Cultivos Tropicales, 2020, vol. 41, no. 2, e07
ISSN impreso: 0258-5936
ISSN digital: 1819-4087

abril-junio
Ministerio de Educación Superior. Cuba
Instituto Nacional de Ciencias Agrícolas

http://ediciones.inca.edu.cu

Original article

Rice plant response to suspension of the lamina of water. Part III

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ABSTRACT

The research was at the Experimental Station of Zaidín, Granada, Spain carried out, with the objective of evaluating the physiological and biochemical response of the rice plant, grown under anaerobic conditions and exposed to a suspension of the water sheet for a period of 15 days in agricultural performance. Rice plants (cv. 'INCA LP-5') were under semi-controlled conditions grown in plastic pots. The suspension of the water sheet was at three moments of its development carried out, at 30, 40 and 50 days after the transplant (DAT). Agricultural performance, the number of panicles per plant, full grains per panicle and the mass of 100 grains, at 147 DAT were evaluated. A performance correlation was with the height of the plant made, fresh aerial mass and roots, foliar water potential, stomatic conductance, foliar content of hydrogen peroxide and oxidative damage to lipids, which were at 122 DAT evaluated. It was obtained that, the number of panicles per plant and the full grains per panicle were favored with the application of water stress, indicators that contributed to the increase of agricultural yield in treatments exposed to water stress (30 DAT=23 %, 40 DAT=20 % and 50 DAT=11 %, compared to the witness). Agricultural yield was correlated with plant height, fresh aerial and root mass, foliar water potential, stomatic conductance, hydrogen peroxide foliar contents and oxidative lipid damage.

Key word: Hydrogen peroxide, *Oryza sativa*, water stress, lipid peroxidation, recovery

INTRODUCTION

Rice (*Oryza sativa* L.) is the food source for 3 billion people worldwide. The world annual production of paddy rice is approximately 650-700 million tons [(1)]. This cereal can grow in a wide range of hydrological situations, soil types and climates. In this crop, the environment where the plants grow is as the conventional flooded rice system known, which leads to using greater amounts of fresh water for its cultivation compared to the dry rice production system [(2)].

In the world, there are about 158 million hectares of flooded rice (including double crops), of which 101 million hectares correspond to irrigated rice crops, constitute 75 % of world rice production; while the remaining 57 million hectares correspond to rainfed rice, and contribute 19 % of world production [(1)].

In Cuba, this cereal is in flood conditions grown, that is, in the presence of a sheet of water for most of its cycle, where the management of irrigation water is a limiting factor in rice production.

The decline in rice productivity in most cases is to various abiotic factors attributed, including drought. Drought stress has become a serious threat to guarantee food security in the developing world [(3)]. Although water is necessary in all periods of growth of the rice plant, there are some critical stages of growth when drought stress severely affects and creates a massive reduction in the quantity and quality of agricultural yield. Crop responses to drought stress and their tolerance level can be by monitoring different physiological and biochemical changes measured after the drought period and plant recovery [(4)]. Plants respond to drought stress at the molecular, cellular and physiological levels that vary between species and the genotype, duration and severity of water stress, the age of the crop and the stage of development [3]. All these elements are essential to be able to understand and work in terms of saving water, while not affecting agricultural performance.

Crop yield is often the attribute that primarily focuses on breeding programs and management decisions. In the case of cereal crops, it is the result of growth and development processes, which are strongly by genetic factors regulated, environmental conditions, and genetic and environmental interactions throughout a growing season [5]. Most crop physiologists work to identify the processes that determine differences in yield due to genetic and environmental factors [5]. Yield is the proportion of the total biomass of the crop assigned to the usable organs. In the case of cereals and other grain crops, it is the biomass assigned to the grains [5].

To maintain crop productivity under drought stress and ensure food security, improved cropping technology must be adopted to increase crop water use efficiency [3]. In Cuba, relevant results have been with the application of water stress achieved (water deficit in the soil) in the cultivation of rice by direct sowing [6]. However, not all the elements that validate the application of water stress in this crop due to transplant technology are available. Therefore, the objective of the present study was to evaluate the physiological and biochemical response of the rice plant, cultivated in anaerobic conditions and exposed to suspension of the water layer for a period of 15 days in agricultural performance.

MATERIALS AND METHODS

The research was at the Zaidín Experimental Station, Granada, Spain (EEZ) carried out, in 2010 under greenhouse conditions with rice plants cv. INCA LP-5. Initially, a rice seedbed was in plastic trays of 0.40x0.80x0.08 m with sterile sand established. To achieve the germination of the rice, the trays were watered until achieving a lamina of water 5 cm above the surface of the sand for a period of 24 hours, after which time the tray was drained, keeping the sand at maximum water holding capacity, until two leaves per plant emerged. Subsequently, the 3 cm lamina of water was until 30 days after the emergency (DAT) restored.

At 30 DAT, a plant was into each 1 kg pot transplanted (0.18 m high and 0.13 m in diameter). It contained a substrate composed of sand (grain size <1 mm) and soil (grain size < 5 mm) in a 1: 1 ratio (v: v). It was previously sterilized; the sand at 120 ºC for 20 min, in a Selecta autoclave, model PRESOCLAVE-II 75 L, and the soil at 95-100 ºC, but for 60 min daily for three consecutive days. The soil that was used was classified as Fluvisol Haplic Calcareous [(7)], which had a pH of 8.1 (measured by potentiometry), 1.81 % organic matter (Walkley and Black method), assimilable phosphorus 6, 2 mg kg^{-1} (P-Olsen) and exchangeable potassium 0.34 cmol kg^{-1} (extraction with NH_4OAc 1 mol L^{-1} at pH 7).

The pots were placed in the greenhouse where the seedbed was established, with temperatures of 26 and 22 ºC (day/night, respectively). The relative humidity was between 50-70; Photoperiod of 16 hours of light and 8 hours of darkness and photosynthetically active radiation of 850 µmol m^{-2} s^{-1}, measured with a portable LICOR (Lincoln, NE, USA, model LI-188B). It was following an experimental design Completely Randomized, with a bifactorial arrangement and five repetitions, for which 15 pots per treatment were used, which allowed evaluations to be made after each period without water lamina.

The water supply consisted of maintaining a lamina of water 5 cm above the surface of the substrate in all treatments (Without E), until the time the lamina of water was suspended, at 30, 40 and 50 DAT (With E), for a period of 15 days. At which time the sheet of water was, which remained until 15 days before harvesting replaced; the group of pots to which the sheet of water was not suspended remained as control treatments (Figure 1).

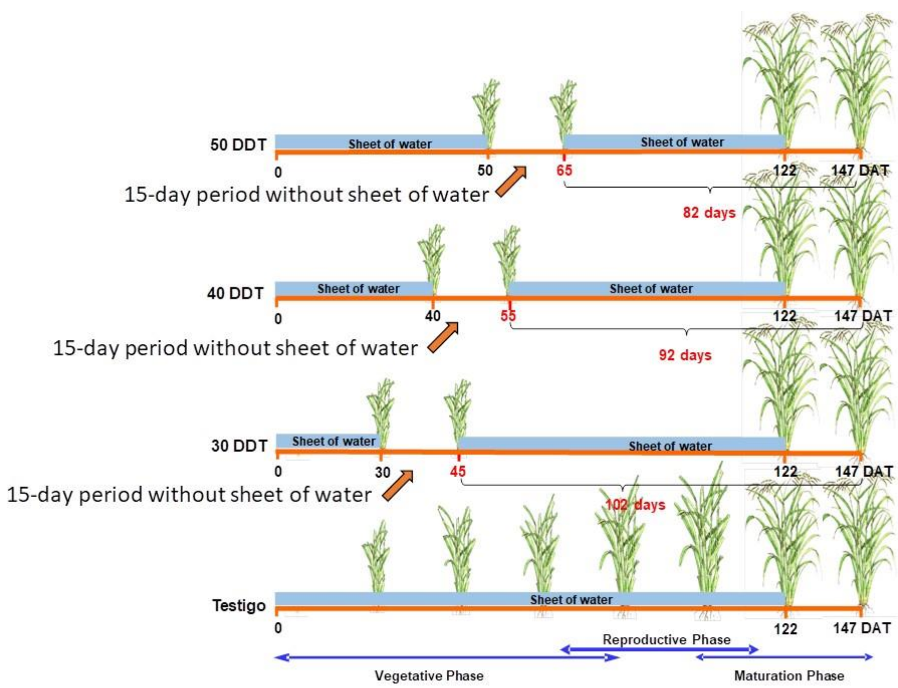

Figure 1. General irrigation scheme developed in the experiment with rice plants cv. INCA LP-5 exposed to water stress at 30, 40 and 50 days after transplantation (DAT) for a period of 15 days

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The total application of nutrients, corresponding to 0.123 g of N; 0.050 g of P_2O_5 and 0.059 g of K_2O per pot, was performed at 20, 35 and 60 DAT, applying at each time 30, 40 and 30 %, respectively, using Urea (46 % N), Triple Superphosphate as carriers. (46 % P_2O_5) and Potassium Chloride (60 % K_2O), respectively.

Sampling and evaluations carried out

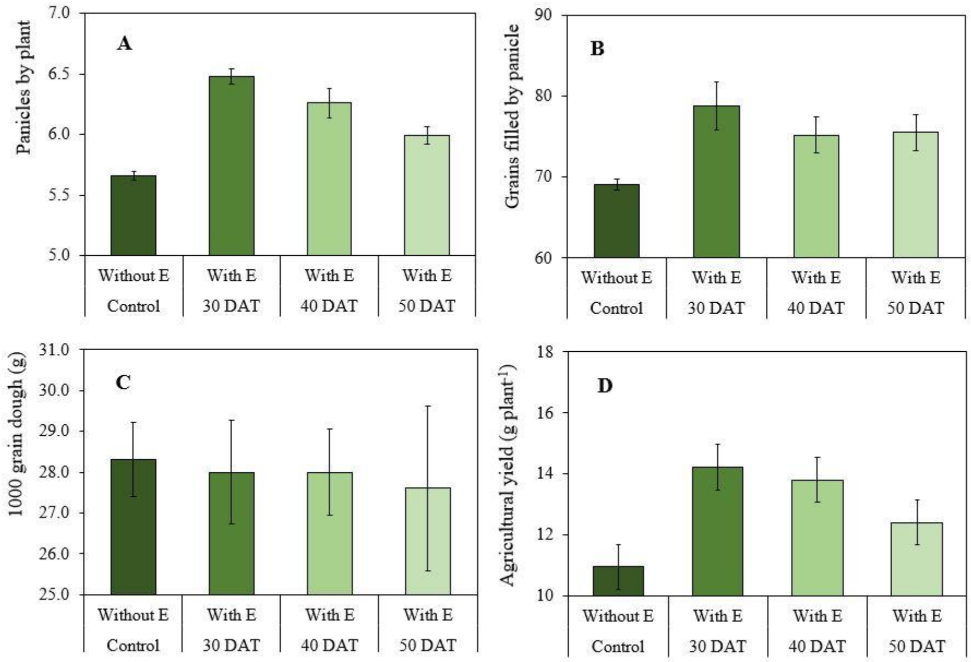
Five plants were taken per treatment at 122 DAT (25 days before the grain harvest), to assess the height of the plants (ALT), the fresh air mass (MFA) and the roots (MFR), the water potential foliar (Ψ_h), stomatal conductance (EC), foliar contents of hydrogen peroxide (H_2O_2) and oxidative damage to lipids (DOL). ALT, MFA, MFR, Ψ_h, CE, H_2O_2 and DOL were determined following the same procedures and protocol that are in the first part of this article described (8).

At the time of harvest (147 DAT) when 85 % of the ears in the plant turned yellow (9). The agricultural yield components [number of panicles per plant (P_p), filled grains per panicle (GLL_p) were evaluated and four panicles were taken to count the filled grains per treatment]. In addition, a mass of 100 grains (M100) was determined per replica and per treatment, and the agricultural yield (YIELD), which was in g plant^{-1} expressed.

The mean values of P_p, Gll_p, M100 and YIELD of each treatment were compared from the Confidence Intervals for $\alpha = 0.05$. With the data of the evaluations of ALT, MFA, MFR, Ψ_h, CE, H_2O_2 and DOL, Pearson Correlations were made based on the effect that the suspension of the water lamina led to at 30, 40 and 50 DAT on performance agricultural rice.

RESULTS AND DISCUSSION

After verifying, that the suspension of the water layer at different times of the vegetative phase of the rice plant for a period of 15 days caused water stress to the plant (8) and demonstrated that these plants recover from it (4) a differential effect was on agricultural yield and its crop components found (Figure 2).



Bars above the columns indicate confidence intervals (α≤0.05)

Figure 2. Panicle per seedling (A), filled grains per panicle (B), mass of 1000 grains (C) and agricultural yield (D) in rice plants without water stress (Without E) and exposed to water stress (With E) for a period of 15 days at 30, 40 and 50 DDT, respectively, evaluated at harvest (147 DDT)

The highest values of panicles per plant, grains filled by panicle and agricultural yield were found in the plants exposed to suspension of the water layer for a period of 15 days during the vegetative phase with respect to the control treatment. In the case of panicles by plants (P_p), the highest values of this indicator corresponded to the suspension of the sheet of water at 30 DDT. However, the applied water management did not lead to variations in the number of panicle-filled grains between the treatments that were exposed to water stress, but these reached a greater number of panicle-filled grains (Gll_p) than the control treatment. On the contrary, no differences were found regarding the mass of 100 grains (M100) between the treatments with E and without E (control).

Agricultural performance increased in treatments exposed to water stress (With E) compared to the control. The increase at 30 DAT compared to the control was 23.03 %, at 40 DDT it was 20.66 % and at 50 DDT it was 11.72 %. However, no differences in performance were found with the application of water stress at 30 and 40 DAT, while there were no differences between the application of water deficit between 40 and 50 DAT.

Exposing the rice plant to reduced water supply at some point in the vegetative phase for a period of 15 days, did not affect the crop and guaranteed the increase in agricultural yield. In this regard, some authors stated that water stress in the vegetative phase in rice plants increases the number of panicles per plant [10], a variable that they identified as a determinant in agricultural performance. However, in this research, it was also found that the number of grains filled by panicles was decisive in agricultural performance. Under field conditions [11], no significant differences were found regarding the mass of 1000 grains and the best response to water stress for a period of 15 days was found when it was applied to 30 DAT [11].

The increase in the number of panicles per plant may be related to the development of the root system, expressed in this case as MFR (4.8). This radical increase was favored by the stimulus for water deficit. Other authors claim that water stress favors radical growth [10,11]. In addition, it may be due to the recovery period, which was greater when the plants were exposed to this condition at the beginning of the vegetative phase, or due to 30 DAT (82 days), 40 DAT (92 days) and 50 DAT (102 days).

The number of panicles per plant and the filled grains per panicle, in the specific case of rice per transplant. They are determining factors in agricultural performance; since, with a smaller number of plants per m^2 and an equal amount of filled grains per panicle, it is possible to equal or increase the agricultural yield, which is achieved in the production of rice by direct sowing [11]. These results show that a similar behavior behaved in this form of production (rice per transplant).

The evaluation that was carried out on the 122 DAT of the physiological and biochemical variables (ALT, MFA, MFR, Ψh, CE, H_2O_2, DOL, PRO) in correlation with performance, explained its effect on variable, induced by the imposed water deficit at 30, 40 and 50 DAT (Table 1).

Table 1. Pearson's correlation coefficients of philological and biochemical variables, with respect to agricultural performance, due to the effect of suspending the sheet of water at 30, 40, and 50 DAT, for a period of 15 days at the time of harvest at 147 DAT. n = 8

Variables	**Agricultural yield (YIELD)**		
	30 DDT	**40 DDT**	**50 DDT**
ALT	-0.811**	-0.786**	-0.881**
MFA	-0.928**	-0.890**	-0.871**
MFR	-0.967**	-0.970**	-0.846**
Ψ_h	0.161	-0.466	-0.794**
CE	0.945**	-0.848**	-0.882**
H_2O_2	-0.970**	0.949**	0.885**
DOL	-0.333	0.824**	0.696*
PRO	-0.716*	0.937**	0.867**

V Altura de las plantas (ALT); Masa fresca aérea (MFA); Masa de las raíces (MFR); Potencial hídrico foliar (Ψ_h); Conductancia estomática (CE); Contenidos foliares de peróxido de hidrógeno (H_2O_2); Daño oxidativo a lípidos (DOL);Prolina (PRO); y Rendimiento agrícola (REND); Días después del trasplante (DDT)

*La correlación es significativa en el nivel 0,05 (2 colas)

**La correlación es significativa en el nivel 0,01 (2 colas)

The water stress applied to the 30 DAT for a period of 15 days led to a positive or negative response regarding the correlations found between the YIELD and the rest of the physiological and biochemical variables evaluated. This was positively related to CE and negatively related to ALT, MFA, MFR, H_2O_2 and PRO. At the same time, it did not show a relationship with Ψh and DOL. The water stress imposed on 40 and 50 DAT at harvest time showed a similar response. Agricultural performance (YIELD) was correlated with all variables evaluated inversely with ALT, MFA, MFR, Ψh, CE and positively with H_2O_2, DOL and PRO.

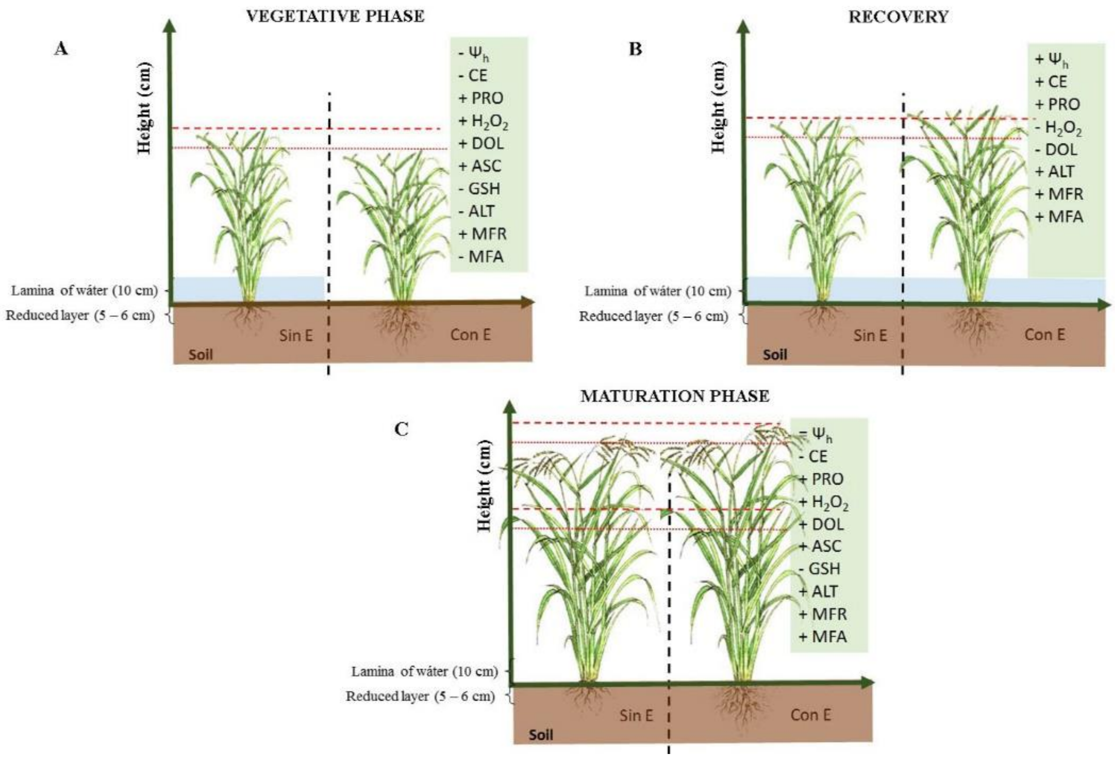
That no correlation was found between the REND and the Ψh at 30 DAT, could be indicating that, at the end of the crop cycle, a stability of this variable in potential values in the order of - 0.93 MPa [(4)]. In addition to this, it is possible that in the phenophase (within the grain maturation phase, it would be at the end of the pasty phenophase) that the plant was, the movement of water decreases, that is, the absorption and, occurs with greater frequency the loss of the same by transpiration and evapotranspiration. However, the correlation found when stress was at 40 and 50 DAT applied; it is possible that it responds to the decrease in water potential that had a marked effect on yields, from a possible decrease in the translocation of photoassimilates. Due to water deficit in the stage that the plant begins to prepare for the reproductive phase. Results that were evident in terms of Ψ_h values [(8)] and the relationship of this with biochemical variables showed the negative effect at this time due to the water deficit caused (4).

The positive correlation found at 30 DAT between the REND and the EC, indicated the dependence of a transpiration system and gas exchange on the production of P_p, the filling of the grains in the translocation process and therefore with the yield agricultural. In this regard, it was reported that plants exposed to water stress closed their stomata to maintain their internal moisture content and, consequently, their transpiration and photosynthetic rates, and productivity decreased (4, 12). Therefore, it is assumed that the decrease in EC during the period of water stress and at the end of the cycle indicated a stomatal closure that caused a reduction in the evapotranspiration of rice (12), which decreases with water stress and is linearly related to grain yield. The positive correlations that were found between agricultural performance and biochemical variables correspond directly to the vegetative phase of the plant, where senescence processes are accelerated at the end of the biological cycle (13) and, therefore, production is increased. H_2O_2, DOL and PRO.

Yield is assumed equally sensitive to changes in biomass at any time during crop growth (14). Instead, another author assures that the effects of water stress in the grain-filling phase could lead to a reduction in panicle-filled grains (2). Therefore, the reduction or decrease in yield when water stress is applied late (to and DAT), although they were not in the grain filling stage, but it was close to the beginning of the reproductive phase.

Water stress for a period of 15 days at 30, 40 and 50 DAT, caused physiological and biochemical changes after the period of water stress, such as decreased growth and air development of the plant, decreased leaf water potential and stomatal conductance On the contrary, the radical mass of the plant was increased (8). From the biochemical point of view, proline content, hydrogen peroxide content, oxidative damage and the content of the antioxidant glutathione increased, while reduced ascorbate decreased (8). After the recovery period at 122 DAT, the water potential and stomatal conductance increased, the content of hydrogen peroxide and oxidative damage to lipids also increased (4). At the time of harvest the best results were found when the water stress was applied to the 30 DAT and the response of the plant was evaluated in the three moments previously exposed, which are summarized in Figure 3.

In this treatment after plant rice stress with respect to the control, the Ψ_h, CE, MFA, ALT, GSH are decreased, while the content of PRO, ASC, H_2O_2 and DOL was increased during the vegetative phase (Figure 3A).



Plant height (ALT), Fresh air mass (MFA), Root mass (MFR), Leaf water potential (Ψh), Stomatal conductance (EC), Foliar contents of hydrogen peroxide (H2O2), Oxidative damage to lipids (DOL), proline (PRO). Sign (+), increase with respect to the previous condition. Sign (-), decreased with respect to the previous condition. Sign (=), equal to the previous condition

Figure 3. Mechanism of physiological and biochemical response in rice plants without water stress (Without E) and exposed to water stress (With E) for a period of 15 days at 30 DAT

After the recovery period at 122 DAT (Figure 3C), the water stress imposed in the vegetative phase caused physiological and biochemical changes, the Ψ_h, CE, MFA, MFR, ALT increased, while H_2O_2 and DOL decreased. (Figure 3 B). At the time of harvest (maturation phase), the igh were equalized, with a tendency to decrease, EC and GSH decreased, while PRO, H_2O_2, DOL and ASC increased.

From the results achieved, it can be assured that the management of irrigation water in rice cultivation is essential for the growth and development of the rice plantation. Because this leads to physiological and biochemical changes that determine the crop's agricultural yield at the end of the cycle [4, 8]. The relationship between the variables of water relations (Ψ_h and CE) with the physiological ones (ALT, MFA and MFR) and biochemical ones (H_2O_2, DOL and PRO) demonstrate the sensitivity of the rice plant to variations in its water status. That is why the rice plant can be used as a model for agricultural, physiological, biochemical and molecular research; from which the response of the same to changes in the management of irrigation water in this crop is evident.

In general, from the effect caused by the water stress imposed on 30, 40 and 50 DDT on the plant [(8)], their response after the recovery period [(4)] and at the time of maturation (harvest) water can be saved, when it is cultivated for the purpose of applying a water deficit in the soil and in the plant with the suspension of the sheet of water at 30 DDT.

It was obtained that the number of panicles per plant and the grains filled per panicle were favored with the application of water stress, indicators that contribute to the increase in agricultural yield in treatments exposed to water stress (30 DDT=23 %, 40 DDT=20 % and 50 DDT=11 %, with respect to the control).

Agricultural performance is correlated, both with physiological and biochemical variables, that is, it is dependent on the water state of the plant and its transformation due to a water deficit. On the other hand, it should be noted that the positive effect of water stress on yield translates into an improvement in the industrial quality of the grain [(11, 14, 15)].

CONCLUSIONS

- The rice plant can be used as a model for agricultural, physiological, biochemical and molecular research from which the response of the same to changes in the management of irrigation water in this crop is evidenced. In addition to the fact that when faced with a water deficit, they show their adaptive potential in the face of this condition of abiotic stress.
- The water stress applied to the rice plant for a period of 15 days to 30 days after transplanting contributes to the increase in agricultural yield with respect to that applied at 40 and 50 days after transplanting.

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