

Studies on the constituents of sclerotia and fermented mycelia of *Grifola umbellata* and the quality analysis

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Grifola umbellata belongs to the family of Polyporaceae, whose sclerotia are used as a folk medicine for diuresis and other kidney diseases for centuries and its bio-activities on immune system, anti-cancer and liver-protection are discovered in the past decades. Nowadays, studies on *G.umbellata* are mainly focused on the polysaccharides, while our research is carried out on the minor components in order to illuminate its chemical background .

Systematic isolation of the 95% alcohol extract of the sclerotia of *G.umbellata* on the basis of modern chromatography and methods gave twenty four compounds, twenty two of which were identified by the spectroscopic analysis (including IR, ¹H-NMR, ¹³C-NMR, HMQC, HMBC, NOESY, EI-MS, FAB-MS and HREIMS), with two new compounds and eleven isolated from the fungus for the first time.

Two new ones were (20*S*,22*R*,24*R*)-16,22-epoxy-3 β ,14 α ,23 β ,25-tetrahydroxyergost-7-en-6-one(**P-2**) and (23*R*,24*R*,25*R*)-23,26-epoxy-3 β ,14 α ,21 α ,22 α -tetrahydroxyergost-7-en-6-one(**P-4**). The others were known to be ergosterol (**P-1**), 22,23-epoxy-3 β ,14 α ,20 β ,24 β -tetrahydroxyergost-7-en-6-one (**P-3**), polyporusterone B (**P-5**), polyporusterone A (**P-6**), N-(2'-hydroxytetracosanoyl)-1,3,4-trihydroxyl-2-octodecanine (**P-7**), 5-hydroxymethylfurfuraldehyde(**P-8**), mannitol (**P-9**), arabinitol (**P-10**), α -hydroxytetracosanoic ethyl ester (**P-11**), friedelin (**P-12**), physcion (**P-13**), chrysophanol (**P-14**), ergosterol peroxide (**P-15**), ergosta-7,22-dien-3-one (**P-16**), ergosta-7,22-dien-3,5,6-triol (**P-17**), 2-acetyl-1,3,8-trihydroxyxanthone (**P-18**), nicotinic acid (**P-19**), uridine (**P-20**), adenosine (**P-21**), uracil (**P-23**).

Chemical constituents of the fermented mycelia of *G.umbellata* were systematically studied for the first time with various adsorption material and isolated methods. Fifteen compounds were isolated from this material and twelve of them were identified by spectra analysis (including IR, UV, ¹H-NMR, ¹³C-NMR, EI-MS and FAB-MS). Apart from the compound butanedioic acid (**MP-E-3**), the rest were also found in the sclerotia, which were ergosterol (**MP-P-1**), ergosta-7,22-dien-3,5,6-triol (**MP-P-2**), ergosta-7,22-dien-3-one (**MP-P-3**), α -hydroxytetra-cosanoic ethyl ester (**MP-P-4**), polyporusterone B (**MP-D-1**), polyporusterone A (**MP-D-2**), N-(2'-hydroxytetracosanoyl)-1,3,4-trihydroxyl-2-octodecanine (**MP-D-4**), adenosine (**MP-E-1**), nicotinic acid (**MP-E-2**), uracil (**MP-B-1**),

uridine (MP-B-2).

Based on the studies on the chemical constituents of the sclerotia of *G.umbellata*, five polyporosterones isolated from the sclerotia were selected as the marker compounds, and a HPLC-DAD method was firstly developed for simultaneous quantitative determination of these five polyporosterones in the sclerotia of *G.umbellata*. The method can be used as an efficient method for quality control of *G.umbellata*. Furthermore, the cultured sclerotia and fermented mycelia were analyzed compared with the wild sclerotia with this method.

Key words: *Grifola umbellata*, Sclerotia, Fermented mycelia, Chemical constituents, Polyporosterones, HPLC-DAD, Quality control, Fingerprint