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SHORT COMMUNICATION

# ISOLATION OF APIGENIN FROM ILLUMINATED CELL SUSPENSION CULTURES OF SOYBEAN, *GLYCINE MAX*

K. HAHLBROCK

Institut fur Biologie II der Universität, Lehrstuhl fur Biochemie der Pflanzen, 78 Freiburg/Br., Germany

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**Abstract**—The flavone apigenin has been isolated and identified from batch-propagated cells of *Glycine max* L. Its synthesis in cell cultures is greatly stimulated by light.

## INTRODUCTION

ANTHOCYANINS, aurones, chalcones, and isoflavones have been identified as flavonoid constituents of soybean (*Glycine max*) [1,2] Some of the enzymes related to the biosynthesis of these compounds have also been demonstrated in cell-free extracts from either intact seedlings[3,4] or cell suspension cultures of this plant [5,6] Recently, glycosides of daidzein have been isolated from callus tissues of this plant after treatment with auxins and cytokinins [7] Culture conditions for cell suspensions of *Glycine max* in liquid media containing mineral salts, vitamins, 2,4-dichlorophenoxyacetic acid, and sucrose have been well elaborated [8,9]

## RESULTS

Upon illumination with high intensities of white light, suspension cultures of soybean root cells produced a compound which was isolated and identified as apigenin (5,7,4′-trihydroxyflavone), as described below This product was formed only during a limited period of time near the end of the growth cycle of the cell cultures. This observation is interesting with regard to an apparent correlation between the formation of apigenin and the activity of two of the enzymes related to its biosynthesis, phenylalanine ammonia-lyase (E.C 4 3.1.5) and *p*-coumarate: CoA ligase.[6]

## EXPERIMENTAL

Cells originally obtained from roots of soybean seedlings were grown in 40 ml of Gamborg's B5-D medium[8] in 200-ml Erlenmeyer flasks at 25–28° for 6 days in the dark Samples of 2 ml were transferred to fresh medium every 5–6 days Under these conditions, fresh weight of cells increased from approx 0 3 to

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[6] K HAHLBROCK, E KUHLEN and T LINDL, in preparation
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5 g per culture within 6 days After illumination with white light from fluorescent lamps (Philips TL 40 W/18, 27,000 lx) for 40 hr, cells were harvested by vacuum filtration and extracted with boiling 80% EtOH (5 ml/g fr wt of cells) Insoluble material was removed by centrifugation Apigenin was identified in the supernatant by co-chromatography with authentic material (6 solvents, paper and TLC) and by UV analysis [10]

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