ELSEVIER

Fitoterapia 73 (2002) 288–291

FITOTERAPIA

www.elsevier.com/locate/fitote

Structure of a novel diterpenoid ester, fritillahupehin from bulbs of *Fritillaria hupehensis* Hsiao and K.C. Hsia

Hanli Ruan[a], Yonghui Zhang[a], Jizhou Wu[a,*], Shukai Deng[a], Handong Sun[b], Tetsuro Fujita[c]

[a] *Faculty of Pharmaceutical Sciences, Tongji Medical College of Huazhong University of Science and Technology, Wuhan 430030, PR China*
[b] *Kunming Institute of Botany, Chinese Academy of Sciences, Kunming 650204, PR China*
[c] *Faculty of Pharmaceutical Sciences, Kyoto University, Kydo 606-01, Japan*

Received 5 December 2001; accepted in revised form 2 April 2002

Abstract

A new diterpenoid ester, fritillahupehin (**1**) together with three known fatty acids: palmitic acid (**2**), lignoceric acid (**3**) and azelaic acid (**4**) were isolated from the bulbs of *Fritillaria hupehensis* Hsiao and K.C. Hsia. The structure of fritillahupehin has been established to be *ent*-kauran-16β-yl lignocerate by means of spectroscopic and chemical evidence. Compounds **2–4** were isolated from *Fritillaria* sp. for the first time.

Keywords: *Fritillaria hupehensis*; Diterpenoid ester; Fritillahupehin

1. Introduction

Fritillaria hupehensis is a liliaceous plant growing in the south-west district of Hubei Province, China. Its bulbs have been recorded in the Pharmacopoeia of the People's Republic of China as a principal Chinese traditional medicine named 'Hubeibeimu' [1].

* Corresponding author.
E-mail addresses: ruanhl@27.net (H. Ruan), wujiz@tjmu.edu.cn (J. Wu).

0367-326X/02/$ - see front matter
PII: S0367-326X(02)00085-0

We have previously reported the isolation of ten *C*-nor-*D*-homo steroidal alkaloids from the bulbs of this plant [2–8]. A number of the non-basic constituents were also isolated: sitosterol, *ent*-kauran-16β, 17-diol, *ent*-kauran-16α, 17-diol, fritillebin C and fritillebin D [9,10]. In our continuing studies on the non-basic constituents, a novel diterpenoid ester, fritillahupehin (**1**) and three fatty acids: palmitic acid (**2**), lignoceric acid (**3**) and azelaic acid (**4**) were isolated. The paper describes the structural elucidation of compounds **1**.

2. Experimental

2.1. General experimental procedures

Melting points were determined on X4 apparatus and are uncorrected. Optical rotations were taken on WZZ-1digital polarimeter. IR spectra were taken on Shimadzu IR-460 spectrometer. EI-MS spectra were measured on Auto-Spe mass spectrometer. ^{1}H-NMR and ^{13}C-NMR data were recorded on Bruker-400 spectrometer using $CDCl_3$ as solvent. TLC: silica gel (QingDao, China). Column chromatography: silica gel (100–200 mesh, QingDao, China).

2.2. Plant material

The bulbs of *F. hupehensis* Hsiao and K.C. Hsia were bought from HuBei Institute of Chinese Materia Medeca and identified by PengDeTai, LiChuan Institute of Chinese Materia Medeca, China.

2.3. Extraction and isolation

The powdered bulbs (12 kg) of *F. hupehensis* were extracted with EtOH. The extract (484.75 g) was partitioned between *n*-hexane and water. The *n*-hexane extract was fractionated by CC over silica gel, and eluted with *n*-hexane–EtOAc containing increasing contents of EtOAc to get fraction II, 155 g. Part (86 g) of fraction II was fractionated by CC over silica gel and eluted with petroleum–EtOAc to obtain: fritillahupehin (53 mg) (**1**); palmitic acid (**2**) (158.7 mg); lignoceric acid (69 mg) (**3**); and azelaic acid (**4**) (75.3 mg).

3. Results and discussion

The EtOH extract of *F. hupehensis* was partitioned between water and *n*-hexane. The *n*-hexane extract was fractionated by column chromatography over silica gel to afford (**1**–**4**). Fritillahupehin (**1**), colorless needles mp 183–185 °C (EtOAc) $[\alpha]_D^{20}$ −78.2° (c 0.67, $CHCl_3$); IR bands (KBr): 1738, 1382, 1365, 720 cm^{-1}; FAB-MS *m/z*:M^+ 731 (M − H + Gly). (Found: C, 82.92; H, 12.24. Calc. for $C_{44}H_{80}O_2$: C, 82.50; H, 12.50).

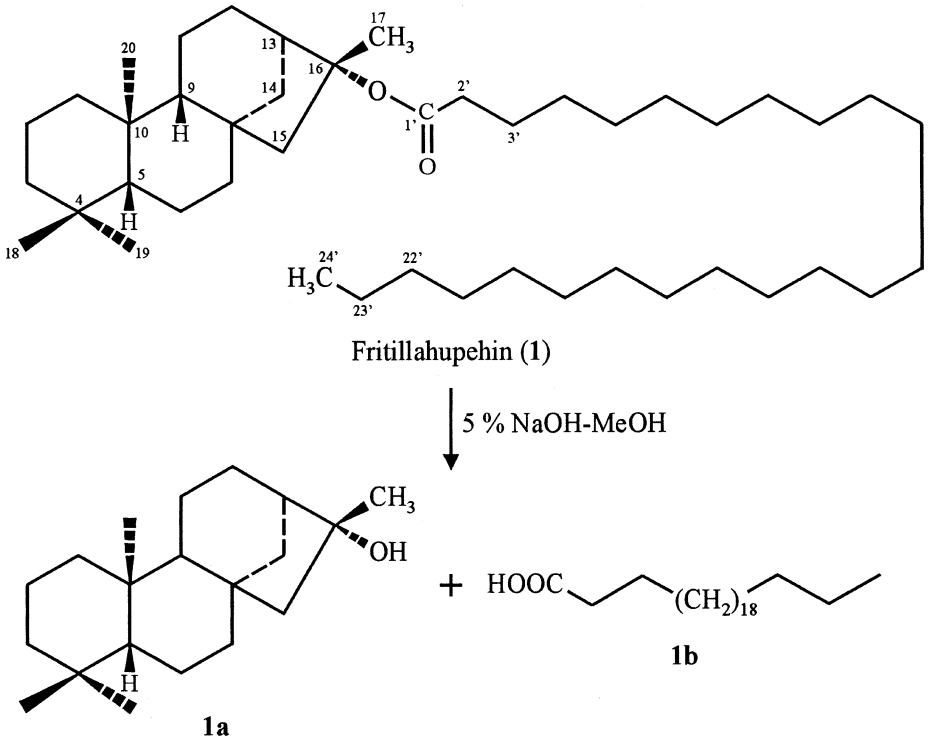

Fig. 1. Fritillahupehin.

Alkaline hydrolysis of fritillahupehin with 5% NaOH–MeOH yielded two compounds. The ^{13}C-NMR [11] and ^{1}H-NMR [12] spectra of the neutral component were in accordance with those of *ent*-kauran-16β-ol (16α-hydroxy-*ent*-kaurane) which had been isolated previously from *Xylopia aethiopica* [13].

The acidic component was identified as lignoceric (**3**) from spectral parameters and direct comparison with the authentic sample.

From the evidence described above, the structure of fritrillahupehin (**1**) was established as *ent*-kauran-16-β-yl lignocerate (Fig. 1).

The other compounds recovered from chromatography were identified as palmitic (**2**), lignoceric (**3**) and azelaic acid (**4**).

References

[1] Pharmacopoeia of the People's Republic of China 2000:286.
[2] Wu JZ. Zhongcaoyao 1982;13:339.
[3] Pu QL, Xu P, Wu JZ. Kexuetongbao 1983;28(18):1145.
[4] Wu JZ, Pu QL. Acta Pharm Sinica 1985;20:372.

[5] Wu JZ, Pu QL. Zhongcaoyao 1986;17:101.
[6] Wu JZ, Wang YY, Ling DK. Acta Pharm Sinica 1986;21:546.
[7] Wu JZ, Pu QL. Zhongcaoyao 1989;20:194.
[8] Wu JZ, Wang MT. Zhongcaoyao 1989;20:530.
[9] Wu JZ, Pu QL, Jiang HM, Jin BL. Zhongcaoyao 1989;20:244.
[10] Ruan HL, Wu JZ, Yao NH, Zhao QS, Sun HD. Zhongcaoyao 2001;32:110.
[11] Fraga BM, Hernandez MG, Fernandez C, Arteaga JM. Phytochemistry 1987;26:775.
[12] Herz W, Serengolam V, Govindan , Watanabe K. Phytochemistry 1982;21:946.
[13] Hanson JR. J Chem Soc C:5061.