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ISOLATION OF THE PYRROLIZIDINE ALKALOID EUROPINE *N*-OXIDE FROM *HELIOTROPIUM MARIS-MORTUI* AND *H. ROTUNDIFOLIUM*

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Key Word Index—*Heliotropium maris-mortui*; *H. rotundifolium*; Boraginaceae; pyrrolizidine alkaloid, europine *N*-oxide.

Heliotropium maris-mortui was collected at Ein Bokek on the shore of the Dead Sea, Israel and *H. rotundifolium* was collected in the Judean Mountains, Israel. No previous work has been reported on these plants. The air-dried above ground plant (*H. maris-mortui* or *H. rotundifolium*) was defatted with light petrol then extracted with MeOH. The black residue remaining after removal of the solvent was partitioned between $CHCl_3$–H_2O. The organic layer contained almost no material and gave no Mattocks test [1] for pyrrolizidine alkaloids. Evaporation of the H_2O layer gave a brown semicrystalline gum which was strongly positive. Chromatography of this residue on basic alumina activity III gave in the EtOAc–MeOH (4:1) eluent almost pure europine *N*-oxide [2]* *ca* 0.1% dry wt. Mp 171–172° [2]; TLC on Si gel: 0.1N NaOH showed one major spot, R_f 0.34* with I_2 or Mattocks reagent [1]. IR 1725 cm^{-1}; $[\alpha]_D$ + 27 [2] (*N*-oxide from *H. maris mortui*: calc. for $C_{16}H_{27}O_7N{\cdot}H_2O$: C, 52.88; H, 8.04; N, 3.86. Found: C, 52.58; H, 7.75; H, 4.01%).

The ^1H and ^{13}C NMR spectra of the alkaloids isolated from the two species were identical and the observed signals could be assigned as illustrated (spectra run in D_2O reference TMS). The ^1H NMR spectrum was likewise consistent with that reported for europine [3]. Likewise the MS of an authentic sample of europine *N*-oxide and the alkaloids from the two species of *Heliotropium* were identical within experimental error. While no molecular ion could be observed, a small signal at M^+-16 (329) was observed [4] and intense peaks were observed at *m/e* 138, 93, 80 and 59 as reported for heliotrine [5].

Reduction [2] of the natural *N*-oxides from *H. maris mortui* and *H. rotundifolium* with Zn/H_2SO_4 gave europine as an oil [2] with identical optical rotation, within experimental error, to that previously reported (+12° *H. maris-mortui*, +10° *H. rotundifolium*, +11° europine [2]). Similarly hydrogenolysis of the *N*-oxides from the two plants using 10% Pd/C in 5% HCl gave lasiocarpic acid of identical melting point (mp 96–98°) to that reported [2] for the pure compound.

The related alkaloid indicine *N*-oxide has shown significant activity against several tumor systems and is presently undergoing human clinical trials [6]. Preliminary screening of europine *N*-oxide with P388 lymphocytic leukemia in mice has shown a similar but somewhat lower activity [7]. Further screening is underway.

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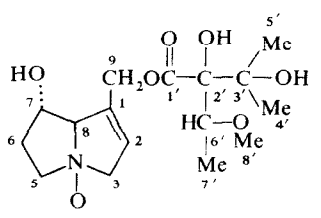
* On Si gel, elution with $CHCl_3$–MeOH, detection with I_2.



Europine *N*-oxide

^1H and ^{13}C NMR of europine *N*-oxide

	1	2	3	4	5	6	7	8	9
			β4.5						
^1H	–	5.9	α3.8	–	3.8	2.2	4.5	4.5	4.9
^{13}C	132.2	124.4	68.6	–	61.9	33.6	97.1	72.3	77.9
	1′	2′	3′	4′	5′	6′	7′	8′	
^1H	–	–	–	1.20	1.25	3.8	1.20	3.2	
^{13}C	174.4	86.1	74.3	26.4	25.7	79.7	13.5	57.1	