133. *Marrubiin, the Bitter Principle of Horehound* (Marrubium vulgare).

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Analytical results for marrubiin and its derivatives favour the formula $C_{20}H_{28}O_4$ rather than $C_{21}H_{28}O_4$ previously suggested. The substance contains a hydronaphthalene nucleus, as is shown by the formation of 1 : 2 : 5-trimethylnaphthalene by dehydrogenation with selenium. Marrubiin is probably a diterpene lactone. A number of derivatives are described.

MARRUBIIN, the bitter principle of horehound (*Marrubium vulgare*), has been examined by several workers (Harms, *Arch. Pharm.*, 1842, **83**, 144; 1851, **116**, 141; Kroymeyer, *ibid.*, 1849, **108**, 258; Hertel, *Amer. J. Pharm.*, 1890, 273; Morrison, *ibid.*, 1890, 327; Matusow, *ibid.*, 1897, 201; Gordin, *J. Amer. Chem. Soc.*, 1908, **30**, 265), but little insight into its constitution obtained. Some derivatives and degradation products are now described.

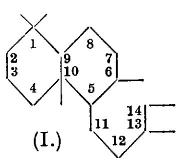
The marrubiin was obtained by Mercier and Mercier's method (*Compt. rend.*, 1932, **195**, 1102) from the flowering tops of horehound of the previous season's crop, the yield being about 0·4% of the dried material. Analyses of the carefully purified substance and its derivatives indicate that the formula should be $C_{20}H_{28}O_4$ and not $C_{21}H_{28}O_4$ as suggested by Gordin (*loc. cit.*). Moreover, the isolation of 1 : 2 : 5-trimethylnaphthalene from the products of selenium dehydrogenation of marrubiin indicate a relationship with the diterpene group and support the C_{20} formula. For these and other reasons it is concluded that marrubiin may be classed with agathic acid, manoyl oxide, and sclareol. The last-named substance is present in *Salvia sclarea*, a member of the *Labiatæ* to which marrubium itself belongs.

In addition to the lactone group in marrubiin, which can be readily hydrolysed by alcoholic potassium hydroxide to give marrubic acid, there is also present a tertiary hydroxyl group. Together with a neighbouring hydrogen atom, this group may be removed as water on careful treatment with thionyl chloride or phosphorus trichloride. Marrubiin is very susceptible to the action of acids or acidic reagents in general, being converted into resinous material. Marrubic acid is scarcely affected by prolonged treatment with alcoholic potassium hydroxide.

On treatment with hydrogen in the presence of platinum-black an acetic acid solution of marrubiin absorbs the equivalent of four atoms of hydrogen, indicating the presence of two double bonds in the molecule. In ethyl acetate solution, no hydrogenation takes place, from which it is concluded that the double bonds are present in the cyclic part of the structure. The *tetrahydro*-derivative resinifies much less readily under the action of acids than does marrubiin.

The fourth oxygen atom in the molecule appears to be present as an ether linkage, since the original material is unchanged by treatment with reagents for the detection of hydroxyl and carbonyl groups. In attempts to split such a linkage by the action of acid reagents, the *hexahydro*-compound, prepared by hydrogenation of the *substance* obtained by the action of phosphorus trichloride on marrubiin, was used; after treatment with anhydrous hydrogen chloride or hydrogen bromide in boiling acetic acid solution or with 98% formic acid at 120°, however, the bulk of the initial material was recovered unchanged.

The above findings and the isoprene rule being taken into consideration, the carbon skeleton (I) is suggested for marrubiin. In the case of agathic acid (Ruzicka and Hosking, *Helv. Chim. Acta*, 1931, **14**, 203), sclareol (Ruzicka and Janot, *ibid.*, p. 645), and manoyl oxide (Hosking and Brandt, *Ber.*, 1935, **68**, 37), which contain this skeleton, the presence or introduction of a double bond in the side chain attached to the hydronaphthalene nucleus enables isomerisation to take place by the action of formic acid, giving a phenanthrene derivative. Neither with marrubiin itself nor with its hydro-derivatives could this cyclisation be realised. After prolonged treatment with formic acid only 1 : 2 : 5-trimethylnaphthalene was isolated from the products of selenium dehydrogenation.



(I.)

The ether link in manoyl oxide is situated between the tertiary atoms C_6 and C_{13} (I), and can be readily ruptured by hydrogen chloride at 0°. In view of the inertness of the fourth oxygen atom in marrubiin, it seems unlikely that the oxygen bridge is at this position: it may perhaps connect two secondary carbon atoms.

EXPERIMENTAL.

Extraction of Marrubiin.—The dried flowering tops of horehound were percolated with acetone for 48 hours. After removal of the acetone from the extract by distillation the residue was boiled with ether. The solution, on cooling, deposited an impure product, which was washed with ether to remove oil and colouring matter, dissolved in boiling 5% alcoholic sodium hydroxide, and poured into water. The recovered solid was repeatedly crystallised from alcohol; m. p. 160° (Found: C, 72·3; H, 8·4; active H, 0·34; *M*, Rast, 377. Calc. for $C_{20}H_{28}O_4$: C, 72·3; H, 8·4; 1 active H, 0·30%; *M*, 332).

Marrubic acid, obtained by the action of boiling 10% alcoholic sodium hydroxide on marrubiin, crystallised from aqueous alcohol or ethyl acetate in fine felted needles, m. p. 205° (decomp.) (Gordin, *loc. cit.*, records m. p. 173—174°) (Found: C, 68·2; H, 8·8; active H, 0·78; *M*, 361. Calc. for $C_{20}H_{30}O_5$: C, 68·6; H, 8·6; 3 active H, 0·87%; *M*, 350). Marrubic acid is very readily lactonised, and on heating to 200°/15 mm., marrubiin distils.

Acetylmarrubic Acid.—A solution of marrubic acid (1 g.) in dry pyridine (10 c.c.) was treated at 0° with acetyl chloride (0·3 c.c.), the reaction completed on the water-bath, and the solution poured into water. After evaporation of an ethereal extract of the acidified solution, the residue crystallised from aqueous alcohol in flat prisms, m. p. 112° (Found: C, 64·3; H, 8·3; *M*, 411. $C_{22}H_{32}O_6,H_2O$ requires C, 64·4; H, 8·3%; *M*, 410).

Dehydrogenation of Marrubiin by Selenium.—A mixture of marrubiin (5 g.) and selenium (25 g.) was heated at 280—300° for 10 hours. The product was extracted with ether and fractionally distilled. The main portion, b. p. 160—180°/14 mm., was a colourless oil, which was converted into the picrate. This crystallised from alcohol in orange-red needles, m. p. 138°, not depressed by 1 : 2 : 5-trimethylnaphthalene picrate, kindly supplied by Professor I. M. Heilbron (Found: C, 57·5; H, 4·4; N, 11·3. Calc. for $C_{13}H_{14},C_6H_3O_7N_3$: C, 57·1; H, 4·3; N, 10·5%). The styphnate crystallised from alcohol in yellow needles, m. p. 131°.

Tetrahydromarrubiin.—Marrubiin (5 g.) was dissolved in glacial acetic acid (200 c.c.) and hydrogenated in the presence of Adams's platinum-black catalyst (0·25 g.); after 3 hours, almost 4 atoms of hydrogen had been taken up. The catalyst was filtered off, the acetic acid removed under reduced pressure, water added to the residue, and the solution neutralised. After extraction with chloroform and evaporation of the solvent, the residue was crystallised several times from ethyl acetate, giving flat prisms, m. p. 134°, in 60% yield (Found: C, 70·8; H, 9·4. $C_{20}H_{32}O_4$ requires C, 71·4; H, 9·5%). The acid obtained by hydrolysis with alcoholic sodium hydroxide crystallised from ethyl acetate in needles, m. p. 185°.

Action of Hydrogen Chloride on Tetrahydromarrubiin.—Hydrogen chloride was passed through a solution of tetrahydromarrubiin (2 g.) in absolute alcohol (100 c.c.) at 0° for 2 hours. After removal of the alcohol in a vacuum, the residue was distilled at 14 mm. The distillate crystallised from light petroleum in long needles, m. p. 124° (Found: C, 75·2; H, 9·4. $C_{20}H_{30}O_3$ requires

C, 75·4; H, 9·4%). This *substance* was also prepared by the action of phosphorus trichloride or thionyl chloride on tetrahydromarrubiin in benzene solution. It was heated for 10 hours at 120° with 98% formic acid. The residue after removal of the formic acid was distilled, and the distillate dehydrogenated with selenium as described in the case of marrubiin. 1 : 2 : 5-Trimethylnaphthalene was the only hydrocarbon isolated.

Hydrogenation of the Substance $C_{20}H_{30}O_3$—The substance was dissolved in glacial acetic acid and hydrogenated in the presence of Adams's platinum-black catalyst; 2 atoms of hydrogen were absorbed. The product, isolated as in the previous case, crystallised from light petroleum in long needles, m. p. 89° (Found: C, 74·3; H, 10·0. $C_{20}H_{32}O_3$ requires C, 75·0; H, 10·0%). This *substance* appears to be a stereoisomer of the hexahydro-derivative (m. p. 106°) of the compound obtained by the action of phosphorus trichloride on marrubiin.

Action of Phosphorus Trichloride on Marrubiin.—To a boiling solution of marrubiin (5 g.) in benzene (100 c.c.), phosphorus trichloride (1 c.c.) in benzene (10 c.c.) was slowly added. When no more hydrogen chloride was evolved, the mixture (which contained a considerable amount of resinous material) was poured into water. The benzene layer was removed, shaken with sodium hydroxide solution, dried, and evaporated. The product crystallised from alcohol in flat prisms, m. p. 98° (Found: C, 75·3; H, 7·8. $C_{20}H_{26}O_3$ requires C, 76·4; H, 8·2%). This *substance* was unchanged by distillation over zinc dust at 150°/1 mm. On treatment with alcoholic sodium hydroxide the lactone ring was hydrolysed; the corresponding acid, m. p. 152°, was obtained in fine needles from ethyl acetate.

Hydrogenation of the Substance $C_{20}H_{26}O_3$.—The substance was hydrogenated as described in the previous cases. The *product* crystallised on neutralisation of the acetic acid and was recrystallised from light petroleum; m. p. 106° (Found: C, 74·7; H, 9·7. $C_{20}H_{32}O_3$ requires C, 75·0; H, 10·0%). The corresponding *acid*, obtained by hydrolysis with alcoholic sodium hydroxide, crystallised from ethyl acetate in flat prisms, m. p. 210° (Found: C, 71·3; H, 9·9. $C_{20}H_{34}O_4$ requires C, 71·0; H, 10·0%).

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