



University of Zabol
Graduate school
Faculty of Agriculture
Department of Agronomy and Plant Breeding

**The Thesis Submitted for the Degree of M.Sc (in the field of
Biotechnology)**

**Effect of drought stress on physiological,
biochemical traits and flavonolignin
content (silymarin) in milk thistle**

Supervisor:

Dr. M. Alahdou

Advisors:

Dr. L. Mehravaran

Dr. H. Piri

By:

Mostafa Sarani malak

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Abstract

Drought stress is the most important factor limiting the yield of crops and medicinal plants in arid and semi-arid regions. Milk thistle (*Silybum marianum*) is a medicinal plant containing secondary metabolites (flavonolignans) with antioxidant properties. The synthesis and accumulation of flavonolignans in plants is influenced by different environmental conditions. The accumulation of secondary metabolites in this plant was studied under drought stress with respect to the antioxidant defense system at the physiological and biochemical level. In this study, evaluation of the effect of drought stress containing 4 different levels of irrigation (100 (non-stress), 75 (mild stress), 50 (moderate stress) and 25% (sever stress) of water capacity) and in 3 growth stages (6, 13 and 20 weeks) on agronomic, physiological and biochemical traits including fresh weight, dry weight, the contents of photosynthetic pigments, proline, carbohydrates, protein, malondialdehyde, total phenol and total flavonoids, relative water content, electrolyte leakage and the activity of antioxidant enzymes (catalase, ascorbate peroxidase, guaiacol peroxidase, super oxide dismutase and poly phenol oxidase) was performed in a factorial design in the greenhouse. The results showed that the effect of different irrigation levels, harvest time and their interaction on the traits were significant. Wet weight, dry weight, photosynthetic pigments content and relative water content decreased under drought stress conditions, so that in severe drought stress conditions (25% of water capacity) the lowest amount them was observed. The contents of proline, carbohydrates, protein, malondialdehyde, total phenol and total flavonoids, the activity of antioxidant enzymes and electrolyte leakage increased with increasing stress intensity, so that in the severe drought stress condition, the highest amount of them was found. These results indicated that milk thistle responded to drought stress using enzymatic and non-enzymatic antioxidant defense systems. With increasing plant growth, the content of total phenols and flavonoids increased, so that in the final growth stage (20 weeks) the highest content was observed. Therefore, drought stress can be used as a improving factor the active ingredients of this plant, and also the final growth stage is the best time to harvest due to the more accumulation of secondary metabolites in this growth stage.

Keywords: Drought stress, Milk thistle, Silymarin, Biochemical traits, Physiological traits