with low PR are: B 76, Adt 27, Tkm 6, IR 8, Ratna, Pankaj, Vijaya. Leaf thickness (SLWD), leaf vein frequency (LVF), stomatal conductance (Cs), Chl_{LA}, N_{LA} and leaf protein showed high association with Pn while leaf area and yield had a tendency for negative association. LAI increased with duration of the variety with N rate and under low light with consequent reduction in Pn and its associated characters, Cs, stomatal frequency (SF), N_{LA} etc. Nevertheless, cultivars like Swarnaprabha showed high LAI, combined with moderate Pn recording high canopy photosynthesis and grain yield. In general, short types were more efficient in translocation efficiency than talls. Cvs. Bala, Swarnaprabha (early), IET 1145 (medium) and Jagannath (late) are efficient in translocation. Translocation was not affected and even marginally increased under slight reduction in light intensity up to 30% normal light, while at lower light intensities, it was considerably impaired. High leaf N (beyond 2.5%) though enhanced Pn, reduced translocation of photosynthates. Maximum translocation of ^{14}C assimilates to panicle was evident 2 weeks after flowering. Assimilates produced during milk stage contributed 75% of total carbohydrates in grain. Varieties with high panicle Pn (~~eg.~~ Pallavi) exhibited better translocation. Varieties with high Pn are not always that efficient in translocation. Photosynthetic rate per se (Pn) is not that associated with yield while the product of photosynthetic surface (LAI) and Pn invariably showed high association with yield indicating the predominance of leaf area over leaf Pn in determining yield. Swarnaprabha though moderate in Pn recorded high photosynthetic productivity/yield due to larger leaf area, lower maintenance respiration and higher P/R ratio and translocation efficiency. In another study on photosynthesis of rice hybrids Baig et al (1998) reported F1 hybrid like IR62829A x Vajram showed higher readings in PN, PN/RM and PN x LAI at the flowering stage than IR62829A x Swarna. The parents Swarna and Vajram although moderate in Pn had highest Pn x LAI at flowering stage due to greater LAI. There was an increase in seedling stage photosynthesis in the paclobutrazol treated plants over the control as there was an increase in leaf chlorophyll content in the treated plants

Screening large number of cultivars/germplasm lines resulted in identification of a few elite line with high photosynthesis rate.

Table 1. List of elite rice cultivars/germplasm having high photosynthetic efficiency.

Type of variety	Duration	Name of varieties
High Yielding Varieties	Early	C 3383, CR 110-174, Pallavi, Kalinga I and Kalinga II, Saket-4, Co 41, Ratna.
	Medium	IET 734, Jayanti, Sona, Vijaya, Indira
	Late	Jagannath
Traditional Varieties	Early	Ptb 10, Asd 5, Mtu 15
	Medium	AC 4491
	Late	Mahsuri, SR 26B, GEB 24, Mtu 16, Latisail, CR 1014, Sigadis
Mutants		Club, Tainan 3 M, Brittle culm, Saturn striata.
Drought		CR 125-12-30, CR 143-2-2.
Tetraploid Lines		Sita, Indrasail.
Hybrids		IR 54752A/Vajram
Restorers		Anamica, IR 58, IR 9761-19-1
Maintainers		IR 19806-8-1-3-2

4.2.1. Efficient Utilisation of Solar Energy

Besides enhancement of single leaf photosynthesis, efficient solar energy utilization (Eu%) for biological yield (BY) and economic yield (EY) enhancement was studied in the division under PL-480 Project during 1980-91. The elite entries for Eu% (BY) are given below.

- HYV: Kalinga I and II, Saket 4, Vaigai, Suphala, Adt 32, Jagriti, Aswati, Co 41, IR 36, Prabhat, Madhu, Swarnaprabha (early), Savitri, Mahsuri, Madhukar, Jalgaon-5, Deepa (medium and late) (Rao et al 1985).
- Traditional: Ptb 10, Co 29, Ch 1039, N 22, Mtu 15, B 76, BR 34, Tkm 6, AC 3905 (early), T 141, GEB 24, Bamia, Bam 9, T 90, Belamancha, Naukoili (late) (Rao and Murty 1984).

It was found that solar energy utilization is higher in *Kharif* season than in *Rabi*. It increased up to flowering stage and was highest during the reproductive stage (PI to flowering). A tendency for increase in Eu% with duration of variety was apparent because of corresponding increase in leaf area index (LAI). The Eu% (BY) is associated with LAI, dry matter partitioning, amount of light (PAR) intercepted by the canopy and with yield while Eu% (EY) is related with post flowering DMP. The Eu% (BY) could be further enhanced in a cultivar by higher planting density and N rate through increase in LAI.

4.2.2. Development of C4 rice

Rice is a model C3 plant which operates Calvin cycle for fixation of atmospheric CO_2 into carbohydrate. It is well known fact that C4 plants are better in carbon fixation than C3 plants specially under limited CO_2 and nitrogen condition as well as under high temperature and drought. Therefore, C4 cycle operating plants are suitable for the changing climatic scenario to address the global food security issue. Hence, we took an initiative to transform rice into C4 plant with an objective to improve its photosynthetic efficiency and yield. We found that the highest expression level of Carbonic Anhydrase (*CA*), a key enzyme for C4 pathway in *Sorghum bicolour* and *PEPC* in *Setaria italica* plant. The highest expression level of *PPDK* and Malic Enzyme (*ME)* was found in *Seitaria italica* plant. Cloning of *Setaria italica* PPDK, *Setaria italica* ME, *Sorghum bicolour* CA and *Setaria italica* PEPC was done in pCambia binary vector. Transformation of *SiPPDK, SiME, SbCA* and *SiPEPC* and expression constructs to *Agrobacterium* strain *LBA4404* and their subsequent transformation into rice genome was done *E. coli* glycolate metabolic pathway genes in rice, (Glycolate dehydrogenase), *GDH* (3 subunit-*glcD*, *glcE*,& *glcF*), *(*Glyoxylate carboligase glyoxylate) *GCL* and *(*Tartronic semialdehyde reductase) *TSR* gene were PCR amplified from *E. coli* genomic DNA and sequence confirmed. Out of 5 genes, 3 were cloned in plant transformation vector for rice transformation.

4.3. Rice Production Physiology

Our research related to rice production physiology showed that timely top dressing of N (30% of total) just prior to 50 DAS helped in maintaining leaf N above 2%, enhanced N uptake and reduced tiller mortality. Thus application of N in 3 splits (40%, 30%, 30% at 20 and 50 DAS and at booting respectively) increased tillering, reduced tiller mortality and enhanced panicle weight and ultimate grain yield/tiller. It was found that grain filling in rice mostly depends on optimum source to sink ratio and climatic condition during ripening phase. Results indicated that per spikelet the optimum source is 2.5 cm^2 leaf area and 14 mg of stem tissue (fresh weight basis) at flowering stage, especially for early varieties like Ratna or Pallavi. Major constraints for productivity in wet season were low grain number per panicle in early duration type cultivars; high spikelet sterility (%) in medium duration type cultivars and low panicle number per unit area in late duration type cultivars. It was also shown that grain development mainly depends on accumulated reserve dry matter at flowering stage in *Kharif* season. Partitioning of dry matter to panicle is also poor due to high sterility resulting in low HI. Both leaf and panicle senescence is also faster resulting in lower post flowering photosynthate production and its mobilization to panicle. Nevertheless, taller genotype Pallavi with high panicle photosynthesis as compared to semi dwarfs like Ratna, showed high translocation and low sterility in low light monsoon (Debata and Murty 1982).

Net photosynthetic rate (Pn) was found to be low during wet season (WS) partly due to low leaf area, nitrogen (N_{LA}) stomatal conductance and high respiration losses. Yield during WS was obviously associated with LAI and DM at flowering (source size and efficiency) while spikelet or grain number/ m^2 (sink size) determined yield in dry season (DS). Varieties with high dry matter production or crop growth rate (CGR) at reproductive /ripening stages rather than at vegetative stage were more productive as dry matter produced at this stage is directly associated with sink potential (spikelet number and its filling). Besides efficient grain filling, high density grain% (HDG %) was also reported to be a desirable trait for higher production. The HDG (%) was generally higher in late than in early or medium duration types. Some of the genotypes identified for HDG (%) were Rasi, IR 50 (early), Jaya (medium), Swarnadhan or IET 5656 (late). Within a panicle, HDG % was higher in top portion than in middle or bottom portion and in primary than in secondary or tertiary rachis branches (Janardhan and Murty, 1977). Foliar spray of growth regulators found to enhance HDG (%) and the order being kinetin >GA_3> IAA >mixtalol.

Besides photosynthetic efficiency and dry matter production potential, grain filling or spikelet sterility was found to be one of the key for higher productivity. This trait was widely varied (16%-74%) among ~2500 genotypes tested during wet season. Some of the identified rice cultivars with low spikelet sterility were Pallavi, Selection TN1 x T 65, Sakti, Mahsuri (HYV); B 76, Bam 12, Ptb 18 and 21, H 4 (traditional). Spikelet sterility was mostly a post fertilization phenomena and was reported to be higher in wet than in dry season (2-3 fold variation). Low Light during flowering to 10 DAF, especially on the day of anthesis was critical. Variation in duration of different phenological phases was observed under direct seeded and transplanted conditions. The reproductive phase under direct seeded condition was 27 to 29 days whereas it was 22 to 25 days under transplanted condition. Transplanting of 45 days old seedlings exhibited higher dry matter accumulation during vegetative phase and also higher grain yield in comparison to transplanting of 30 days old seedlings. Biomass partitioning study revealed a similar partitioning pattern in leaf but variation in stems. In general, the rate coefficient (¡) in biomass partitioning in leaves was less than in stems. The trend indicated slow yet progressive decrease from panicle initiation (PI) to maturity and becoming almost negligible at maturity (Krishnan et al. 1998).

In general, partitioning from source to sink (panicle growth rate) was faster under direct seeded than in transplanted condition, but larger sink size emanated from transplanted conditions. Contribution pre-flowering stored carbohydrate to grain was more during the milky white stage > 43% that reduced to 35% towards ripening stage. However, pre and post flowering carbohydrate contributions were 39% and 61% respectively. A significant positive relation (r=0.89**) between dry matter accumulated between PI to flowering stage was associated with spikelet number under close spacing (10 x10 cm) with moderate nitrogen level (40-60 kg ha^{-1}) that was highly advantageous to exploit more number of filled grains (high density grain). Leaf area index (LAI) at flowering was linearly related with grain number (r=0.79**) indicating larger and functional source size at flowering eventually resulted in higher yield (sink size). Positive correlation of maintenance respiration with biomass production at early stages and negative correlation at post flowering stages indicated the possibility to identify cultivars having high biomass with less maintenance expenses even at high level of N supply and could be further exploited for breeding elite varieties (Swain et al. 2000).

4.3.1. Physiology of Hybrid Rice

Hybrids exhibited variation in terms of sink capacity and higher sink capacity in terms of panicle no. m^{-2} (CRHR 5), fertile grains $panicle^{-1}$ (CRHR7 and KRH2), low sink capacity but high partitioning rate (KRH-2 and PHB-71), high sink capacity but less partitioning rate (DRRH-1). 1000 grain weight and high translocation efficiency (HI > 0.50) with better grain filling rate (spikelet fertility > 80%) contributed to higher grain yield. Foliar application of GA_3 (30 ppm) in hybrid rice was found more effective in reducing spikelet sterility (20-30%) followed by IAA (17-22%) and Brassinolide (14-21%). Also GA_3 application being expensive, combination with other cheaper chemicals like Brassinolide was found equally effective in seed setting and grain yield, thus reducing the cost of seed production in hybrid rice (Baig et al. 1995, IRRN).

4.3.2. Physiological Efficiency of New Generation Rice (NGR)

The highest erecto-foliage leaf orientation coupled with higher LAI (5.0-6.3), highest leaf photosynthetic rate (35.2 - 49.1µ mole CO_2 $m^{-2}s^{-1}$), maximum photosynthetic quantum yield efficiency of PS II (Fv/Fm ratio of 0.770 - 0.808) with high performance index (2.21 - 3.84), high biomass (10-11 t ha^{-1}), high HI (0.52), high panicle number (340) and higher translocation efficiency with high grain filling percentage (>85 %) are key traits contributing for higher yield potential (more than 7-8 t ha^{-1}) with yield advantage of 0.5 - 1.0 t ha^{-1} over the checks in NGR lines.

Table 2. Elite Rice varieties identified for physiological efficiency.

Useful trait	Growing Condition	Elite Genotypes
High photosynthetic efficiency	Under normal light	Ptb 10, Mtu 15, Mahsuri, Co 41, Ratna, Saket-4, IR 58.
	Under low light	Ptb 10, Swarnaprava, Vijaya, Vajram
Low photorespiration		TKM 6, Triveni, Kanchan, Pusa 33, T(N) 1.
Low maintenance respiration		Rasi, T(N)1, Kanchan, Swarnaprava.
High translocation efficiency		Swarnaprava, IR 50.
High solar energy utilization	Biological Yield	Ptb 10, AC 4491, Bam 9, Swarnaprava, Saket 4.
	Economic Yield	Ptb 10, Pallavi, AC 4491
Nitrogen use efficiency at low inputs		Pallavi, Swarnaprava
Slow leaf and panicle senescence		Pallavi, Vijaya.
Low spikelet sterility		Pallavi, Saket.
High density grain (%)		IR 50, Swarnadhan.
Stresses	Low light	Ptb 10, Swarnaprava, Vajram
	Moisture deficiency	CR 143-2-2, Annada (MW10)
	Salinity	CSR 1,2,3.
	Water-logging	Utkalprava, Gayatri, Suresh, Tulashi
General high physiological efficiency		Ptb 10, (traditional) Swarnaprava (HYV)

4.4. Identification of unique rice germplasm and novel genetic information

Screening and evaluation of large numbers of genotypes for tolerance to different abiotic stresses, photosynthetic efficiency and grain and nutritional qualities led to identification of a few unique germplasm lines, which were registered through Plant Germplasm Registration Committee (PGRC), Indian Council of Agricultural Research, New Delhi. Some of the lines thus registered are,

- Unique rice germplasm Cherayi Pokkali (ICAR-NRRI Gene Bank Accession No. AC39416A) was identified and registered by PGRC (Registration number INGR19004) of ICAR. This rice germplasm is very unique as it possesses multiple abiotic stress tolerance, which is an utmost important character and can be used for developing climate resilient rice cultivars. AC 39416A is tolerant to drought, salinity and stagnant flooding at vegetative stage and moderately tolerant to germination stage oxygen deficiency (anaerobic seeding) and tolerance to combined stress of salinity and drought and water stagnation and salinity.
- Three rice germplasm lines Mahulata (AC No. 35186), Brahman Nakhi (AC- 35678) and Sal-kaiin (AC- 34992) as new sources of vegetative stage drought tolerance and one genotype CR 143-2-2 for both vegetative and reproductive stage drought tolerance having desirable root traits with high water use efficiency (WUE) are registered by PGRC, ICAR, New Delhi and are being utilized in different breeding programme.
- Unique Germplasm of Rice Khora-1 (ICAR-NRRI Gene Bank Accession No. AC41620) was identified and registered by PGRC (Registration number INGR19006) of ICAR. This rice germplasm is having exceptionally high anaerobic germination potential (AGP), a trait most important for rain-fed direct seeded rice. Also, a detailed analysis of underlying mechanism suggests existence of novel mechanism of AGP other than known role of AG QTLs in rice. This genotype is a potential source for developing novel AG (anaerobic germination) QTL(s) useful for direct seeded rice.
- Land races 'Bhundi' (INGR 14005) for Elongation ability and "Kalaketki" (INGR 14202) for Submergence tolerance, one unique germplasm AC41620 for high anaerobic germination ability and another unique germplasm AC39416 for multiple abiotic stress (salinity and stagnant flooding at vegetative stage and moderately tolerant to germination stage oxygen deficiency) were registered by PGRC, New Delhi.

Besides this few new gene sequences were identified and registered in NCBI gene bank, which can be used as basic source of information for transforming rice into a C4 plant. The sequences are

- *Sitaria etalica* Malic enzyme (NCBI Acc. No.MG999525)
- *Sitaria etalica* PPDK (NCBI Acc. No.MF593307)
- *Sitaria etalica* PEPC (NCBI Acc. No.MF967570)
- *Sorghum bicolor* Carbonic Anhydrase (NCBI Acc. No.MF593306)
- *Zea mays* PPDK (NCBI Acc. No.MF593305)

Other than this, we have generated global transcriptome database for multiple abiotic stress tolerance in rice. The gene expression data were submitted to GEO (Gene Expression Omnibus) of NCBI (National Center for

Biotechnology Information, NIH, USA) for its global public access. This database would help to understand the differential mechanism of tolerance in rice under waterlogging and salinity stresses by looking into expression profile of DEGs and uniquely expressed genes.

4.5. Rice Biochemistry

The prime determinant of consumer choice and marketability of a variety is its grain quality aspects. Thus, improving the nutritional quality of produce is imperative in the times of climate change to cater to the diverse dietary requirements of millions of people primarily dependent on rice. The division worked towards estimation and identification of germplasms with good nutritional profile for further breeding and improvement programs.

4.5.1. Grain Nutritional Quality

4.5.1.1. High Protein Rice: The nutritional status of a crop majorly depends on its protein content. Rice protein, when compared to other grains, is considered to be one of the highest quality proteins but although this cereal contributes to the diet of people around the world, milled rice is generally low in grain protein (6-7%). Since enrichment of rice grains with protein would have a positive effect on the nutritional profile, breeding programs for enhancing protein content was urgency.

To this effect, the Division contributed in evaluating around 3000 rice germplasm for grain protein content since 2004 and also found wide diversity for the trait (5-15%). Two low yielding germplasms from Assam rice collection (ARC10075, ARC10063) with high (13-15%) grain protein content in brown rice were identified. Cultivars with high protein content namely ARC 10075 (13%) and Heera (11.5%) were found to richer in nearly all amino acids. Heera was found to have highest amount of threonine, which is known to help maintain the health of the digestive system lining. Heera also had the highest amount of lysine, an essential amino acid. CR 2819-1-3 (12.08% CP), CR 2821-1-8 (11.9% CP), CR 2820-1-8 (11.8% CP) were identified as high protein F3 population along with high protein donor, ARC 10075-6 (11.75% CP). CR2821-1-5 (10% CP), CR2821-1-9 (10.8% CP) and CR2821-1-3 (10.0% CP) showed higher protein yield of 68 g m^{-2}, 46 g m^{-2} and 34 g m^{-2}, respectively than Naveen (30 g m^{-2}), Swarna (31 g m^{-2}) and IR 64 (24.1 g m^{-2}) (Govandaswamy et al. 1973).

The Institute has developed protein rich lines in high yielding backgrounds such as Naveen and Swarna suited for irrigated and favourable rainfed system namely CRDhan 310 (in Naveen Background) and CRDhan 311 (in Swarna background). The Twelve lines with high GPC and protein yield were subjected to protein fractionation. The glutelin fraction was enhanced compared to Swarna, while the Prolamin/Glutelin ratio was maintained. This indicated that the quality of grain protein also enhanced together with the amount of protein in high GPC breeding lines. Subsequently, a rapid method to distinguish between low and high protein rice grains was developed (Bagchi et al. 2016).

4.5.1.2. Glycaemic Index of Rice: Rice is nearly 90% carbohydrate on dry weight basis. Glycemic Index concept, as developed by David Jenkins, Thomas Wolever and colleagues ranks the quality of individual carbohydrate-rich foods on a scale of 1-100 by measuring how blood glucose levels rise after someone eats an amount of food that contains 50 grams of available carbohydrate. Foods are classified as low GI (GI, 55 or less), medium GI (GI, 56-69), and high GI (GI, 70 or more) types, when D-glucose is given a GI of 100. Refined, processed starches/ fruits have a higher GI. Whole grains, high fiber foods, whole fruits vegetables and legumes have lower GI. The GI value of rice varies widely (48-92) with an average value of 64 including the brown and milled rice. Rice contains less than 3% Resistant Starch or RS (mainly of type 5), which escapes digestion almost entirely and therefore its calories are unavailable for cells to use. The more the RS, the slower the digestion of rice and consequently the lower is the GI.

There is evidence to suggest that low GI diets reduce the incidence of diabetes, hyperlipidaemia and cardiovascular disease. GI values of milled rice of popular Indian varieties are higher (70-77) compared to those of brown rice (50-87) as per the 2008 international GI table. In this respect, an *in vitro* method for estimation of glycaemic index was developed and validated by the Division of Biochemistry. 102 varieties/germplasm from different ecologies were screened for glycaemic index (GI) using this in vitro method. Genotype PB177 was found to have the lowest GI (57.91). Large variation in the value of GI (57.50-76.40) and resistant starch (RS) (0.28-2.94%) was observed among 100 NRRI varieties. Among the genotypes studied, Shaktiman showed lowest GI (57.50) with relatively high RS (2.11%) while the highest value for GI (76.40) was found for Hue with lowest RS (0.28%).

4.5.1.3. Antioxidant Value of Rice: Antioxidants, the substances found in foods and dietary supplements help protect cellular constituents like proteins, lipids and DNA against the damage caused by free-radicals including reactive oxygen species (ROS), which are routinely produced during aerobic energy metabolism in our body. Brown rice (BR) or dehusked rice, which is obtained when paddy (rough rice) is subjected to hulling is rich in bioactive components such as dietary fiber, functional lipids, amino acids, vitamins, phytosterols, phenolic compounds, gamma-aminobutyric acid (GABA), minerals and many antioxidant molecules. To satisfy consumers' needs, the rice grain is usually milled into white rice, while the bran and husk are discarded. Most of the antioxidants are confined to the bran layer and endosperm and are absent from the milled rice. Pigmented rice is now gaining popularity because of its documented health benefits. In addition to its high protein, vitamin, and fiber content, it is a good source of a variety of phytochemicals including polyphenols, isoflavones, phytosterols, and anthocyanidins that have several beneficial functions in human health. The nutritional advantages offered by both brown and pigmented rice necessitate their inclusion in the daily diet to a greater extent. Hence, characterization of the colored and other rice for their antioxidant value needs to be a priority.

Research carried out in the division showed that the total anthocyanin content (TAC), total phenolic content (TPC) and antioxidant activity (ABTS) differed significantly among the pigmented genotypes with highest concentration of these parameters in the purple grain (Mamihungar), whereas no significant difference between the colour groups (red and purple) was observed for total flavonoid content (TFC), gamma-oryzanols and phytic acid content indicating that value of these parameters depends on genotypes and not on kernel color. A high correlation of TAC with TPC and ABTS suggests that the major phytochemicals responsible for the tested antioxidant activities are phenolic acids and anthocyanins. Estimation of the total phenolic content was found to be maximum in Lalbora (0.27mg/g GAE) and minimum in Mornodoiga (0.10 mg/g GAE). The free radical scavenging activity (RSA) of colored rice extracts was determined by the DPPH method to assess the antioxidant activity. The RSA was found to be highest in the rice Saathi and was lowest in Mugai.

4.5.1.4. Mineral Bioavailability: Fe and Zn are essential trace elements in human nutrition and their deficiencies are major public health threats worldwide. Unfortunately, rice does not furnish minerals adequately, because it contains only small amounts of Fe and Zn, and the loss of minerals (particularly Fe) during milling is high. In addition, rice contains phytic acid (PA), the most important anti-nutritional factor impeding availability of divalent cations. As an anti-nutrient, high levels of PA can affect the bioavailability of essential minerals such as Zn, Fe and Ca, as it is a strong chelator of divalent cations. The anti-nutritional properties of PA can be further extended to human health as it is considered to be the most important anti-nutritional factor contributing to the iron deficiency suffered by over 2 billion people worldwide. The undesirable properties of PA make the development and characterization of low phytate rice crops a high priority in agricultural research.

Studies carried out in the Division identified the NRRI rice cultivar CR Dhan 907 to be the richest in iron (20 ppm) followed by CR 3704 (14.1 ppm), the latter was also found to be rich in zinc (27.7 ppm). Studies were undertaken to enrich rice grains with iron and zinc, by which Fe content could be increased by 2-10 times and Zn by up to 2.6 times depending on variety. Rice variety Naveen was soaked in 1000 ppm of iron and zincs each and subjected to parboiling. The content of the two elements was determined in brown, milled and well washed milled rice prepared from the parboiled paddy grains. The washed milled rice grains of treated samples had 3-4 times more iron and zinc than the untreated control; for Fe, Control: 4.70 ppm and treated:19.05 ppm (in 1000 ppm Fe) and 19.47 ppm (in 1000 ppm Fe and Zn each); likewise, for Zn, control:14.5 ppm and treated 73.7 ppm (in 1000 ppm Zn) and 49.3 ppm in 1000 ppm Fe and Zn treated samples). The presence of iron and zinc was found to cause reduction in the absorption of the other element. A simple colorimetric protocol was also developed for mass screening of Fe in rice grain. Iron was first extracted from grain samples in dilute acid solution followed by heating on a boiling water bath. An aliquot of this solution was mixed with a colour forming reagent and the developed colour was measured in a spectrophotometer. The results were validated with a standard colorimetric method for iron estimation (R^2=0.739).

A simple colorimetric protocol was also developed for mass screening of phytate in rice grain. Phytate was first extracted from grain powder in dilute acid solution followed by heating on a boiling water bath. An aliquot of this solution was mixed with a colour forming reagent and the resulting colour was measured with a spectrophotometer. The results were validated with a standard colorimetric protocol of phytate estimation (R^2= 0.702). An inverse relationship was also found between PA content and Fe/Zn bioavailability among different varieties. The brown rice of Bindli, which had the lowest PA (0.82%) showed highest Zn bioavailability (12.51 ppm), while PB267, which had the highest amount of PA (2.62%) showed low bioavailability of Zn (8.94 ppm) and Fe (4.04 ppm).

4.5.1.5. Phytic acid content of rice including pigmented rice in brown rice grains: Iron and zinc are essential micronutrients for humans; their deficiency affects metabolism considerably with adverse effect on health. Rice does not provide these micronutrients adequately because the processing decreases their content in rice grains significantly. Not only this, presence of phytate in grain aggravates the problem as the interaction of phytic acid with proteins, vitamins and several minerals (Fe, Zn, Ca) further restricts their bioavailability. Phytic acid content was determined by an assay procedure specific for the measurement of phosphorus from phytic acid, myo-inositol (phosphate) and monophosphate esters by phytase and alkaline phosphatase using the phytic acid/total phosphorus assay kit (Megazyme International Ireland Limited). The 32 rice varieties already screened for Zn, Fe contents and the 22 colored rice varieties available in the institute were analyzed for phytic acid in the brown rice. The highest phytic acid content (2.83 g/100 g) was found in PB267 and lowest in Bindli (0.82 g/100 g) among the non- pigmented rice. In case of colored rice, lowest phytic acid was found in Mornodoiga (0.34 g/100 g), while the highest amount was found in Manipuri Black rice (2.97 g/100 g) followed by Mamihungar.

4.5.2. Grain Physico-Chemical Quality

The concept of rice grain quality varies with the consumer preference and the purpose (end use). But normally physical qualities like (grain size, shape and appearance), milling quality (the capacity to withstand the pressure of milling) and chemical quality which determines cooking characteristics and nutritional quality are the main determinants of rice grain quality. Though, the quality attributes are determined mainly by the genetic constitution, the environmental conditions and cultural practices have profound role in shaping the final product. Grain quality characteristics assume much more importance for rice compared to other food grains and are the prime determinants of market price, because most of the rice (almost 95% of production) is consumed as *cooked whole grain*. The current emphasis on quality rice is because of the increase in per capita income, which led to the increased demand for quality rice. Hence, breeding for improvement of grain quality has become a priority area of rice research.

Forty four promising aromatic, semi dwarf, high yielding breeding lines having medium slender grain developed from the crosses Swarna/Geetanjali (CR2937), CR689-116-2/Kalanamak (CR2938), Tillak Chandan/Kalanamak (CR2936), IR36/Basmati-370 (CR-2939), CRM 2203-4/Dubraj (CR2947) and BPT5204/Kalanamak (CR2941) were evaluated under Advance Yield Trial with Kalanamak and Badshabhog as checks and five cultures gave more than 5.00 t ha^{-1} yeild. The NRRI rice varieties Sarala and Gayatri were found to be the best for making popped rice as each of them exhibited ten times volume expansion.

Some popular rice cultivars from Nagaland and West Bengal were assessed for grain quality traits. It was found that the cultivar Nyakmok-V4 (Nagaland) and Kalabhat (W.B.) had very low amylose (9.59% and 5.32%, respectively). Long slender (LS) grain cv. Banskathi, which is very popular in the eastern parts of India and commands high market price showed an AC of 26.74% (high amylose rice) with GC = 38.2 mm.

Forty-four promising aromatic, semi dwarf, high yielding breeding lines having medium slender grain developed from the crosses Swarna/Geetanjali (CR2937), CR689-116-2/Kalanamak (CR2938), Tillak Chandan/Kalanamak (CR2936), IR36/Basmati-370 (CR-2939), CRM 2203-4/Dubraj (CR2947) and BPT5204/Kalanamak (CR2941) were evaluated under Advance Yield Trial with Kalanamak and Badshabhog as checks and five cultures gave more than 5.00 t ha^{-1}. Three promising aromatic genotypes CR2947-1, CR2738-2, CR2713-35, CR2934-39 and CR2934-35 having desirable quality traits were nominated for evaluation under IVT-ASG as new short grain entries for all India testing. One hundred single plants and 26 bulk populations belonging to F5 -F7 generations have been harvested in wet season. Duration of those lines varied from 120- 150 days. Average single plant yield was 40 g. Mean seed protein content of these genotypes was significantly higher (11.1%) than high yielding parents (9%). The F1 seeds from ARC10075/Swarna and ARC10075/ Naveen contained 14.5% crude protein. Highest seed yield/plant (71 g) was recorded in a line derived from Naveen/ARC10063 cross, but its protein content was low (8.85%). On the other hand, highest seed protein content (15.18%) was recorded in a line derived from ARC10075/Swarna cross with 36g seed yield. High protein yield/plant were recorded in CPL-C-2 (7.49 g), a line derived from IR64/ARC10063 and CPL-H-11 (7.29 g), a line derived from Naveen/ ARC10063 as compared to Swarna (4.27 g). Some genotypes such as CPLH-4 (5 and 12.04%) and CPL-B-3 (6 and 11.8%) were observed to have good alkali spreading value as well as high crude protein content. They could be preferred for their good cooking and nutritional quality. During the third cycle of purification, 153 panicle to row progenies of Kalajeera (three lines each) transplanted in 2011 wet season and no morphological variation was observed in the population. Six uniform Kalajeera pure lines with more than 19% amylose content were identified for seed production purpose.

The rice Aghoni was identified as a soak n eat rice earlier. With repeated cultivation at Cuttack, its soaking time increased from 40 min in 2008 to 90 min in 2010 at this institute. Later, two more *soak-n-eat* rice namely, Nalbora and Asham Biroin were identified in 2011 out of 32 Assam rice germplasm tested. A multi-location trial was initiated during 2012-13 in six states viz., Odisha, West Bengal, Assam, Bihar, Jharkhand and Meghalaya to identify regions most suited for cultivation of soak n eat rice, so that the harvested grains do not show increase in soaking time in subsequent generations. Out of the surplus received from four sites, increase in soaking time was noticed in samples of Aghoni and Nalbora obtained from Pusa (Bihar) and Ranchi (Jharkhand); there was no increase in the samples grown at Cuttack (Odisha) and Gerua (Assam). The grains of Asham Biroin not only showed increase in soaking time after the first harvest, but also retained a hard core and hence have been discarded.

4.5.3. Plant molecular biochemistry related to grain quality

Over the last few decades, the improvement in human nutrition and health in Asia has largely been attributable to stable and affordable rice supply. With almost sufficient production of rice, now the research focus is on better nutritional quality of rice with respect to micronutrient rich rice or rice with low anti-nutritional factors like Phytic acid (PA). Knowledge of the biosynthetic pathways related to micronutrients/anti-nutritional factors will permit genetic engineering of metabolic pathways to enhance the availability of micronutrients or to reduce anti-nutritional factors and hence help in improving the nutritional quality of rice grain. Phytic acid (PA) is the principle storage form of phosphorus in cereal grains including rice. PA acts as a strong chelator of metal cations to form phytate and is considered an anti-nutrient as it reduces the bioavailability of important micronutrients. With an aim to lower the amount of PA to an optimum level, this basic study has been carried out.

The enzyme IPKI plays a key role in the last step of PA biosynthesis. The rice *IPKI* gene (Os04g0661200) is highly expressed in developing rice grain and activation or suppression of this gene may result in alteration of PA biosynthesis and change in total phosphorus content in rice seeds. Expression analysis was done in developing grains of three rice cultivars (Bindli, Heera and PB267) having different levels of PA in grains. Lower level expression of *IPKI* was detected at the initial and final stages of grain development in all the three cultivars suggesting that the process of synthesis and accumulation of PA occurs during the mid-stage of grain filling. There was several fold increase in the expression level of *IPKI* in the middle stage in all the three genotypes.

Among these genotypes, PB267 showed highest expression of *IPKI* at the mid-stage of grain filling, which was positively correlated with their phytate content.

In an another study with an objective to enrich the micronutrient in rice grain the accumulation of ferritin protein and expression of corresponding gene was studied in two rice cultivars that differ in grain Fe content. Both Sharbati and Lalat, accumulated maximum ferritin protein in the flag leaf at 5 ppm of Fe in the growth medium beyond which the concentration declined; while the decline was gradual in Lalat up to 50 ppm, it was abrupt in Sharbati, showing almost complete inhibition at 15 ppm of Fe. Differential response of the cultivars to higher level of Fe might be due to the fact that the low Fe cultivar, Lalat, was perhaps inefficient in absorbing and translocating the element within the plant. Further the study was extended where the candidate genes of Fe homeostasis were studied for their expression in rice genotypes having different grain Fe content. Expression of the genes varied among the cultivars as well as different tissues within a cultivar.

Recently molecular analysis of 227 lines of Machhakanta and 234 lines of Haladichudi was done with 24 highly variable Rice Microsatellite (RM) markers. Out of 24 markers, fifteen and seven markers could detect polymorphism in Machhakanta and Haladichudi populations, respectively.

5. IMPACTS

5.1. Rice physiology

Newer technologies in respect of development of rice cultivars tolerant to submergence, multiple abiotic stress tolerance are arising day by day through sharing of materials and knowledge developed by the division through in-house and out-house project. Technologies developed in relation to seed and seedling qualities are being applied in farmers' field through IRRI-ICAR-NRRI collaborative programme namely Stress Tolerant Rice for Poor Farmers in Africa and South Asia (STRASA) as well as through National Initiative on Climate Resilient Agriculture (NICRA), ICAR, India. Swarna-Sub1 developed by IRRI, however, it was tested and released by this institute in India. Swarna-Sub1 now occupies millions of hectares of land in India.

Rice variety CR Dhan 206 was released in 2014 by SVRC Odisha for aerobic ecosystem, using Brahman-nakhi, a drought tolerant donor. This variety is of 115 days duration having yield potential of 3.95 t ha^{-1} with moderately resistant to leaf blast, brown spot, Sheath rot, stem borer and leaf folder. GEB-24 a traditional variety identified for high photosynthesis was used as a parent in breeding programme for development of high yielding variety Krishna in 1970. Another elite variety T 90 efficient for photosynthesis under low light was used as parent in development of high yielding variety Vijaya and Jayanti in 1970s and CR 1014 in 1988. TKM 6 identified for low photorespiration was used as a parent for development of variety Saket 4. Tinan 3 M- a mutant with high photosynthesis was used for development of Indira in 1980. Late variety Jagannath with high photosynthesis and Pankaj for low photorespiration was used for development of Savitri, Gayatri, Dharitri etc in 1980s which are still popular in many states. MTU 15 used for Annada in 1987, Vijaya for Kshira (1988) and Pooja (1999) still prevailed in seed chain as a best popular variety in Odisha. CR 1014 was used to develop Sarala, Durga, Tulasi, Tara, Panidhan etc. during 1988s. Sigadis, Latisail, CR1014, PTB10, CN 540, etc. were used in many crosses for development of high yielding varieties.

Drought tolerant genotypes C22 and Kalakeri were crossed to develop Vandana (1992 and 2000). Lalnakanda 41 used to develop CR 143-2-2, a unique breeding line having both vegetative and reproductive stage drought tolerance which also got registered by PGRC in 2017. Brahman-nakhi used for development of CR Dhan 206 (2014) which is getting popular in rainfed upland areas. Some of the drought tolerant identified lines viz, Annapurna, Selumpikit, CR 143-2-2, C 22, Lalnakanda 41, Browngora, Blackgora, Rasi, Kalinga III, MTU 17, Dular, two accessions of *O. nivara* (AC 100374, AC 100476) were used in many crosses for development of drought tolerant varieties. Some mutants were also developed from the identified lines for different traits. Mutant of identified lowlight tolerant variety Swarnaprabha was developed as Radhi in 1996, high photosynthesis variety CR 1014 was developed as Padmini in 1988 and drought tolerant variety Ch-45 was developed as Chandan in 2008.

Strategic research for identification of markers/traits associated with submergence, multiple abiotic stresses (salinity, stagnant flooding, anaerobic germination, and drought) could address the need of the day in developing suitable cultivars / technologies for flood prone as well as coastal areas of South and Southeast Asian countries. Several publications were made which were quoted by several workers working on the aspects as evident from citation indices. Cloning and introgression of *SUB1* into mega varieties provided a good protection against short-term submergence. Greater efforts are needed to further enhance tolerance to flash-floods as well as to stagnant long-term flooding, predominant in most flood-prone areas. Cultivars with multiple abiotic stresses tolerant are needed to address climate change effects. The new genetic resources are now the sources of identification of new genes/ technologies and for mechanistic studies.

Characterization of floodwater and impact of each parameter on survival improved our understanding and helped in developing better screening methodology and explaining reasons for variability in survival/mortality of a cultivar at different places. We have standardized screening technique based on the floodwater characteristics. Now it is possible to identify tolerant cultivars even in the laboratory and their performances at different locations could be predicted with greater accuracy. Dry- and wet-seeding methods are becoming more popular not only with rice farmers of rain-fed lowlands but also with the farmers of irrigated ecology as because they require less labour and time than transplanting. However, direct seeding has some inherent problems due to which sometimes farmers are reluctant to adopt this technique. Anaerobic germination tolerance can solve this problem to an great extent. Pioneer work in India on this aspect was done in the Division (Sarkar et al. 1999).

Physiological basis of tolerance to different abiotic stresses have made wider our understanding based on which rice cultivation technology is being developed. Different ecosystems require different types of cultivars. Now it is feasible to identify cultivars suitable for flash-floods/water-logging/anaerobic germination/salinity/multiple abiotic stresses tolerant based on the understandings and identification of physiological markers.

5.2. Rice biochemistry

Research conducted in the division aims at understanding the basic mechanisms underlying physiological and biochemical responses of crop. In addition, evaluation and intensive screening of germplasms for various nutritional parameters are carried out routinely to identify suitable donors for various quality traits for further improvement programs. Likewise, the division has contributed immensely to the varietal development program of the institute. Two varieties were released in 2016 by NRRI, Cuttack with high protein content using donors identified by Biochemistry division. High Protein rice CR DHAN 310 was released by Central Variety Release Committee (CVRC) in 2016. This is the first high protein (10.2%) rice variety in national level and has medium slender grains. This is an introgression line (CR 2829-PLN-37) in Naveen background. The average grain yield in national level was 4.5 t ha^{-1}. Another nutrient rich rice MUKUL (CR Dhan 311) (IET 24772: CR2829-PLN-100) was released by SVRC, Odisha in 2016. It has high protein (10.1%) and moderately high level of Zn (21 ppm) in Naveen background. It is medium early (125 days) with long bold grain. The average yield at national level was 4.1 t ha^{-1} and in Odisha- 5.5 t ha^{-1} (in AICRIP trials) and 4.6 t ha^{-1} (farmers' fields in Odisha).

6. PUBLICATIONS

Since inception our division had published numbers of peer reviewed research articles in journal of International and National repute. More than 400 research articles were in different journals, which promoted the basic concepts, newer knowledge and strategic researches in the field of abiotic stress tolerance in rice, enhancement of photosynthetic efficiency in rice, growth and production physiology, etc. The last ten years' publication trend of the division highlighted almost equal numbers of research papers in journals of NAAS score more than 8 (30), score of 6-8 (33) and score of less than 6 (34).

7. HUMAN RESOURCE DEVELOPMENT

In addition to the research work this division is also engaged for training and guidance to Junior Scientists/Research Scholars for PhD/MSc programs and professional trainings. Since inception a total of 22 research scholars obtained their PhD degree by working under the supervision of different scientists of this division. Also 9 student carried out their dissertation work under the guidance of the scientists of this division.

8. LINKAGES

Since inception Crop Physiology and Biochemistry attracted fairly good external linkages with different National and International research organization/ institute and/or funding bodies. The division received financial help in terms research grants, which strengthen and intensified the research work of the division. The Rockefeller Grant (prior to 1960), Colombo Plan (aid from Japan in 1974), PL-480 Project for Photosynthesis (1980-90), Canadian line of credit (1978), IRRI-Collaborative Program under IRWYN (1982-84), National Fellow Award (1984-88), Emeritus Scientist Award (1989-92) were few notables which helped in developing divisional infrastructure and also established good research linkages outside this institute. Besides, in last few decades this division had good research collaboration with International Rice Research Institute (IRRI), Philippines in terms of active participation in different IRRI-ICAR collaborative projects *viz*. STRASA and others.

9. ASPIRATIONS

From the beginning, the division is engaged in both basic and strategic research in relation to abiotic stress tolerance, photosynthetic efficiency, crop Ideotype concept, enhancement of nutritional and grain quality of rice. Till date, considerable progress had been made in these areas. However, we feel there are still plenty of research opportunities remained to be explored in the above mentioned aspects. In next few years our division would like to focus on some of the emerging and most important areas of research such as.

- Reduction of energy cost for various abiotic stress tolerance in plants which include stresses like salinity, submergence, drought, heat, germination stage oxygen deficiency etc.
- Understanding the plant behaviour and underlying mechanism of simultaneous multiple abiotic stress tolerance.
- Development of C4 rice for better photosynthetic efficiency and yield enhancement.
- Identification and characterization of CO_2 responsive and thermo-tolerant rice genotypes suitable for climate resilient rice cultivation.
- Identification and development of Diabetic Rice with low Glycaemic Index.
- Development of nutrient rice with high iron (Fe), zinc (Zn) and phytate content.

References

Bagchi TB, Sharma S and Chattopadhyay K (2016) Development of NIRS models to predict protein and amylose content of brown rice and proximate compositions of rice bran. Food Chemistry 191:21-27.

Baig MJ, Swain P, Pradhan SB, Jachuck PJ and Murty KS (1995) Photosynthesis and respiration of some F1 hybrid rice. IRRN 20(4):15-16.

Baig MJ, Swain P and Murty KS (1998) The photosynthetic efficiency of some elite rice hybrids and resoters. Photosynthetica 35(2): 241-245.

Dash GK, Barik M, Debata AK, Baig MK and Swain P (2017) Identification of most important rice root morphological markers in response to contrasting moisture regimes under vegetative stage drought. Acta Physiol Plant 39:8.

Das KK, Panda D, Sarkar RK, Reddy JN and Ismail Abdelbagi M (2009) Submergence tolerance in relation to variable floodwater conditions in rice. Environ Exp Bot 66:425-434.

Das KK, Sarkar RK and Ismail Abdelbagi M (2005) Elongation ability and non-structural carbohydrate levels in relation to submergence tolerance in rice. Plant Sci 168:131-136.

Debata A and Murty KS (1982) Panicle senescence in rice. Current Science 51:296-297.

Govandaswamy S, Ghosh AK, Sinha NK, Dey RN, Dash AB (1973) Genetic variability of protein content in rice. Ind J Agric Sci 43:805-809.

ICAR-NRRI Annual Report 2014-15.

ICAR-NRRI Annual Report 2015-16.

Janardhan KV and Murty KS (1970) Effect of sodium Chloride treatment in leaf injury and chloride uptake by young rice seedlings. Ind J Plant Physiol 13:225:232.

Janardhan KV and Murty KS (1977) Association of some leaf characters with photosynthesis in rice. Curr Sci 47:367-369.

Krishnan P, Swain P, Nayak SK (1998) Effect of nitrogen levels on pattern of incremental biomass partitioning in rice (*Oryza sativa*) at different growth stages. Indian J Agric Sci 68(4):44-48.

Murty KS and Sahu G (1987) Impact of low light stress on growth and yield of rice. In Weather and Rice International Rice Research Institute Los Banos Philippines p 93-101.

Nayak SK, Murty KS (1979). Effect of low light intensity on chlorophyll content and RUBP carboxylase activity in rice. Plant Biochem 6:102-106.

Panda D, Rao DN, Sharma SG, Strasser RJ and Sarkar RK (2006) Submergence effects on rice genotypes during seedling stage: Probing of submergence driven changes of photosystem 2 by chlorophyll *a* fluorescence induction O-J-I-P transients. Photosynthetica 44:69-75.

Pradhan B, Chakraborty K, Prusty N, Deepa, Mukherjee AK, Chattopadhyay K and Sarkar RK (2018) Distinction and characterisation of rice genotypes tolerant to combined stresses of salinity and partial submergence, proved by a high-resolution chlorophyll fluorescence imaging system. Functional Plant Biol 46(3):248-261.

Rao Ch N and Murty KS (1984) Solar energy utilization by traditional rice varieties. Indian J Plant Physiol 27:1-7.

Rao ChN, Pattanaik RK and Murty KS (1985) Solar energy utilization in high yielding rice varieties. Oryza 22: 119A-119F.

Reddy MD and Mitra BN (1985) Effect of complete submergence on vegetative growth, grain yield and biochemical changes in rice plants. Plant Soil 87:365-374.

Sarkar RK, Bera SK and De RN (1999) Rice (*Oryza sativa*) cultivars for anaerobic seeding. Indian J of Agric Sci 69:73-76.

Sarkar RK and Bhattacharya B (2011) Rice genotypes with *SUB1* QTL differ in submergence tolerance, elongation ability during submergence and re-generation growth at re-emergence. Rice 5:7.

Sarkar RK, Das S and Ravi I (2001) Changes in certain antioxidative enzymes and parameters as a result of complete submergence and subsequent re-aeration of rice cultivars differing in submergence tolerance. J Agro Crop Sci 187:69-74.

Sharma AR (1994) Effect of seed rate and row spacing on the erformance of early and late rice cultivars in mixed crop systems under intermediate deep water conditions(15-50cm). J Agric Sci (Cambridge) 122:201-205.

Singh DP and Sarkar RK (2014) Distinction and characterisation of salinity tolerant and sensitive rice cultivars as probed by the chlorophyll fluorescence characteristics and growth parameters. Functional Plant Biol 41(7): 727-736.

Swain P, Baig MJ and Nayak SK (2006) Diurnal dynamics of photosynthesis and light response pattern at different growth stages of rice cultivars. Oryza 43 (2): 143-147.

Swain P, Baig MJ and Nayak SK (2000). Maintenance respiration of rice leaves at different growth stages as influnced by nitrogen supply. Biologia Plantarum 43(4):587-590.

Swain P, Raman A, Singh SP and Kumar A (2017) Breeding drought tolerant rice for shallow rainfed ecosystem of eastern India. Field Crops Res 209:168-178.

Vijayan J, Senapati S, Ray S, Chakraborty K, Ali Molla K, Basak N, Pradhan B, Yeasmin L, Chattopadhyay K and Sarkar RK (2018) Transcriptomic and physiological studies identify cues for germination stage oxygen deficiency tolerance in rice. Environ Exper Bot 147:234-248.❄