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Development of rare flavonoid as a cosmetic ingredient by using GPR (Glycoside pattern remodeling) technology

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Objectives

Glycosides are diverse group of natural products that are ubiquitous in nature and are present in virtually all kind of plants, microorganisms and animals. They comprise several important classes such as hormones, alkaloids, flavonoids, etc. Among them, kaempferol and its glycosides, kaempferol-3-O-rutinoside (nictoflorin) and Kaempferol-3-O-glucoside (astragalol), are representative of the green tea flavonoid. Because of their broad biological activities, there are strong needs to apply them to cosmetic ingredient. However, many kinds of plants contain these compounds including green tea, these are in very small quantities. So these components cannot be prepared in high degree of purity using purification method and simple extract has been used for cosmetic application. Herein we prepared these rare flavonoids in high degree of purity using GPR (Glycoside pattern remodeling) technology.

Materials and Methods

1. Materials

Green tea seed (*Camellia sinensis* (L.) O. Kuntze, Theaceae) was purchased at Zhejiang chemicals, China. Kaempferol authentic was purchased from Sigma chemical Co. (St. Louis, MO, USA). The following enzyme preparations were used: hesperidinase, -glucosidase, cellulase, -glucuronidase, pectinase, -galactosidase, amyloglucosidase, -amylase, dextranase. All organic solvents were used of analytical grade and purchased from Fisher scientific UK (Loughborough, Leics, UK.)

2. Extraction of Green tea seed

GTS was ground for extraction in a grinder (Food Mixer, FM-680T, Han il, Korea) and crushed GTS was defatted three times with n-hexane. 100g of defatted and dried GTS was extracted with 2 l of 70% ethanol in a Soxhlet apparatus for approximately 6 h and was then filtered and concentrated using vacuum evaporator. Final evaporation yielded 25.4 g of a yellow powder.

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3. Enzymatic hydrolysis of green tea seed extract

GTSE (0.5 g) in 8 ml 0.02 M sodium-acetate buffer (pH 5.0) with 2 ml of several glycolytic enzyme solutions was incubated with stirring at 37°C for 24hr. Each sample and blank was used as reaction controls. All samples were prepared in duplicate. The enzyme reaction was conducted solely or mixed properly. After incubation, each aliquot was extracted with Ethanol and it was centrifuged at 4°C for 10 min. To determine the material changing, it was stored frozen for HPLC analysis.

Results and Discussion

We verified that green tea seed has relatively high amounts of kaempferol glycosides, kaempferol-3-O-[2-O- β -D-galctopyranosyl-6-O- α -L-rhamnopyranosyl- β -D-glucopyranoside (camelliaside A) and kaempferol-3-O-[2-O- β -D-xylopyranosyl-6-O- α -L-rhamnopyranosyl- β -D-glucopyranoside (camelliaside B). Three rare flavonoids (kaempferol, nictoflorin and astragalin) were prepared in high purity using enzymatic selective deglycosylation. Selective hydrolysis of camelliaside A and camelliaside B was occurred controlling reaction temperature, pH, concentration and species of enzyme. We also evaluated their activities in the skin. Each compound shows their own effects according to combining sugar although they have identical mother compound. Kaempferol, alycone of camelliaside A and camelliaside B, is an efficient scavenger of 1,1-diphenyl-2-picrylhydrazyl radicals and an inhibitor of xanthine/xanthine oxidase. Another flavonol, nictoflorin shows an outstanding promoting effect in hair growth after topical application it on to the back of mice. Finally, astragalin was found to possess antiinflammatory action by inhibiting histamine release from human basophilic cell line. These results suggest that we can be available kaempferol, nictoflorin and astragalin in large quantities by enzymatic deglycosylation and apply for them as an active ingredient in cosmetic.

Reference

Park JS, Rho HS, Kim DH, Chang IS (2006) Enzymatic Preparation of Kaempferol from Green Tea Seed and Its Antioxidant Activity. *J. Agric. Food. Chem.* 54: 2951-2956.