Novel Sesquiterpenes from the Fungus *Lactarius piperatus*

by **Ying Wang**, **Sheng-Ping Yang**, and **Jian-Min Yue***

State Key Laboratory of Drug Research, Institute of Materia Medica, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, 294 Taiyuan Road, Shanghai, 200031, P.R. China (tel: 86-21-64311833, fax: 86-21-64370269, e-mail: jmyue@mail.shcnc.ac.cn)

and

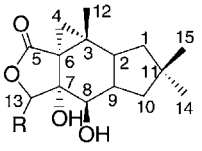
Sharon Chow and **William Kitching***

Department of Chemistry, The University of Queensland, Brisbane, 4072, Australia (e-mail: kitching@chemistry.uq.edu.au)

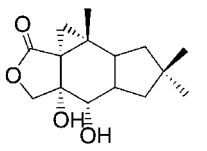
Four novel sesquiterpenes, namely 7α,8β,13-trihydroxy-5,13-marasmanolide (**2**), isoplorantinone (**5**), 4,8,14-trihydroxyilludala-2,6,8-triene (**6**), and 8-hydroxy-8,9-secolactara-1,6-dien-5,13-olide (**10**), together with six known ones, 7α,8β-dihydroxy-5,13-marasmanolide (**1**), 7α,8α-dihydroxy-5,13-marasmanolide (**3**), isolactarorufin (**4**), blennin A (**7**), blennin D (**8**), and lactarorufin (**9**), were isolated from the ethanolic extract of *Lactarius piperatus*. The structures of these sesquiterpenes, representing diversified structural types, were determined mainly by spectroscopic methods, especially 2D-NMR techniques. The structure of **6** was further confirmed by a single-crystal X-ray-diffraction determination.

Introduction. – The fungus *Lactarius piperatus* (Fr.) S. F. GRAY (family Russulaceae, Basidiomycotina) is widely distributed in China. The sample of *L. piperatus* used in this study was collected from the Kunming area of Yunnan province, China. The whole body of this inedible fungus exudes a large amount of milky juice when the mushroom is cut or broken, and has a very hot taste (called 'hot milk mushroom' by local residents). The ethanolic extract has been reported to inhibit the growth of several tumor cell lines [1]. *L. piperatus*, growing in Europe and Japan, has been investigated, and a new amino acid and a few sesquiterpenes were isolated [2], but that growing in China has not previously been investigated chemically. A series of sesquiterpenes, belonging to the marasmane, lactarane, isolactarane, and secolactarane types, has been isolated from the genus of *Lactarius* [2–4]. These sesquiterpenes provide a chemical defense system against parasites and predators [3][4]. In the current project, four new sesquiterpenes, namely 7α,8β,13-trihydroxy-5,13-marasmanolide (**2**), isoplorantinone (**5**), 4,8,14-trihydroxyilludala-2,6,8-triene (**6**), and 8-hydroxy-8,9-secolactara-1,6-dien-5,13-olide (**10**), along with six known sesquiterpenes, 7α,8β-dihydroxy-5,13-marasmanolide (**1**), 7α,8α-dihydroxy-5,13-marasmanolide (**3**), isolactarorufin (**4**), blennin A (**7**), blennin D (**8**), and lactarorufin (**9**), were isolated, and their structures were determined mainly by spectroscopic methods, especially 2D-NMR techniques. The structure deduced for compound **6** was confirmed by a single-crystal X-ray determination. It is of interest that the compounds isolated from this fungus represent diverse types of sesquiterpenes, including marasmane (**1**–**3**),

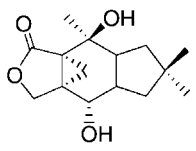
lactarane (**7**–**9**), isolactarane (**4**), secolactarane (**10**), protoilludane (**5**), and illudane (**6**). The protoilludane and illudane types of sesquiterpenes have been found in the genus of *Lactarius* for the first time in the present work. The diversified structural types from this mushroom can be rationalized biogenetically as shown in the *Scheme* (see below). Herein, we report the isolation and structural elucidation of ten sesquiterpenes from the fungus *L. piperatus.*



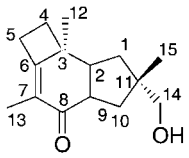
1 R=H
2 R=OH



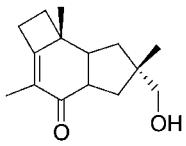
3



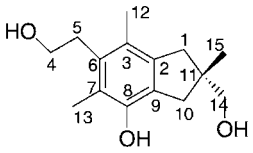
4



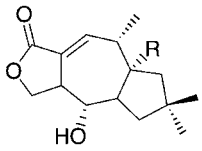
5



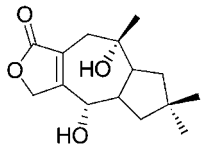
5a



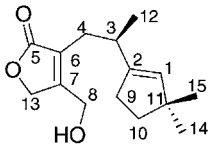
6



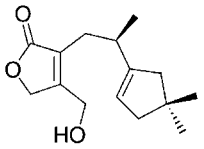
7 R=H
8 R=OH



9



10



10a

2. Results and Discussion. – 7α,8β,13-Trihydroxy-5,13-marasmanolide (**2**) with the molecular formula $C_{15}H_{22}O_5$ as determined by HR-EI-MS (*m/z* 264.1341 ($C_{15}H_{20}O_4$ $[M-H_2O]^+$; calc. 264.1362), was obtained as a gum. The ^{1}H- (*Table 1*) and ^{13}C-NMR (*Table 2*) spectra showed signals of three Me (δ 1.29 (*s*, 3 H); 1.18 (*s*, 3 H); 1.08 (*s*, 3 H)), three CH_2, four CH groups, and five quaternary C-atoms. A broad IR band at 3354 cm^{-1} supported the presence of OH groups. Analysis of the ^{1}H- and ^{13}C-NMR spectra of **2**, and those of 7α,8β-dihydroxy-5,13-marasmanolide (**1**), which was also isolated from this fungus, indicated that the structure of **2** was very similar to that of **1**, but with possibly a OH group at C(13) to form a hemiacetal in **2**. The ^{13}C signal at δ

105.1 supported the presence of a hemiacetal group. The ^{1}H- and ^{13}C-NMR signals were completely assigned by ^{1}H,^{1}H-COSY and HMQC spectra, and comparison with the data assigned for compound **1**. The structure of **2** was finally confirmed by an HMBC experiment (*Fig. 1*), in which the 'loose ends' resulting from the insertion of quaternary C-atoms C(3), C(5), C(6), C(7), and C(11) into the fragments established by ^{1}H,^{1}H-COSY and HMQC, could be fully connected. Therefore, the structure of **2** was determined to be 7α,8β,13-trihydroxy-5,13-marasmanolide.

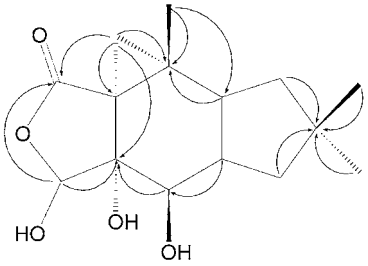


Fig. 1. *Key HMBC correlations of* **2**

Isoplorantinone (**5**), with the molecular formula $C_{15}H_{22}O_2$ determined by HR-EI-MS (*m/z* 234.1610; calc. 234.1620), was obtained as white powder, and all fifteen ^{13}C signals were resolved in the ^{13}C-NMR (*Table 2*). The ^{1}H-NMR spectrum (*Table 1*) showed signals of three angular Me groups (δ 1.78 (*s*, 3 H); 1.54 (*s*, 3 H); 1.24 (*s*, 3 H)). The UV (249 nm) and IR (1659 cm^{-1}) absorptions indicated the presence of a disubstituted, six-membered-ring α,β-unsaturated ketone [5], and this was supported by the signals at δ 204.5, 173.4, and 126.2 in the ^{13}C-NMR spectrum [6]. The ^{1}H- (δ 3.47 and 3.44) and ^{13}C-NMR (δ 72.1), and IR (3447 cm^{-1}) spectra indicated the presence of one CH_2OH group. The ^{1}H,^{1}H-COSY and HMQC spectra were recorded to acquire the major structural fragments of compound **5**. The HMBC spectrum (*Fig. 2,a*) was performed to link the major fragments with the quaternary C-atoms to obtain the planar structure of **5**, which is the same as plorantinone A (**5a**) [6]. The ^{1}H- (*Table 1*) and ^{13}C-NMR (*Table 2*) data of **5** were very different from those of plorantinone A (**5a**) (for the same solvent, $CDCl_3$) reported in [6], suggesting that compound **5** is probably a stereoisomer of plorantinone A (**5a**). A NOESY spectrum (*Fig. 3,b*) was recorded to establish the relative configuration of **5**. The strong correlations between H_α–C(2) and H_α–C(9), CH_3(12)and H_α–C(9), CH_3(12) and H_α–C(4), CH_3(12) and H_α–C(5), and a weak correlation between CH_3(12) and H_α–C(2), clearly indicated the presence of a 12α-Me group. The CH_3(15) correlated with H_β–C(1) and H_β–C(10), and, therefore, the 15-Me group was β-oriented. The structure of **5** was elucidated to be isoplorantinone A. The NOESY spectrum of **5** exhibited strong correlations between CH_3(12) and H_α–C(9), and weak correlation between CH_3(12) and H_α–C(2), indicating that the six-membered ring may adopt a boat conformation. The strong correlation between H_α–C(1) and H_α–C(10) is consistent with an envelope-conformation for the cyclopentane ring.

A 3D structure (*Fig. 2,c*) of **5** generated by the molecular-modeling program (CS Chem3D Pro Version 5.0 with the MM2 force field for energy minimization) provided a low-energy conformation for **5** and close contacts of the atoms in space, especially for

Table 1. ^{1}*H-NMR Chemical Shifts for Compounds* **2**, **5**, **6**, *and* **10**

H-Atom	**2**[a])	**5**[a])	**5**[b])	**6**[a])	**10**[c])
H_α–C(1)	1.62 (*m*, 2 H)	1.87 (*dd*, 12.7, 7.1)	1.70 (*dd*, *J* = 10.8, 7.1)	3.01 (*d*, *J* = 15.8)	5.19 (*s*)
H_β–C(1)		1.39 (*dd*, *J* = 12.7, 12.1)	1.15 (*dd*, *J* = 12.1, 10.8)	2.69 (*d*, *J* = 15.8)	
H–C(2)	2.60 (br. *d*, *J* = 5.4)	2.41 (*ddd*, *J* = 13.2, 12.1, 7.1)	2.19 (*ddd*, *J* = 13.2, 12.1, 7.1)		
H–C(3)					2.53 (*m*)
H_α–C(4)	1.12 (*d*, *J* = 4.4)	2.04 (*dd*, *J* = 17.3, 9.0)	1.80 (*dd*, *J* = 17.2, 9.0)	3.73 (*t*, *J* = 8.3, 2 H)	2.17 (*dd*, *J* = 13.6, 8.3)
H_β–C(4)	1.40 (*d*, *J* = 4.4)	2.14 (*dt*, *J* = 9.0, 2.5)	1.89 (*dt*, *J* = 9.0, 2.7)		2.37 (*dd*, *J* = 13.6, 6.6)
H_α–C(5)		3.33 (*m*)	3.08 (*m*)	3.07 (*t*, *J* = 8.3, 2 H)	
H_β–C(5)		2.94 (*ddd*, *J* = 2.5, 7.7, 15.6)	2.69 (*ddd*, *J* = 2.7, 7.9, 15.4)		
H–C(6)					
H–C(7)					
H–C(8)	3.99 (br. *d*, *J* = 11.8)				4.54 (*s*, 2 H)
H–C(9)	1.86 (br. *d*, *J* = 13.8)	2.81 (*ddd*, *J* = 13.3, 11.2, 7.5)	2.54 (*ddd*, *J* = 13.4, 10.1, 7.5)		2.04 (*m*, 2 H)
H_α–C(10)	1.55 (*dd*, *J* = 13.9, 7.4)	1.70 (*dd*, *J* = 13.0, 11.2)	1.51 (*dd*, *J* = 11.2, 7.5)	2.98 (*d*, *J* = 15.8)	2.08 (*m*, 2 H)
H_β–C(10)	1.86 (br. *d*, *J* = 13.8)	1.81 (*dd*, *J* = 13.0, 7.5)	1.68 (*dd*, *J* = 11.2, 10.1)	2.71 (*d*, *J* = 15.8)	
H–C(11)					
H–C(12)	1.29 (*s*, 3 H)	1.54 (*s*, 3 H)	1.31 (*s*, 3 H)	2.32 (*s*, 3 H)	0.94 (*d*, *J* = 6.8, 3 H)
H–C(13)	5.67 (*s*)	1.78 (*s*, 3 H)	1.60 (*s*, 3 H)	2.36 (*s*, 3 H)	4.81 (*s*, 2 H)
H_α–C(14)	1.18 (*s*, 3 H)	3.44 (*d*, *J* = 10.6)	3.2 (*s*, 2 H)	3.60 (*s*, 2 H)	1.01 (*s*, 3 H)
H_β–C(14)		3.47 (*d*, *J* = 10.6)			
H–C(15)	1.08 (*s*, 3 H)	1.24 (*s*, 3 H)	1.1 (*s*, 3 H)	1.33 (*s*, 3 H)	1.02 (*s*, 3 H)

[a]) Measured in CD_3OD. [b]) Measured in $CDCl_3$. [c]) Measured in (D_6)acetone.

Table 2. *^{13}C-NMR Data for Compounds* **2**, **5**, **6**, *and* **10**

C-Atom	**2**[a])	**5**[a])	**5**[b])	**5a**[b])[c])	**6**[a])	**10**[d])
C(1)	45.2	37.9	36.6	35.5	43.8	122.2
C(2)	47.6	57.4	55.4	46.8	141.6	147.9
C(3)	33.3	50.6	48.9	46.1	125.2	34.8
C(4)	25.5	34.3	33.1	35.0	62.5	30.2
C(5)	180.1	31.4	30.3	28.0	34.5	175.3
C(6)	36.4	173.4	169.8	168.6	135.2	125.2
C(7)	78.3	126.2	125.2	125.8	122.9	162.7
C(8)	67.1	204.5	201.7	201.4	150.5	57.6
C(9)	40.8	50.6	49.0	51.5	127.4	47.9
C(10)	43.3	37.4	35.8	38.8	41.1	47.9
C(11)	38.0	43.5	42.1	44.4	45.7	38.6
C(12)	17.9	16.6	16.2	24.0	16.1	19.1
C(13)	105.1	9.7	9.4	10.1	12.4	71.0
C(14)	33.3	72.1	71.5	70.7	71.2	30.2
C(15)	33.3	27.4	26.5	24.4	25.4	30.2

[a]) Measured in CD_3OD. [b]) Measured in $CDCl_3$. [c]) **5a** stands for the data of plorantinone A from [5]. [d]) Measured in (D_6)acetone.

some key H- and C-atoms (see *Exper. Part*), which were consistent with the configuration and conformation of **5** assigned by the NOESY data. The computer modeling also indicated the boat conformation for the six-membered ring and a slightly twisted envelope conformation for the five-membered ring. Comparison of the ^{13}C-NMR data of **5** with those for **5a** (both recorded in $CDCl_3$) showed that the signal for C(2) in **5** was shifted downfield (δ 7.6), and that for C(12) was shifted upfield (δ 7.8), which can be interpreted by the deshielding and shielding effects, respectively, of the α,β-unsaturated ketone group in the slightly twisted boat conformation.

4,8,14-Trihydroxyilludala-2,6,8-triene (**6**) with the molecular formula $C_{15}H_{22}O_3$ as determined from HR-EI-MS (*m/z* 250.1568; calc. 250.1569) was obtained as a white microcrystalline material. The ^{1}H-NMR spectrum (*Table 1*) showed signals of three Me groups (δ 1.33 (*s*, 3 H); 2.32 (*s*, 3 H); 2.36 (*s*, 3 H)). Broad IR absorption bands at 3363 cm^{-1} indicated the presence of OH groups. Two primary alcohol groups were consistent with ^{1}H-NMR signals at δ 3.60 (*s*, 2 H) and δ 3.73 (*t*, $J = 8.3$, 2 H), and ^{13}C-NMR signals at δ 71.2 and 62.5. Signals of six quaternary sp^2-C-atoms appeared in the ^{13}C-NMR (*Table 2*), indicating the existence of a persubstituted benzene, and supporting by typical IR absorption bands at 1587, 1500, and 1450 cm^{-1}. All ^{1}H signals were assigned to the corresponding C-atoms by an HMQC experiment. Analysis of coupling constants and comparison of the ^{1}H- and ^{13}C-NMR data with those of **5** indicated the presence of the structural fragments CH_2CH_2OH and $CH_2C(Me)$-$(CH_2OH)CH_2$ in **6**. The linkage of the two structural fragments and the remaining two Me groups to the aromatic core in **6** was achieved by a high-quality HMBC spectrum (*Fig. 3*,*a*). The remaining OH group was finally assigned to C(8) at δ 150.5. The structure of **6** was, thus, elucidated to be 4,8,14-trihydroxyilludala-2,6,8-triene, and was finally confirmed by a single-crystal X-ray-diffraction study (*Fig. 3*,*b*).

8-Hydroxy-8,9-secolactara-1,6-dien-5,13-olide (**10**), with the molecular formula $C_{15}H_{22}O_3$ as deduced from HR-EI-MS (250.1570; calc. 250.1569), was obtained as white

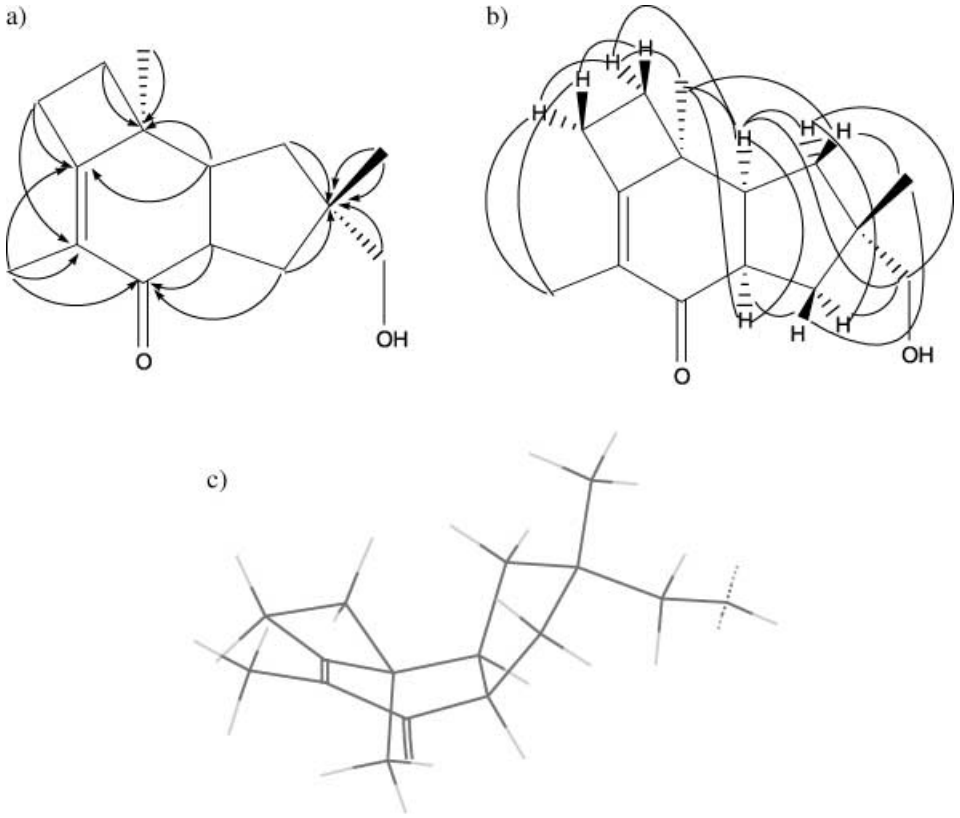


Fig. 2. a) *Key HMBC correlations of* **5**. b) *Key NOE correlations of* **5**. c) *3D View of* **5** *generated from computer modeling.*

powder. The ^{1}H-NMR spectrum indicated the presence of three Me groups (δ 1.02 (*s*, 3 H); 1.01 (*s*, 3 H); 0.94 (*d*, $J = 6.8$, 3 H)), and an olefinic H-atom (δ 5.19 (*s*)). An IR absorption band at 1672 cm^{-1}, and ^{13}C-NMR signals at δ 175.3, 162.7, and 125.2 indicated the presence of an α,β-unsaturated lactone [7]. One ^{1}H signal at δ 5.19 (*s*) in the ^{1}H-NMR and two ^{13}C signals at δ 147.9 (quaternary sp^2-C-atom) and 122.2 (tertiary sp^2-C-atom) in the ^{13}C-NMR spectra suggested a trisubstituted C=C bond. Fifteen ^{13}C signals appeared in the ^{13}C-NMR corresponding to the fifteen C-atoms of the molecular formula. The complete assignments of ^{1}H and ^{13}C signals, and identification of major structural fragments were achieved from the ^{1}H,^{1}H-COSY and HMBC spectra. Comparison of the ^{1}H- and ^{13}C-NMR data of compound **10** and blennin C (**10a**) [7] indicated that the two compounds had very similar structures, and the only difference was the location of the trisubstituted C=C bond. The singlet ^{1}H signal at δ 5.19 suggested the presence of the C(1)=C(2) rather than the C(2)=C(9) bond. The structure of **10** was finally confirmed by HMBC spectrum (*Fig. 4*). The structure of **10** was, therefore, elucidated to be 8-hydroxy-8,9-secolactara-1,6-dien-5,13-olide.

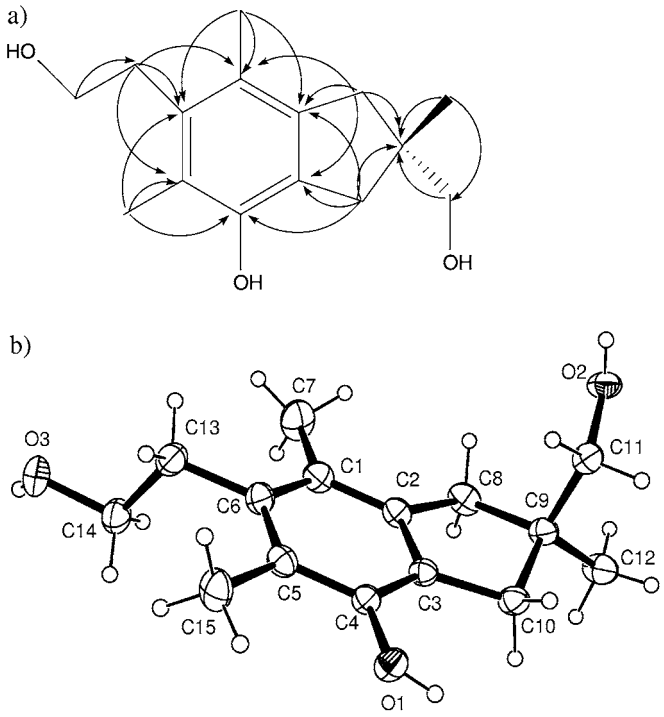


Fig. 3. a) *Key HMBC correlations of* **6**. b) *Single-crystal X-ray structure of* **6**.

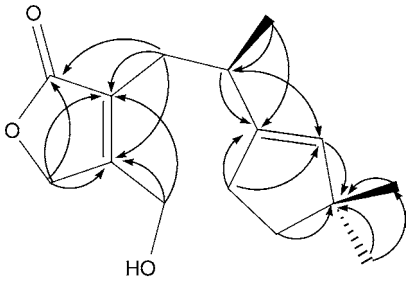


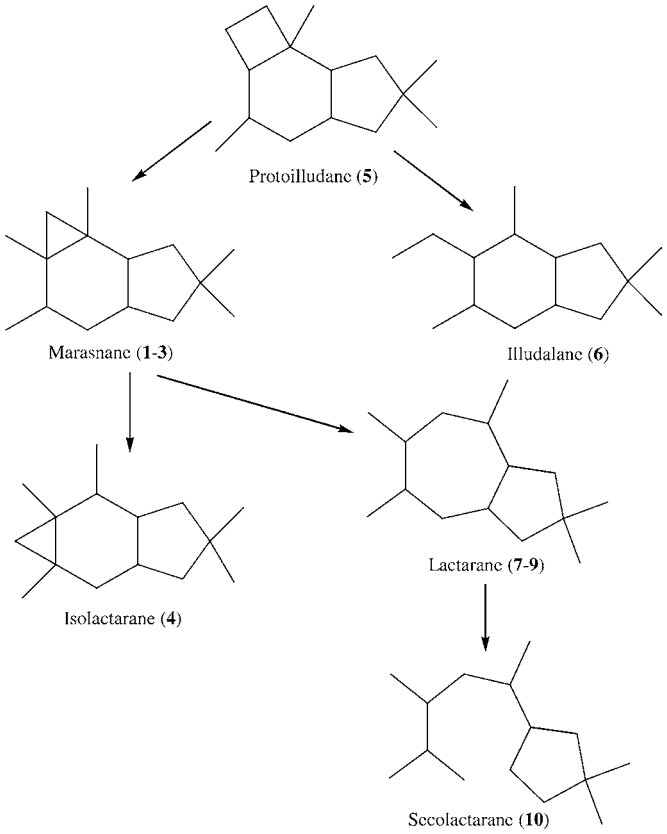
Fig. 4. *Key HMBC correlations of* **10**

The known compounds **1**, **3**, **4**, **7**, **8**, and **9** were identified as 7α,8β-dihydroxy-5,13-marasmanolide [8], 7α,8α-dihydroxy-5,13-marasmanolide [8], isolactarorufin [9], blennin A [10], blennin D [11], and lactarorufin [12] on the basis of ^{1}H- and ^{13}C-NMR, EI-MS, HMQC, and HMBC data.

The co-occurrence of this diverse group of structural types, comprising marasmane (**1**–**3**), lactarane (**7**–**9**), isolactarane (**4**), secolactarane (**10**), protoilludane (**5**), and illudane (**6**) systems, in one species of the genus *Lactarius* is unique, and it is of considerable interest from a biogenetic view. To our knowledge, none of the protoilludane and illudane types of sesquiterpenes have been in the genus of *Lactarius*

before this work. A biogenetic pathway from protoilludane to marasnane (illudalane) and then to isolactarane was proposed by the isolation of marasmic acid from *Marasmius conigenus* and isolactarorufin from *Lactarius rufus* [9][13]. Finding sesquiterpenes belonging to marasnane, lactarane, and secolactarane types in the genus of *Lactarius* has led to the suggested operation of a biosynthetic pathway from marasnane to lactarane and then to the secolactarane type of sesquiterpenes [3][14]. The isolation of diversified structural types of sesquiterpenes from *L. piperatus* has provided the opportunity to rationalize the biogenetic conversions for these types of sesquiterpenes (*Scheme*).

Scheme. *A Possible Biosynthetic Pathway for All Six Types of Sesquiterpenes*



Experimental Part

General. All solvents used were of anal. grade (*Shanghai Chemical Plant*). Column chromatography (CC): silica gel (200–300 mesh). TLC: precoated silica-gel *GF 254* plates (*Qingdao Haiyang Chemical Plant*). The *MCI Gel CHP20P* (75–150 μ) (*Mitsubishi Chemical Industries Ltd.*) and *C18* reversed-phased silica gel (150–200 mesh; *Merck*) were also used for CC. Optical rotations: *Perkin-Elmer 341* polarimeter. UV Spectra: *Shimadzu UV-210A* spectrometer. IR Spectra: *Perkin-Elmer 577* spectrometer. NMR Spectra: *Bruker AM-400*

spectrometer with TMS as internal standard. EI-MS (70 eV): *Finnigan MAT 95* mass spectrometer. In X-ray crystallography, cell constants were determined by a least-squares fit to the setting parameters of 25 independent reflections measured on an *Enraf-Nonius CAD4* four-circle diffractometer with graphite monochromated MoK_α radiation (0.71073 Å) and operating in the ω-2θ scan mode. Data reduction and empirical absorption corrections (ψ-scans) were performed with the XTAL package.

Plant material. Lactarius piperatus (Fr.) S. F. GRAY was collected from the Kunming area in Yunnan province of China and authenticated by Prof. *Mu Zang*, Kunming Institute of Botany, where a voucher specimen labeled as HKAS 30213, was deposited.

Extraction and Isolation. The fresh mushroom (5 kg) was extracted with 95% EtOH and yielded 91 g of crude extract, which was then suspended in 2 l of H_2O. The suspension was partitioned with AcOEt acetate (4 × 200 ml) to give an AcOEt-soluble portion, and a H_2O-soluble fraction. After removal of the AcOEt under reduced pressure, 49 g of dark residue was obtained, and this was subjected to silica-gel chromatography, eluted with a stepwise gradient solvent system of petroleum/acetone 10:0 to 5:5, followed by MeOH, to afford four major fractions (monitored by TLC). *Fraction 1* consisted mainly of fatty acids. *Fr. 4* was much smaller and complex. The separation and purification were focused on *Fr. 2* and *3*, in which the sesquiterpenes were concentrated. *Fr. 2* (2.62 g) was subjected to a *MCI* gel column eluting with 55% MeOH in H_2O, followed by silica gel eluting with $CHCl_3$/acetone 40:1, and then a reversed-phase *C18* silica-gel column, eluting with 65% MeOH in H_2O. This procedure afforded compounds **3** (6.4 mg), **5** (14 mg), **7** (38 mg), and **10** (6.4 mg). *Fr. 3* was subjected to a MCI gel column eluting with 50% methanol in water, followed by a silica-gel column eluted with petroleum/EtOAc (1:1), and then extensively separated on reverse phased C-18 silica-gel columns eluting with MeOH in H_2O with a changeable ratio. This afforded compounds **1** (10 mg), **2** (5 mg), **4** (40 mg), **6** (20 mg), **8** (30 mg) and **9** (90 mg).

7α,8β,13-Trihydroxy-5,13-marasmanolide (**2**). White powder. $[\alpha]_D^{20} = +27.9$ ($c = 0.19$, MeOH). UV (MeOH): no absorption band between 200–400 nm was observed. IR (KBr): 3354, 2949, 2872, 1757, 1443, 1363, 1175, 1082, 1055, 1018, 945, 754. ^{1}H- and ^{13}C-NMR: see *Tables 1* and *2*. EI-MS: 264 (12, $[M - H_2O]^+$), 249 (17), 246 (43), 236 (75), 235 (31), 231 (22), 220 (20), 219 (51), 218 (51), 217 (41), 208 (20), 203 (26), 190 (38), 189 (25), 175 (34), 153 (27), 152 (59), 135 (68), 130 (21), 123 (100), 122 (25), 121 (24), 109 (24), 107 (47), 105 (25), 95 (44), 93 (23), 91 (45), 81 (44), 79 (26), 77 (24), 55 (27). HR-EI-MS: 264.1341 ($C_{15}H_{20}O_4^+$, $[M - H_2O]$; calc. 264.1362).

Isoplorantinone (**5**). White powder. $[\alpha]_D^{20} = -71.2$ (c = 1.25, MeOH). UV (MeOH): λ_{max} 249. IR (KBr): 3447, 2952, 2886, 1659, 1454, 1375, 1317, 1240, 1043. ^{1}H- and ^{13}C-NMR: see *Tables 1* and *2*. EI-MS: 234 (36), 219 (24), 205 (100), 201 (54), 187 (92), 173 (76), 159 (53), 145 (38), 131 (30), 119 (37), 105 (45). HR-EI-MS: 234.1610 ($C_{15}H_{22}O_2$, M^+; calc. 234.1620). The close contacts of atoms in space calculated for compound **5** [Å]: C(6) ⋯ C(9): 2.818; C(8) ⋯ H_β–C(10): 2.484; H_α–C(1) ⋯ C(14): 2.239.

4,8,14-Trihydroxyilludala-2,6,8-triene (**6**). Microcrystalline solid. $[\alpha]_D^{20} = +1.2$ ($c = 1.14$, MeOH). UV (MeOH): λ_{max} 216. IR (KBr): 3363, 2953, 1587, 1500, 1450, 1317, 1292, 1261, 1101, 1041, 690. ^{1}H- and ^{13}C-NMR: see *Tables 1* and *2*. EI-MS: 250 (58), 219 (100), 201 (62), 187 (22), 173 (20). HR-EI-MS: 250.1568 ($C_{15}H_{22}O_3^+$, M^+; calc. 250.1569).

8-Hydroxy-8,9-secolactara-1,6-dien-5,13-olide (**10**). White powder. $[\alpha]_D^{20} = -2.0$ (c = 0.60, MeOH). IR (KBr): 3435, 2953, 2926, 1738, 1672, 1456, 1362, 1086, 1045. ^{1}H- and ^{13}C-NMR: see *Tables 1* and *2*. EI-MS: 250 (10), 235 (60), 219 (16), 149 (43), 123 (100), 95 (25), 81 (44). HR-EI-MS: 250.1570 ($C_{15}H_{22}O_3^+$, M^+; calc. 250.1569).

Financial support of the *National Scientific Foundation* (for *J. M. Yue*, 30025044) and from the *Ministry of Science and Technology* (2002CB512807) is gratefully acknowledged. We thank Prof. *Mu Zang*, Kuming Institute of Botany, for the identification of the fungus and Dr. *Paul Bernhardt*, University of Queensland, for the X-ray crystal-structure determination of **6**. We gratefully acknowledge the support from the exchange program between the *Chinese Academy of Sciences and Australian Academy of Science.*

REFERENCES

[1] X. Mo, J. Wang, J. Mo, J. Mo, T. Liu, S. Xue, Z. Zhang, H. Zhang, Y. Zhao, X. Guo, C. Geng, X. Han, 'Economic fungi of China', Scientific Press: Peking, 1995, pp. 403.

[2] a) S. Fushiya, F. Watari, T. Tashiro, G. Kusano, S. Nozoe, *Chem. Pharm. Bull.* **1988**, *36*, 1366; b) O. Sterner, R. Bergman, C. Franzen, B. Wickberg, *Tetrahedron Lett.* **1985**, *26*, 3163; c) Y. Yaoita, K. Machida, M. Kikuchi, *Chem. Pharm. Bull.* **1999**, *47*, 894.

[3] a) M. De Bernardi, L. Garlaschelli, L. Toma, G. Vidari, P. Vita-Finzi, *Tetrahedron* **1993**, *49*, 1489; b) O. Sterner, T. Anke, W. S. Sheldrick, W. Steglich, *Tetrahedron* **1990**, *46*, 2389.
[4] O. Sterner, R. Bergman, J. Kihlberg, B. Wickberg, *J. Nat. Prod.* **1985**, *48*, 279.
[5] D. H. Williams, I. Fleming, 'Spectroscopic Methods in Organic Chemistry', McGraw-Hill Book Company, London, 1989; pp. 15.
[6] M. Clericuzio, J. Fu, F. Pan, Z. Pang, O. Sterner, *Tetrahedron* **1997**, *53*, 9735.
[7] G. Vidar, D. Bernardi, P. Vita-Finzi, G. Fronza, *Phytochemistry* **1976**, *15*, 1953.
[8] W. M. Daniewski, M. Gumulka, K. Ptaszyna, P. Skibicki, J. Krajewski, P. Gluzinski, *Phytochemistry* **1992**, *31*, 913.
[9] W. M. Daniewski, M. Kocor, S. Thoren, *Polish Journal of Chemistry* **1978**, *52*, 561 – 572.
[10] M. D. Bernard, G. Fronza, G. Mellerio, G. Vidari, P. V. Finzi, *Phytochemistry* **1980**, *19*, 99.
[11] L. Garlaschelli, L. Toma, G. Vidari, D. Colombo, *Tetrahedron* **1994**, *50*, 1211.
[12] W. M. Daniewski, M. Gumulka, E. Pankowska, K. Ptaszynska, E. Bloszyk, U. Jacobsson, T. Norin, *Phytochemistry* **1993**, *32*, 1499.
[13] J. J. Duggan, P. De Mayyo, M. Nisbet, J. R. Robinson, M. Anchel, *J. Am. Chem. Soc.* **1966**, *88*, 2838.
[14] M. De Bernardi, G. Fronza, A. Scilingo, G. Vidari, P. Vita-Finzi, *Tetrahedron* **1986**, *42*, 4277.

Received March 19, 2003