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3-METHYLTHIOPROPYLAMINE AND (*R*)-3-METHYLSULPHINYLPROPYLAMINE IN *IBERIS AMARA*

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Abstract—3-Methylthiopropylamine and (*R*)-3-methylsulphinylpropylamine have been isolated from *Iberis amara* and identified by PC, high voltage electrophoresis, GLC, MS and PMR. The configuration for (*R*)-3-methylsulphinylpropylamine was established from the optical rotation, and the content of this compound in different parts of the plant has been determined.

Glucosinolates, well known from a number of different plant families, yield nitriles, thiocyanates, isothiocyanates, or derivatives thereof, by enzymatic hydrolysis [2, 3]. The occurrence of amines in plants containing structurally related glucosinolates indicates a metabolic relation between these compounds [1, 4]. As previously reported, some plants contain both glucosinolates and the structurally related amines: *Moringa oleifera* Lam. (syn. *M. pterygosperma* Gaertn.) (Moringaceae), benzylglucosinolate [5] and benzylamine [6]; *Sinapis alba* L. (Cruciferae), *p*-hydroxybenzylglucosinolate [7], and *p*-hydroxybenzylamine [4]; *Lunaria annua* L. (Cruciferae), isopropylglucosinolate [8] and the γ-glutamylderivative of isopropylamine [9]. In addition *Reseda odorata* L. (Resedaceae) contains *o*-hydroxybenzylamine, and the corresponding rhamnoside [10], and also glucosinolates of unknown structures but presumably with a carbohydrate moiety in the side chain [1, 10, 11].

However, it is known that benzylisothiocyanate by chemical treatment gives small amounts of benzylamine [2]. Similarly it may be assumed that other amines may be chemically derived from isothiocyanates, but only when the corresponding carbonium ion is stabilized, as for example in the case with benzyl derivates. Therefore the simultaneous occurrence of glucosinolates and structurally related amines in cases where there is no question of carbonium ion stabilization is strong support for a metabolic relation.

The present paper describes the isolation and identification of 3-methylthiopropylamine (**1**) and (*R*)-3-methylsulphinylpropylamine (**2**) from *Iberis amara* L. (Cruciferae). This plant also contains (*R*)-3-methylsulphinylglucosinolate [1, 11, 13, 14] and 3-methylthiopropylglucosinolate [1, 3, 14].

The green parts, roots, and flowers of *I. amara* grown outdoors were harvested in the autumn, and kept at −20° until isolation was performed. Ethanolamine (**3**), **2**, and **1** were isolated from defatted plant material by ion-exchange chromatography, preparative PC and high voltage electrophoresis (HVE). For comparison **1** and **2** were synthesized by previously described methods (see Experimental). The identities of the compounds were established by comparing PC R_f-values, HVE mobilities (Table 1), GLC R_ts, MS, and PMR spectra of the amines with those of synthetic material.

Semiquantitative estimation of **2** showed the following concentrations (in mg/g fr. wt): roots (0.06), stems and leaves (0.2), inflorescence (0.62), seeds (0.14). The amounts of **1** present were too small to make quantitative estimates. Both **1** and **2** were also found in inflorescences after homogenization of freshly harvested material in 0.1 M HCl or boiling H_2O [15, 16].

The optical rotation of **2** isolated from the inflorescences was similar to that reported in the literature for (*R*)-3-methylsulphinylpropylamine [13, 17]. In glucosinolates with a sulphoxide group in the aglucone part, the

Table 1. R_f-values and ionic mobilities towards the cathode of basic amino acids and amines

Compounds	R_f-values in solvent*			Distance in cm obtained by HVE in buffer system*	
	1	2	3	(a) pH 3.6	(b) pH 6.5
Histidine	0.15	0.78	0.18	12.5	5.2
Lysine	0.14	0.85	0.14	10.8	17.5
Arginine	0.16	0.92	0.13	10.0	16.1
Ethanolamine (3)	0.34	0.95	0.53	16.2	29.3
3-Methylthiopropylamine (1)	0.61	0.95	0.45	13.3	24.2
3-Methylsulphinylpropylamine (2)	0.38	0.95	0.45	12.4	22.8

* For solvent and buffer systems, see Experimental.

configuration at the sulphur atom is (*R*) [11, 13], while the opposite seems to be the case for naturally occurring amino acids containing a sulphoxide group [18]. Therefore, the (*R*)-configuration in **2** indicates that it is a catabolic product of the structurally related glucosinolate and makes it unlikely that it is either produced by a nonspecific oxidation of **1** or is a decarboxylation product of methionine sulphoxide.

EXPERIMENTAL

Iberis amara was purchased from J. E. Ohlsens Enke, Copenhagen.

General methods and instrumentation. PMR spectra were measured at 60 MHz. Amines were dissolved in D_2O, chemical shifts are in ppm downfield from Na-2,2,3,3-tetradeuterio-3-(triMe) propionate. MS were determined at 70 eV and optical rotations in a photoelectric polarimeter using 10 cm tubes.

GLC was carried out isothermally at 110° using a glass column (1.5 m × 4 mm id) packed with 3% SE 30 (He flow rate 40 ml/min). PC was performed in *n*-BuOH–HOAc–H_2O (12:3:5) (solvent 1), PhOH–H_2O–13 M NH_3 (120:30:1) (w/v/v) (solvent 2), and *iso*-PrOH–13 M NH_3–H_2O (8:1:1) (solvent 3) by the descending technique on Whatman No. 1 paper. Prep–PC was performed on Whatman No. 3 MM paper. HVE was carried out on Whatman No. 3 MM paper using a flat-plate unit and the following systems: (a) Buffer pH 3.6 (Py–HOAc–H_2O) (1:20:200), 30 min at 3.2 kV and 90 mA; (b) Buffer pH 6.5 (Py–HOAc–H_2O) (25:1:500), 30 min at 5 kV and 90 mA.

Synthesis. **1** and (*R/S*)-**2** were synthesized as described in the lit. [17, 19].

Isolation of **1**, **2**, *and* **3**. Whole flowering plants (2.2 kg) were homogenized in CH_2Cl_2 and defatted by Soxhlet extraction with CH_2Cl_2 (24 hr). After air drying, the residue (360 g) was extracted twice with MeOH–H_2O (7:3, 5 l. each time) by refluxing (3 hr), cooling and filtering. The filtrates were combined with the H_2O-phase from the CH_2Cl_2 extraction and concd to dryness (90 g). The residue was suspended in H_2O–EtOH (3:1, 400 ml), filtered and applied to a strongly acid ion-exchange resin (Amberlite IR 120, H^+, 10 × 80 cm). The column was washed with H_2O–EtOH (4:1, 5.5 l.) and H_2O (7.5 l.). Amino acids and amines were eluted with M NH_3. The amines **1** and **2** were eluted together with lysine in the eluate at 20–25 l., **3** and arginine appeared in the eluate at 22–29 l. The evaporation residue containing the basic amino acids, **1**, **2**, and **3** (410 mg) was purified by prep-PC (solvent 3; Table 1). Final purification of **1**, **2**, and **3** was accomplished by prep-PC (solvent 1; Table 1) and prep-HVE (pH 3.6; Table 1). The yields of the acetates were 7 mg **1**, 73 mg **2**, and 82 mg **3**. Optical rotation was determined for the free amine **2** isolated from the inflorescences $[\alpha]_D^{20}$ −108° (*c* 0.21, H_2O). Lit. value for (*R*)-3-methylsulphinylpropylamine $[\alpha]_D^{18.5}$ −115° (*c* 0.735, EtOH) [13, 17]. Identity of the compounds was established by PC, HVE (Table 1), GLC R_t, MS and PMR; the results obtained were identical with those obtained for synthetic **1** and (*R/S*)-**2**, and for commercial **3**. GLC R_t were 2 min for the free amine **1** and 3 min for the free amine **2**. MS of free amine **1** [20], *m/e*(%): M^+ 105(13), 88(30), 73(9), 61(13), 57(26), 56(16), 30(100), and for the free amine **2**: M^+ 121(15), 120(10), 106(10), 105(100), 104(5), 103(8), 102(5), 79(23), 78(8), 77(17), 53(12), 52(5), 51(10), 50(5), 44(25), 43(11), 42(18), 30(4), PMR spectrum of the HCl of **1**: δ 3.17 (*t*, 2H), 2.68 (6, 2H), 2.13 (*s*, 3H), 2.2–1.8 (*m*, 2H), and for the HCl of **2**: δ 3.4–2.9 (*m*, 4H), 2.80 (*s*, 3H), 2.5–1.9 (*m*, 2H).

Isolation and semiquantitative determination of **2**. Plant material (10 g) was homogenized in H_2O at 0–2°, centrifuged (2 hr, 2° at 10 000 *g*), and then the supernatant was lyophilized. The residue was suspended in H_2O, filtered and applied to Amberlite IR 120 (H^+, 1.6 × 30 cm). Fractions (25 ml) were collected at 80 ml/hr. After washing with H_2O (fractions 1–10), acid and neutral amino acids were eluted from the column with M Py (fractions 11–40). The column was subsequently eluted with 2 M NH_3 to give the fractions with the basic amino acids and amines (fractions 60–80). **2** was isolated from these fractions by prep-PC and HVE as described above for the isolation of **1**, **2**, and **3**. The amount of **2** was determined by PMR using the ratio between the intensity of the Me singlet from **2** (δ 2.80) and the singlet from formic acid added in known amounts.

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