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**The effect of biotic and abiotic elicitors on biochemical traits and silymarin content of milk thistle in cell suspension culture conditions**

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***Abstract:***

Milk thistle (*Silyban marianum*) is a dicotyledonous plant from the Asteraceae family, which is very important in the pharmaceutical industry. The active substance of this plant is called silymarin, which is a combination of flavonolignans. This combination is effective in the treatment of various liver diseases, hepatitis, diabetes, cardiovascular diseases, cancer, blood fat. In the present research, the aim of this research is to optimize the tissue culture of thistle plant, increase the production of its secondary metabolites by using different elicitors in cell suspension conditions. Therefore, in this research, in order to produce sterile seedling, after disinfection, the seeds were cultivated in MS medium and four weeks after cultivation from the hypocotyl and cotyledon of these seedlings to produce callus in MS medium containing growth regulation hormones (BAP and NAA). Two consecutive cultivations were done to increase the callus and the produced fibrous calluses were transferred to the liquid culture medium containing the previous regulation hormones. Callus treatment at a concentration of 40 mg/ml and control treatment showed the highest and lowest increase in total phenol content, total flavonoid, total tanning and antioxidant activity, respectively. The content of total proline also showed the highest increase at the concentration of 80 µg/ml, which is due to the increase in tension created by this elicitor. The mannitol elicitor shows different reaction on the traits in different content at the conservation such that total phenol content at a concentration of 120 µg/ml, total flavonoids and carbohydrate content at the concentration of 240 µg/ml, total tannin and proline content at a concentration of 60 µg/ml and antioxidant activity showed the highest increase in the control treatment. The amount of silymarin is also. Therefore, if the cellulase elicitor with a concentration of 80 micrograms per ml is added to the suspension in the growth phase (eighth day), it will have the best result in increasing the secondary metabolites of this plant and antioxidant activity.

Keywords: marigold, silymarin, hormone tissue culture, isolated piece, flavonolignan culture.