# Morphological Mechanisms of Drought Tolerance in Cutleaf Medic [*Medicago laciniata*(L.) Mill]

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## Abstract

Cutleaf medic [*Medicago laciniata* (L.) Mill] is an annual medic native of Mediterranean that grows naturally in many semi-arid regions. In the throughout south-west and south of Iran areas with annual rainfall lower than 200 mm, recognized some ecotypes of cutleaf medic that were drought tolerant. In order to study of some of morphological mechanisms of drought tolerance in sensitive and tolerant ecotype of cutleaf medic, a greenhouse experiment was conducted in 2004-5, at the Agricultural Biotechnology Research Institute of Iran, using a factorial arrangement in RCBD with three replications. Two ecotypes were treated in four levels of water stress include -0.1, -0.2, -1 MPa as low, medium and high stress levels respectively and normal condition (FC = -0.03 MPa). Morphological traits were measured in first day to reach per stress level and ten days after to remain in stress levels. Results indicated that tolerant ecotype had a significant superiority to sensitive ecotype in most studied morphological traits such as, shoot height and leaf area. Drought stress reduced La in both ecotypes, but reduction of La in tolerant ecotype was related to increase the dept of cut of leaves rather than size, length, desiccation of leaves and decreased transpiration in tolerant ecotype. Moreover, shoot height in sensitive ecotype was higher than tolerant ecotype in normal condition, but tolerant ecotype continued its growth and development under stressed condition which final height was higher than sensitive ecotype. This experiment indicated that leaf area is one of the major concerns of tolerance.

## Media summary

Assessment of important morphological mechanisms in ecotypes of cutleaf medic under drought stress will result for fast selection of tolerant ecotype of annual medic.

## Key Words

Cutleaf medic, morphological traits, water stress, leaf area, plant height

## Introduction

*Medicago laciniata* (L.) Miller (cutleaf medic) is widely distributed throughout semi-arid south and south western provinces of Iran with annual rainfall lower than 200 mm that occurs during winter and early spring. Cutleaf medic occurs mainly on sandy-surfaced red earths in a wide variety of vegetation communities. The species is thought to be native to the North African countries that border the Mediterranean Sea. Its natural habitat is dry sandy or stony desert environments, where it is often the only *Medicago* species that survives. Francis (1981) suggested that it was a most promising species for range improvement in the subdesert regions of north-western Libya. In New South Wells, it is an aggressively colonizing species and continues to advance into new areas, with its rhizobia following some distance behind. Ghanavati *et al*. (2005) were collected thirty-two accessions of cutleaf medics throw 12 provinces of south and sought-west and western provinces of Iran. Moradi *et al*. (2005) reported that among the thirty-two collected accessions, the most tolerant ecotype was belong to Bosheher province (south of Iran) and the most sensitive ecotype was belong to Lorestan province (west of Iran) with average annual rainfalls 173 mm 530 mm respectively. Young *et al*. (1992) reported that dry matter and seed production are usually less than *Medicago truncatula* Gaertner, under ideal conditions, *M. laciniata* yields may be greater when the season is short, which very common condition in southern provinces of Iran. One of the most important factors limiting the growth, development, productivity, and dispersion of plants in the biosphere is desiccation induced by decreasing environmental water potential ($\psi_w$) (Sanchez, *et al.,* 1998). As a result of water limitation, plants express various responses and have developed a wide diversity of drought tolerance mechanisms from both morphological and physiological aspects (Blum, 1996). Studies have shown that drought stress can affect the growth of plant organs differently (e.g. Spollen *et al*., 1993). The change in root to shoot dry mass ratio has been considered as one of the mechanisms involved in the adaptation of plants to drought stress (Turner, 1997). Drought stress reduced both root and shoot growth. However, root growth seems to be less affected (Liu and Stutzel, 2004). Zhang *et al* (2004) indicated significant differences in plant height, total biomass, total leaf area, root/ shoot ratio in *Populus davidiana* under drought stress. Thus, it is important to study on the mechanisms of drought response in tolerant ecotypes of *M. laciniata*.

## Methods

Certified ecotypes of cutleaf medic (*Medicago laciniata* (L.) Miller), 'drought tolerant' (Pic.1) and 'drought sensitive' (Pic.2) gifted from National Plant Gene Bank of Iran. The two ecotypes were classified as drought tolerant or sensitive according to water stress period. A greenhouse experiment was conducted in 2004-5, using a factorial arrangement in RCBD with three replications. Two ecotypes of cutleaf medic, were treated in four levels of water stress (Soil water potential) include -0.1, -0.2, -1 MPa as low, medium and high stress levels respectively and normal condition (FC = -0.03 MPa). The water stress regimes were applied in 50 d after sowing (usual time of water deficit in real habitat) by weigh each pot and add water to the weight calculated for the desired water regime (Va´zquez *et al.,* 2001). Soil gravimetric water content was defined as $\theta = Ww / DWs \times 100$ (where Ww is the weight of the water contained in a soil sample, and DW is the dry weight of the sample (Martýnez *et al.,* 2004) and the pots were weighed twice a day (800 and 1600 O'clock) and water loss replaced by top watering. Evaporation from the soil surface and transpiration water loss and increases in plant weight were estimated by weight difference of pots of without plant and main pots. Morphological traits such as, plant height was measured in first day to reach per stress level and 4, 7, 10 days after to remain in stress levels and leaf area, shoot and root dry matter and shoot/root dry weight were measured in first day to reach per stress level and ten days after to remain in stress levels.

## Results

Tolerant ecotype had a significant superiority to sensitive ecotype in the most of studied morphological characteristics i.e., Shoot dry weight (Shoot DW), Root dry weight (Root DW), Soot/Root ratio (Sr), total leaf area (La) (Table 1) and Shoot height (Ht) (Fig.1) under four watering regimes (soil water potentials). Ecotypes from the Lorestan province (sensitive ecotype, moderate climate) in control condition (-0.03, MPa) had greater Shoot DW, Root DW, Sr, and La than ecotype from the Bosherher province (tolerant ecotype, dry climate), But these traits had increased in tolerant ecotype under drought condition. The watering regimes significantly affected all these morphological properties ($P<0.01$). Drought stress reduced leaf area in both ecotypes, but reduction of La in tolerant ecotype was related to increase the dept of cut of leaves rather than size, length, desiccation of leaves and decreased transpiration in tolerant ecotype. Moreover, shoot height in sensitive ecotype was higher than tolerant ecotype in normal condition, but tolerant ecotype continued its growth and development under stressed condition which final height was higher than sensitive ecotype. It seems High root DW in tolerant ecotype showed root development that maybe, leads to water absorption from soil depth under drought stress.

## Conclusion

Drought affected on growth and development in treated plants. Changes in plant growth and structure in response to progressive drought stress exhibited the primary signals for drought adaptation. The existence of a large number of species and varieties of *Medicago* growing in many diverse habitats of Iran enables the selection of species and seed sources for almost any environmental condition, including severe drought regions. Physiological adaptation, plant structural modifications and growth pattern adjustments are useful indices of the consequences of water deficit. Differences in drought adaptations among *M. lacinita* ecotypes were demonstrated and attributed to differences in these morphological responses to water availability. Significant differences among ecotypes were found in; Shoot DW, Root DW, Sr, La (Table 1) and Ht (Fig.1) under four water regimes. The effect of drought stress and interaction between drought and ecotypes were also highly significant in measured physiological and morphological properties. Apparently, in the tolerant ecotype several adaptations strategies have been evolved to cope with adverse environmental conditions.

Sensitive ecotype had good performance only in well-watered condition. Nonetheless, the dry climate ecotype (tolerant) which grows under habitat with prolonged annual drought, grew better under water stress regimes. All these responses enabled the tolerant ecotype to overcome with prolonged drought condition. This study provided evidence that plant surface structure, form and composition carry a major impact on the plant interaction with the environment. Plant surface absorbs solar energy part of which is used for photosynthesis and most of which must be dissipated. However, plant surface structure determines the hydraulics of leaf surface and the boundary layer conditions, which affect the rate of water removal from the leaf surface, upon transpiration. As, this experiment indicated that leaf area is one of the major concerns of tolerance.

Table 1. Shoot dry weight, root dry weight, shoot to root ratio, and leaf area of two contrasting cutleaf medic ecotypes in different soil water potentials 10 d after exposure to the treatments.

| | Shoot d. wt (g) | | Root d. wt (g) | | Shoot / Root Ratio | | Leaf Area ($cm^2$) | |
|---|---|---|---|---|---|---|---|---|
| **Soil water potential (Mpa)** | **Sensitive** | **Tolerant** | **Sensitive** | **Tolerant** | **Sensitive** | **Tolerant** | **Sensitive** | **Tolerant** |
| -0.03 | 1.054 | 0.942 | 0.138 | 0.134 | 7.51 | 7.34 | 160 | 154 |
| -0.1 | 0.634 | 0.83 | 0.119 | 0.133 | 5.34 | 6.23 | 105 | 118 |
| -0.2 | 0.512 | 0.753 | 0.115 | 0.128 | 4.48 | 5.91 | 69 | 115 |
| -1 | 0.483 | 0.649 | 0.1 | 0.118 | 4.84 | 5.5 | 53 | 97 |
| **Mean** | **0.671** | **0.793** | **0.118** | **0.128** | **5.54** | **6.24** | **97** | **121** |
| **Significance** | | | | | | | | |
| Stress (S) | *** | | *** | | *** | | *** | |
| Genotype (G) | ** | | *** | | *** | | *** | |
| S*G | * | | *** | | * | | *** | |
| **LSD 0.05** | | | | | | | | |
| Stress (S) | 0.008 | | 0.048 | | 0.048 | | 5.53 | |
| Genotype (G) | 0.006 | | 0.034 | | 0.34 | | 3.91 | |
| S*G | 0.071 | | 0.011 | | 0.772 | | 8.68 | |
| C.V | 5.54 | | 5.33 | | 6.63 | | 4.11 | |

Data are means of three replications. *, **, *** significant at P<0.05, 0.01 and 0.001, respectively. ns= non significant.

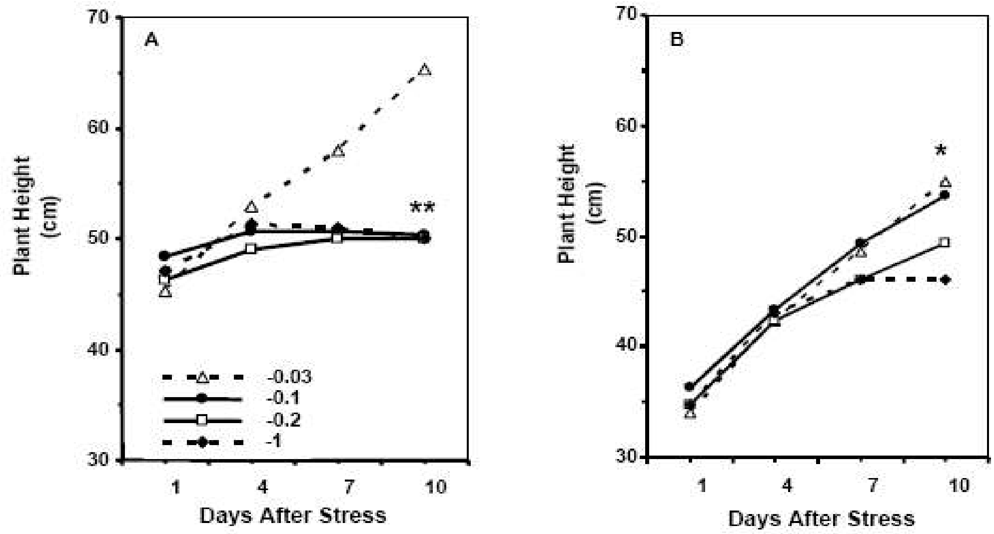


Fig. 1. Shoot length (cm) of sensitive (A) and tolerant (B) genotypes of cutleaf medic under different water potential. Measurements were done at 1, 4, 7 and 10 d after stress. * and ** indicating significance at P<0.05 and 0.01, respectively at day 10, using duncan multiple test.

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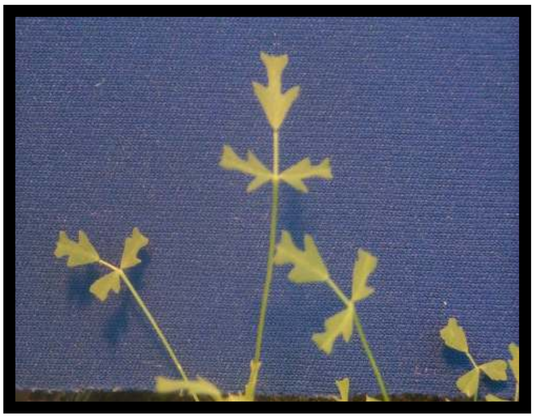

Pic.1. Drought tolerant ecotype

Pic.2. Drought sensitive ecotype