

A Review on: Photo protective role of non-photosynthetic pigments and leaf pubescence against UV-B radiation in the traditional rice varieties

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Abstract:

Origin of life was never being thought without considering the role of UV radiation. Once the “boon” is slowly becoming “curse” for life, similarly enhanced UV-B radiation have harmful effect on rice productivity. Rice (*Oryza sativa* L.) plants are exposed to many biotic and abiotic factors but the problem of enhanced UV-B radiation created by the anthropogenic activities resulted in Ozone layer depletion. Focusing on this, the main objective of study is to summarize the various response of traditional and wild rice varieties of western part of Odisha against the enhanced UV-B radiation. However no screening of traditional rice varieties for UV-B resistance hitherto. Traditional rice varieties have contains high amount of anthocyanin, flavonoides and pubescent hair which are act as quencher of UV-B radiation. Rice plants are performing well to cope up with such problems but depletion of stratospheric Ozone layer has created the threat of enhanced UV-B radiation which causes the reduction of productivity by reducing the rate of photosynthesis, plant growth and overall yield in most of the rice cultivars. So, the scope of further research should be focused on the selection of tolerant species for improving knowledge, scope of genetic improvement and other protective response of Rice varieties against UV-B radiation and provide insight for the breeders to develop stress resistance rice genotype in the near future.

1) INTRODUCTION:

Rice (*Oryza sativa* L.) is one of the most staple foods throughout the world since the mankind learned farming. It is the second highest produced cereal grain after wheat that serve as the important food source for more than one-third of the population (Islam,2009). Rice is currently grown in over a hundred countries that produced more than 715 million tons of paddy rice annually. Fifteen countries account for 90% of the world’s rice harvest where as China and India alone account for 50% of rice grown (Muthaya et.al, 2012). Rice is a semi-aquatic annual grassplant that includes approximately 22 species of the genus *Oryza*, of which 20 are wild varieties. Two species of rice are important for human consumption: *O. sativa* and *O. glaberrima*. Rice is considered as model monocotyledon for research because it possesses a large scale analysis of expressed sequences tags (ESTs), highly saturated molecular map, genetic stock and resources. Cultivated rice is believed to have evolved from one of the

wild species (*Oryza rufipogon*) through long term domestication (**Chang et.al, 1976**). Two species termed *japonica* and *indica* are speciated in cultivated rice. This suggests that these two subspecies were generated by humans.

1.1) IMPORTANCE OF RICE:

Rice has shaped the culture, diets and economic of thousand of millions of peoples. For more than half of the humanity “Rice is Life”. Considering its importance position, the united nation designated year 2004 as the ‘International year of Rice’. The demand for consumption, production and distribution of rice is growing rapidly in almost all the countries of the world.

1.1.1) Rice as food source:

Rice is an important staple food crop for more than 60% of the world people. In 2008, more than 430 million metric tons of rice was consumed worldwide, according to the USDA. Today rice is an important food source for half of the world’s population because it is healthy, versatile, affordable and easy to prepare. Rice is a nutrient rich food with over 15 vitamins and minerals. It is an excellent source of complex carbohydrates hence; rice is most important food crop for developing world.

1.1.2) Rice in culture:

Rice has been associated with the Indian people from time immemorial. It holds a great spiritual and ritual significance in the society and considered as a potent symbol of auspiciousness, prosperity and fertility. It is used extensively in Hindu rites and rituals. Rice plays a central role in the festival of Nuakhai and Makar sankranti in Odisha. Festivals and rituals associated with rice cultivation are celebrated in most of the part of the India with different local name but with the same motive.

1.1.3) Rice in industries:

Increasing the demand of rice in various purposes, the utilization of the byproduct of rice is used in various useful utilities like in cottage industries; rice straw is used for preparation of hats, mats, ropes, sound absorbing, straw board and litter material. Rice husk or hulls are used for fuel, packing materials, industrial grinding and fertilizer and in manufacture of industrial chemicals. Rice bran oil and rice bran wax are manufactured in oil industries from rice husk (chaff).

1.1.4) Rice as animal feed:

Rice hand harvested at ground level to be used as livestock feed. Rice straw, husk and bran is used as cattle feed. Defatted rice bran (DRB) is rich in protein and mostly used in preparation of biscuits and cattle feed. The byproducts of milling including bran and rice polish are used as livestock feed and are used to feed the domesticated animals like cows, pigs, bulls and buffalos etc.

1.2) UV-B IRRADIANCE IN THE ENVIRONMENT:

UV radiations are measured in two terms i.e. irradiance and fluence rate. Irradiance is the radiation falling on a flat surface per unit area per second whereas fluence rate is the radiation falling on sphere per unit cross section per second (Agrawal et. al., 2017). Ozone is a form of oxygen which plays a vital role in the atmosphere and provides protection of life on the earth from UV-B radiation. However due to rapid anthropogenic activities the protective Ozone layer is depleting day by day and form Ozone hole which allowing maximum irradiance of UV-B radiation.

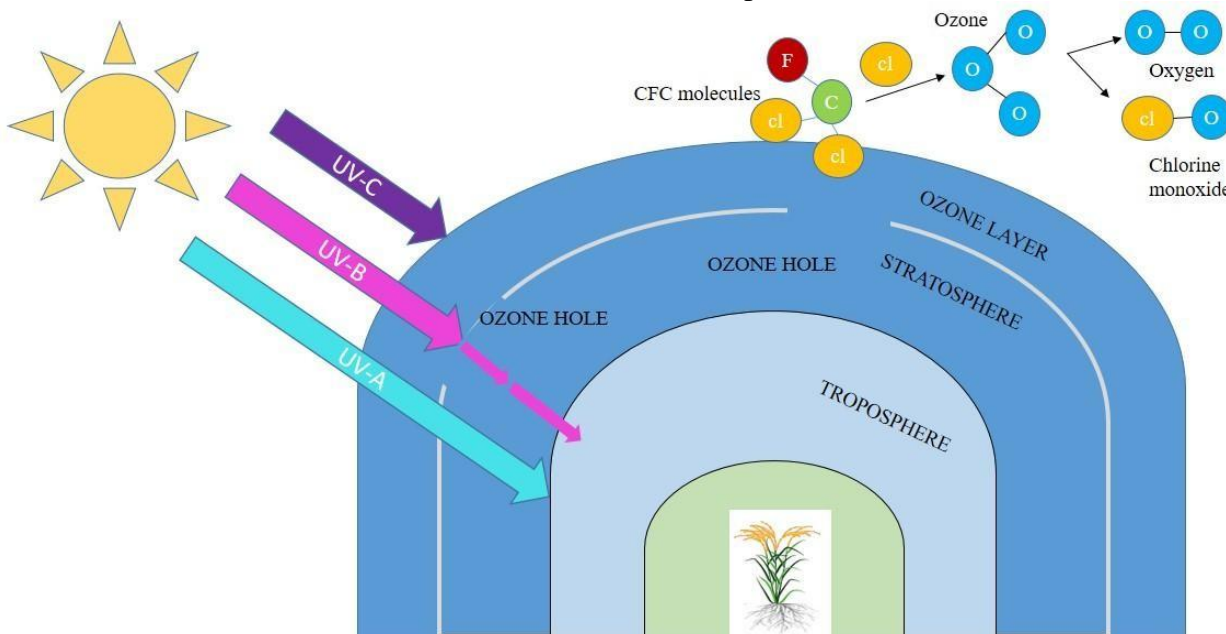
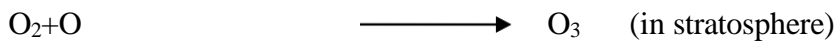
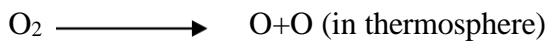


Fig.1 (Ozone layer depletion and penetration of unwanted UV-B radiation)

1.2.1) Ozone layer depletion facilitates UV-B irradiance:

The Ozone layer depletion process begins when CFCs, NO_x and other Ozone depleting substances (ODS) are emitted into the atmosphere (Solomon et al., 1986). In the presence of strong UV-B radiation, ODS molecules

break apart into chlorine atoms and other halogens and these atoms are actually involved in destruction of Ozone layer and allowing maximum amount of UV-B radiation entering into earth surface.



1.3) DELETERIOUS EFFECT OF UV-B RADIATION ON RICE:

Rice is one of the world's most important food crop and grown mostly in tropical and subtropical countries. It is known that UV-B radiation is highest in tropical regions where rice is grown because the stratospheric Ozone layer is high latitudes and solar angles are higher. Increased UV-B radiation induces a significant reduction in the total biomass in a number of rice cultivars, accompanied by a reduction in tiller number and Photosynthetic capacity of plants. The

prolonged exposure of UV-B light affects plant height, leaf area, dry weight, net assimilation rate and relative growth rate in some rice cultivars (**Ziska and Teramura, 1992**).

The stresses imposed by UV-light irradiation can cause reactive oxygen species (ROS) generation such as O_2^- and H_2O_2 (**Shibata et al., 1991**). H_2O_2 cause extremely deleterious effect to cellular constituents such as DNA damage.

1.4) EFFECTS OF UV-B ON MORPHOLOGY, PHYSIOLOGY, BIOCHEMICAL AND MOLECULAR ASPECTS IN RICE:

Increase of UV-B radiation will alter the growth and metabolisms of rice plants thus UV-B radiation acts as an environmental stress factor on rice plants.

1.4.1) Morphological changes induced by UV-B:

Morphology of rice plants are considered to be very effective indicator of UV-B damage. Solar radiation comprised of various radiations among which the percentage of UV-B is very less but the range of its morphological effects on rice plants is very diverse. Increased level of UV-B radiation also induces some common morphological changes such as reduction in leaf area, thickening of leaves, reduced tiller number and decreased in the number of fruits, flower and seedling (**Kakani et. al., 2003**).

1.4.2) Physiological changes induced by UV-B:

Prolongated exposure of UV-B radiation damaged to ultra structure of chloroplast and light harvesting complex

(LHC). It also reduced Rubisco activity, decline in oxygen evolving, CO₂ fixation, reduced chlorophyll and starch contents. The main target of UV-B is PS-II (Fiscus and Booker, 1995). PS-II comprised of two proteins; D1 and D₂ which are very sensitive to UV-B radiation. Some indirect effects of UV-B are altering the rate of Photosynthesis such as stomata closure, changes in leaf thickness and anatomy, decrease in total canopy and individual leaf area.

1.4.3) Biochemical and Molecular changes induced by UV-B:

The stress imposed by UV-B light irradiation can cause reactive oxygen species (ROS) generation such as O₂⁻ and H₂O₂ which affect to cellular constituents such as DNA damage. It's also activated the membrane localized NADPH oxidase which than leads to the generation of ROS (Rao et.al, 1996). Enough doses of UV-B have the potential to affect proteins, lipids and nucleic acids by knocking electrons out of their orbital with breakage of chemical bonds which results the formation of non-functional proteins, leaky membranes, mutated DNA and ribotoxicity (i.e. non-functional translation due to malfunctioning ribosome).

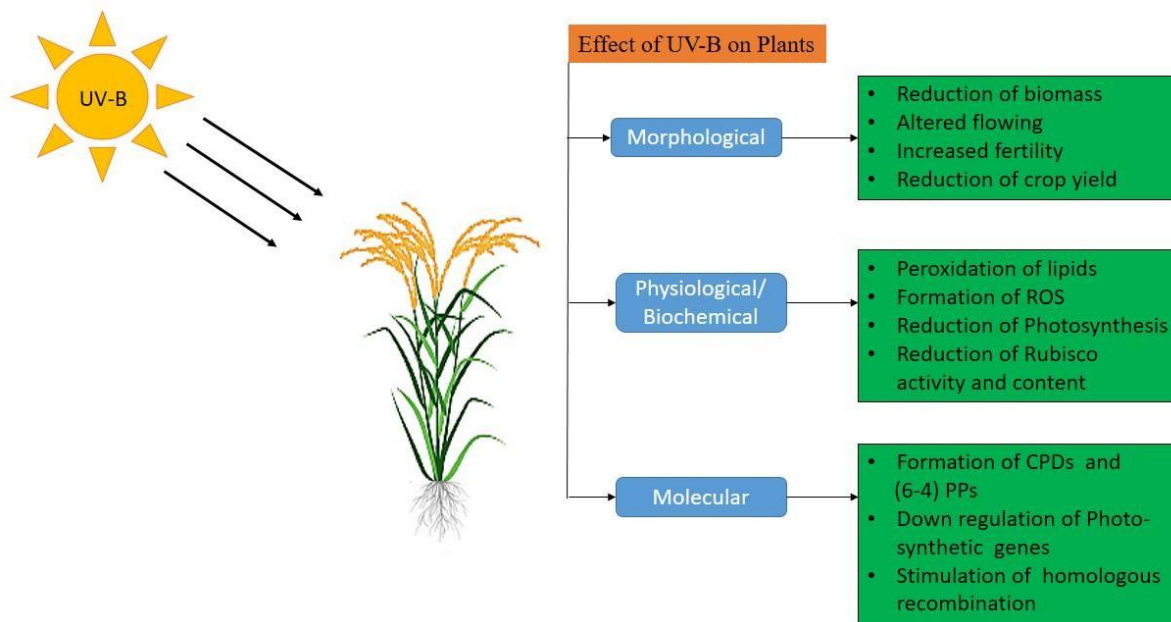


Fig.2 (Harmful effect of UV-B radiation on morphological, physiological and molecular aspects)

1.5) GENE REGULATION UNDER UV-B RADIATION IN RICE:

Over expression of rice gene WRKY89 enhances UV-B tolerance and disease resistance in Rice plants (Wang et

al., 2007). OSWRKY89 gene led to growth retardation at the early stage and reduction of internodal length. Scanning electron microscopy revealed an increase in wax deposition on leaf surfaces of OSWRKY89 over expression lines. W285F mutant gene in rice plant provides proper strategies against UV radiation (Kimberley et al., 2013) and found that UVR8 wild type (WT) and W285F mutant type absorb light at maximum 282 nm with a profile

that is similar to tryptophan absorption whereas WT dimer was fluorescence peak at 332nm and W285F dimer was at 355nm which is longer than the emission peak of tryptophan in water at 350nm. Over expression of a small heat shock protein, sHSP 17.7 confers both heat tolerance and UV-B resistance to rice plant (Murakami et al., 2012). Traditional rice has evolved certain mechanisms to counteract the toxicity of Ultraviolet by synthesizing the UV-absorbing/screening compounds such as Mycosporine like amino acids (MAAS) that repairs UV-induced damage of DNA and accumulating carotenoids and antioxidant (Singh et al., 2008).

UV-B radiation stress causes alterations in whole cell protein profile and expression of Certain genes in the rice phyllospheric bacterium *Enterobacter cloacae* and genes of hypothetical proteins (CP011650 and CP002886) showed over expression under UV-B stress and provide UV-B resistant mechanisms (Kumar et al., 2016). UV-B resistance were detected by QTL analysis, using backcross inbred line (BILs) derived from a cross between japonica cultivar 'Nipponbare' and an indica cultivar, 'kasalath'. Among them found qUVR-10, a QTL for UV-B resistance on chromosome 10 (Sato et al., (2004).

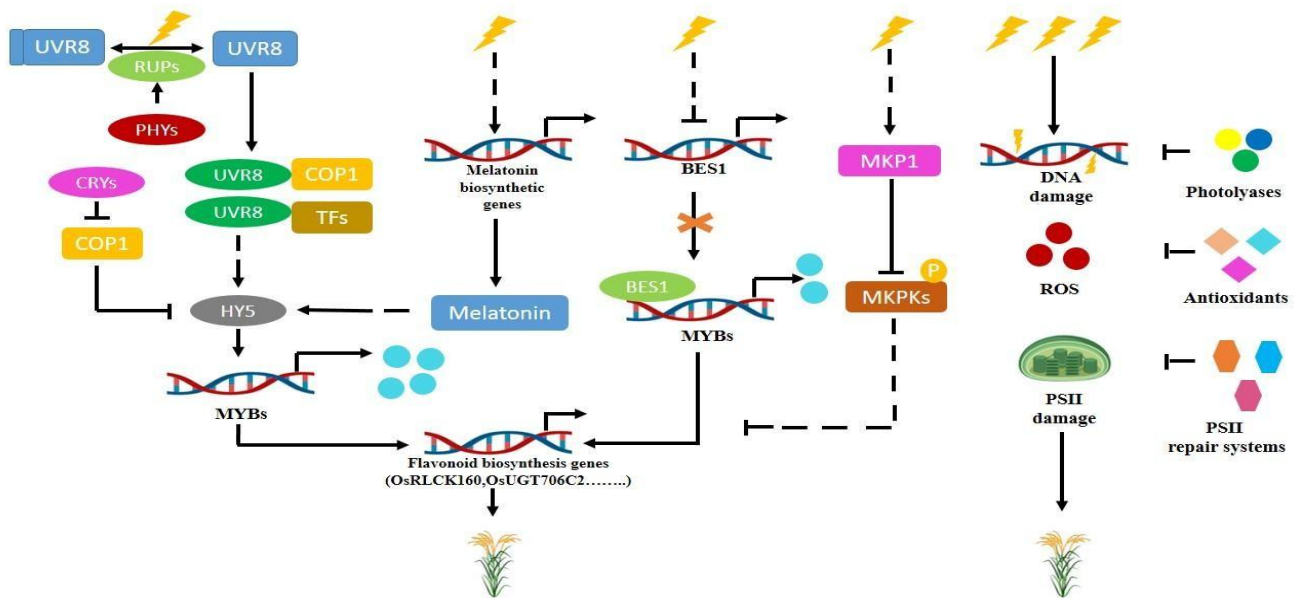


Fig.3 (molecular approach of rice cultivars for UV-B signaling and flavonoid biosynthesis)

flavonoides indicated possible role in UV-B resistance and act as Quencher and effective scavenger of ROS. The presence of pubescent hair in the leaf surface is act as a reflectance of UV radiation. **Teranishi et al., (2004)** examined the UV-B resistance mechanisms between the 17 japonica rice cultivars. Out of these rice cultivars they found Sasanishiki rice is highly resistance to UV-B radiation. These rice cultivars containing higher CPD photorepair ability as compared to Norin 1 and Surjamkhi which are sensitive varieties. He arranges these three rice cultivars according to UV-B resistance, Sasanishiki> Norin1> Surjamkhi. **Reddy et al., (1994)** classified the seedling of 17 rice (*Oryza sativa L.*) cultivars on the basis of anthocyanin pigment into three groups: an acyanic group with 9 cultivars, a moderately cyanic group with 5 cultivars and a cyanic group with 3 cultivars. Seedlings of the cyanic group were deep purple in color, possessing copious amounts of anthocyanin in shoots which result cyanic group of purple puttu seedling is mediated exclusively UV-B resistance.

Special type of pubescent hair present in both adaxial and abaxial surface of BRRI DHAN29 are act as reflectance of UV-B light and don't allow UV-B radiation enter into cell for damage (**Islam et al., 2009**). Japanese lowland and upland rice cultivars resistance to UV-B radiation due to accumulation of high phenolic content (**Sato et al., 1994**). Three rice cultivars "Koshikari", 'IR 45' and 'IR 74' showed no UV-B effects on plant height, tiller number and grain yield due to natural abundance of ^{13}C in the flag leaves and high accumulation of UV-B absorbing compound in leaf epidermis (**Kim et al., 1996**).

1.7) MECHANISMS OF UV-B RESISTANCE IN TRADITIONAL RICE CULTIVARS:

UV absorbing compounds are accumulating in the epidermal cell layers which have traditionally being considered as UV filters and to play an important role in countering the damaging effects of UV-B radiation. Traditional rice varieties have developed certain morphological, physiological, biochemical and molecular modification for UV-B resistance. In morphological aspects they reduced leaf area, thickening of leaves, curling or cupping of leaves and develop the pubescent hair in the epidermal surface of leaves (**Kakani, 2003**). In biochemical aspects they increased soluble phenolic pigments such as flavonoids and anthocyanins which strongly absorb UV-B radiation. Plants comprised of several strategies to acclimatized and metabolized ROS. These includes active defense systems using low molecular weight antioxidants such as ascorbic acid, phenols, flavonoids, glutathione, carotenoids etc. and high molecular weight enzymes such as superoxide dismutase (SOD), ascorbate peroxidase (APX), peroxidase (POD), catalase (CAT) etc. (**Miller, 2002**).



Fig.4 (High yielding, Traditional and wild rice cultivars of Western Odisha)

2) REVIEW OF LITERATURE:

2.1) INTERNATIONAL STATUS:

Mohammed et al., (2013) investigated that elevated UV-B radiation deleteriously affects rice yields. It decreases photosynthetic growth rate (17%), quantum yield (8%), electron transport rate (9%), total chlorophyll concentration (8%), plant height (12%), and number of leaves (17%), pollen viability (6%), phenolic concentration (46%) and yield (21%). The applications of alpha- tocopherol, glycine-betaine (GB) and salicylic acid (SA) increased the yield by 23%, 18% and 29% respectively and also increased leaf phenolic content thus rendering protection against elevated UV-B. **Kumagai et al., (2006)** investigated the effects of supplementary UV-B radiation on the growth and yield of Japanese rice cultivars in paddy field. They found that supplementary UV-B radiation inhibited growth, yield and grain filling in rice and also influence protein, starch, sugar status and taste of the grain. **Teranishi et al., (2004)** examined the core relation between CPD photorepair in 17 japonica rice cultivars that were progenitors of the UV resistant (Sasanishiki) and UV sensitive (Norin1) strain. Cultivars are dividing into two groups, one is UV resistant with high CPD photorepair ability similar to Sasanishiki and other is UV sensitive strain with lower CPD photorepair ability similar to Norin1. The higher activity of CPD photo repair is

due to single amino acid change. The relative rate of CPD photo repair among three varieties are Sasanishiki>Norin1>Surjamkhi.

Huyskens et.al, (2007) had study on phenol content and phenolic composition (Flavonols, anthocyanins, hydroxycinnamic and hydroxyl benzoic acids) increased to a large extent during UV-B treatment. Anthocyanin

absorbed the UV radiation while flavonols and phenolic acids are assumed to have an impact on antioxidant protection of UV-B mediated tissue damage. **Kimberley et al., (2013)** stated that UVR8 is a recently discovered UV-B photo receptor with a homodimer form and they study about the absorbance and fluorescence spectra of the UVR8 wild type (WT) and W285F mutant type. Both the dimers absorb light at maximum 282 nm with a profile that is similar to tryptophan absorption whereas WT dimer was fluorescence peak at 332nm and W285F dimer was at 355nm which is longer than the emission peak of tryptophan in water at 350nm. **Murakami et al., (2004)** stated that over expression of a small heat shock protein, sHSP 17.7 confers both heat tolerance and UV-B resistance to rice plant. Exposure of rice (*Oryza sativa* L.) seedlings to a high temperature (42°C) resulted in a significant increase in resistance to UV-B damage. UV-B resistance was enhanced in parallel with the period of heat treatment. **Islam et al., (2009)** investigated the presence of special type of pubescent hair in both adaxial and abaxial surface of traditional rice cultivars which are act as reflectance of UV-B light and don't allow radiation enter into cell for damage.

Barabas et al., (1998) investigated the effect of excess UV-B irradiation on the antioxidant defense mechanisms in Rice seedlings. It was found that catalase (CAT), peroxidase (POD) and superoxide dismutase (SOD) activities decreased while level of glutathione disulfide (GSSG) and glutathione reductase (GR) activity increased under excess UV-B irradiation. **Hidema et al., (2006)** describes about the mechanism of UV-B resistance in higher plants, with emphasis on rice (*Oryza sativa*). UV absorbing compounds accumulating in the epidermal cell layers have traditionally been considered to function as UV filters and counting the damaging effects of UV-B radiations. **Sato et al., (1994)** studied on the rice cultivars by crossing between UV-resistant (Sasanishiki) and UV-sensitive (Norin1) strains. It showed that UV-B sensitivity is controlled by more than two major recessive genes. **Sato et al., (2004)** stated that UV-B resistance were detected by QTL analysis, using backcross inbred line (BILs) derived from a cross between japonica cultivar Nipponbare' and an indica cultivar, 'Kasalath'. Among them found qUVR-10,

a QTL for UV-B resistance on chromosome 10. **Yano et al., (2000)** quantitative trait locus (QTL) analysis is a powerful tool for analyzing genes affecting traits under multiple gene control and has been employed in many studies investigating complex trait inheritance. **Sato et al., (2003)** detected 3 QTLs associated with UV-B resistance in rice, on chromosomes 1,3 and 10, using backcross inbred lines (BILs) derived from a cross of a UV-sensitive indica cultivar (Kasalath) and a UV-resistant japonica cultivar (Nipponbare). Among the QTLs, qUVR-10 (the QTL for UV-B resistance on chromosome 10) showed the target allelic difference (the percentage of variance explained was approximately 40%).

Bornman and Teramura (1991) together they experimentally found that the accumulation of certain

phenylpropanoid compounds (such as flavonoids and anthocyanin), in the vacuoles of the epidermal and sub epidermal cell layers, plays a role in mitigating UV-B induced damage. **Kang et al., (1998)** showed that UV-absorbing compounds in rice leaves were effective in reducing susceptibility to CPD induction by challenge UV-B exposure, but had no effect on steady state CPD levels during growth under chronic exposure to supplementary UV-B radiation. **Maekawa et al., (2001)** produced near isogenic lines (NILs) for three rice purple leaf genes, PI, PI^w and PIⁱ

, with a genetic background of Taichung 65 (T-65), a normal rice variety from Taiwan. They found that total biomass of all the irradiated NILs decreased with the increasing dose of UV-B irradiation and the decrease was negatively correlated with the increase in anthocyanin accumulation in the dose range of UV-B irradiation. **Hada et al., (2003)** examined using two strains (T-65 and T-65 PI) for different perspective such as the relationship between changes in steady state CPD levels and accumulation of flavonoids (anthocyanins and other UV-absorbing compounds) in leaves relative to leaf age. And the ability to photorepair CPDs and the susceptibility to CPD induction by UV-B radiation. And found that flavonoids functioned by effectively reducing susceptibility to CPD levels during growth under chronic exposure to elevated levels of UV-B radiation.

Kim et al., (1996) experimented on three rice cultivars “Koshikari”, ‘IR 45’ and ‘IR 74’ were irradiated with enhanced UV-B. These results showed no UV-B effects on plant UV-B effects on plant height, number of tillers and panicles, dry weight of the plant parts or the grain yield for any of the three cultivars. **Haung et al., (1993)** experimental on detached leaves of two rice cultivars (Er Bai Aai and Lemont) to evaluate whether the short term supplementary UV-B might

be an initial method for screening for UV-B susceptibility. As a result they found that rapid short term responses of detached leave allow early screening of relative sensitivity of rice cultivars to UV-B. **Dai et al., (1997)** UV-B radiation also produces ROS such as superoxide, H₂O₂ and singlet oxygen. Enhancement of the ROS scavenging system may produce UV-B resistance in rice. **Chow et al., (1993)** studied the effects of supplementary UV-B radiation on rice and pea plants. The multiplicity of photosynthetic responses and of different protective strategies that may account for the differential sensitivity of plants to supplementary UV-B radiations.

Wang et al., (2007) state that over expression of rice gene WRKY89 enhances UV-B tolerance and disease resistance in Rice plants. Scanning electron microscopy revealed an increase in wax deposition on leaf surfaces of OSWRKY89 over expression line. **Wang et al., (2013)** investigated that UVR8 wild type (WT) and W285F mutant type absorb light at maximum 282 nm with a profile that is similar to tryptophan absorption where as WT dimer was fluorescence peak at 332nm and W285F dimer was at 355nm which is longer than the emission peak of tryptophan in water at 350nm. **Chang et al., (1976)** state that Wild rice species *O. nivara* and *O. rufipogon* have

contains high amount of anthocyanin, flavonoides and pubescent hair in epidermal surface which act as quencher against UV-B radiation.

2.2) NATIONAL STATUS:

Sharma (2001) investigated cyanic rice cultivar, Purple putt u which has high amount of anthocyanin pigmentation and is more resistant to UV-B induced damage and this cultivar is associated with PAL biosynthesis. **Singh et al., (2008)** stated that traditional rice have allowed certain mechanisms to counteract the toxicity of Ultraviolet by synthesizing the UV- absorbing/screening compounds such as Mycosporine like amino acids (MAAS) that repairs UV- induced damage of DNA and accumulation of carotenoids, radical quenchers and antioxidant. **Sharma (2011)** stated that moderate and high light grown rice plants showed relatively less photo inhibition and also showed higher non-photochemical quenching which indicated better resistance to light stress mostly UV light stress and provides the relationship between xanthophylls cycle and non-photochemical quenching in rice plants in response to UV light stress. **Nayudu et al., (1997)** examined the relative levels of phenolic compounds produced in extract of rice after UV-B exposure to the plants. Tolerant cultivars accumulated relatively higher levels of phenolics compounds which provide protection against the UV radiation.

Reddy et al., (1994) classified the seedling of 17 rice (*Oryza sativa L.*) cultivars on the basis of anthocyanin pigment into three groups: an acyanic group with 9 cultivars, a moderately cyanic group with 5 cultivars and a cyanic group with 3 cultivars. Seedlings of the cyanic group were deep purple in color, possessing copious amounts of anthocyanin in shoots. Sunlight (SL) mediated anthocyanin and Phenyl-alanine ammonia lyase (PAL) induction in a cyanic cultivar, purple putt u was compared with an acyanic cultivar, black putt u. The anthocyanin induction in purple putt u seedling is mediated exclusively UV-B resistance.

Kumar et al., (2016) show that UV-B radiation stress causes alterations in whole cell protein profile and expression of Certain genes in the rice phyllospheric bacterium *Enterobacter cloacae* and genes of hypothetical proteins(CP011650 and CP002886) showed over expression under UV-B stress and provide UV-B resistant mechanisms. **Ambasht et al., (1998)** in a field experiment with rice (*Oryza sativa L. cv. Saket 4*) grown ambient and supplemental UV-B radiation. They found the decline in photosynthesis was associated with prorogated exposure of UV-B radiation.

4) CONCLUSION AND VIEW:

Increase of UV-B radiation will alter the growth and metabolism of rice plants, thus UV-B radiation act as an environmental stress factor. But certain traditional rice varieties have developed various morphological,

physiological, biochemical and molecular modification for UV-B resistant. For their protection they reduced leaf area, thickening of leaves, curling or cupping of leaves and developed pubescent hair in the epidermal surface of leaves which are considered as UV-filters. These traditional rice varieties have accumulated higher amount of anthocyanin, flavonoides and other phenolic compounds which are act as UV-absorbing /screening compounds.

Increased anthropogenic activities allow gradual depletion of Ozone layer and providing a suitable platform for irradiation of maximum amount of UV-B radiation into the earth surface. In near future two problems will arise i.e., the rapid population growth and the less productivity of rice as compared to it because of more irradiation of UV-B light. To enhance the yield of rice in UV radiated environment there should be a need of develop UV-B resistant genotype which easily grow in UV stress condition and provide maximum productivity.

3) FUTURE PROSPECTS:

Increased anthropogenic activities allow gradual depletion of Ozone layer and providing a suitable platform for irradiation of maximum amount of UV-B radiation into the earth surface. Increased UV-B radiation affects rice crop growth adversely because of significant decline in leaf photosynthesis due to damage of photosynthetic apparatus and poor biomass accumulation attributable to reduction in stomatal conductance. Hence the study on morphological, physiological, biochemical and also molecular basis of UV-B resistance in rice will be of high significance and will definitely provide insight for breeders to develop stress resistant rice genotypes in the near future. Future prospects are to find out gene of traditional rice cultivars that provide resistant mechanism and interpret these genes into sensitive varieties by the application of genetic engineering to develop more UV resistance rice cultivar.

AUTHOR CONTRIBUTIONS

PKS concept, manuscript writing, review and correspondence.

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CONFLICT OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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