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Physiological and biotechnological approach to improve water use efficiency in rice (*Oryza sativa* L.): Review

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Abstract

Rice is the widely cultivated cereal crop in the world. But low water use efficiency of rice (*Oryza sativa* L.) led rice cultivation more water expensive. Due to the climate change, drought and high temperature are the major environmental stress experienced by the plants. In this situation to sustain the yield of rice under limited water consumption is the major concern. Therefore physiological, biotechnological and agronomical approaches are adopted to improve water use efficiency (WUE) of rice. This review focused more about physiological and biotechnological approach to improve water use efficiency of rice.

Keywords: Rice (*Oryza sativa* L.), drought, water use efficiency, biotechnology

Introduction

World population mainly depend on rice as their staple food source. Even though rice is widely adapted cereal crop, its production and productivity is affected by extreme climatic condition. Drought, flood, and land degradation are some of the impact of climate change. Increase of world population added the scarcity of water and land resources. Reports revealed that prediction had been made about the rise in global temperature from 2.4-4.3 °C, this will be leading to the water scarcity and drought (IPCC 2007) [17]. According to IPCC fifth assessment report warming was recorded 1.5 °C due to human activity as compared to preindustrial revolution. The increment was around 0.2 °C per decade (Allen *et al.*, 2018) [2]. In some study showed that 50% of the rice yield is reduced due to drought. Therefore improvement of water use efficiency is the one of the strategy that can be used to tackle this situation.

In plants 90% of the water lose through transpiration (Morrison *et al.*, 2008) [23] so that reducing transpiration without compromising CO₂ uptake is the major concern. Efficiency of water improved through agricultural practices and genetic improvement are widely studied. Landraces harbored many genes responsible for stress tolerance so that it can be used as candidate gene identification especially for improving water use efficiency. Genetic manipulation mainly focused on genes responsible for stomatal density, ABA signaling and root character through breeding and biotechnological tools like genetic transformation.

Water use efficiency

Water use efficiency is the ratio of biomass or grain yield to the water supplied or evapotranspiration which expressed as Kg m⁻³ water (Sinclair *et al.*, 1984) [27].

$$WUE = \frac{\text{Biomass or grain yield}}{\text{Evapotranspiration}}$$

Flexas, (2016) [12] studied genetic improvement of water use efficiency in C₃ plants and generated equation for intrinsic water use efficiency, it is the ratio of assimilation to stomatal conductance.

$$WUE_i = \frac{\text{Assimilation(intrinsic)}}{\text{Stomatal conductance}}$$

Water productivity is another term which also used instead of water use efficiency in many studies. Water productivity is the ratio of agriculture benefit to the water use, the unit is same as that of water use efficiency, which give the holistic water use efficiency of a cultivation (Kijre *et al.*, 2003) [20]. One component of water use efficiency is the agricultural product which measured in terms of yield (Kg, mg, t), Energy equivalent (Kcal) or income (\$).

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Evapotranspiration is another component of water use efficiency which can be measured by using equation

$$ET = P + I + G \pm Q - AS$$

Where, P-Precipitation, I- Irrigation, G-net ground water flow, Q-Run on or off ΔS change in soil water content (Allen *et al.*, 1998) [1]. In crop plants, C₃ plants like rice, shows lower water use efficiency than C₄ plants like sugar cane and maize. Crassulacean acid metabolism (CAM) plants shows higher water use efficiency than C₄. Irrigation and N₂ fertilizer application increases water use efficiency at certain level, after that it decreases which experimented by Gajri *et al.*, (1993) [13] in wheat with varying levels of water and nitrogen fertilizer. Some studies revealed that aquaporin also involve in water use efficiency of rice its inadequate expression leads to limited water use efficiency (Nada and Abogadalla, 2014) [24]. Rice cultivation require large amount of water especially low land rice so that its water use efficiency very low. Many method can be adopted to improve water use efficiency of rice such as wet seeding, mulching, genetic transformation, plant breeding methods and aerobic rice are some of them.

Carbon isotope discrimination

Extent of carbon isotope discrimination in leaves can be correlated with water use efficiency of the crop. The relationship between water use efficiency and ¹³C/¹²C ratio experimented in a study conducted by Winter *et al.*, (2004) [29] using C₃ and CAM plants (Fig.1). In a study, 28 rice cultivars including Indica, Japonica and AUS types were analyzed for carbon discrimination (Δ) using the equation $\Delta = Ra/Rp - 1$, [where Ra-Isotopic ratio of atmospheric CO₂ Rp- Isotopic ratio of In plant CO₂] which is negatively correlated with WUE (Farquhar and Richards 1984, Wright *et al.*, 1988) [9, 30]. Result showed that carbon isotope discrimination and Water use efficiency correlated in Japonica and Aus types but not in Indica group (Dingkuhn *et al.*, 1991) [7]. Another study revealed that double haploid and recombinant imbred lines generated from IR64 X Azucena showed genetic variation in the carbon isotope discrimination which is contributed by genetic variation in the carbon metabolism and genetic variation in the stomatal conductance (This *et al.*, 2010) [28].

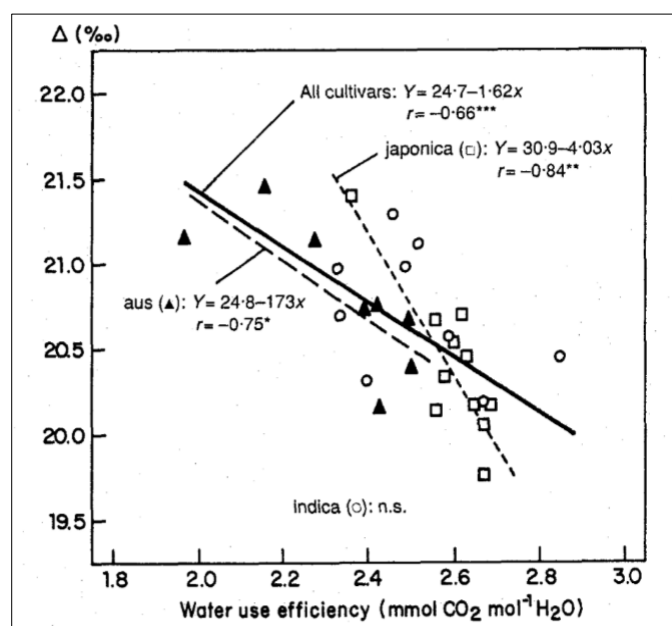


Fig 1: Correlation graph between WUE and Δ (This *et al.*, 2010) [28].

Methods to improve water use efficiency

Water use efficiency of rice can be improved through various methods such as cultural, biotechnological, breeding and physiological approaches. Many studies were conducted under these approaches and results showed improvement in water use efficiency of rice

A. Breeding and Molecular approaches

1. Genetic transformation

Arabidopsis drought and salt tolerant gene HARDY introgressed to rice genome to improve water use efficiency (WUE) in rice (*Oryza sativa* var. Japonica). The rice cultivar Nipponbare was used for the study, results showed that there did not have significant difference in germination, growth and yield but the transformed plants showed increase in leaf canopy (Fig 2) with more number of tillers and significant increase in number of bundle sheath cell (Fig 3). HRD gene plays a role in stomatal conductance as it reduced the stomatal conductance led to increase in water use efficiency (Karaba *et al.*, 2007) [18]. A multifunctional protein target of Rapamycin (TOR) present in all eukaryotic organism. In a study TOR gene from Arabidopsis thaliana introgressed in to widely cultivated indica rice variety. The over expression of AtTOR gene in transformed lines showed higher photosynthetic efficiency with lower water use efficiency which measured in term of $\Delta^{13}C$ which was low(17‰) in transformed plants (Bakshi *et al.*, 2017) [4].

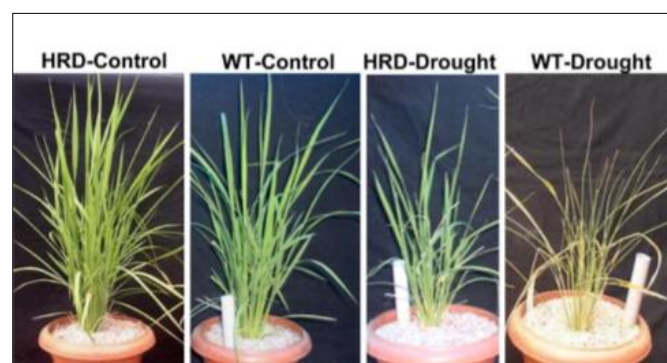


Fig 2: Phenotype of HRD overexpression in rice

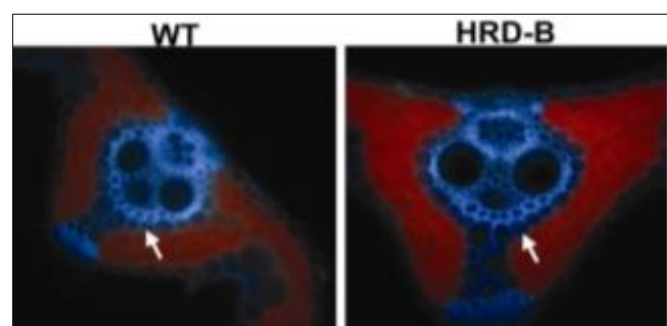


Fig 3: Leaf cross section of HRD over expressed line (Karaba *et al.*, 2007) [18].

2. Activation tagging

Gain of function mutagenesis through activation tagging used as alternative approach to loss function mutagenesis. Activation tagging is suitable for Japonica than Indica due to recalcitrant nature of Indica seed. Tagging of ribosomal protein is a new category of activation tagging which studied by Moin *et al.*, (2016) [21] to understand the stress tolerance capacity of mutant indica rice varieties under water limiting condition. The vector used for the study was PSQ5 and

obtained enhancer sequence from CaMV35S promoter. Callus transformation method used for the transfer of vector in to the rice. The results showed that mutants recorded better yield, tiller number, number panicle and seed yield than wild type. Percentage of $\Delta^{13}\text{C}$ measured was high for wild type (22.05%) compared to mutant, which indicate lower water use efficiency of wild type than mutants. As a continuation of this study Moin *et al.*, (2017) [22] identified ribosomal protein large (RPL) gene family can be manipulated to improve water use efficiency, fresh weight, root length, increase in proline and chlorophyll content of rice under drought and salinity stress.

3. Backcross breeding

Maintaining leaf turgor and carbon metabolism are two adaptive mechanism developed by tolerant plants under water limited condition. This was evidently showed in a study conducted by Dharmappa *et al.*, (2019) [6]. Introgression of water mining root and water use efficiency trait in to IR-64 rice variety. AC39020 was the donor parent used for root mining trait whereas for water use efficiency IET16348 was selected. Introgression lines developed through marker assisted back cross breeding. The desired plant achieved after 3 backcross and selfing. The resultant plant showed 53% and 23.5% increment in root weight and root length respectively and 53% and 63% higher yield obtained at 100% and 60% field capacity. Carbon isotope discrimination was very low for introgressed line indicate water use efficiency and yield were enhanced in trait introgressed lines which is quite evident in water limiting condition (Fig 4). In a study, association mapping of 173 diverse rice lines were evaluated to find out QTL for root and water use efficiency characters. The results showed that 9 and 16 markers each responsible for carbon isotope discrimination and root length respectively (Raju *et al.*, 2016) [25].

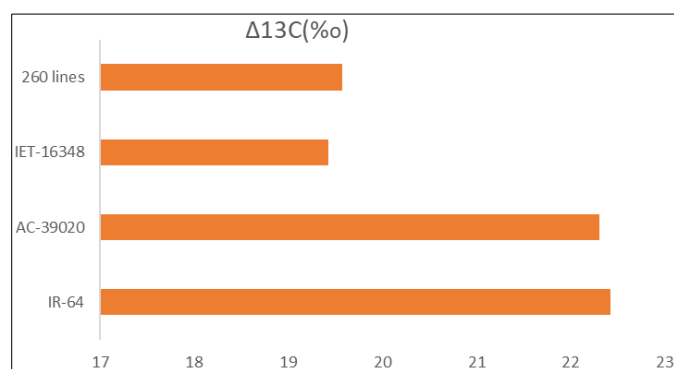


Fig 4: Carbon isotope discrimination between parents and TIL (Dharmappa *et al.*, 2019) [6].

B. Physiological approaches

1. Endophytic bacteria

Recent study revealed that Salicaceae endophytes enhanced water use efficiency of rice. Rho *et al.*, (2018) [26] isolated the strains of diazotrophic endophytic bacteria and yeast such as WP1, WP5, WP9, WP19, WP13, WW5, WW7C and PTD from plants like cotton wood, wild willow and hybrid poplar. These strains have the capacity to amplify nifH marker gene which is responsible for the enzyme nitrogenase reductase in

the nitrogen fixation led more biomass accumulation. These strains inoculated plants also reduced stomatal conductance and stomatal density of rice during day time which in turn increased water use efficiency, this effect pronounced under water stress condition.

2. Polyamine, Selenium and *Trichoderma* sp application

Among extreme climate condition, drought is a serious stress this will hamper soil and plant water status. In a study to alleviate the negative impact of drought stress, polyamines like putresine, spermine and spermidine were applied exogenously (seed priming, foliar application). Results showed that net photosynthetic rate and biomass yield were increased, this is because of maintenance of leaf water status of the plant through water use efficiency Fig.5 (Farooq *et al.*, 2009) [10]. In another study exogenous application of Selenium (Se) at the rate of 2mgKg^{-1} increased water use efficiency, net photosynthetic rate and antioxidant activity of rice grown under water stress condition (Andrade *et al.*, 2018). Effect of different *Trichoderma* sp on rice growth were evaluated in a study revealed that *Trichoderma* sp treated plants showed higher growth and physiological activity compared to control. Among various isolates of *Trichoderma* sp, local isolate SL2 treated plants showed higher root biomass and intrinsic water use efficiency (Doni *et al.*, 2014) [8]. Gypsum application improved water use efficiency and soil properties compared to control in saline sodic soil under rice cultivation (Hafez *et al.*, 2015) [14].

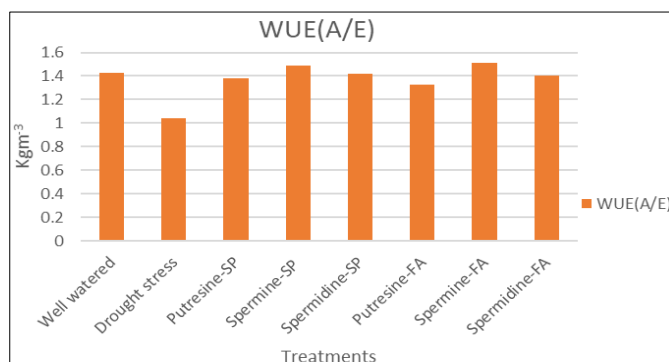


Fig 5: Water use efficiency under polyamine treatment (Farooq *et al.*, 2009) [10].

3. Leaf hair modification

One of the morphological character which changes leaf water use efficiency is leaf hair. In a study plant breeders developed an introgression line through back cross breeding called IL-hairy, which possess more leaf hair. IL-hairy was the progeny of parental line IR-24 (hair less) and IRGC105715 is the wild accession of *Oryza nivara* have hairs on the leaf. Scientist identified the gene responsible for the hairiness using simple sequence repeat (SSR) markers and named as BLANKET LEAF (BKL) and it was located between RM 3567 and RM 7243 in chromosome number 6 in BC_5F_3 population. IR-lines have significantly higher water use efficiency under high and moderate light intensity than parental line however its photosynthetic rate was lesser compared to IR 24 this can be used for water saving cultivation system (Fig 6) (Hamaoka *et al.*, 2017) [15].

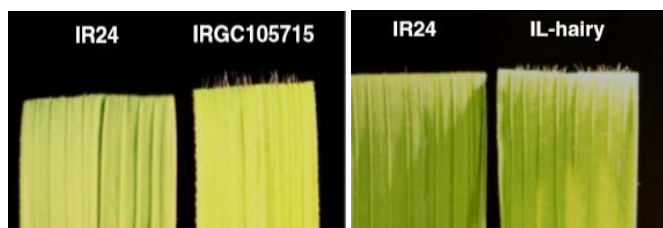


Fig 6: Comparison of leaf hair between parental line and Introgressed Line (Hamaoka *et al.*, 2017) ^[15].

4. Aerobic rice

Kato *et al.*, 2009 studied the difference in yield potential between aerobic and flooded high yielding rice varieties in Japan. The results showed that aerobic rice sustained yield under less water than flooded condition and water productivity measured for aerobic rice was 0.8-1Kgm⁻³ of water. This study added one more option for water conservation system of cultivation

B. Cultural practices

1. Wet seed cultivation

Improvement in rice production technology led the farmers to expense major portion of irrigation water for rice cultivation. Under such circumstances, need arisen to use water effectively for sustaining rice cultivation. Scientist in Philippines developed a wet seed culture method which was superior to traditional transplanted rice culture and the water use efficiency of the method was significantly higher (Table.1) (Bhuniyan *et al.*, 1995) ^[5]. Alternate wetting and drying method also reduced water usage in the rice cultivation even though it reduced yield compared to continuous flooding system (Zhang *et al.*, 2012) ^[32]. System of rice intensification is an another agronomical practice which improved both yield and water use efficiency (Zhao *et al.*, 2010) ^[33].

Table 1: Water use efficiency of wet seeded rice and transplanted rice (Redrawn from Bhuniyan *et al.*, 1995) ^[5].

Water regime	Water use efficiency(kgm ⁻³)	
	Wet seeded rice	Transplanted rice
5-7cm standing water	5.13	5.28
2cm every alternate days	6.18	6.12
2cm every 4 days	7.56	5.75
5cm every 5 days	3.93	3.32
Average	5.7	5.11

2. Mulching

Flooding cultivation is the common, widely accepted cultivation practice in rice production. The disadvantage of this system is that, it require ample amount of water during entire life cycle of rice. Non flooded mulching cultivation was experimented in Indica rice hybrid Shanyou, revealed that straw mulched plants showed similar yield as traditional flooding cultivation. Mulching reduced tiller number in early stages of growth but later stage it performed better than non-mulched plants. This is due to increase in chlorophyll content, increased photosynthetic rate of flag leaf and root growth which retained in the last stage of plant growth therefore this method can be approached as water saving cultivation in rice (Xu *et al.*, 2007) ^[31]. In another study Fawibe *et al.* (2020) ^[11] evaluated water use efficiency of rice cultivar Koshihikari and Norin 24, low land and upland rice cultivars respectively grown under continuous flooding and drip irrigation with plastic mulch(DPD). The results showed that the water use efficiency of Koshihikari and Norin 24 increased by 50% and

70% respectively under DPD compared to continuous flooding, indicate Fe deficient and water limited condition drip irrigation with plastic mulch in rice is a promising approach to sustain yield. Similar study of plastic mulching with drip irrigation on rice plants were conducted by he *et al.*, (2013) ^[16].

Conclusion

There is a wide scope to improve water use efficiency in rice to sustain its yield under water limited condition. Key strategies used to improve water use efficiency are genetic transformation, wet seed cultivation, polyamine application, mulching, using endophytic bacteria, improve morphological adaptation like leaf hair, aerobic rice cultivation, activation tagging and backcross breeding. But care should be taken during water storage, transportation and irrigation to reduce wastage of water. The holistic approach of agronomy, physiology, breeding and biotechnology will help to conserve more water that can be used efficiently for rice production.

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