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Panax Saponin C의 부분화학 구조

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Partial Structure of Panax Saponin C

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전보에서 저자들은 인삼의 anti-inflammatory activity를 추적하여 Panax saponin A 및 C로 명명된 dammalene계 glycoside를 분리하였고 A에 대해서는 이미 그 화학구조를 밝혀 보고한바 있다. 본보에서는 Panax saponin C(PS-C)의 부분 화학구조를 밝혀 보고코자 한다. PS-C는 산분해하면 panaxatriol 1 mol, glucose 2 mol 및 rhamnose 1 mol을 생성하고, acetylation하면 dodeca-acetate를 형성한다. 따라서 protopanaxatriol의 20[s]-수산기는 glycoside결합에 참여하고있다. PS-C는 6 mol의 HIO_4를 소모하고 permethylate에 대한 methanolysis product를 GLC로 분석한 결과 2, 3, 4-trimethoxy-*methyl-rhamnoside 1 mol*과 *2, 3, 4, 6-tetramethoxy-α-methyl glucoside 2 mol*이 생성되므로 PS-C 중에 존재하는 3 mol의 sugar는 oligoside 결합에 의하지 않고 monoside결합에 의하여 연결되어 있고 glucose는 β-glycoside 결합을 하고 있음을 의미한다.

In our previous report,[1] the pure isolation of Panax Saponin C, a new triterpene-glycoside of Korean ginseng, possessing anti-inflammatory activity, was described together with the partial structure of Panax Saponin A which was obtained by concomittant process. On accetylation, Panax Saponin C, which is chromatographically single on any chromatographic solvent, dissociated into two acetate components on thin layer chromatogram with equall chromatographic strength. Both acetates were returned to original Panax Saponin C by deacetylation, therefore the possibility of molecular degeneration during the acetylation process of Panax Saponin C was excluded. Furthermore, such dissociation of Panax Saponin C was not observed with the sample which was obtained from stem part of korean ginseng, therefore both components were designated tentatively Panax Saponin C_1 and C_2 by order of their decreasing R_f-value of acetate (Benzene: Ethylacetate). Satisfactory separation of these two acetates was achieved by fractional column chromatography over silica-gel column, C_1-acetate, amorphous powder, mp. 136-140°, anal.: found: C, 59.20, H, 7.54, calcd. for $C_{72}H_{106}O_{30}$: C, 59.59, H, 7.31% and C_2-acetate, amorphous powder, mp. 143-145°.

In this report, the partial structure of Panax Saponin C_1 will be described. Panax Saponin C_1 was obtained in a pure state, amorphous powder, mp. 190-2°, by oridinary deacetylation process.

On acid hydrolysis with 2.5% H_2SO_4 in 75% dioxane solvent, PSC_1 afforded panaxatriol, glucose and rhamnose whose identity were proved by comparison with the authentic standard on TLC and GLC. The number of glucose and rhamnose in the molecule were found to be

2 : 1 by chemical and gas-chromatographic assay. Peracetylation was also proved by the disappearance of hydroxyl absorption in the ir-spectrum of PSC_1-acetate and the number of acetyl function in the acetate was found to be 12 by the careful assay on its acetyl value. This result will suggest C-20 glycoside structure, since the C_{20}-hydroxyl function of protopanaxatriol is already known to have strong resistance to acetylation.[2] One mol of PSC_1 consumed 6 mol periodic acid, therefore the structure of PSC_1 will be suggested to be either the monoside structure or 1-6 type oligoside structure. Permethylation of the saponin was accomplished by repeated methylation with Hakomori's process.[3] From the methanolysate of the permethylate 2 mol of methyl-α-2, 3, 4, 6-tetra-methoxy-glucopyranoside and 1 mol of methyl-2, 3, 4-tri methoxy-rhamnopyranoside were detected by comparison with the authentic samples on the gas-chromatogram. This result excludes the possibility of any oligoside bond for the structure of PSC_1. Considering the inversion of C_1-configuration during the methanolysis, the anomeric structure for the 2 mol glucose moiety will be concluded to be β-glycoside bond.

Experimental

Acetylation of PSC. : PSC was acetylated with Ac_2O and pyridine at room temperature overnight. The reaction mixture was poured into ice water and the ppt was taken up in $CHCl_3$. The $CHCl_3$ soln was washed with water repeatedly and concentrated to dryness. Column chromatography of the acetate on silica gel (solvent:$CHCl_3 \cdot Et_2O$ 1:1) afforded PS-C_1 acetate and PS-C_2 acetate as amorphous powder of single state respectively. PSC_1-acetate, m.p. 136-140°, (Calc. for $C_{72}H_{106}O_{30}$ C, 59.59 H, 7.31. Found C, 59.20, H, 7.54%), IR: (in KBr) strong OAc absorption and no OH band acetyl value: 12 Mol. PS-C_2-acetate, m.p. 143-145°, IR: (in KBr) strong OAc absorption and OH band.

PS-C_1: A soln of PS-C_1 acetate in MeOH/sod. methoxide was refluxed for 30 minutes. After concentration and addition of water, the reaction mixture was extracted with n-BuOH. The BuOH layer was deionized by passing through a column of ion exchange resin (IR-120 and IR-410) and evaporated *in vacuo* to dryness, affording PS-C_1 as a colorless powder, m.p. 190-2°.

Hydrolysis of PS-C_1: A) With sulfuric acid in aquous dioxan: A soln of PS-C_1 (20mg) in 2.5% H_2SO_4 (dioxan:H_2O 3:1) (2ml) was refluxed for 1 hr. The reaction mixture was diluted with water (2 ml) and washed with $CHCl_3$. The aqueous layer was neutralized carefully by Ba $(OH)_2$ solution, and centrifuged. Glucose and rhamnose were detected by TLC (silica gel, $CHCl_3$: MeOH 30:18) and GLC (TMS treated, column 2% OV17 0.3×200 cm, Temp. 120°C, Programming 5°C/min).

Chromatogram of GLC gave rhamnose and glucose Peaks with the strength of 1 by 2.

B) With dil H_2SO_4 in aquous EtOH: PSC_1 was hydrolyzed by refluxing with 5% H_2SO_4-in 50% aqueous EtOH for 4 hr. The soln was diluted with water, and the resulted ppts were taken up in ether. The hydrolysate was chromatographed on silica gel to afford panaxatriol.

Periodate consumption of PS-C_1 : (Fluery-Lange method)[4]: To a cooled soln of PS-C_1 51.1 mg in water (10 ml) containing $NaHCO_3$ 100 mg, a soln of $NaJO_4$ 544 mg in 30 ml water gradually added with stirring. Total volume of this mixture solution was made up to 50 ml. Exactly, 5 ml of this solution was titrated with 0.02 N I_2 solution. After 168 hrs, the solution showed 6 mole-consumption of periodate.

Methylation of PS-C_1 : NaH (50%, 1.0g) was washed with petroleum ether and dissolved

in DMSO (15 ml) by heating 1.5hr. at 60~70° undre nitrogen stream. To this reagent was gradually added a soln of PS-C_1 (0.7g) in DMSO (5 ml) at room temp. and the mixture was stirred at room temp. for 3 hr. After addition of CH_3I (7 ml), the mixture was allowed to stand at room temp. for one day. The entire reaction was carried out under N_2. The reaction mixture was then diluted with water and extracted with ether. The ether layer was washed with water repeatedly and evaporated to dryness. The residue was methylated again under the same conditions as above. The products were chromatographed on silica gel to give the dodecamethyl ether, which was proved to be homogeneous on TLC (silica gel, solvent, petroleum ether: ether=1 : 1), and which showed no OH absorptions band.

Methanolysis of the dodecamethyl ether: A soln of the dodecamethyl ether (10 mg) in 5% MeOH HCl (2ml) in a sealed tube was heated in a boiling water bath for 5hr. GLC of the product (Column 2% OV 17, 0.3×200 cm, temp. 150°C) revealed the presence of methyl 2,3,4-tri-omethyl-l-rhamnoside and α-methyl 2,3,4,6-tetra-o-methyl-d-glucoside (rato 1:2).

References

1. B.H. Han, Y.N. Han, L.K. Woo, *J. Pharm. Soc.* (Korea) **16**, in press (1972).
2. S. Shibata, T. Ando, and O. Tanaka, *Chem. Pharm. Bull.*, **14**, 1157-61 (1966).
3. S. Hakomori, *J. Biochem.*, **55**, 205 (1964).
4. P.F. Fleury, J. Lange, *J. Pharm. Chim.*, **17**, 107, 196 (1933).

제 3 회 정기총회 및 학술대회 예고

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