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ROOT SYSTEM OF MAIZE PLANTS CULTIVATED UNDER WATER DEFICIT CONDITIONS WITH APPLICATION OF CHITOSAN

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Abstract – Low water availability is regarded as an abiotic stress that limits the agricultural production. Due to the physical and chemical characteristics of chitosan (CHT), this substance might stimulate physiological responses on plants with respect to water deficit tolerance. In this sense, maize plants were submitted to water deficit and leaf application of chitosan (140 mg/L) during pre-flowering stage. Two maize hybrids, presenting different water deficit tolerance levels, were used: DKB 390 (tolerant) and BRS1010 (sensitive). Evaluations of the root system and production components were conducted. Maize plants submitted to leaf application of chitosan at the dose of 140 mg/L presented specific behavior in regard to hybrids. The tolerant type presented a more developed root system and an expressive agronomical yield when compared to the sensitive type. These results were probably obtained due to the stimulating effect of CHT on plant growth, enhancing the root system development and contributing to increase the availability and absorption of water and nutrients. Chitosan presents potential to reduce the harmful effect of water deficit on the root system, without negatively compromising the agronomical yield.

Keywords: Biopolymers, Water deficit, WinRhizo.

SISTEMA RADICULAR EM PLANTAS DE MILHO CULTIVADAS EM CONDIÇÃO DE DÉFICIT HÍDRICO SOB APLICAÇÃO DE QUITOSANA

Resumo - A baixa disponibilidade hídrica caracteriza-se como um estresse abiótico limitante para a produção agrícola. Em virtude das características físicas e químicas da quitosana (CHT), tal químico pode estimular respostas fisiológicas para tolerância ao déficit hídrico. Nesse sentido, as plantas de milho foram submetidas ao déficit hídrico e aplicação foliar de quitosana (140 mg/L) no pré florescimento, em híbridos de milho contrastantes em relação a tolerância ao déficit hídrico, tolerante (DKB 390) e sensível (BRS1010). Para tal foram realizadas avaliações do sistema radicular e componentes de produção. As plantas de milho submetidas a aplicação foliar de quitosana na dose de 140 mg/L apresentaram um comportamento específico, em relação aos híbridos, as plantas tolerantes, apresentaram um sistema radicular mais desenvolvido, um rendimento agronômico expressivo, quando comparado as plantas sensíveis. Provavelmente os resultados foram obtidos devido ao efeito estimulante da CHT no crescimento das plantas, promovendo o desenvolvimento do sistema radicular, contribuindo com um aumento na disponibilidade, absorção de água e nutrientes. A quitosana apresenta potencial para reduzir o efeito nocivo do estresse hídrico, sobre o sistema radicular, sem comprometer negativamente o rendimento agronômico.

Palavras-chave: Biopolimeros, estresse hídrico, WinRhizo.

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The low water availability is regarded as a limiting factor for maize crop, which reduces plant growth and productivity in the farms (Boyer et al., 2013). The reposition of soil water through irrigation, in the correct moment and amount, is the key to obtain maximization of production. Two days of water deficit during flowering period are sufficient to decrease grain yield in 20%, whereas four to eight days of water deficit result in a decrease of 50% (Magalhães et al., 2008).

In this sense, it is necessary to comprehend the mechanisms regarding plant adaptation to water deficit, as well as alternative ways to increase plant tolerance to water deficit. An important process of adaptation is the maintenance of root growth during periods when water availability is lower, which allows the plant to access water at deeper soil profile (Zhu et al., 2010). Root characteristics, especially related to length, superficial area and volume, are key features to improve crop adaptation under low water availability conditions. In this situation, it is necessary to have a greater knowledge of the strategies that aim at increasing the tolerance to water deficit, thus influencing the development of the root system without compromising the performance of the crops.

The usage of chemical antitranspirants that act as biostimulators is a strategy widely used nowadays (Katiyar et al., 2014). The chitosan (CHT) is considered a biostimulant substance, which induces physiological responses on plants to tolerate water deficits. Results from the past decades indicate that this biopolymer has the potential to be developed as an antitranspirant in situations of agricultural stress, inducing the tolerance to droughts by enhancing the defenses against oxidative stress, but without compromising the agricultural yield. As it can be noticed, the stimulating effect of the CHT on growth and development of crops, mainly the increase of water and nutrient availability and absorption, is due to osmotic cellular pressure (Lavinsky et al., 2015). This study intends to clarify the influence of this biopolymer on the root system of maize plants. The objective of this work was to evaluate the influences of application of chitosan on leaves and its effects on root system of contrasting maize plants under water deficit.

Material and Methods

Two contrasting maize hybrids regarding drought tolerance, DKB390 (tolerant) and BRS1010 (sensitive), were cultivated in a greenhouse at Embrapa Maize & Sorghum, located in the municipality of Sete Lagoas, Minas Gerais State (19º28’S, 44º15’08’’W, 732m altitude). Both maize hybrids were cultivated in plastic pots with 20 dm^3 of typical dystrophic Red-Yellow Latosol, with medium to moderate texture, from a plateau area.

The amount of water in the soil was daily monitored in the morning and afternoon (9am and 3pm), with the use of a Watermark moisture sensor (tensiometer), model 200SS

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– 5” (Irrometer Company, California – USA), installed in the center of each repetition under 20 cm depth. The sensors detect water tension in the soil, based on the electric resistance. They were coupled to digital indicators (Watermark meter) from the same company. Water reposition was performed based on the results obtained with the sensor and the water was replaced until field capacity (FC) during the period that preceded the imposition of treatments. These calculations were performed with the assistance of an electronic spreadsheet, made as a function of the curve of water retention in the soil.

Imposition of the treatments

Maize plants were submitted to water deficit before they reached the pre-flowering growth stage. This condition was imposed when the soil water potential reached approximately -138 kPa (Souza et al., 2014). The treatment types were: irrigated, water deficit and water deficit + application of CHT in the concentration of 140 mg/L.

The applied CHT concentration was previously determined in experimentation performed under greenhouse conditions, where it was possible to observe that this concentration was the one that provided more benefits to maize plants submitted to water deficit. The CHT concentration was prepared by using the solution according to Dzung et al. (2011), that is, by diluting the CHT in 100 mL of acetic acid at 0.5% during a 12h period. This was necessary due to the chemical and physical characteristics of the CHT and because it is a polymer that is insoluble in water. The solubility can be reached when there is a pH adjustment by using acetic acid dilution (Hadwirger, 2013). After that, the solution was diluted to the correspondent concentration. In order to impose the treatments, a costal sprayer pressurized by CO_2 (2.15 kgf cm^2) was used, coupled with a XR Teejet 110.02 VS nozzle, applying an equivalent to 120 l ha^{-1} of spray solution.

All measures were taken to prevent the CHT solutions from reaching neighbor plants. The treatments were applied on the first day after the imposition of water deficit, which had duration of 15 days. After that, the water supply was reestablished and maintained to field capacity.

Evaluation of root system

The WinRhizo Pro, Regente Inc. Instr., Canadá system was used to measure the length, superficial area and volume of the roots according to diameter classes as follow: very thin roots (Ø lower than 0,5 mm), thin roots (> 0,5 Ø < 2,0 mm) and thick roots (Ø > 2,0 mm) (Magalhães et al. 2011). In order to perform these analyses, roots were sampled from four repetitions in each treatment.

Production variables

At harvesting season, plant height was

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measured with the use of a graduated ruler. In addition to that, the following characteristics were evaluated: ear weight, ear length and diameter, number of grains per row, number of grains per ear, harvest index (grain dry mass / plant dry mass + grain dry mass) x100, and biomass of the canopy parts (Durães et al., 2008). The total dry biomass was estimated by drying the plants in a forced air circulation oven at 70ºC during 72h.

Data analysis

The mean values and standard error (SE) were calculated for all the parameters analyzed. Analysis of Variance (ANOVA) was used, in addition to Scott-Knott test with significance of 0.05% ($P \leq 0.05$) (Scott & Knott, 1974), with the use of Sisvar software version 4.3 (Ferreira, 2011).

Results and Discussion

The results showed that the application of CHT in the concentration of 140 mg/L resulted in specific response regarding hybrids. The drought sensitive hybrid BRS1010 presented lower total root length, and length of the very thin, thin and thick roots, when compared to the drought tolerant DKB390 hybrid. In fact, this was true only for the treatments submitted to water deficit plus CHT application. It was possible to notice that the drought tolerant hybrid DKB390 had a significant increase in root length with CHT application (Figure 1).

Similar responses could be observed regarding other variables of the root system (Figures 2 and 3). The presence of CHT resulted in an increase of the total superficial root area, superficial area of the very thin and thin roots in maize plants that are tolerant to water deficit (DKB 390) (Figure 2).

DKB390 plants performed similarly to the other variables with respect to total root volume, volume of the very thin, thin and thick roots, especially when cultivated under water deficit and with CHT application.

Therefore, CHT application on leaves produced a higher root volume when compared to the other treatments and to the sensitive hybrid BRS1010 under water deficit (Figure 3).

Under water deficit, a well-developed root system can absorb more water to maintain the moisture stable. CHT application on leaves might reduce the inhibition of the roots by promoting a development of the root system and ensuring that they absorb water, thus enhancing the tolerance to drought periods. This corroborates with the results observed for wheat plants in a research carried out by Zeng & Luo, 2012.

The size, properties and distribution of the root system determine the access of the plants to water, then establishing tolerance limits concerning water deficit condition (Lavinsky et al., 2015). The root system of maize plants is composed by thick and thin roots, where the thick roots act like an anchorage system and typically establish the general architecture of the root system by controlling the depth and capacity of the plants to grow towards deeper layers of

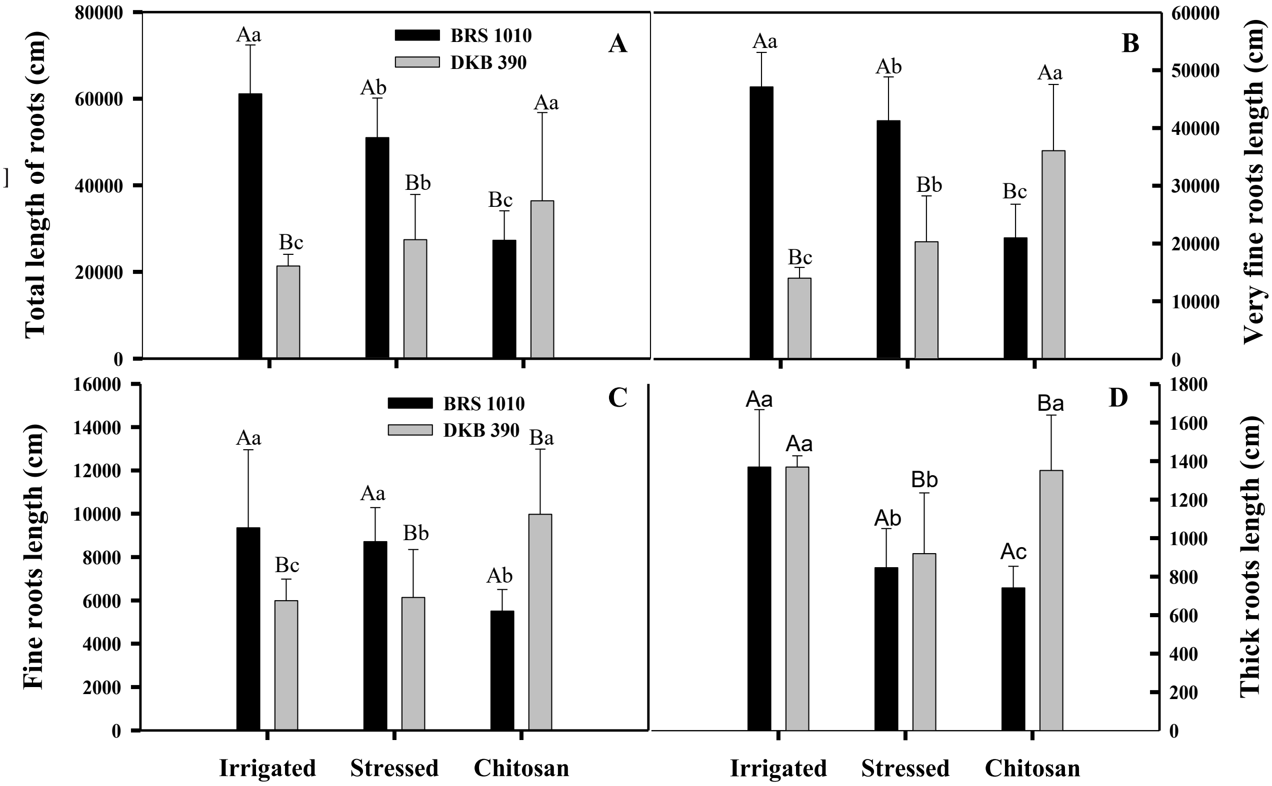

Figure 1. A: Total size of the roots (cm); B: length of the very thin roots (cm); C: length of the thin roots (cm); D: length of the thick roots (cm) submitted to different treatments. The mean values followed by the same letters are not statistically different. Uppercase letters refers to comparisons between hybrids, while lowercase letters refers to comparisons between the same genotype according to Scott-Knott test with 5% significance ($P \leq 0,05$).

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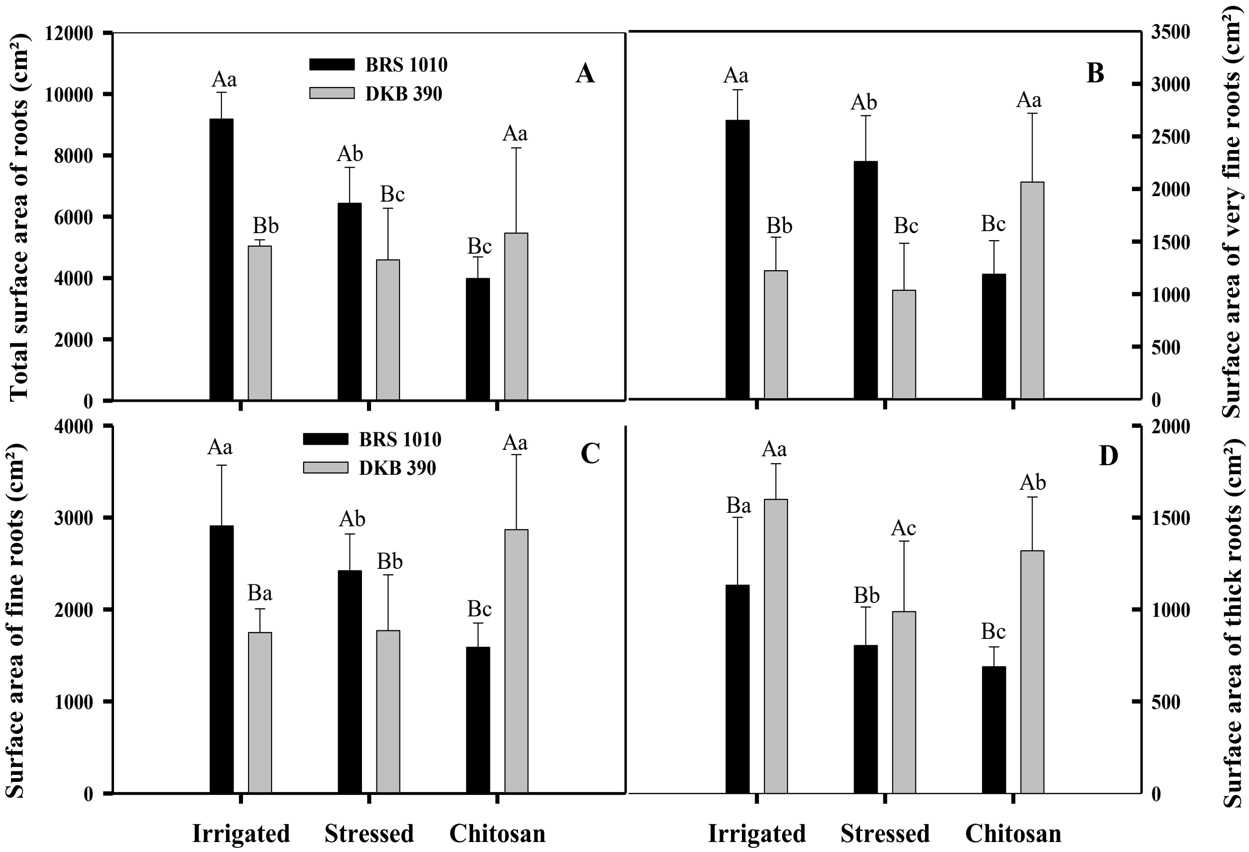

Figure 2. A: Total superficial area of the roots (cm²); B: Superficial area of the very thin roots (cm²); C: Superficial area of the thin roots (cm²); D: Superficial area of the thick roots (cm²) submitted to different treatments. The mean values followed by the same letters are not statistically different. Uppercase letters refers to comparisons between hybrids, while lowercase letters refers to comparisons between the same genotype according to Scott-Knott test with 5% significance ($P \leq 0{,}05$).

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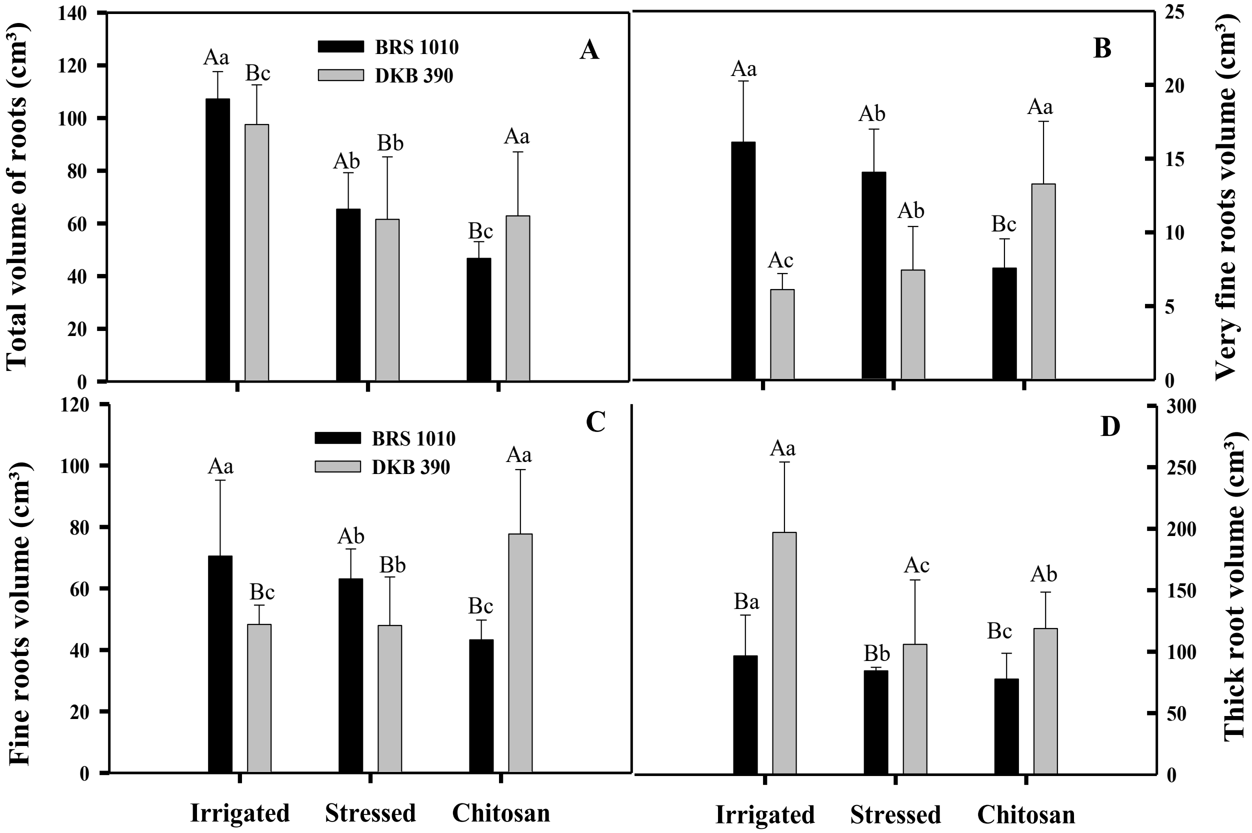

Figure 3. A: Total volume of the roots (cm³), B: volume of the very thin roots (cm³) , C: Volume of the thin roots (cm³) D: Volume of the thick roots (cm³) submitted to different treatments. The mean values followed by the same letters are not statistically different. Uppercase letters refers to comparisons between hybrids, while lowercase letters refers to comparisons between the same genotype according to Scott-Knott test with 5% significance ($P \leq 0,05$).

the soil (Henry et al., 2012). Thin roots are the active part of the root system in the sense that they effectively absorb water and comprehend the major part of length and surface area of those systems (Comas et al., 2013). It can be noticed that the application of CHT on leaves significantly influenced all the variables of the root system analyzed in this study. This can justify the CHT capacity to promote a higher water absorption and maintenance of the physiological metabolism during unfavorable growth conditions.

According to the functional equilibrium theory, plants increase the allocation of biomass to the shoot parts if the carbon gain is affected by limited resources, like sun light and CO_2. In the presence of low levels of underground resources, such as, water and nutrients, plants tend to enhance their biomass allocation to the roots (Brouwer, 1962; Pooter & Nagel, 2000). These alterations on the root system structure aim at providing an increase in the root growth angle, becoming an important strategy to prevent damages caused by water deficit. This enables the roots to reach deeper layers in soil exploration, thus generating great improvement on production and quality of maize grains (Lavinsky et al., 2015).

Those relations justify the results found in this study, where it can be observed that the sensitive hybrid (BRS1010) presented a larger root system in conditions without CHT application, smaller biomass shoot parts and a non-significant grain production (Table 1). The drought tolerant plants (DKB390), under CHT application, presented a more developed root system when compared to the other treatments. The results found for the drought tolerant type (DKB390) confirm a higher tolerance when compared to the sensitive hybrid (BRS1010). It can be noted that the maize plants cultivated under water deficit and without CHT application did not result in an expressive grain production. It was demonstrated, through the analyzed parameters, that the maize plants cultivated with application of CHT at 140 mg/L and under irrigated treatments presented similar responses.

These results were probably obtained due to the stimulating CHT effect on plant growth, where the root system was able to contribute with the increase of availability and absorption of water and essential nutrients, by adjusting the cellular osmotic pressure. The decrease in accumulation of free radicals by increasing the activity of antioxidant enzymes (Pirbalouti et al., 2017, Pichyangkura and Chadchawan, 2015), provides a balance between allocation of photoassimilates to shoot and root parts, without decreasing the agronomical yield.

Conclusions

The application of chitosan on the leaves, at a 140 mg/L dose, presents beneficial effects on the development of the root system of maize plants under water restriction conditions, without negatively affecting the agricultural performance of the crop.

The differences observed between hybrids are related mainly to their genetic characteristics,

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Table 1. Characteristics of production in corn hybrids contrasting towards drought tolerance cultivated under different treatments.

Parameters	Tolerant DKB 390			Sensitive BRS 1010		
	Irrigated	Chitosan	Stressed	Irrigated	Chitosan	Stressed
PH	2,47 Aa	2,46 Aa	2,02 Ab	2,18 Ba	2,19 Ba	1,88 Bb
ED	49,10 Aa	49 Aa	35 Ab	43,31 Ba	43,1 Ba	*
NGR	26 Aa	26,8 Aa	10,8 Ab	17,2 Ba	16,9 Ba	*
NGE	15,80 Aa	15,8 Aa	8,4 Ab	13,2 Ba	13,1 Ba	*
TDB	175,84 Aa	173,9 Aa	91 Ab	129 Ba	128,1 Ba	37,5
TGB	61,89 Aa	61,11 Aa	28,2 Ab	25,3 Ba	25 Ba	*
HI	35% Aa	35% Aa	30% Ab	31 % Ba	31 Bb	*

Drought tolerant hybrid (DKB390), drought sensitive hybrid (BRS1010) PH: Plant Height (m); ED: Ear Diameter (mm); NGR: Number of Grains per Row; NRE: Number of Rows per Ear; TDB: Total Dry Biomass (g); TGB: Total Grains Biomass (g); HI: Harvest Index ($g.g^{-1}$). * There was no production.
The mean values followed by the same letters are not statistically different. Uppercase letters refers to comparisons between genotypes, while lowercase letters refers to comparisons between the same genotype according to Scott-Knott test with 5% significance ($P \leq 0,05$).

which allow them to be characterized as tolerant and sensitive to the water deficit condition.

References

Boyer, J.S.; Byrne, P.; CASSMAN, K.G.; COOPER, M.; DELMER, D.; GREENE, T. GRUIS, F. The U.S, 2013. Drought of 2012 in perspective: a call to action. **Global Food Security**, Japão, v.2, n.3, p. 139-143. DOI: 10.1016/j.gfs.2013.08.002.

BROUWER, R. Distribution of dry matter in the plant. **Journal of Agriculture Science**, Netherlands, v.10, p.399-408, 1962.

COMAS, L.H.; BECKER, S.R.; CRUZ, V.M.R.; BYRNE, P.F.; DIERIG, D. Root traist contributing to plant productivity under drought. **Frontier of Plant Science,** v.4, p.442, 2013. DOI: 10.3389/fpls.2013.00442.

DURÃES, F.O.M.; MAGALHÃES, P.C.; OLIVEIRA, A.C. Índice de colheita genético e as possibilidades da genética fisiológica para melhoramento do rendimento do milho. **Revista Brasileira de Milho e Sorgo**, Sete Lagoas, v.1, p.33-40, 2008. DOI: 10.18512/1980-6477/rbms.v1n1p33-40.

DZUNG, N. A.; KHAN, T. P.; DZUNG, T. T. Research on impact of chitosan oligomers on biophysical characteristics, growth, development and drought resistance of coffee. **Carbohydrate Polymers**, v. 84, p. 751–755, 2011. DOI: 10.1016/j.carbpol.2010.07.066.

FERREIRA, D. F. Sisvar: a computer statistical analysis system. **Ciência e Agrotecnologia**, Lavras, v.35, n.6, p. 1039-1042, 2011. DOI: 10.1590/S1413-70542011000600001.

HADWIGER, L. A. Multiple effects of chitosan on plant systems: Solid Science or hype. **Plant Science,** 208, 42-49, 2013. DOI: 10.1016/j.plantsci.2013.03.007.

HENRY, A.; CAL, A.F.; BATOTO, T.C.; TORRES, R.O.; SERRAJ, R. Root attibutes affecting water uptake of rice (Oryza sativa) under drought. **Journal of Experimental Botanic**, Lancaster, v.63, n.13, p.4751-63, 2012. DOI: 10.1093/jxb/ers150.

KATIYAR, D., HEMANTARANJAN, A., SINGH, B. Chitosan as a promising natural compound to enhance potential physiological responses in plant: a review. **Indian Journal of Plant Physiology**, India, v. 20, p. 1-9, 2014. DOI: 10.1007/s40502-015-0139-6.

LAVINSKY, A.O.; MAGALHÃES, P.C.; ÁVILA, R.G.; DINIZ, M.M.; SOUZA, T.C. Partitioning between primary and secondary metabolism of carbon allocated to roots in four maize genotypes under water deficit and its effects on productivity. **The Crop Journal**, China, v.3, p.379-386, 2015. DOI: 10.1016/j.cj.2015.04.008.

MAGALHÃES, P.C.; SOUZA, T.C.; CANTÃO, F.R. Early evaluation of root morphology of maize genotypes under phosphorus deficiency. **Plant Cell Enverinomental**, v.57, p.135-138, 2011. DOI: 10.17221/360/2010-PSE.

MAGALHÃES, P. C.; DURÃES, F. O. M. Ecofisiologia. In: CRUZ, J. C. (Ed.). **Cultivo do milho**. 4. ed. Sete Lagoas: Embrapa Milho e Sorgo, 2008. p. 64-87. (Embrapa Milho e Sorgo. Sistema de Produção, 1).

PICHYANGKURA, R.; CHADCHAWAN, S. Biostimulant activity of chitosan in horticulture. **Scientia Horticulturae**, Georgia, v.196, p.49-65, 2015. DOI: 10.1016/j.scienta.2015.09.031.

PIRBALOUTI, A.G.; MALEKPOOR, F.; SALIMI, A.; GOLPARVAR, A. Exogenous application of chitosan on biochemical and physiological characteristics, phenolic contente and antioxidante activity of two species of basil (*Ocimum ciliatum* and *Ocimum basilicum)* under reduced irrigation. **Scientia horticulturae,** v, 201, p.114-122, 2017. DOI: 10.1016/j.scienta.2017.01.031.

POOTER, H.; NAGEL, O. The role of biomass allocation in the growth response of plants to differente levels of light, CO_2, nutrients and water: a quantitative review. **Australian Journal of Plant Physiology**, Rockville, v.27, n.12, p.1191-1191, 2000. DOI: 10.1071/PP99173_CO.

SCOTT, A. J., KNOTT, M. A. Cluster analysis method for grouping means in the analysis of variance. **Biometrics**, Arlington, v.30, n.3, 507-512, 1974. DOI: 10.2307/2529204.

SOUZA, T. C.;MAGALHÃES, P. C.;CASTRO, E. M.; CARNEIRO, N. P.; PADILHA, F. A. ABA application to maize hybrids contrasting for drought tolerance: changes in water parameters and in antioxidant enzyme activity. **Plant Growth Regulation,** China v.73, p.205-217, 2014. DOI: 10.1007/s10725-013-9881-9.

ZENG, D.; LUO,X. Physiological Effects of Chitosan Coating on Wheat Growth and Activities of Protective Enzyme with Drought Tolerance. **Journal of Soil Science**, Hubei Province, v.2, p.282-288, 2012. DOI: 10.4236/ojss.2012.23034.

ZHU, J.; BROWN, K.M.; LYNCH, P.J. Root cortical aerenchyma improves the drought tolerance of maize (*Zea mays* L.). **Plant Cell Envirinomental**, v.33, p.740-749, 2010. DOI: 10.1111/j.1365-3040.2009.02099.x.