ABSTRACT

Charles University

Faculty of Pharmacy in Hradec Králové

Department of Pharmacognosy

Student: Nikola Urbanová

Supervisor: PharmDr. Marie Kašparová, Ph.D.

Title of diploma thesis: The study of secondary metabolites in plant tissue cultures II

The aim of this work was to determine the effect of the potential elicitor chitosan on production of podophyllotoxin in callus and suspension cultures of Juniperus virginiana Glauca variety and suspension cultures of Juniperus virginiana Hetzii variety. Schenk and Hildebrandt medium supplemented with α -naphtylacetic acid (3.0 mg/l), kinetin (0.2 mg/l) and ascorbic acid (15.0 mg/l) was used for the cultivation. Chitosan solutions of various concentrations (0.001; 0.01; 0.1; 1 g/100 ml) were affecting the cultures for 6, 24, 48 and 168 hours. The content of chitosan was determined by HPLC.

Higher values of podophyllotoxin content were measured in cultures derived from the Glauca variety. The best chitosan effect on podophyllotoxin production was manifested in callus cultures after 24 hours application of 0.001 g/100ml concentration. A podophyllotoxin content of 0.210 % was determined, which was about 320 % higher in comparison with the control. The maximum content (0.140 %) in suspension culture was induced by 24-hours application of a 0.1 g/100 ml concentration; this was about 211 % higher than the control. The callus culture derived from Hetzii variety compared to the Glauca variety had a lower average content of podophyllotoxin in control samples and in most cases, there was no positive effect on production by elicitor. Chitosan can be used as a potent elicitor and especially is able to increase podophyllotoxin production in Juniperus virginiana Glauca variety callus and suspension cultures in vitro.

Keywords: Anti-cancer lignans, podophyllotoxin, Juniperus virginiana, suspension cultures.