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9. ABSTRACT

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LEAF WATER POTENTIAL AND STOMATAL ACTIVITY IN SORGHUM AS INFLUENCED BY SOIL MOISTURE STRESS*

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ABSTRACT

Five sorghum (*Sorghum bicolor* Moench) varieties were grown in soil in a growth chamber and submitted to two consecutive soil drying cycles separated by a 3-day recovery period. During each drying cycle measurements were made on stomatal pore width, leaf diffusion resistance (R_l), leaf water potential (ψ_l) and soil water potential (ψ_s). In previously stressed plants stomata were less sensitive to reduction in ψ_s, and ψ_l was lower at a given ψ_s, as compared to non-previously stressed plants. In non-previously stressed plants stomata responded sensitively to an initial reduction in ψ_l, but once closure was initiated (at ψ_l of about -8 bars) stomata remained partly closed ($^1/_4$-$^1/_3$ of maximum aperture) even when ψ_l dropped to -19 bars. During this phase control over transpiration was apparently non-stomatal.

Sorghum (*Sorghum bicolor* Moench) is considered a drought resistant plant, but information is lacking on the control over transpiration under water stress in this plant.

Sorghum stomatal activity was investigated by Glover (1959) using an air flow porometer. He found sorghum stomatal activity was impaired by a period of drought, but activity was restored quicker than in maize. Ehrler and Van Bavel (1967) observed an increased day time leaf diffusion resistance in sorghum (to 8 s cm^{-1}) when soil moisture was limited. Pallas and Bertrand (1966) using percent open stomata as an index to stomatal activity found that stomata of several sorghum varieties began to close at a soil moisture tension of -0.3 bars and closed completely at -6 to -7 bars. Stomatal response to soil moisture tension was linear. By using an air flow porometer it was found (Sullivan, Biggs, Eastin, Clegg and Maranville, 1968) that sorghum stomata closed at a leaf water potential of about -15 bars. In research done with sorghum and corn it was found, by using a diffusion porometer (Sanchez-Diaz and Kramer, 1971), that in sorghum an abrupt stomatal closure occurred at a leaf water potential of -8.5 to -8.8 bars

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It was also found that for the same leaf water potential, stressed sorghum maintained a smaller water saturation deficit than stressed corn.

The research described below does not attempt to investigate the nature of drought resistance of sorghum but rather to obtain further information on leaf water status and stomatal activity in response to soil moisture stress. Since sorghum is grown also under conditions where alternate soil drying and wetting occurs, this experiment included two consecutive soil drying cycles.

MATERIALS AND METHODS

Five genotypes of different sorghum varieties were used: RS-610 hybrid, H-610 (a similar hybrid to RS-610 from seed produced in Israel), M-35-1 (an Indian variety found to have high heat tolerance (Sullivan and Blum, 1971)), 1136-1 and 1136-3. The two latter varieties were F_6 lines from a T - 7078 × M-35-1 cross.

The experiments were carried out in a growth chamber, 20/25 °C night/day temperatures, 50-60% relative humidity, 0.55 ly/min radiant energy and a 12 h day. Plants were grown from seed in metal flats containing 1/3 soil, 1/3 sand and 1/3 peat moss, a mixture hereafter referred to as "soil". The experimental design was a randomized block with three replications. Each block (replication) consisted of one flat. Within each flat each of the varieties was planted in one row, thinned to 7-8 plants. Flats were irrigated daily with complete nutrient solution and tap water alternately. Plant growth and appearance was normal.

Thirty days after emergence irrigation was stopped and the first drying cycle commenced. Four days later irrigation was resumed and continued for three days before the next drying cycle.

Lights switched on at 7.00 am and during each drying cycle measurements were made daily at 10.00 am and 3.00 pm. Measurements in plants were made on fully expanded and illuminated top leaves. *Leaf diffusion resistance* was measured on the upper leaf surface with a horizontal sensor (Kanemasu, Thurtell and Tanner, 1969). On each sampling date four measurements were made in each variety/replication, or a total of 12 measurements per variety/date. *Stomatal pore width* was determined microscopically in upper epidermal prints made with Acta-seal plastic spray (Brown and Rosenberg, 1970). Results thus obtained compared well with silicon rubber prints especially where print films were taken at the edges of the sprayed spot, where the spray was very thin. Prints were made on each leaf measured for leaf diffusion resistance. Five random microscopic fields were scanned in each print, and about 5-10 stomates per field were measured. Visual detection of small stomatal apertures is quite difficult in many plant species. However, with appreciable experience and continuous manipulation of the depth of field, small stomatal apertures could be delineated in sorghum and, at times, completely closed stomata could be distinguished. It is stressed that stomatal

pore width measurement is not a routine technical chore that can be readily assigned to a technician. It requires a great deal of experience in evaluating a good print and in the determination of the actual pore. Leaf samples (excluding midrib) were taken with a cork borer for the measurement of *leaf water potential* with a Spanner type thermo-couple psychrometer (nine samples per variety/date). Three soil samples were taken from each flat on each date for measurement of *soil water potential*. Each sample was taken with a cork borer, mixed rapidly and two subsamples put into the psychrometers.

RESULTS

Results are presented as arithmetic means for the five varieties. Leaf water potential (Ψ_l) decreased (became more negative) with the decrease in soil water potential (Ψ_s) (Fig. 1). Sorghum leaves maintained a Ψ_l of about -5 bars at the highest Ψ_s, indicating a rather high evaporative demand in this experiment. As Ψ_s decreased to below approximately -7 bars, Ψ_l of 1st cycle plants was lower (more negative) at a given Ψ_s than that of 2nd cycle plants.

Maximum mean aperture (width) obtained in sorghum leaf stomata in well-watered plants prior to the onset of drought cycle was about 3 μ (Fig. 2). Individual samples reached about 4 μ. An increase in radiant energy might have further increased aperture. In first cycle plants stomata closed abruptly at a rather defined water stress, at a Ψ_s of about -3 bars and Ψ_l of about -8 bars. However, complete closure was not observed even when Ψ_l dropped to -19 bars. In the second cycle plants stomata remained nearly closed at an aperture of about 1 μ, throughout the Ψ_l range of -5 to -19 bars.

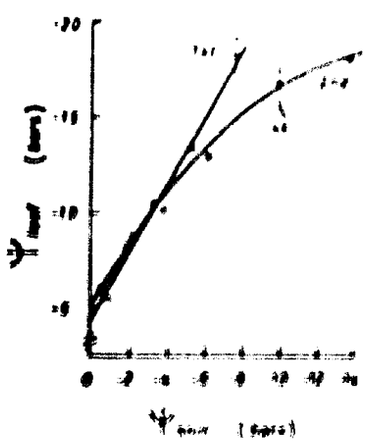

Fig. 1. Relations between Ψ_s and Ψ_l in sorghum leaves during the first (○) and second (●) soil drying cycles (S.E. = Standard Error)

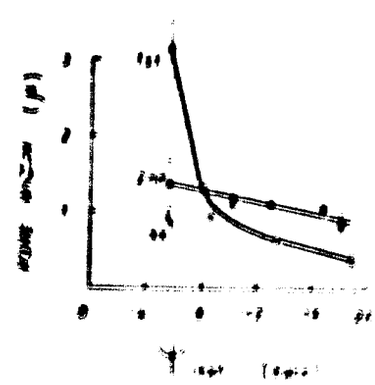

Fig. 2. Relations between Ψ_s and stomatal pore width in sorghum leaves during the first and second soil drying cycles (S.E. = Standard Error)

Although stomatal aperture, in relation to Ψ_l, greatly differed between first and second cycle plants, no differences in this respect were found in R_l (compare Fig. 2 with Fig. 3). Furthermore, in second cycle plants, in spite of practically unchanged stomatal aperture in response to Ψ_l (Fig. 2), R_l still increased steadily with decreased Ψ_l. Irrespective of stomatal aperture or drying cycle, R_l was related almost linearily to Ψ_l.

In first cycle plants (Fig. 4) stomatal closure to approximately $^1/_4$–$^1/_3$ of maximum aperture had only a small effect on R_l. Further slight closure appeared to be associated with a very marked increase in R_l. In second cycle plants, while stomata remained at an aperture of about 1 μ, R_l still increased from about 6 to 25 s cm^{-1}. It can therefore be seen in plants subjected to the second drying cycle that while stomatal pore width remained at about 1 μ during the decrease in Ψ_s and Ψ_l, control over transpiration (R_l) was still in evidence. Varieties did not exhibit significant differences in most measurements taken.

DISCUSSION

Sorghum plants were subjected to two consecutive soil drying cycles with three days recovery period in between. Two days recovery period was found to be sufficient for full recovery of sorghum (Sanchez-Diaz and Kramer, 1971).

The water relations of sorghum plants differed greatly between the two drying cycles. A major effect of the first drying cycle on plant response to stress during the second drying cycle was a markedly reduced response of stomatal aperture to change in leaf water stress. This confirms previous results in sorghum (Glover, 1959) and other crops (Fischer, Hsiao and Hagan, 1970). A second effect concerned the relations between Ψ_s and Ψ_l. As Ψ_s decreased, plants in the second cycle maintained a higher (less negative) Ψ_l than in the first drying cycle. A

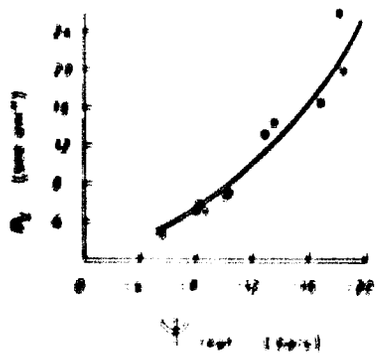

Fig. 3. Relations between Ψ_s and R_l in sorghum leaves during the first and second drying cycle.

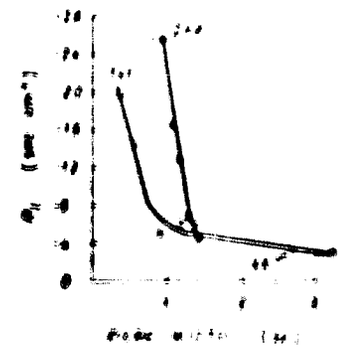

Fig. 4. Relations between R_l and stomatal pore width in sorghum leaves during the first and second drying cycle (S.E. = Standard Error).

previous drought may, therefore, apparently reduce the soil-to-leaf water potential gradient developed along the recovered plant, and thus imply (Slatyer, 1967) increased adaptation to soil moisture stress, or a hardening effect.

Sorghum stomata responded sensitively to an initial decrease in Ψ_l (and Ψ_s). In non-previously stressed plants stomata closure occurred at a very narrow Ψ_l range, as found also in other plants (Ehlig and Gardner, 1964; Kanemasu and Tanner, 1969). In sorghum the abrupt stomatal aperture closure occurred at Ψ_l of about −8 bars. This is in good agreement with observations by Sanchez-Diaz and Kramer (1971) who found a sharp decrease in leaf diffusion resistance at a Ψ_l of −8.5 to −8.8 bars.

However, interpretation of results regarding stomatal activity should take into account the method involved – whether visual measurements are made directly on the stomata or an indirect method such as the diffusion porometer is employed. Taking visual aperture measurements as a measure of apparent stomatal activity, it was indeed observed that the abrupt stomatal closure at Ψ_l of −8 bars occurred concurrently with an abrupt increase in R_l. However, below Ψ_l of −8 bars, while stomata pore width remained at about 1 μ, R_l still increased appreciably with the continued decrease in Ψ_s and Ψ_l. This was most outstanding in previously stressed plants, where R_l changed from about 7 to 25 s cm^{-1} (Fig. 4) while stomatal aperture hardly changed. This phenomenon is indicated also from Sullivan's measurements (Sullivan et al., 1968) with a diffusion porometer vs. air flow porometer. When leaf porosity was reduced from 2.5 to 0.5 cm^{-3} cm^{-2} s^{-1}, diffusion resistance changed very little. When porosity was further reduced by increased stress from 0.5 only 0.2 cm^{-3} cm^{-2} s^{-1}, diffusion resistance changed appreciably from about 5 to 21 s cm^{-1}.

It appears, therefore, that in sorghum under imposed soil moisture stress, initiation of stomatal closure occurs at Ψ_l of about −8 bars, and then stomata remain partly open at $^1/_4$–$^1/_3$ of maximum aperture in spite of further reduction in Ψ_l, even down to −19 bars. This is supported by Sullivan's data (Sullivan et al., 1968) and also evidenced from Carlson's data (1969) where (under relatively low light intensity) CO_2 fixation by intact sorghum leaves was not reduced until Ψ_l dropped to −19.6 bars. This is not quite in agreement with Sanchez-Diaz and Kramer (1971), who concluded, by observation made with a diffusion porometer, that sorghum stomates fully close at −16 bars.

Changes in R_l (especially in previously stressed plants) occurring during the phase when stomata remain open to only $^1/_4$–$^1/_3$ of their maximum aperture, apparently result from non stomatal resistances (Shimshi, 1963; Ehlig and Gardner, 1964; Jarvis and Slatyer, 1970).

It is possible, though, that changes in R_l at very small stomatal apertures resulted from changes in aperture that were below the visual resolution of the method employed. However, it is pointed out that although 1st and 2nd cycle

plants differed in the relations between stomatal aperture and Ψ_l (Fig. 2), the relation between R_l and Ψ_l was similar in both cycles (Fig. 3). This would support the conclusion that in sorghum control over transpiration below the threshold of Ψ_l of about -8 bars may be non-stomatal.

REFERENCES

BROWN, K.W. AND N.J. ROSENBERG. 1970. Influence of leaf age, illumination, and upper and lower surfaces differences on stomatal resistance of sugar beet (*Beta vulgaris*) leaves. Agron. J. 62: 20–24.

CARLSON, L.L. 1969. The effect of water stress on CO_2 exchange and fixation in sorghum. M.Sc. thesis, University of Nebraska.

EHLIG, C.F. AND H.R. GARDNER. 1964. Relationship between transpiration and internal water relations of plants. Agron. J. 56: 127–130.

EHRLER, W.L. AND C.H.M. VAN BAVEL. 1967. Sorghum foliar response to changes in soil water content. Agron. J. 59: 243–246.

FISCHER, R.A., T.C. HSIAO AND R.M. HAGAN. 1970. After effect of water stress on stomatal opening potential. I. Techniques and magnitudes. J. Exp. Bot. 21: 371–385.

GLOVER, J. 1959. The apparent behaviour of maize and sorghum stomata during and after drought. J. Agric. Sci. 53: 412–416.

JARVIS, P.G. AND R.O. SLATYER. 1970. The role of the mesophyll cell wall in leaf transpiration. Planta 90: 303–322.

KANEMASU, E.T. AND C.B. TANNER. 1969. Stomatal diffusion resistance of snap beans. I. Influence of leaf water potential. Plant Physiol. 44: 1547–1552.

KANEMASU, E.T., G.W. THURTELL AND C.B. TANNER. 1969. Design, calibration and field use of a stomatal diffusion porometer. Plant Physiol. 44: 881–885.

PALLAS, J.E. AND A.R. BERTRAND. 1966. Research in Plant Transpiration: 1963. Prod. Res. Rep. No. 89, ARS, USDA, Georgia Agric. Exp. Stn. and Meteorol. Dept. US Army Elect. Res. Dev. Activ.

SANCHEZ-DIAZ, MANUEL F. AND P.J. KRAMER. 1971. Behaviour of corn and sorghum under water stress and during recovery. Plant Physiol. 48: 613–616.

SHIMSHI, D. 1963. The effect of soil moisture and phenylmercuric acetate upon stomatal aperture transpiration and photosynthesis. Plant Physiol. 38: 713–721.

SLATYER, R.O. 1967. Plant-Water Relationships. Academic Press, London, New York.

SULLIVAN, C.Y., W.W. BIGGS, J.D. LASHS, M.D. CLEGG AND J. MARANVILLE. 1968. Heat and drought stress studies of sorghum, millet and corn. In: The Physiology of Yield and Management of Sorghum in Relation to Genetic Improvement. Ann. Rep. No. 2, University of Nebraska, The Rockefeller Foundation, CRD, ARS, USDA. pp. 14–31.

SULLIVAN, C.Y. AND A. BLUM. 1971. Drought and heat resistance of sorghum and corn. Proc. 25th Ann. Corn & Sorghum Res. Conf. pp. 55–56.