

Abstract

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Title of Thesis: The Influence of Elicitation on the Production of Suspension Culture of *Trifolium pratense* L.

The production of secondary metabolites in the *Trifolium pratense* L. suspension culture (red clover) is low and therefore there was an attempt to increase it by elicitation. Elicitation is a method making use of protective mechanisms of plants to increase the production of secondary metabolites in plants and *in vitro* cultures. The jasmonic acid and its precursors and derivatives are some of the endogenous signal substances of the plants' defensive responses. In the exogenic application case, they may also function as elicitors.

This diploma thesis examined the effect of jasmonic acid and the effect of jasmonic acid in combination with calcium ions and verapamil on the production of flavonoids and isoflavonoids by the *Trifolium pratense* L. suspension culture. The culture was cultivated in Gamborg medium to which 2 mg.l⁻¹ of 2,4-dichlorophenoxyacetic acid and 2 mg.l⁻¹ of 6-benzylaminopurine were added, at the temperature of 25°C, 16-hr light/8-hr dark period. The flavonoids were determined spectrophotometrically according to the Czech Pharmacopoeia 2009 and the isoflavonoids by the HPLC method. The best elicitation effect of the jasmonic acid was observed after the 24-hour application of the 500 μmol concentration. The stimulation of elicitation with extracellular calcium ions mostly took effect after the 24-hour application of the 10 mmol concentration CaCl₂. An application of verapamil decreased production of secondary metabolites by blocking calcium channels.