was worked-up as usual and the product was further purified by PLC over silica gel plates using C_6H_6–hexane as eluents. The purified demethylated product (yield 6 mg) was found to be completely identical to lansiol (**1**) isolated from *C. lansium*, on TLC, mp, mmp and superimposable IR spectrum.

Reduction of lansiol. A mixture of (**1**) (50 mg) dissolved in $CHCl_3$ (20 ml) and Adam's catalyst (25 mg) was stirred for 2 hr at room temp. in an atmosphere of H_2. The solvent was removed and the product (**5**) was crystallized with MeOH–$CHCl_3$; yield, 48 mg; mp 198–200°; M^+ *m/z* 470; $[\alpha]_D^{25}$ +51.5 ($CHCl_3$; *c* 0.8).

Acknowledgement—We are grateful to Dr D. Prakash for his association in early stages of this work.

REFERENCES

1. Manandhar, M. D., Shoeb, A., Kapil, R. S. and Popli, S. P. (1977) *Experientia* **33**, 153.
2. Prakash, D., Raj, K., Kapil, R. S. and Popli, S. P. (1978) *Phytochemistry* **17**, 1194.
3. Prakash, D., Raj, K., Kapil, R. S. and Popli, S. P. (1980) *Indian J. Chem.* **19**, 1075.
4. Lakshmi, V., Raj, K., Kapil, R. S. and Patnaik, G. K. (1987) *Indian Drugs* **24**, 285.
5. Lakshmi, V., Prakash, D., Raj, K. and Kapil, R. S. (1984) *Phytochemistry* **23**, 2629.
6. Lakshmi, V., Raj, K., Kapil, R. S. and Patnaik, G. K. (1988) *Indian Drugs* (in press).
7. Prakash, D., Lakshmi, V., Raj, K. and Kapil, R. S. (1989) *Fitoterapia* (in press).
8. Ritchie, E., Senior, R. G. and Taylor, W. C. (1969) *Aust. J. Chem.* **22**, 2371.
9. Hui, W. H., Luke, K., Arthur, H. R. and Loo, S. N. (1971) *J. Chem. Soc.* (*C*) 2826.
10. Vorbruggen, H., Pakrashi, S. C. and Djerassi, C. (1963) *Annalen* **57**, 668.
11. Wyllie, S. G. and Djerassi, C. (1968) *J. Org Chem.* **33**, 305.

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ERGOSTA-7,22-DIEN-3β-OL GLYCOSIDE FROM *TYLOPILUS NEOFELLEUS*

YOSHIHISA TAKAISHI,* TAKASHI OHASHI and TOSHIAKI TOMIMATSU

Faculty of Pharmaceutical Sciences, The University of Tokushima, Shomachi 1-78, Tokushima 770, Japan

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Key Word Index—*Tylopilus neofelleus*; Basidiomycete; higher fungi; glycoside; ergosta-7,22-dien-3β-*O*-glucopyranoside.

Abstract—A new glycoside of ergosta-7,22-dien-3β-ol was isolated from the fresh fruit body of *Tylopilus neofelleus* and its structure was established by chemical and spectroscopic means. Basidiomycetes produce many ergosterol analogues, but the presence of the glycoside of an ergosterol derivative is the first finding from a natural source.

INTRODUCTION

We have investigated the constituents of Basidiomycetes and elucidated the structure of three new lanostane triterpenes having a δ-lactone in the side chain from *Astraeus hygrometricus* [1], and some ergosterol analogues from *Inonotus mikadoi* [2]. Now, we have carried out an investigation of the constituents of *Tylopilus neofelleus* (Boletaceae). In this paper we wish to report the structural elucidation of a new glycoside, ergosta-7,22-dien-3β-*O*-glucopyranoside (**2**).

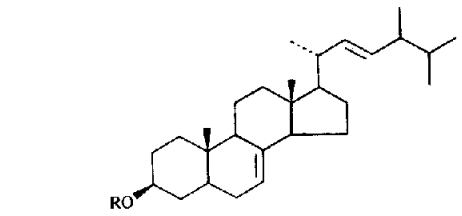
RESULTS AND DISCUSSION

The methanol extract from the fresh fruit bodies of *T. neofelleus* Hongo was partitioned with water and ethyl acetate. The ethyl acetate extract was separated using silica gel column chromatography to give compounds **1**–**3**.

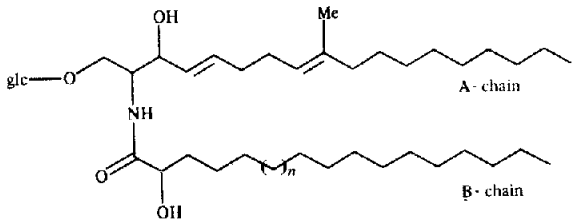
*Author to whom correspondence should be addressed.

Compound **1**, $C_{28}H_{46}O$, showed the presence of two tertiary methyls, four secondary methyls, one proton attached to an oxygen bearing carbon and three protons attached to double bond in the ^1H NMR spectrum. Its decoupled ^{13}C NMR spectrum contained 28 peaks (Table 1). In the mass spectrum of **1**, fragment ion peaks were very similar to those of an ergosterol derivative [3]. These facts indicated that compound **1** was ergosta-7,22-dien-3β-ol, confirmed by direct comparison with literature data [4].

Compound **2**, $C_{34}H_{56}O_6$, showed absorption at 3500 (OH) cm^{-1} in the IR spectrum. The ^1H NMR spectrum of **2** showed the presence of two tertiary methyls, four secondary methyls, and a glycoside moiety. The decoupled ^{13}C NMR spectrum (Table 1) of **2** contained 34 signals, six of which were assigned to a glucose moiety [5]. The remaining signals were very similar to those of compound **1** except the signals at δ 30.1 (*t*), 77.5 (*d*), and 34.9 (*t*). This suggested that the structure of compound **2** was ergosta-7,22-dien-3β-*O*-glucoside, and the ^{13}C NMR differences (a downfield shift of the signal at δ



1 R = H
2 R = glc



3a n = 1
3b n = 2

Table 1. ^{13}C NMR spectral data of compounds **1** and **2** (50.1 MHz, C_5D_5N)

C	1	2	C	1	2
1	37.7 *t*	37.5 *t*	18	12.3 *q*	12.3 *q*
2	32.3 *t*	30.1 *t*	19	13.2 *q*	13.0 *q*
3	70.3 *d*	77.5 *d*	20	40.8 *d*	40.6 *d*
4	38.9 *t*	34.9 *t*	21	21.4 *q*	21.4 *q*
5	40.7 *t*	40.5 *t*	22	136.2 *d*	136.2 *d*
6	30.1 *t*	30.1 *t*	23	132.1 *d*	132.3 *d*
7	118.0 *d*	117.9 *d*	24	43.1 *d*	43.1 *d*
8	139.6 *s*	139.6 *s*	25	33.3 *d*	33.4 *d*
9	49.8 *d*	49.9 *d*	26	19.9 *q*	19.8 *q*
10	34.6 *s*	34.7 *s*	27	20.1 *q*	20.1 *q*
11	21.8 *t*	21.8 *t*	28	17.8 *q*	17.9 *q*
12	39.7 *t*	39.8 *t*	1′		102.4 *d*
13	43.5 *s*	43.6 *s*	2′		75.3 *d*
14	55.4 *d*	55.4 *d*	3′		78.1 *d*
15	23.3 *t*	23.3 *t*	4′		72.0 *d*
16	28.5 *t*	28.4 *t*	5′		78.5 *d*
17	56.2 *d*	56.4 *d*	6′		63.1 *t*

77.5 and upfield shifts of signals at δ 30.1 and 34.9, see Table 1) from those of compound **1** were explained by glycosylation shifts on C-3 [6]. The mass spectrum of compound **2** indicated the molecular ion at m/z 560.4060 ($C_{34}H_{56}O_6$) and fragment ions at m/z 381 [M − glucose + H], 273 [M − glucose − side chain (C_9H_{17}) + H], and 125 [C_9H_{17}]. The presence of glucose and ergosta-7,22-diene-3β-ol moieties in compound **2** was confirmed by TLC of the acid hydrolysis products of **2**. In the ^1H NMR spectrum, the coupling constant of the anomeric proton at δ 4.92 (1H, *d*, J = 7.6 Hz) indicated that the glucose was attached to the remainder of the molecule via a β-linkage. Thus the structure of compound **2** was deduced to be ergosta-7,22-diene-3β-*O*-β-glucopyranoside as shown. Basidiomycetes contain many ergosterol analogues. It has been suggested that glycosides of ergosterol analogues might be responsible for the beneficial effects of certain yeast and plant extracts in rats with experimental rickets [7]. Synthetic ergosterol glucoside was found to have similar effects. To the best of our knowledge, the isolation of glycosides of ergosterol analogues from natural sources have not been reported in the literature. This is the first finding of the presence of the glycoside from a natural source.

Compound **3** showed absorptions at 3360 (OH), 1640 and 1530 (CONH) cm^{-1} in the IR spectrum. Its ^1H NMR spectrum (in C_5D_5N) showed the presence of three methyls [δ 0.88 (6H, *br t*), 1.63 (3H, *s*)] and many methylenes [δ 1.27 (ca. 42H), 2.20 (2H, *br t*), and 2.17 (4H, *br t*)]. The signals at δ 5.99 (2H, *m*) and 8.37 (1H, *d*, J = 7.8) were assigned to the protons on a double bond and an amide proton, respectively. The ^{13}C NMR spectral data of **3** showed the presence of three methyls, many methylenes, eleven methines and two quaternary carbons. The signals at δ 105.6 (*s*), 74.6 (*d*), 77.9 (*d*), 71.2 (*d*), 77.9 (*d*), and 62.4 (*t*) were assigned to glucopyranose carbons [8]. The signal at δ 175.5 (*s*) was assigned to amide carbon and the signals at δ 124.0 (*d*), 131.5 (*d*), 132.2 (*d*), and 135.5 (*s*) were assigned to olefinic carbons. The remaining low field signals at δ 69.7 (*t*), 72.1 (*d*), and 72.3 (*d*) were attributed to carbons bearing oxygen atoms. These spectral data were in good agreement with the structure of cerebroside, [(4*E*,8*E*)-*N*-D-2′-hydroxypalmitoyl-1-*O*-β-D-glucopyranosyl-9-methyl-4,8-spingodienine] isolated from *Schizophyllum commune*, in which the compound has a fruiting-inducing activity [8]. The FABMS of compound **3** showed peaks at m/z 778 [M_{II} (727) + Na], 750 [M_I (755) + Na], 576 [M_{II} − glucose], 548 [M_I − glucose], 294 [$M_{I(II)}$ − glucose − B-chain], 276 (294 − H_2O), and 180 [glucose]. These results indicated that the compound **3** was a mixture (**3a**: M_I = 727, **3b**: M_{II} = 755) due to differences in the length by chain B (**3a**: palmitoyl, **3b**: stearoyl). The compound **3** may be related to the induction of fruiting in *T. neofelleus* as in *S. commune*.

EXPERIMENTAL

Mps: uncorr. ^1H NMR: 200 MHz, ^{13}C NMR: 50.1 MHz, with TMS as int. standard; CC: silica gel Merck 60.

Material. The fresh fruit body of *T. neofelleus* were collected from Wajiki-Chyo, Tokushima, Japan in July 1986.

Isolation of compounds (**1–3**). Fresh fruit bodies (2.47 kg) of *T. neofelleus* were cut and extracted with hot MeOH until the MeOH extract was colourless. The MeOH soln was evapd to dryness (148.5 g), dissolved in H_2O and extracted with EtOAc. The EtOAc extract was evapd under red. pres. to give a residue (4.82 g), which was chromatographed on a silica gel column (150 g) and eluted successively with $CHCl_3$–MeOH (9 : 1) and MeOH to afforded 10 fractions. Fr. 2 (0.241 g), fr. 4 (0.160 g), and fr. 6 (0.103 g) were crystallized from MeOH to give compounds **1** (38 mg), **2** (9 mg), and **3** (93 mg), respectively.

Compound **1**. Colourless needles, mp 168–170°, $[\alpha]_D^{22}$ −25.0° ($CHCl_3$; *c* 0.2), ^{13}C NMR (C_5D_5N): Table 1, was identical with ergosta-7,22-diene-3β-ol [4].

Compound **2**. Colourless needles, mp 272–274°C, $[\alpha]_D^{22}$ +31.1° (pyridine; *c* 0.9), IR ν_{max}^{KBr} cm^{-1}: 3500, 1650, 1370, 1080, 1030. EIMS m/z (rel. int.): 560 $[M]^+$ (11), 398 (17), 381 $[M - glu + H]^+$ (80.8), 273 $[M - glu - side\ chain\ (C_9H_{17}) + H]^+$ (35.5), 271 (26.6), 255 (62.9), 253 (22.8), 229 (27.6), 213 (10), 125 [C_9H_{17} (side chain)]$^+$ (97.3), 69 (100). HRMS m/z: 560.4060 for $C_{34}H_{56}O_6$.

required: 560.4077. ^{1}H NMR (C_5D_5N): δ 0.59 (3H, *s*), 0.73 (3H, *s*), 0.88 (3H, *d*, $J = 6.6$ Hz), 0.89 (3H, *d*, $J = 6.6$ Hz), 0.99 (3H, *d*, $J = 6.8$ Hz), 1.08 (3H, *d*, $J = 6.6$ Hz), 4.01 (1H, *m*, 3-H), 4.05–4.32 (4H, m, 2′-H, 3′-H, 4′-H, and 5′-H), 4.39 (1H, *dd*, $J = 11.5$ and 5.3 Hz, 6′-H), 4.60 (1, *dd*, $J = 11.5$ and 2.4 Hz, 6′-H), 5.30 (3H, *m*, 7-H, 22-H, and 23-H). ^{13}C NMR (C_5D_5N): Table 1.

Compound **3**. Colourless plates, $[\alpha]_D^{22}$ +19.8° (EtOH; *c* 0.2), IR ν_{max}^{KBr} cm^{-1}: 3360 (OH), 1640 and 1530 (CONH), 1460, 1080. EIMS *m/z* (rel. int.): 276 $[C_{18}H_{46}N]^+$ (57), 258 (28), 180 [glu]$^+$ (10). FABMS *m/z* (rel. int.): 778 $[M_{II}+Na]^+$ (18), 750 $[M_I+Na]^+$ (25), 576 $[M_{II}-glu+H]^+$ (12), 548 $[M_I-glu+H]^+$ (26), 294 $[C_{18}H_{49}NO]^+$ (10), 276 $[C_{18}H_{47}N]^+$ (13). ^{1}H NMR (C_5D_5N): δ 0.89 (3H × 2, *br t*), 1.29 (*ca* 42H), 1.63 (3H, *s*), 2.04 (2H, *br t*), 2.19 (4H, *br t*), 3.90 (1H, *m*), 4.04 (1H, *br t*), 4.1–4.8 (9H, *m*), 4.94 (1H, *d*, $J = 7.6$ Hz, anomeric H), 5.28 (1H, *m*), 6.00 (2H, *br t*), 6.02 (2H, *m*), 8.39 (1H, *d*, $J = 7.8$ Hz, CONH). ^{13}C NMR (C_5D_5N): δ14.0 × 2 (*q*), 16.0 (*q*), 22.7 × 2 (*t*), 25.7 (*t*), 28.0 (*t*), 28.2 (*t*), 29.5 × 3 (*t*), 30.1 × 10 (*t*), 32.1 × 2 (*t*), 33.0 (*t*), 35.5 (*t*), 39.9 (*t*), 54.4 (*d*), 62.4 (*t*), 69.5 (*t*), 71.2 (*d*), 72.0 (*d*), 72.3 (*d*), 74.6 (*d*), 77.9 × 2 (*d*), 105.6 (*d*), 124.0 (*d*), 131.5 (*d*), 132.2 (*d*), 135.5 (*s*), 175.5 (*s*).

REFERENCES

1. Takaishi, Y., Murakami, Y., Ohashi, T., Nakano, K., Murakami, K. and Tomimatsu, T. (1987) *Phytochemistry* **26**, 2341.
2. Takaishi, Y. Ohashi, T., Murakami, Y., and Tomimatsu, T. (1987) *Bull. Inst. Chem. Res., Kyoto Univ.* **65**, 114.
3. Yokoyama, A. and Natori, S. (1975) *Phytochemistry* **14**, 487.
4. Lu, W., Adachi, I., Kano, K., Yasuta, A., Torizuka, K., Ueno, M., and Horikoshi, I. (1985) *Chem. Pharm. Bull.* **33**, 5083.
5. Bock, K. and Pederson, C. (1983) *Adv. Carbohydr. Chem. Biochem.* **41**, 27.
6. Tori, K., Yoshimura, Y., Arita, H. and Tomita, Y. (1978) *J. Am. Chem. Soc.* **100**, 3331.
7. Maccorquodale, D. W., Steenbock, H. and Adkins, H. (1930) *J. Am. Chem. Soc.* **52**, 2512.
8. Kawai, G. and Ikeda, Y. (1983) *Biochim. Biophys. Acta* **754**, 243.

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p-HYDROXY ACETOPHENONE DERIVATIVES FROM *DIOSCOREA BULBIFERA*

DIPTI GUPTA and J. SINGH

Natural Products Laboratory, Department of Chemistry, University of Allahabad, Allahabad - 211 002, U. P., India

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Key Word Index—*Dioscorea bulbifera*; Dioscoreaceae; bulbs; *p*-hydroxy acetophenone derivatives.

Abstract—From the bulbs of *Dioscorea bulbifera*, two new *p*-hydroxy acetophenone derivatives, namely 4-hydroxy-[2-*trans*-3′,7′-dimethyl-octa-2′,6′-dienyl]-6-methoxyacetophenone and 4,6-dihydroxy-2-*O*-(4′-hydroxybutyl)acetophenone have been isolated.

INTRODUCTION

The genus Dioscorea comprises 600 tropical and subtropical species; three species are distributed in the Pyrenees, Balkan Penins and Caucasus [1]. *Dioscorea bulbifera* is common throughout India ascending up to 6000 ft in the Himalayas. Its bulbs are used to treat piles, dysentry, syphilis and are applied to ulcers [2]. Poisonous glucosides have already been reported from bulbs of Dioscorea [3]. This paper describes the isolation of *p*-hydroxy acetophenone derivatives from *D. bulbifera*; their structures were established by spectroscopic methods.

RESULTS AND DISCUSSION

A series of conventional extraction and separation procedures yielded compounds **1** and **2**. Homogeneity and purity of these compounds was established by chromatography.

Compound **1** shows a $[M]^+$ in the mass spectrum at *m/z* 302 in agreement with the formula $C_{19}H_{26}O_3$. Its UV spectrum is characteristic of acetophenone derivatives. Acetylation of **1** yielded a monoacetate (**1a**), showing the presence of one hydroxyl group in the molecule. The bathochromic shift (10 nm) induced in the UV spectrum