

A Mustard Oil of *Hesperis matronalis* Seed, 6-Methylsulphinylhexyl Isothiocyanate *

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The rocket, *Hesperis matronalis* L., of the family Cruciferae, is a biennial or perennial herb, cultivated in temperate regions as an ornamental. A few years ago, Schultz and Wagner,^{1,2} on basis of paper chromatography, concluded that *Hesperis matronalis* seed contained one minor and three major thioglucosides; none of these have been chemically identified.** A similar chromatographic pattern was observed in this laboratory.

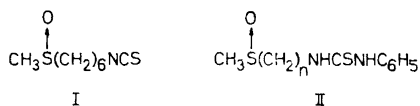
We now wish to report that one of the thioglucosides in *H. matronalis* seed on enzymic hydrolysis gives rise to the production of a previously unknown, optically active 6-methylsulphinylhexyl isothiocyanate (I).

When the crude thioglucoside mixture, isolated from *H. matronalis* seed through ion exchange in the customary way (*cf. e.g.* Ref.⁴), was subjected to enzymic hydrolysis, and the resulting isothiocyanates in turn treated with aniline, a mixture of phenylthioureas was obtained. Upon recrystallization, a homogeneous, levorotatory phenylthiourea, $C_{14}H_{22}N_2OS_2$, separated; the second sulphur atom was present in a sulphoxide-grouping as apparent from an intense absorption band at 1000 cm^{-1} (KBr) in the infrared spectrum.

In view of the many straight, ω -methylsulphinylalkyl side-chains formerly encountered in natural products of the present type,⁴ it appeared likely that the new phenylthiourea possessed the structure (II, $n = 6$). In fact, this was experimen-

tally borne out by reduction of the sulphoxide-grouping with titanous chloride, a reagent introduced by Barnard and Hargrave for similar purposes.⁵ On critical comparison, the reduction product proved identical with an authentic specimen of 1-(6-methylthiohexyl)-3-phenylthiourea, prepared in this laboratory several years ago.⁶ Hence, one of the thioglucosides in *H. matronalis* seed is enzymically hydrolyzed to 6-methylsulphinylhexyl isothiocyanate (I); this represents an addition to a homologous series of mustard oils derivable from thioglucosides in higher plants and characterized as phenylthioureas (II, $n = 3, 4, 5, 8, 9, 10$).⁴ The molecular rotation of the new derivative is comparable in sign and magnitude to that of other phenylthioureas of this series;⁴ therefore, it almost certainly belongs to the same stereochemical series.*

It is of interest that a closely related glucoside, glucolesquerellin, undergoing enzymic hydrolysis to 6-methylthiohexyl isothiocyanate, was reported recently by Daxenbichler *et al.*⁵ as a constituent of seeds of the crucifer *Lesquerella lasiocarpa*. The same group subsequently demonstrated glucolesquerellin to be present in eight additional *Lesquerella* species, mostly accompanied by a trace constituent giving rise to a thiourea with a much lower R_{ph} -value.⁹ It appears likely that the latter derives from the non-volatile 6-methylsulphinylhexyl isothiocyanate (I), produced in this case as an artefact by oxidation of the parent sulphide mustard oil during manipulations and chromatography. In *H. matronalis* seed, on the other hand, the thioglucoside, whence (I) derives and for which we suggest the name *glucohesperin*, appears to be genuinely present, since no spot attributable to glucolesquerellin is observable on paper chromatograms of seed extracts prepared under mild conditions.



* Part XLIX of a series of papers on naturally derived isothiocyanates; Part XLVIII: *Acta Chem. Scand.* 17 (1963) 561.

** The same authors^{2,3} indicated, yet without details, that one of the thioglucosides, 'glucomatralin', forms a crystalline heptaacetate.

* Configurational identity of the other members of this series has been supported by rotatory dispersion studies.⁷

Experimental. *Hesperis matronalis* seed* was milled, defatted with carbon tetrachloride, and thoroughly extracted with 70 % methanol. The concentrated extract was dissolved in water, filtered, and passed through an anion exchange resin (Amberlite IR-4B) on the chloride form. After washing with water, the glucoside fraction was eluted with a 5 % potassium sulphate solution. The brown, crystalline glucoside mixture** was employed without further purification for the enzymic hydrolysis, performed in a citrate buffer (pH 6.5) with a cell-free, crude myrosinase solution and a trace of ascorbic acid.¹⁰ After 4 h, the resulting isothiocyanates were extracted with chloroform, the dried extract was concentrated to a small volume, and a few drops of aniline were added. Next day, the solution was taken to dryness and the residue was recrystallized, first from a mixture of ethyl acetate and petroleum ether and then from acetone, to give the colourless, crystalline phenylthiourea. This proved homogeneous on thin-layer chromatography (alumina, chloroform with 5 % ethanol, spray: Grote's reagent).

An analytical specimen was produced by two additional recrystallizations from ethyl acetate, separating as flat prisms, m.p. 135° [α]_D²⁶ - 51° (c 0.8, chloroform). (Found: C 56.57; H 7.56; N 9.35. Calc. for C₁₄H₂₂N₂OS₂: C 56.33; H 7.43; N 9.39). Besides the expected bands, a very strong band at 1000 cm⁻¹ in the IR-spectrum (KBr) was indicative of a sulphoxide-grouping.

Reduction. The reduction of the sulphoxide-grouping was carried out with titanous chloride, essentially as described elsewhere.⁵ To a solution of the above phenylthiourea (13 mg) in glacial acetic acid (5 ml), from which air was expelled by addition of sodium hydrogen carbonate (750 mg), a 0.11 N solution of titanous chloride was added, excess reagent was removed with ferric ammonium sulphate, and water was added (25 ml). The solution was

neutralized with solid sodium carbonate to pH 7 and extracted thrice with 25 ml-portions of chloroform. On removal of the solvent an oil remained which crystallized on seeding with 1-(6-methylthiohexyl)-3-phenylthiourea, previously synthesized in this laboratory.⁶ Two recrystallizations from aqueous ethanol afforded a product (5 mg) with the m.p. 50-53°, undepressed on admixture with the authentic specimen (m.p. 55°). The infra-red spectra of the two preparations were perfectly identical.

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** Paper chromatography indicated its contents of two major and two minor thioglucosides.