*iso*Thiocyanates XXXIII. * An *iso*Thiocyanate Glucoside (Glucobarbarin) of *Reseda luteola* L.

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Phylogenists are agreed that the family *Resedaceae*, comprising six genera and about 70 species, belongs in the order *Rhoeadales*. Within the latter its close affinity to the *Cruciferae* and, particularly, the *Capparidaceae* has been generally accepted. Hence, it is not surprising that several literature references can be found to the occurrence of mustard oils in species of the genus *Reseda*.

Reseda luteola L. (dyer's weed, weld) was once grown as a source of the flavonoid dye luteolin. In 1871, Hirschberg [1] reported on the appearance of a smell similar to that of horse-radish in disintegrated root material of *R. luteola* and *R. odorata*. This observation was confirmed by Spatzier [2] who further demonstrated the presence of myrosinase in seeds and epidermic tissue of various *Reseda* species.

Pietschmann [3] later adduced microchemical evidence for the occurrence of an unidentified mustard oil in the leaf, stem, root and seed of *R.luteola*. In their paper-chromatographic survey of *iso*thiocyanate glucosides in numerous plants, Schultz and Gmelin [4] recorded the presence in roots, as well as other parts of fresh plants, of *R. luteola* of a single, unidentified glucoside travelling on paper at a rate (R_F 0.20) considerably lower than that of glucotropaeolin (R_F 0.31), the glucoside of *Tropaeolum majus*.

A closer study of the mustard oil glucoside in leaves and inflorescences of *Reseda luteola* L. has now resulted in the isolation of (−)-5-phenyl-2-oxazolidinethione as a product of enzymic fission. Paper chromatography in two solvent systems revealed the presence of only one glucoside in the employed material, migrating at a rate similar to that of glucotropaeolin.

Leaves and inflorescences of *Reseda luteola* (fresh weight 300 g) were added to hot methanol (3 l) and the mixture was refluxed for 1 h. After filtration, the pressed filter cake was extracted with hot 50 % methanol (3 l) and the filtrate collected and combined with the first extract. The solution was stripped of methanol, and water (1 l) was added to the residue. The precipitate was removed by filtration through Celite and 10 % lead acetate solution was added to the filtrate until no further precipitation took place. Excess lead ions were removed as lead phosphate by adding excess of 1/15 M phosphate buffer (pH 6.5) to the filtrate. After filtration, a cell-free myrosinase preparation (10 ml) was added and the buffered mixture set aside at room temperature for 3 h. The solution was then extracted with three 200 ml portions of chloroform. The chloroform extract was washed thrice with a 5 % $NaHCO_3$-solution and then shaken with about 10 g of acid-washed alumina in order to remove traces of flavonoid impurities. On evaporation of the dried chloroform extract a crystalline fraction remained which was recrystallized from ethyl acetate to give well-formed prisms. An additional recrystallization from 70 % ethanol afforded a pure specimen of colourless material (496 mg), m. p. 126—127° (capillary tube, uncorrected), $[\alpha]_D^{23}$ −69.3° ± 1.5° (c=2.0, MeOH).

The identity of the isolated compound with (−)-5-phenyl-2-oxazolidinethione (I) was suggested through the above physical constants and established beyond doubt upon comparison with an authentic specimen of the latter, previously synthesized in this laboratory [5]. Undepressed mixed melting point along with coinciding infra-red spectra served to establish the identity of the compound isolated from *Reseda luteola* and (I).

The formation of (I) upon enzymic hydrolysis suggests the presence in *R. luteola* of glucobarbarin, formerly encountered in seeds of *Barbarea vulgaris* R.Br.[5] As briefly pointed out previously [5], (I) depicts the absolute configuration of the naturally derived, *levo*rotatory heterocyclic compound as a consequence of its synthesis from (−)-2-amino-1-phenylethanol, accomplished in the following way.

(−)-2-Amino-1-phenylethanol (2.0 g) (m. p. 62—3°, $[\alpha]_D^{23}$ −43.5° (c = 2.0, abs. EtOH)), prepared by $LiAlH_4$ reduction of (−)-mandelamide as described by Pratesi and Grassi [6], was dissolved in anhydrous dioxane (6 ml), con-

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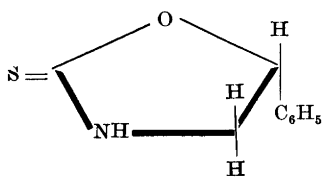
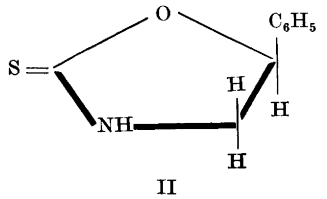


I



II

taining triethylamine (1.55 g), and the solution was cooled to —10°. CS_2 (0.90 ml) was added with stirring, and the solution was allowed to come to room temperature. After cooling again to —10°, ethyl chloroformate (1.78 g) was introduced, resulting in separation of triethylammonium chloride. Chloroform (12 ml) and triethylamine (1.50) were added and the stirred reaction mixture was allowed to come to room temperature. After the evolution of COS had ceased, HCl (1 N) was added to acid reaction, and the aqueous phase was extracted thrice with chloroform. On evaporation of the dried chloroform solution a partly crystalline product resulted, which could be recrystallized from a mixture of ethyl acetate and hexane to give prisms of (—)-5-phenyl-2-oxazolidinethione (I) (1.48 g), m. p. 125°. An analytical specimen was produced by two additional recrystallizations from 50 % ethanol as colourless prisms, m. p. 126° (uncorr.), $[\alpha]_D^{23}$ —71.0° ± 1.5° (c = 2.0, MeOH). (Found: C 60.10; H 4.84; N 7.68. Calc. for C_9H_9NOS: C 60.31; H 5.06; N 7.82).

Similarly processed, (+)-2-amino-1-phenylethanol (m. p. 63°, $[\alpha]_D^{23}$ +44.2° (c = 2.0, abs. EtOH)) afforded the enantiomeric (+)-5-phenyl-2-oxazolidinethione (II), m. p. 126° (uncorr.), $[\alpha]_D^{23}$ +71° ± 1.5° (c = 2.0, MeOH) (Found: C 60.00; H 5.00; N 7.66).

A solution of equal amounts of the antipodes in hot aqueous ethanol crystallized on cooling and seeding as the racemic compound, m. p. 139° (uncorr.), undepressed on admixture with an authentic specimen [5].

The employed synthesis procedure represents an extension of the method developed by Hodgkins and Ettlinger [7] for the synthesis of *iso*thiocyanates *.

Glucobarbarin appears to be a glucoside of the general structural type [8], containing a 2-hydroxy-2-phenylethyl-side chain. The configuration of the latter is likely to be the same as that established above for (—)-5-phenyl-2-oxazolidinethione (I), which results from spontaneous cyclization of the primarily formed 2-hydroxy-2-phenylethyl *iso*thiocyanate without participation of bonds extending from the asymmetric carbon atom.

Paper chromatography and spectrophometric assays have disclosed the presence of glucobarbarin also in seeds of *R. luteola* L. The results of further investigations of *Resedaceae* species will form the subject of a forthcoming communication.

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* Applied similarly by Dr. Ettlinger for the synthesis of (±)-4-vinyl-2-oxazolidinethione (private communication).

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