

OSMOTIC ADJUSTMENT IN SUNFLOWER (*Helianthus annuus* L.): DIFFERENCES BETWEEN GENOTYPES.**C.A. Chimenti and A.J. Hall.**

Depto. Ecología, Facultad de Agronomía (UBA), Av. San Martín 4453, 1417 Buenos Aires, Argentina.

SUMMARY

A collection of genotypes of different origin was exposed to water stress at three developmental stages: seedling (8 leaves), pre-anthesis (appearance of star) and post-anthesis (last anthesis). The changes in water status of these plants during a period of gradual drying out of the soil were defined in terms of leaf relative water content (RWC) and osmotic potential (π). The $\ln RWC/\ln \pi$ relationship (Morgan, 1983) was used to estimate the degree of osmotic adjustment. Degree of osmotic adjustment was expressed as the estimated value of RWC for a π of -1.7 MPa (RWC_g). Thus, a high value of RWC_g indicates a high degree of osmotic adjustment. The responses of 33 genotypes were examined in seedling stages, for these genotypes the relationship could be described by either a single or by two intersecting straight lines. Irrespective of the form of the response, all genotypes showed some degree of osmotic adjustment and significant differences ($P=0.05$) in RWC_g were shown to exist between some genotypes. In the pre and post-anthesis stages, seven genotypes were examined, all showed osmotic adjustment. Both forms of the $\ln RWC/\ln \pi$ relationship were found in genotypes tested in these stages. Significant ($P=0.05$) linear relationships were found between RWC_g measured in seedlings and that measured in later developmental stages (pre-anthesis/seedling $r=0.81$, post-anthesis/seedling $r=0.87$). We conclude that there are differences between sunflower genotypes in degree of osmotic adjustment and that it is possible to use measurements made on seedlings as a means of selection for osmotic adjustment in later developmental stages.

INTRODUCTION

The generation of low osmotic potential, is one of the mechanisms that can determine increased tolerance to water stress. Osmotic adjustment is the physiological process that arises from a net increase in solutes through uptake of ions (Munns *et al.*, 1983) or by the production of osmotically active solutes, for example sugar or proline (Jones *et al.*, 1980). Osmotic adjustment permits the partial or total maintenance of leaf turgor potential when the water potential of the tissue decreases and can reduce the effects of water stress on processes such as photosynthesis or leaf expansion (Turner and Jones, 1980). It has been shown in crops such as wheat (Morgan, 1983) and sorghum (Jones and Turner, 1978) that cultivars that have osmotic adjustment show a greater tolerance to water stress. This paper reports a study of intraspecific variability for the capacity for osmotic adjustment in sunflower. The hypotheses tested were: A) genotypes differ in their capacity for osmotic adjustment and B) there exists an association between the capacity for osmotic adjustment measured in seedlings and that manifested in later developmental stages.

MATERIALS AND METHODS**Osmotic adjustment in seedlings**

A collection of 33 genotypes of different origin were grown in the field in 12-l pots, in a mixture of soil and sand (2:1 v/v) well provided with water and nutrients. There were 10 plants for each genotype and 1 plant per pot. When the plants reached the eight-leaf stage the pots were enclosed in plastic bags, to avoid a rapid drying out of the soil and the effects of fortuitous rains, and watering was withheld. During the period of exposure to water stress, the changes in water status were defined in terms of leaf relative water content (RWC) and osmotic potential (π). Measurements were made on the uppermost expanded leaves, each leaf was sampled twice. Sampling commenced before day-break and 3 samples for each genotype were taken on each sampling date. For π measurements 0.5 cm² discs were used. It were frozen at -20°C and after thawing osmotic potential was measured in psychrometer chambers (Wescor, C-52) calibrated using ClK solutions. RWC was determined according to the method described by Slatyer and Barrs (1965) on 3.2 cm² discs taken from the laminae on the opposite side to that of the π sample. When the seedling showed signs of wilting at day-break for three consecutive days sampling was concluded. The resulting period of exposure to water stress was usually ten days. The degree of osmotic adjustment was determined from the $\ln RWC/\ln \pi$ relationship (Morgan, 1983).

Osmotic adjustment in pre and post-anthesis

A sub-set of seven genotypes was grown in the field in 30-l pots, in a mixture of soil and sand (2:1 v/v) with

adequate water and nutrient supply. There were 15 plants for each genotype and 1 plant per pot. Plants were exposed to water stress in two developmental stages: pre-anthesis (appearance of the star) and post-anthesis (last anthesis). To avoid a rapid drying out of the soil after regular watering was terminated, the stressed plants received a daily watering equivalent to 20% of the potential evapotranspiration of the previous day, calculated as described by Priestley and Taylor (1979). For both treatments, the period of water stress was about 20 days. For the pre-anthesis stage, the end of exposure to stress coincided with initiation of anthesis; and for the post-anthesis stage it coincided with the end of the linear phase of embryo growth (Hall *et al.*, 1985) of mid-radial file cypselae. RWC and π were measured as described above on samples of leaf tissue were taken before day-break (4 plants of each genotype). Samples were taken from the last 3 uppermost expanded leaves.

RESULTS

Osmotic adjustment in seedlings

When the concentration of cell solutes changes in response to gain or loss of water, these changes can be described by the RWC/ π relationship ($\pi = \pi_0 RWC_0 / RWC$, where the subscript 0 signifies full turgor). For convenience, the linear form of the equation, $\ln \pi = \ln (\pi_0 RWC_0) - \ln RWC$ is used where the intercept is $\ln (\pi_0 RWC_0)$ and the negative slope has a value of 1 (Morgan, 1984). By evaluating the slope of the observed response of $\ln RWC$ to $\ln \pi$ it is possible to determine the extent to which changes in π , as the tissue loses water, is due to concentration by water loss. If the results obtained can be explained by the above equation (*i.e.* the slope is equal to -1) the genotype

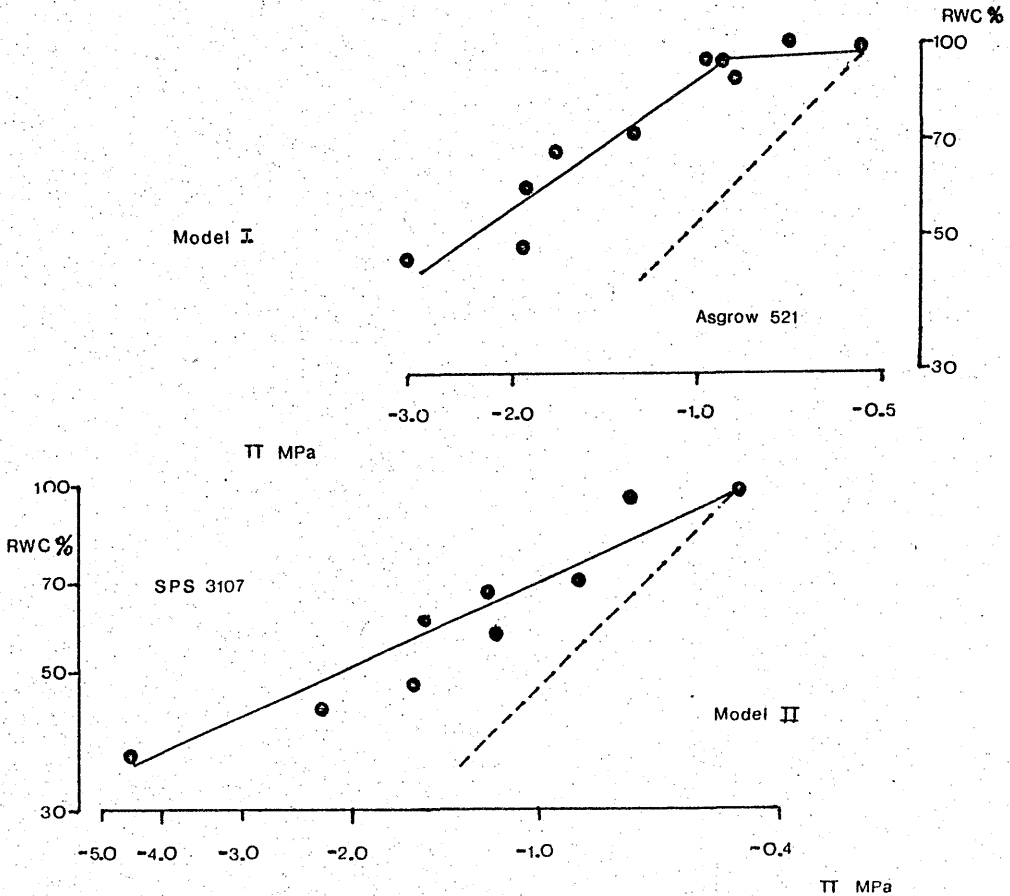


Fig.1 Examples of the $\ln RWC / \ln \pi$ relationship. The dotted line represents the relationship expected when osmotic adjustment is lacking.

does not exhibit osmotic adjustment. Our results could not be so explained, thus confirming the existence in this species of osmotic adjustment (Fig. 1). As in wheat (Morgan, 1983) our results were best described by either two intersecting (Model I) or a single straight line (Model II) (Fig. 2). With Model I (Fig. 2), in the first phase there are changes in π but not in RWC. The RWC starts to change after a threshold osmotic potential is reached. Threshold π and the slope of the second phase can be different among genotypes. In Model II (Fig. 2) the changes in RWC start at high values of π and the slope of the relationship in RWC/ $\ln\pi$ changes with the genotypes. The degree of osmotic adjustment was expressed by the predicted value of RWC at an osmotic potential of -1.7 MPa (RWC_0). This value is useful in describing osmotic adjustment because it incorporates the changes in the slope of the response as well as the differences in initial π and the value of π at which the second phase commences (Model I). For this estimation the linear regression of $\ln RWC$ on $\ln \pi$ was used. For Model I responses, linear regressions were fitted to values of the second phase. The correlation coefficient (r) for all regressions was significant ($P=0.05$) (data not presented). The confidence intervals ($P=0.95$) were calculated for RWC_0 . The results indicate the existence of intraspecific variability for capacity for osmotic adjustment (Table 1).

Osmotic adjustment in pre and post-anthesis

The $\ln RWC / \ln \pi$ relationships in the later developmental stages could again be described by Model I or II. The results (data not presented) indicate that the genotypes exhibit different degrees of osmotic adjustment. The value of RWC_0 tended to increase from pre to post-anthesis stage. This modification could be explained by the changes

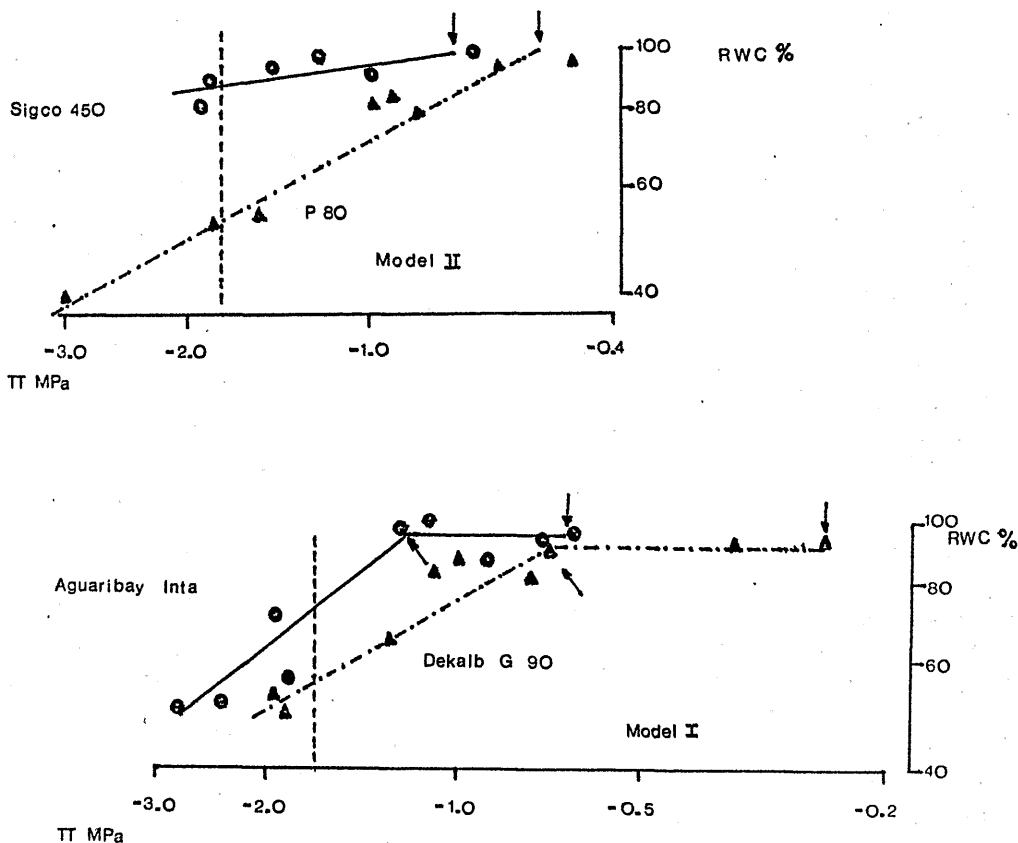


Fig. 2. Model II (above) and Model I (below) responses of π to changes in RWC. Examples show differences between cultivars for initial osmotic potential (π_i) \downarrow , and for osmotic potential at which RWC starts to fall (π_c) \downarrow . Vertical dotted lines indicate value of π used for RWC_0 estimates.

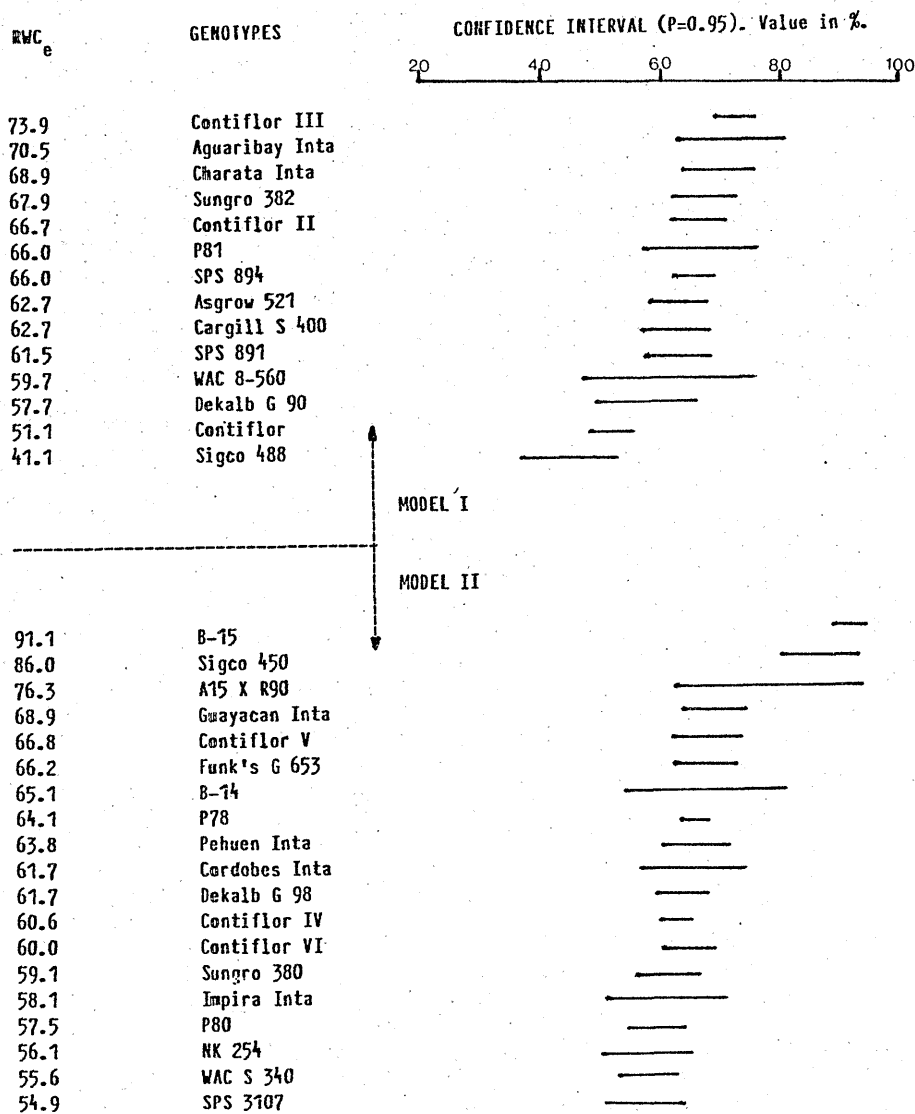
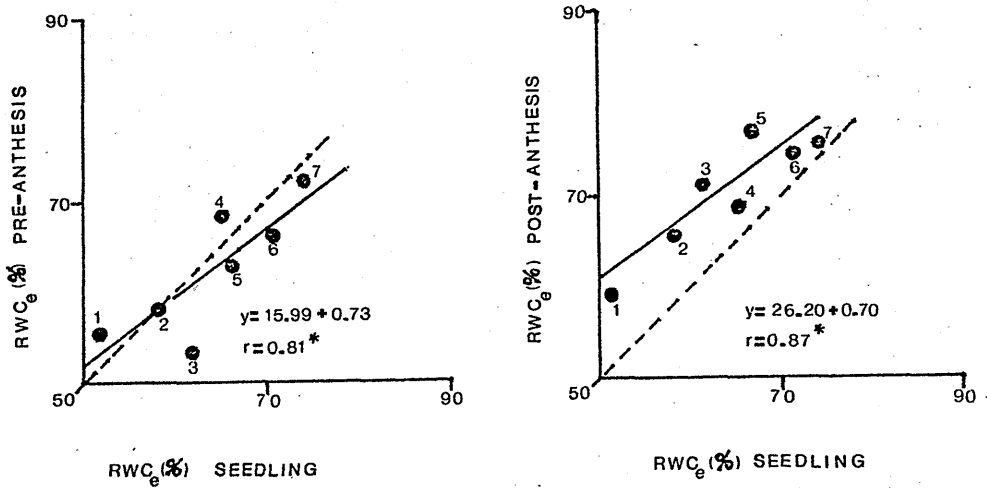


Table 1. Cultivar differences in relative water content at an osmotic potential of -1.7 MPa (RWC_e). Horizontal lines show confidence intervals (P=0.95).

observed in the initial π (data not presented). Turner *et al.* (1979) have observed similar changes with ontogeny in sorghum and sunflower, as have Hall *et al.* (unpublished) in sunflower.

The RWC_e values for the different genotypes measured in the seedling stage and those observed in later developmental stages were linearly related ($P=0.05$) (Fig. 3).



- 1. Contiflor
- 2. Dekalb G 90
- 3. Dekalb G 98
- 4. P 81
- 5. Contiflor II
- 6. Aguaribay Inta
- 7. Contiflor III

Fig. 3. Relationship between RWC_e measured in seedlings and in pre- (left) and post- (right) anthesis stages. Dotted line shows the 1:1 relationship. * Significant ($P = 0.05$).

CONCLUSIONS

Sobrado and Turner (1983) and Turner *et al.* (1979) have shown that osmotic adjustment occurs in sunflower. Our results coincide with theirs and extend them to show that intraspecific variability for this attribute exists. An essential pre-requisite for selection for physiological and biochemical attributes for tolerance to water stress is that the variability for these attributes should be reasonably easy to detect. The possibility of carrying out selection for osmotic adjustment in the seedling stage permits the use of controlled or partially controlled environments (such as glasshouse) where selection can be more efficient and allows the handling of a greater number of populations.

ACKNOWLEDGEMENTS

Financial support by CONICET and Compañía Continental S.A. are gratefully acknowledged. We thank A. Vazquez for much assistance during the experiments.

REFERENCES

- HALL, A.J.; CHIMENTI, C.A.; VILELLA, F. and FREIER G. 1985. Timing of water stress on yield components in sunflower. Proceedings 11th. International Sunflower Conference, Mar del Plata, Argentina. pp.131-136.
- JONES, M.M. and TURNER, N.C. 1978. Osmotic adjustment in leaves of sorghum in response to water deficits. *Plant Physiology* 61:122-126.
- JONES, M.M.; OSMOND, C.B. and TURNER, N.C. 1980. Accumulation of solutes in leaves of sorghum and sunflower in response to water deficits. *Australian Journal of Plant Physiology* 7:193-205.
- MORGAN, J.M. 1983. Osmoregulation as a selection criterion for drought tolerance in wheat. *Australian Journal of Agricultural Research* 34:607-614.
- MORGAN, J.M. 1984. Osmoregulation and water stress in higher plants. *Annual Review of Plant Physiology* 35:299-319.
- MUNNS, R.; GREENWAY, H. and KIRST, G.O. 1983. In LANGE, O.; NOBEL P.; OSMOND, C. and ZIEGLER, H. (Eds.) *Encyclopaedia of Plant Physiology, volume 12C: Physiological Plant Ecology III*. Springer-Verlag: Berlin & Heidelberg. pp. 59-136.
- PRIESTLEY, C. and TAYLOR, R. 1972. On the assessment of surface heat flux and evaporation using large-scale parameters. *Monthly Weather Review* 100:81-92.
- SLATYER, R. and BARRS, H. 1965. Modifications to the relative turgidity technique with notes on its significance as an index of internal water status of leaves. *UNESCO Arid Zone Research* 25:331-342.
- SOBRADO, M.A. and TURNER, N.C. 1983. Influence of water deficits on the water relations characteristics and productivity of wild and cultivated sunflowers. *Australian Journal of Plant Physiology* 10:195-203.
- TURNER, N.C.; BEGG, J. and TONNET, M. 1979. Osmotic adjustment of sorghum and sunflower crops in response to water deficits and its influence on the water potential at which stomata close. *Australian Journal of Plant Physiology* 5:597-608.
- TURNER, N.C. and JONES, M.M. 1980. Turgor maintenance by osmotic adjustment: a review and evaluation. In TURNER, N.C. and KRAMER, P.J. (Eds.). *Adaption of plants to water and high temperatures stress*. Wiley-Interscience, New York. pp. 87-103.