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Effects of Methyl Jasmonate and Salicylic Acid on the Production of Bilobalide and Ginkgolides in Cell Suspension Culture of *Ginkgo biloba*

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Objectives

In an attempt to increase productivity, the effects the elicitors methyl jasmonate (MJ) and salicylic acid (SA) on the production of bilobalide (B), ginkgolide A (GA), and ginkgolide B (GB) were studied in cell suspension cultures of *G. biloba*.

Materials and Methods

The callus of *G. biloba* were obtained from embryo were transferred into MS liquid medium supplemented with 3% sucrose and 3.5 mg/L NAA. Sterilized MJ and SA were added to flasks cultured for 2 weeks at concentrations of 0.01, 0.1, 1.0 and 2.0 mM. The extract samples were performed HPLC using a Lichrospher®100 RP-18 (4.6 mm × 25 cm, 5 μm, Merck) column and a UV detector operating at a wavelength of 250 nm. The isocratic mobile phase is a mixture of MeOH and H₂O (50:50 v/v). Retention times of B, GA and GB were 6.89, 9.92 and 10.26 min, respectively.

Results and Discussion

The concentrations and exposure times of both MJ and SA were factors that strongly affected the production of B, GA, and GB. Our experiments showed that MJ and SA increased the content of B, GA and GB in cell cultures of *G. biloba*. The effect of MJ on the production of B was maximal at 0.01 mM whereas the effect of MJ on the production of GA and GB was maximal at 1.0 mM. Treatment of exogenous MJ not only increased the production of B, GA and GB in cells, but also markedly stimulated their secretion into the culture medium. The 1.0 mM MJ treatment produced a maximal secretion of B after 12 h of exposure and increased the concentration of B in the culture medium up to 6.25 fold compared to the controls. Also, 0.1 mM MJ stimulated the secretion of GA and GB which reached the highest levels after 48 h (2.12 and 12.4 fold, respectively) compared with the controls. Treatment with 1.0 mM SA transiently enhanced GA and GB production up to 3.1 and 6.1 fold, respectively, compared to the control. The release of GA and GB into the culture medium significantly increased 12 h after treatment with 0.1 mM SA and then rapidly decreased. The stimulatory effects of MJ and SA on B, GA and GB production may be due to stimulation of the biosynthetic enzymes. Further work will be required to investigate the related enzymes involved in biosynthesis of B, GA and GB.

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