comparison with authentic samples. 12-*Acetyl jativatriol* (**1**) Mp 206–208° (Et_2O–*n*-hexane); $[\alpha]_D^{20°}$ −17 3° (EtOH; *c* 0·23). IR (KBr) ν_{max} cm^{-1} 3440, 3380 (–OH); 3055, 770 (olefin), 1710, 1260 (–OAc). NMR (60 MHz, $CDCl_3$) δ 5·95 (2H, AB *q*, *J* 6 Hz, C-15 and C-16 vinylic protons), 5·18 (1H, *m*, $W_{1/2}$ 8 Hz, C-12 equatorial proton), 3·28 (1H, *m*, $W_{1/2}$ 18 Hz, C-1 axial proton), 3·15 (2H, AB *q*, *J* 12 Hz, C-17 methylene), 2 11 (3H, *s*, –OAc), C–Me singlets at 0·88, 0 83 and 0 76 MS (70 eV) *m/e* (rel int.)· 344 M^+-18 (8), 302 M^+-60 (100, base peak), 284 (36), 269 (20), 267 (18), 242 (22), 161 (20), 106 (90), 92 (96) (Found· C, 72·61, H, 9·67 $C_{22}H_{34}O_4$ requires C, 72·89, H, 9·45%)

Acetylation of **1** Treatment of compound **1** (50 mg) with Ac_2O–Py 24 hr at room temp. gave **2** (51 mg) mp 124–125° (aq EtOH), $[\alpha]_D^{25°}$ −41·3° ($CHCl_3$; *c* 0 61). IR (KBr) ν_{max} cm^{-1} 3080, 3070, 770 (olefin), 1735, 1250 (–OAc) NMR δ 5 86 (2H, AB *q*, *J* 6 Hz, C-15 and C-16 vinylic protons), 4 97 (1H, *m*, $W_{1/2}$ 7 Hz, equatorial C-12), 4 53 (1H, *m*, $W_{1/2}$ 18 Hz, axial C-1), 3·94 (2H, AB *q*, *J* 12 Hz, C-17), 1·99 (3H, *s*, –OAc), 2 03 (6H, *s*, two –OAc), C–Me singlets accumulated at 0·86 (9H) (Found C, 69·79; H, 8·61 Calc. for $C_{26}H_{38}O_6$: C, 69·93, H, 8·58%) Compound **2** was identical in all respects with jativatriol triacetate[3]

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(-)-(*TRANS*-4′-HYDROXYCINNAMOYL)LUPININE, A NEW ALKALOID IN *LUPINUS* SEEDLINGS

ISAMU MURAKOSHI, KAEKO SUGIMOTO and JOJU HAGINIWA
Faculty of Pharmaceutical Sciences, University of Chiba, Chiba

and

SHIGERU OHMIYA and HIROTAKA OTOMASU
Hoshi College of Pharmacy, Ebara 2-4-41, Shinagawa-ku, Tokyo, Japan

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In connection with our studies on the lupin alkaloids[1, 2], variations in alkaloid content at various stages of seedling growth of *Lupinus luteus* were examined and a new alkaloid was observed in varying concentration at different times in young seedlings. The present report describes its isolation and characterization as a *trans*-4-hydroxycinnamic acid ester of (-)-lupinine, i.e. (-)-(*trans*-4′-hydroxycinnamoyl)-lupinine (**1**).

No detectable amount of **1** was found in the mature and immature seeds, and in the later stages of the plant's growth. However, its concentration increased rapidly during the first 4–8 day's growth of seedlings; during further growth, the concentration fell gradually to a very low level

The structure of **1** was determined by spectrometric (IR, MS and NMR) data and by direct comparison with a synthetic sample, prepared as described in the Experimental. Both the natural and synthetic samples showed the presence of a

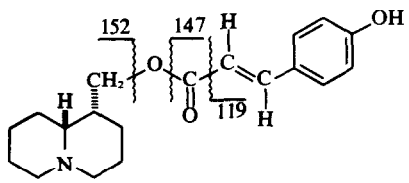
trace amount of the *cis*-isomer as a contaminant, formed during a treatment of **1** in daylight. Actually, the transformation of **1** into the cis-isomer was more rapid by irradiation in EtOH in UV light. **1** is possibly an intermediate in the biosynthesis of ω-feruloyloxylupinane, isolated previously from the young leaves of the same plant by Podkowinska *et al.*[3].

EXPERIMENTAL

NMR spectra were recorded in acetone-d_6 with TMS as internal standard at 60 MHz, MS at 70 eV, and ORD in 95% EtOH.

Plant material. Seeds of *Lupinus luteus* were collected in June at the Kashima area, Japan. *Lupinus* seedlings were grown in moistened vermiculite in the dark for 7–8 days at 30° The testas were removed and then the whole seedlings were extracted immediately for the alkaloids

Isolation of **1**. Freshly harvested *Lupinus* seedlings, grown from 2 kg of the seeds, were homogenized in 95% EtOH and left overnight at 5–10°, crude alkaloids (7·64 g) were obtained from the supernatant as a viscous pale yellow oil which crystallized partly on standing in refrigerator. The total alkaloid fraction (4 1 g) was chromatographed on a column of Si gel 60(400 g, 70–230 mesh, Merck) with CH_2Cl_2–MeOH–conc NH_4OH(90·9 1), 10 ml fractions being collected. R_f's on Si gel TLC for **1**, lupinine and sparteine, developed with the same solvent, were 0·62, 0·31 and 0·1, respectively, whilst weak spots with R_f's of 0 94 and 0 18 were also observed. After monitoring by TLC, the fractions were variously combined, from which **1** (0·15 g, about 4–5% of total alkaloids) was obtained as a highly viscous oil which showed 1 spot on TLC in 4 solvents. Additional amount of I was further separated from the intermediate fractions by preparative TLC MS: *m/e* 315 (M^+, 8%), 168(5), 152(100) [4, 6], 147(9) and 119(6)[7] (see Fig. 1) IR: $\nu_{max}^{CCl_4}$ cm^{-1}, 3630,



1

Fig 1. Characteristic fragment ions in the MS.

3370(OH), 2807, 2767(*trans*-quinolizidine[8]), 1715(ester), 1637(CH=CH), 1610, 1515(aromatic), 1270(ester) NMR: (δ, ppm) 4·40 (2H, *d*, *J* 6·5 Hz, =CH–CH$_2$–O–CO–), 5·40 (1H, *b*, OH), 6·39 (1H, *d*, *J* 16 Hz, –CO–CH=CH–), 6·96 (2H, *d*, *J* 9 Hz, P-sub aromatic), 7·58 (2H, *d*, *J* 9 Hz, P-sub aromatic) and 7·68 (1H, *d*, *J* 16 Hz, –CO–CH=CH–) The *p*-nitrobenzoate pale-yellow plates, mp 130–131° (from Et_2O). IR: ν_{max}^{KBr} cm^{-1}, 1750, 1710, 1640, 1530, 1345 (NO_2) MS: *m/e* 464 (M^+, 5%), 152(100). (Found: C, 67·34; H, 6 08, N, 5 88 $C_{26}H_{28}O_6N_2$ requires C, 67 22, H, 6·08; N, 6·03%).

Hydrolysis to (-)-*lupinine and* trans-4-*hydroxycinnamic acid.* Heating of **1** (11 mg) in 5% NaOH (5 ml) at 85–90° for 30 min gave equimolecular amounts of (-)-lupinine (5 mg) and *trans*-4-hydrocinnamic acid (5·5 mg): (-)-lupinine and *trans*-4-hydroxycinnamic acid were identified by means of mp's, colour reactions, and TLC, and by comparison of the IR and MS with those of authentic samples.

Synthesis. **1** was synthesized from (-)-lupinine (38 mg) and *trans*-*p*-acetoxycinnamoyl chloride (60 mg, m.p 119–121°)[9] according to a modification of the procedure of Boido *et al.*[10] for the lupinine ester, involving the acetyl -derivative of **1** as intermediate. since the purification of the acetate of **1** was difficult, it was treated with 2% HCl–Me_2CO[11] to remove the acetyl group. Synthetic **1** was purified by Si gel column chromatography, developed with CH_2Cl_2–MeOH–conc NH_4OH(98·3 1 5.0·2). A pale yellow oil (63 mg) was obtained. The synthetic product and its *p*-nitrobenzoate were found to be completely identical with those of the natural product in their IR, MS, NMR, and chromatographic behaviour

Trans–cis *inter-conversion.* When the EtOH soln of **1** was irradiated by UV lamp (365 mn) for 5 min, the formation of the *cis* isomer was clearly observed, as previously described by Kahnt[12] for hydroxycinnamic acid derivatives, on cellulose TLC developed with: 1, 0·4% AcOH[13]; 2, 1 M $(NH_4)_2SO_4$ in H_2O[3]. The *trans* and *cis* isomers of **1** were easily distinguished by chromatography: the R_f's for *cis*-isomer obtained in these solvents were 0·54 and 0·68, respectively, whilst *trans*-isomer had R_f's 0 38 and 0 51, respectively. The *cis*-isomer eluted from the TLC plates was identified by MS and UV UV(EtOH): *cis*, 314 nm; *trans*, 317 nm (*cis*-isomers in general absorb at shorter wavelengths than *trans* [13, 14]

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