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Original article

Behavior of physiological variables in maize plants (*Zea mays*) under controlled deficit irrigation (CDI) in stages

Comportamiento de variables fisiológicas en plantas de maíz (*Zea mays*) bajo riego deficitario controlado (RDC) por etapas

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ABSTRACT: The behavior of physiological variables in corn plants (cultivar P7928) grown in semi-controlled conditions (concrete containers) and applications of controlled deficit irrigation (CDI) strategies in three stages of crop development were studied. The four treatments applied consisted of suspending irrigation for 15 days in the growth (GT), flowering (FT) and grain filling (GF) stages and a control treatment was maintained (T 100) that always received the corresponding water at 100 % of ETc (standard crop evapotranspiration). At the end of the irrigation suspensions, the following were evaluated: soil moisture, leaf area, aerial dry mass, leaf water potential (Ψf) and its components, total soluble protein content, chlorophylls A, B, A+B and carotenes in leaves. With the application of CDI treatments, the percentage of soil moisture decreased to 60 % of the Field Capacity (C.c.). The results of the evaluations in the plants of each of the treatments (GT, FT and GF) were compared always with the control plants (T 100). Water stress negatively affected growth variables in GT and leaf area turned out to be more sensitive to stress. Regarding the values of Ψf and its components, these indicated that the plants in GF were less sensitive to the suspension of irrigation. Furthermore, water stress enhanced protein content and did not affect carotene content in any of the stages; while chlorophylls had greater degradation in (FT and GF) plants.

Key words: water stress, foliar water potential, growth, proteins, chlorophylls.

RESUMEN: Se estudió el comportamiento de variables fisiológicas en plantas de maíz (cultivar P7928) cultivadas en condiciones semicontroladas (contenedores de hormigón) y aplicaciones de estrategias de riego deficitario controlado (RDC) en tres etapas del desarrollo. Los tratamientos aplicados consistieron en suspensión del riego 15 días en la etapa crecimiento (TC), de floración (TF) y de llenado del grano (TLL) y un tratamiento control (T 100) que recibió siempre el agua correspondiente al 100 % de la ETc (evapotranspiración estándar del cultivo). Al concluir las suspensiones del riego se evaluaron: humedad del suelo, el área foliar, la masa seca aérea, el potencial hídrico foliar (Ψf) y sus componentes, el contenido de proteínas solubles totales, las clorofilas A, B, A+B y los carotenos en las hojas. Con la aplicación de los tratamientos de RDC disminuyó la humedad del suelo hasta el 60 % de la capacidad de campo (C.c.) Los resultados de las evaluaciones en las plantas de cada uno de los tratamientos (TC, TF y TLL) se compararon siempre con las plantas del control (T 100). El estrés hídrico afectó negativamente las variables de crecimiento en TC y el área foliar resultó ser más sensible al estrés. En cuanto los valores de Ψf y sus componentes estos indicaron que las plantas en TLL resultaron menos sensibles a la suspensión del riego. Además, el estrés hídrico favoreció el contenido de proteínas y no afectó el de carotenos en ninguna de las etapas; mientras que las clorofilas tuvieron una mayor degradación en las plantas de (TF y TLL).

Palabras clave: estrés hídrico, potencial hídrico foliar, crecimiento, proteínas, clorofilas.

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INTRODUCTION

The agricultural sector faces the challenge of meeting the nutritional needs of a growing population. Increasing demand for food places a burden on natural resources, including water; although water is the most abundant substance on the planet, with about 70 % of the Earth's surface covered by it, only about 2.5 % is fresh. Generally, irrigated agriculture is the largest consumer of this natural resource, accounting for approximately 70-85 % of its consumption (1).

In light of the current global water scarcity, one strategy to reduce water consumption in irrigated agriculture is to reduce the amount of irrigation water compared to full crop irrigation, in other words, deficit irrigation (2).

Controlled deficit irrigation (CDI) is an irrigation practice in which crops are irrigated with an amount of water below the optimum required for plant growth and productivity. In CDI, plants are intentionally exposed to a certain level of water stress, which can cause a decrease in plant yield; but in the long term, considerable water savings can be achieved (3 and 4). Water saved by the application of these irrigation strategies can be used to irrigate additional land where water is the limiting factor. CDI aims to improve water use efficiency (WUE) either by reducing the amount of irrigation water in each irrigation event or by eliminating irrigation events in periods when irrigation is less productive (5,6).

CDI is considered a sustainable practice and has been adopted to improve water use efficiency, minimize yield losses, and improve product quality (7). Several advantages of CDI include: 1- maximizing water use efficiency, 2- reducing the risk of plant disease spread due to lower moisture, and 3- reducing nutrient loss and leaching out of the root zone, resulting in better groundwater quality and less fertilizer requirements compared to full irrigation (2).

In previous research, crop models have been used to examine the interaction between corn yield and water stress. For example, the MOPECO model and the DSSAT-CERES-Maize model to improve irrigation use efficiency (8).

The variation of irrigation levels with respect to crop growth stages has been little studied in other parts of the world. However, these growth stages have a fundamental importance in the decision related to allow deficit irrigation and, in spite of its importance, as far as it is known, in Cuba, the effects of different levels of reduced irrigation application on the behavior of corn, in different stages of development, has been little studied.

Therefore, the objective of this work was to study the behavior of physiological variables in maize plants (cultivar P7928) grown under semi-controlled conditions and applications of CDI strategies at three developmental stages.

MATERIALS AND METHODS

The work was carried out under semi-controlled conditions in the central area of the National Institute of Agricultural Sciences (INCA) geographically located at 22º58′00″N and 82°09′00″O at 130 m a.s.l. In April 2021, 12 concrete containers 2.60 m long by 0.60 m wide (1.56 m^2) containing Ferrallitic Red Leached Ferrallitic soil from Mayabeque province, an area that is part of the Havana-Matanzas karst plain (9), were sown. In each container were sown corn seeds cultivar P7928 arranged in two rows and with a separation between them of 0.4 m and 0.2 m between plants (26 plants per container).

Four irrigation treatments were studied as described in Table 1 and were distributed following a randomized block experimental design with three replications.

Irrigation was applied by an automated micro-sprinkler system and water delivery was controlled by valves placed on the irrigation laterals of each treatment. The pH and electrical conductivity (E.C. dS m^{-1}) values of the water applied to the crop during the experiment were 7.8 and 0.58, respectively.

Irrigations consisted of replenishing the crop standard evapotranspiration (ETc) daily three times a week (Monday, Wednesday and Friday). At T 100 317.1 mm, at GT 298.1, at F T284.1 and at GF 250.11 mm were applied by irrigation. Plants before and after irrigation suspension (IS) received 100 % of ETc.

The reference evapotranspiration ETo (mm), standard crop evapotranspiration ETc (mm) and irrigation requirements (ETc= ETo*Kc) were obtained using the CropWat.8 Program; this was updated with a historical series of meteorological data for 32 years (1990- 2022) corresponding to Tapaste Meteorological Station which belongs to the National Institute of Meteorology and it is located approximately 200 m from the experimental site and for the calculation of ETo and ETc the monthly mean values were used.

Crop coefficients (Kc) used were: initial Kc = 0.62, average Kc= 1.00 and final Kc= 0.93, proposed for the region (10).

Evaluations performed

All evaluations were carried out at 35, 55 and 75 days after sowing (DAS) coinciding with the culmination of the periods of irrigation suspension in the growth (GT), flowering (FT) and grain filling (GF) phases.

Table 1. Description of the deficit irrigation treatments studied

Treatments	Description
T 100	Control irrigated at 100 % of the crop standard evapotranspiration (ETc) during the whole cycle.
GT	Suspension of irrigation for 15 days in the growth stage (between 20-35 DAS).
FT	Suspension of irrigation for 15 days at flowering stage (between 40-55 DAS).
GF	Suspend irrigation for 15 days at grain filling stage (between 60-75 DAS).

DAS= days after planting. During periods when irrigation was suspended, the concrete containers were covered with a transparent polyethylene roof to prevent rain

Soil moisture was measured at 20 cm depth using a HD2 Precise Moisture Measurement equipment equipped with a Moisture Sensor TRIME®- PICO TDR Technology, Germany calibrated with the internal calibration No. 2 of the probe itself and 15 replicates per treatment were performed.

Leaf area (cm^2)=(length*mean leaf width)*0.75 according to (11) and dry mass of the aerial part (g) were evaluated in nine plants per treatment on the technical balance.

Assessments of leaf water potential (Ψf), current osmotic potential (Ψs) and osmotic potential at maximum saturation (Ψ 100 s) in leaves were performed. All evaluations of water relations were performed on five plants per treatment and on well-developed, sun-exposed leaves of the upper third.

The Ψ f was measured between 10:00 and 11:00 am, with a Scholander type pressure chamber, Soil Moisture Model 3000; Soil Moisture Equipment Co, Santa Barbara, CA, USA. Leaves were pressurized at a rate of 0.03 MPa s^{-1}. To measure Ψs, after Ψf was evaluated, leaves were immediately covered with aluminum foil, frozen in liquid nitrogen, and placed in freezing at -80 °C. For the determination of Ψ100 s, leaves adjacent to those selected for Ψf measurement were taken and placed in hydration chambers with the petiole introduced in distilled water, in the dark and between 6 and 8 ºC for 24 hours. They were then wrapped with aluminum foil for freezing in liquid nitrogen and stored in freezer at -80 °C.

Subsequently, all samples were thawed at room temperature and centrifuged at 3000 rpm for three minutes; the cell juice of the leaves was obtained. From 100 µL aliquots, Ψs and Ψ100 s were determined using a vapor pressure osmometer (Vapro 5520). The turgor potential Ψt was calculated by differences between the Ψs the Ψf.

Evaluations of total soluble protein content were made by the Micro Lowry method and those of chlorophylls A, B, A+B and Carotenes by Wickliff and Aronoff, 1962. For these evaluations, six samples per treatment were taken and the protocols were followed (12).

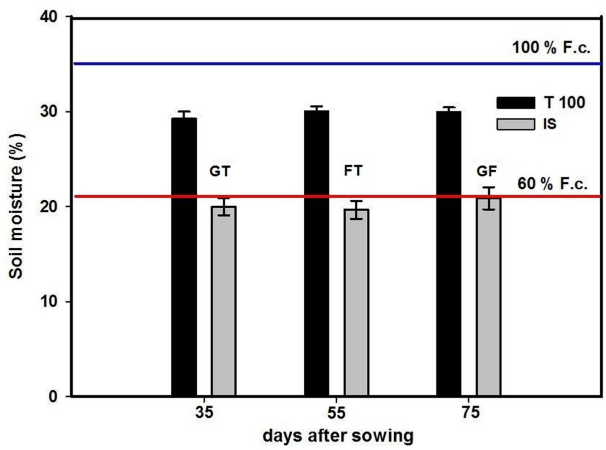
The calculation of confidence intervals of the means and the comparison of these was carried out with the IBM SPSS Statistics 19 program and the graphing of the results by means of the Sigma Plot 11.0 program.

RESULTS AND DISCUSSION

The variations in soil moisture content in the different treatments studied are shown in Figure 1, where it can be seen that, at the end of the irrigation suspension period in each of the phases studied, soil moisture decreased by about 60 % of the soil field capacity, a value that can be considered as a moderate water stress for the crop.

These results suggest that the irrigation scheduling employed was practical in establishing differences in soil moisture content among the treatments used, but only allowed providing the amounts of water to reach the level of approximately 86 % of the field capacity to the T 100 plants.

The effect of CDI treatments on leaf area values (Figure 2 A) shows that the suspension of irrigation for



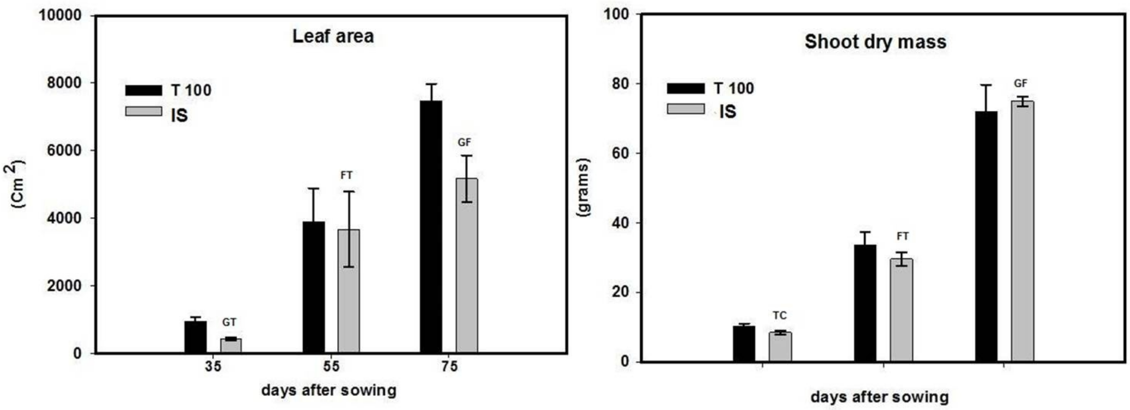
IS= irrigation suspension, GT= suspension of irrigation at growth stage, FT= suspension of irrigation at flowering stage and GF= suspension of irrigation at grain filling stage. C.C = field capacity. Bars above the mean values represent the confidence interval of the means, α= 0.05

Figure 1. Variation of soil moisture percentage in the treatments studied at 20 cm depth

15 days negatively affected the leaf area of the GT and GF treatments with respect to the T 100 plants. On the other hand, the dry mass of the aerial part (Figure 2 B) only decreased slightly with the suspension of irrigation in the GT plants, a behavior that is basically associated with a greater efficiency of nitrogen use in the plants corresponding to FT and GF under conditions of moderate water stress, which allowed maintaining an adequate production of foliar dry matter (13).

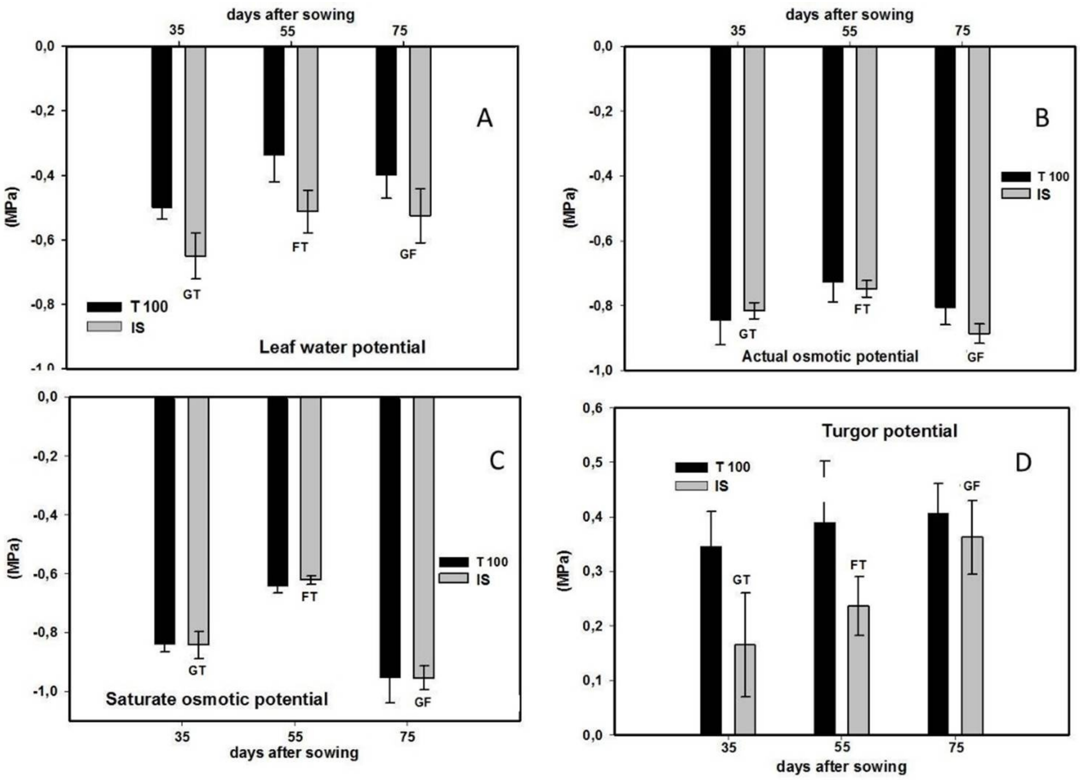
These results indicate that the application of water stress (60 % of the C.C.) in the growth stage (GT) affected plants negatively in both variables (Figure 2 A and B). On the other hand, it should be noted that leaf area was more sensitive to the suspension of irrigation than aerial dry mass. This behavior is in correspondence with that reported by (8) when studying different irrigation methods and levels in two stages of maize plant development.

In the results of the evaluations of Ψf (Figure 3 A) and its components Ψs (B), Ψ100s (C) and Ψt (d) it was found that in the case of Ψf, suspension of irrigation (IS) in GT plants was negatively affected and slightly Ψt, in the case of Ψf with a reduction of 23 %, in relation to control plants (T100). In plants corresponding to FT, only Ψf decreased by 35 % relative to those of T100. However, in the plants with irrigation suspension at the grain filling stage (GF), there were no differences with those of the control, indicating that it was the stage less sensitive to moderate stress. It is noteworthy that in the case of the Ψ100 s in none of the plants of CDI treatments under study was there evidence of the occurrence of the osmotic adjustment process, since in no case was there any difference in the Ψ100 s between the stressed and control plants.



GT= suspension of irrigation at growth stage, FT= suspension of irrigation at flowering stage and GF= suspension of irrigation at grain filling stage. Bars above the mean values represent the confidence interval of the means, α= 0.05

Figure 2. Effect of irrigation suspension (IS) at different stages of maize plant development on leaf area (A) and aerial dry mass (B)



GT= suspension of irrigation at growth stage, FT= suspension of irrigation at flowering stage and GF= suspension of irrigation at grain filling stage. Bars above the mean values represent the confidence interval of the means, α= 0.05

Figure 3. Effect of suspension of irrigation (IS) at different stages of maize plant development on Ψf (A), Ψs (B), Ψ100 s (C) and Ψt (D)

Evidently, the decrease in foliar water status in plants in the growth (GT) and flowering (FT) stages compared to T100 is a characteristic symptom of the effect of water stress in maize and the fact that in GF these differences were not manifested with respect to T100, indicates that this stage is less sensitive to water stress, results that corroborate those reported (14,15) working with different maize hybrids under water stress conditions. On the other hand, although the existence of the osmotic adjustment process in stressed plants was not effectively demonstrated, it is noteworthy, that the non-occurrence of differences in Ψt between GF and T100 plants, indicates the occurrence of some adjustment that allowed maintaining similar levels of turgor in plants of both treatments.

Table 2 shows the values of total soluble protein, chlorophyll and carotene content of plants affected or not by the suspension of irrigation at three stages of their development, where it was observed that the total soluble protein content was always higher in plants corresponding to the treatments with suspension of irrigation, which shows once again that adequate water stress favors plant quality.

Table 2. Effect of irrigation suspension at three stages of maize plant development on total soluble protein, chlorophyll and carotene content

Variables-Treatments	Proteins Soluble Totals (µg mL^{-1})	Chlorophyll A (µg mL^{-1})	Chlorophyll B A (µg mL^{-1})	Chlorophyll A+B (µg mL^{-1})	Carotenes (µg mL^{-1})
T 100	0.2570	10.163	2.546	12.793	4.900
GT	0.3130	14.774	3.717	18.491	6.285
E s. X.	0.0035 *	0.345*	0.0851*	0.450*	0.983ns
T 100	0.2580	12.861	3.496	16.357	2.574
FT	0.2953	11.909	2.941	14.851	2.473
E s. X.	0.0041*	0.2802 ns	0.0871*	0.066*	0.030ns
T 100	0.2535	11.735	3.258	14.993	2.501
GF	0.3015	9.843	2.425	12.268	2.274
Se. X.	0.0049*	0.139*	0.107*	0.3014*	0.032*

(*) means that there are statistically significant differences

Similarly, the carotene content did not differ between the values presented by the plants corresponding to GT and FT, with respect to those of T100. These results are fundamentally associated with the fact that, in general, water stress leads to an increase in carbohydrate content, which is of great importance in preventing cellular dehydration and maintaining turgor, protecting the cell membrane and preventing the destruction of proteins (16). They also constitute a supplementary source of energy.

In the case of chlorophylls A, B and A+B, when irrigation was suspended during the growth stage (GT), the highest values corresponded precisely to the stressed plants. When irrigation was suspended at FT, there were no differences in the chlorophyll A content of the plants with respect to those at T100, while the B and A+B content decreased slightly. However, in the case of plants corresponding to GF, a considerable decrease in chlorophylls (A, B and A+B) was found with respect to those of the T100 treatment. This behavior is fundamentally associated with the fact that plants of the FT and GF treatments received less water applied by irrigation, indicating that the moderate water stress applied to the maize plants in the flowering and grain filling stages led to a decrease in leaf area, associated with greater senescence (as observed in Figure 2A) and a significant degradation of chlorophyll, results that are in line (15).

CONCLUSIONS

- The suspension of irrigation in each of the phases studied decreased soil moisture by about 60 % of the field capacity, which was considered a moderate water stress for the crop.
- Leaf area proved to be more sensitive to soil water stress, mainly in the plants of the treatments with irrigation suspension in the growth (GT) and grain filling (GF) phases.
- The values of Ψf and its components Ψs and Ψt, as well as Ψ100 s, suggest that the most sensitive stages in plants to irrigation suspension with respect to these indicators of water status are the growth and flowering stages.
- Water stress favored leaf protein content and had practically no negative effect on carotene content.
- Moderate water stress applied to plants at the flowering (FT) and grain filling (GF) stages favored a greater degradation of chlorophyll pigment.

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