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Studies on the Suppressive Effect of Antioxidant in Human Placental Extract on Oxidative Stress and its Mechanism

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Introduction

Several antioxidants are used as defence to the oxidative stress resulting from environmental pollutants and ultraviolet rays. We examined antioxidative activity and antioxidants in human placental extract (PLx) because it was reported that PLx had an antiinflammatory effect and an inhibitory effect on melanin production. It is considered that one of the roles of placenta is protection of the embryo from oxidative stress and that placental antioxidants can efficiently scavenge active oxygen species and free radicals. We had already found the radical scavenging activity of PLx in an *in vitro* experiment but the antioxidants are not clarified yet. In this study, we investigated an antioxidant in PLx that is a glucuronide of an amino acid metabolite.

Methods

(1)180 mg/kg of PLx and 86 mmol/kg of ethanol were administered to 6 week old ddY mice. At 24 h after ethanol administration, GOT and GPT activities in serum were measured as the parameters of acute alcoholic liver injury, and thiobarbituric acid reactive substances (TBARS) content, GSH content and antioxidative enzyme activities in liver were measured as parameters of oxidative stress. (2)PLx was applied to gel filtration with Sephadex G-50 and the antioxidative activity, uronic acid and amino acid contents in each fraction was measured with the deoxyribose method, carbazole-sulfuric acid method and the ninhydrin reaction, respectively. The phenolic OH group and the indole ring in acid-hydrolysed PLx were qualitatively identified and amino acids and tryptophan metabolites in acid-hydrolysed PLx were identified by silica gel TLC. Antioxidative activities of hydroxykynurenine, glucuronic acid and others were measured with the deoxyribose and conjugated diene methods.

Results and Discussion

GOT and GPT activities in serum and TBARS content in liver were increased and GSH content and antioxidative enzyme activities in liver were decreased by ethanol administration. However, these changes were suppressed by the pre-administration of PLx. These results suggest that the acute alcoholic liver injury and oxidative stress due to ethanol administration were suppressed by pre-administration of PLx and it decreased the production of active oxygen species and free radicals.[1)]

An antioxidative activity, amino acid and uronic acid were eluted in the same fraction in the gel filtration of PLx. The phenolic OH group appeared by acid hydrolysis of PLx and antioxidative activity was decreased by treatment of acid-hydrolysed PLx with D-amino acid oxidase. However, the indole ring was not identified in PLx. These results and TLC analysis suggest that one of the antioxidants in PLx is 5-OH-kynurenine glucuronide. 5-OH-kynurenine is a tryptophan metabolite but its glucuronide has not been identified. The antioxidative activities of 5-OH-kynurenine and glucuronic acid were about 5 times more than dimethyl sulforide (DMSO) and mannitol, and about 3 times more than tryptophan and 5-OH-tryptophan. It is considered that 5-OH-kynurenine in PLx is one of the most useful antioxidants and that PLx may possibly be used as a preventive agent to diseases resulting from oxidative stress.

Reference

1) Watanabe S., Kawauchi S., *Bitamin*, **68**, 79—85 (1994).