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LEAF EPIDERMIS ANATOMY OF RICE PLANTS (Oriza sativa L.) WITH DIFFERENT SALINITY TOLERANCE DEGREES

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INTRODUCTION

Although the basic anatomy of rice vegetative and reproductive organs have attracted the attention of many investigators (ANGLADETTE, 1968; LISA CLARK & HARRIS, 1981; TAKANE & HOSHIKAWA, 1993; SUZUKI et al., 1993), the works in this field continue to be of great interest, for they

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are related to the adaptative capacity of these plants to live under adverse conditions or different stresses.

Salinity is one of the most common stresses in rice (0. sativa) crop, due to the plant growing conditions, and because saline stress affects large areas at present. Accordingly, the works looking for better adapted varieties should be intensified, specially in Cuba, where research on rice anatomy is scarse, mainly when related to abiotic stress tolerance. The present study was conducted on the leaf epidermis of three rice varieties (0. sativa L.) with different salinity tolerance degrees, growing in a non-saline medium, have in view that leaf epidermis has several interesting agronomic implications, due to the influence of frequency and stomatal size on two important plant physiological processes: photosynthesis and transpiration.

MATERIAL AND METHODS

Rice seeds (Oriza sativa L.) from Amistad-82 (A-82), salt sensitive. Pokkali (Pok) and LP-10, salt tolerant varieties, were put in plastic trays with zeolite and tap water for germination under natural conditions, in September. The epidermis from the mid third of leaf number four was sampled 15 days after plant germination through a plastic trasparent gum; also a similar epidermis was prepared to scanning electron microscopy by means of conventional techniques for Gramineae leaves (Lab. of Ultrastruc ture of CIGB and CNIC).

Table 1 presents some plany growth indicators of the varieties under study. Those values are the means of 15 plants per variety, in order to know plant conditions at sampling time. Stomata were counted per unit area and their length was measured in the gum impressions of adaxial (ADA) and abaxial (ABA) surfaces. For each used to compare each variable, 100 measures were taken. The ${f t}$ test was used to compare each indicator per variety for the same surface, applying Mann Whitney's nonparametric alternative test

Table 1. Mea as ger	Table 1. Mean lengths of pseudostem, leaf sheet number 3 and 4 and the roots, as well as l st order root from three rice varieties 15 days after germination.	udostem, r root f	leaf sheet rom three ri	number 3 and 4 ce varieties 19	and the roots, 5 days after
		Length (cm)	(cm)		
Varieties	Pseudostem	Leaf 3	Leaf Number 3 4	Roots	First-order root number
A-82	4.95	10.20	5.23	3.27	4.25
Pokkoli	6.25	12.47	5.65	3.93	4.00

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4.33

6.67

5.70

12.00

5.20

LP-10

when data did not follow a normal distribution.

RESULTS AND DISCUSSION

Table 2 shows the stomatal length and number per unit of leaf area of the three varieties under study. Τt is appreciated that surface lengths of varieties at the mid leaf zone were different, higher in the ADA surface for LP-10 cv. and ABA for Pokkali. In a unique definite tendency of each variety, it was not directle related to saline stress tolerance. Meanwhile the behaviour of stomatal number per unit area was different and it was observed that this indicator was lower in salt tolerant varieties for ADA surface, which is an important feature to consider when evaluating the salt tolerant genetic material, since there are heritable anatomical features related to leaf tissues (SETHI & ROY, 1985) in rice.

It is interesting to emphasize stomatal density in rice plant leaves from Pokkali and LP-10 varieties in the ADA surface, because they are salinity tolerant; it was observed in different gramineae varying among species and varieties (McCREE & DAVIS, 1974; LIANG et al., 1975; RO-DELLA & ANDRADE, 1983-84, and RODELLA & DE MARINIS, 1990).

Other plant species as Prosopis cineraria and P. tamargo, SERRATO VALENTI et al. (1991 and 1992) showed that anatomical disorders in different seedling organs were the answer to salinity, and in relation to epidermis, it was observed that the number of water storing cells from leaf hypocotiles and in the cortical layers oncreased with salt concentration. The was also a reduction in the epidermal cell size, while by increasing salinity in leaves they tended to lose their dorsiventral organization and increase mesophyl thickness. These results give an idea of what happens in some species, although these same investigators agree to accept these contradictory changes in other spe~ cies submitted to treatments.

However, little is known about the anatomical characteristics favouring salt tolerance. Therefore, it is ne-

Table 2. St an (m Varieties	Stomatal length in the ADA and ABA leaf surfaces of and its amount per mm ² in rice plants 15 days after (mean value and its standard error). Stomatal length (µm) Stomatal number ADA ABA ABA ADA	he ADA and ABA 1 m ² in rice plant standard error). h (μm) ABA	eaf surfaces of le s 15 days after Stomatal number p ADA	af number 4 germination ber mm ² ABA
Amistad-82	28.44 ⁻ 0.46 b	23.04 0.34 b	435 - 11.53 a	448 - 13.92 a
Pokkali	24.10 [±] 0.45 c	26.95 <mark>+</mark> 0.40 a	247 ± 10.48 c	424 + 16.92 a
LP-10	31.42 [±] 0.58 a	21.17 ⁺ 0.36 c	397 ± 12.86 b	283 1 16.26 b
Means with	common letters do	not differ signi	Means with common letters do not differ significantly for p < 0.05.	.05.

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cessary to continue looking for theses characteristics in tolerant plants, when they are not under treatment.

On the other hand, what was observed in this work in relation to stomatal number per unit of leaf area would be an important indicator to consider in the salinity tolerance studies.

Noticeable differences were not observed between the epidermis of the same surface for different varieties: thus, in Figure 1 the epidermis of two leaf surfaces was observed (Pokkali A and A-82 B varieties) by means of scanning electronic microscopy. The generalcharacteristics were appreciated as TAKANE & KOSHIKAWA (1993) described. and it is observed in some other Gramineae a hoghly differenciated epidermis, with different types of cells and sto mata disposed in longitudinal lines along the surface (LEE & LOMMASSON, 1968). Also, it is appreciated the papillae of different sizes, shapes and number in different types of epidermal cells. In the ABA surface (Fig. 1B), longer very big papillae are observed in longer cells: these characteristics are different among varieties and they will be the object of subsequent studies.

In Figure 2 stomata from ABA surface of leaves of only LP-10 (A) and Pok (B) are shown since somewhat similar to leaf epidermis occurred, the papillae surrounding stomata in number of four are noticeable, two in each side of the pore with a regular longitudinal separation and a dense layer of epicuticular waxes at the papilla height that runs parallel to the stomatal pore; these waxes also cover the subsidiary and companion cells in a lower density. These waxes are related to plant conditions since they are hurt by different stresses, where these waxes are less around stomata (TAKANE & KOSHIKAWA, 1993).

Stomata of ADA surface have equal characteristics a lthough different sizes, according to what is showed in **Table 2**.

This study was conducted for the first time under our conditions with rice varieties investigated, in which it



Figure 1. Leaf epidermis of the ADA and ABA surfaces of two varieties under sudy. Preparation for scanning electron microscopy. Estomata, CS - epidermal cells, T-tricoma and D-surface depression, P-papillae, A-ADA surface of Pok and B-ABA surface of A-82. 5000 X.



Figure 2. Stomata of the ABA surface of the A) LP-10 and B) Pok. Preparation for scanning electron microscopy. C-wax epicuticular border. 5000 X.

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was observed the proper characteristics of this genus (Oriza); however, there are differences in some variables resulting interesting in relation to the behaviour of these plants under saline stress conditions; nevertheless, it is necessary to continue deepening on their anatomy and to prove the physiological implications of these characteristics for the development of these plants when submitted to saline stress.

SUMMARY

The study of leaf epidermis of three rice varieties with different salinity tolerance degrees was made in the National Institute of Agricultural Sciences (Cuba). Stomatal length and number per unit area were measured at the adaxial and abaxial epidermis after 15 days of plant germination; also, the morphological characteristics of leaf epidermis were observed by means of a scanning electron microscopy. Differential characteristics between the раpillae of both surfaces, and stomata amount and stomata per unit area were found between the susceptible and salinity tolerant varieties.

Key words: Leaf anatomy, epidermis, salinity, rice.

RESUMO

ANATOMIA DA EPIDERME FOLIAR DE ARROZ (Oriza sativa L.) COM DIFERENTES GRAUS DE TOLERÂNCIA À SALINIDADE

Realizou-se, no Instituto Nacional de Ciências Agrícolas (Cuba), o estudo da epiderme foliar de três variedades de arroz, com diferentes graus de tolerância à salinidade. Aos 15 dias após a germinação, foram determinados o comprimento e o número de estômatos por unidade de área, nas superfícies adaxial e abaxial, bem como observaram-se as características morfológicas da epiderme foliar em microscópio eletrônico de varredura. As variedades susceptíveis e as tolerantes à salinidade apresentaram características diferenciais, entre as papilas de ambas as superfí-

cies foliares e o número de estômatos por unidade de área.

Palavras-chave: Anatomia foliar, epiderme, arroz, salinida de.

BIBLIOGRAPHIC REFERENCES

ANGLADETTE, A., 1968. El Arroz. La Habana, Instituto del Libro. 876p.

- CLARK, LISA H. & W. HARRIS, 1981. Observations on the Root Anatomy of Rice (Oriza sativa L.). Amer. J. Bot., 68(2): 154-161.
- LEE, K.W. & R.C. LOMMASSON, 1968. The Epidermis of the Blades Joint of Sorghum bicolor L. Moench. Proc. Iowa Acad. Sci., 75: 55-64.
- LIANG, G.H.; A.D. DAYTON; C.C. CHU & A.J. CASADY, 1975. Heritability of Stomatal Density and Distribution on Leaves of Grain Sorghum. **Crop Sci.**, **15**(4): 567-570.
- McCREE, K.J. & S.D. DAVIS, 1974. Effect of Water Stress and Temperature on Leaf Size and on Size and Number of Epidermal Cells in Grain Sorghum. Crop Sci., 14 (5): 751-755.
- RODELLA, R.A. & V.M. de ANDRADE, 1983-84. Número e Tamanho dos Estômatos em Folhas de Quatro Cultivares de Sorghum bicolor (L.) Moench. Sep. Garcia de Orta, Ser. Bot. Lisboa, 6(1-2): 7-12.
- RODELLA, R.A. & G. DE MARINIS, 1990. Número de Estômatos em Folhas de Sorghum bícolor (L.) Moench Tratado com Herbicidas. Rev. Agricultura, Piracicaba, 65(1):63-73.
- SETHI, M. & J.K. ROY, 1985. Nature of Inheritance of Some Anatomical Characters in Rice. Indian J. Genet., 45(3): 525-530.
- SERRATO VALENTI, G.; M. FERRO; D. FERRARO & F. RIVEROS, 1991. Anatomical Changes in Prosopis tamargo Phil. Seed lings Growing at Different Levels of NaCl Salinity. Annals of Bot., 68: 47-53.
- SERRATO VALENTI, G.; L. MELONE; O. ORIS & F. RIVEROS, 1992. Anatomical Changes in Prosopis cineraria (L.) Druce Seedlings Growing at Different Levels of NaCl Salinity. Annals of Bot., 70: 399-404.

- SUZUKI, K.; H. MIYAKE; T. TANIGUCHI & E. MAEDA, 1993. Ul trastructural Studies on Rice Globular Embryos with Emphasis on Epidermis Initiation. Japanese J. of Crop Sci., 62(1): 116-125.
- TAKANE, M. & K. HOSHIKAWA, 1993. Science of the Rice Plant Morphology. Tokyo. 886. Ed. Food and Agric. Policy Research Center.

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