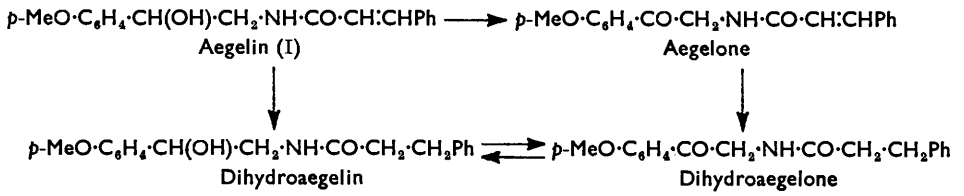
## 315. *The Structure of Aegelin.*

By R. N. Chakravarti and B. Dasgupta.

Details are provided for the proof of structure and synthesis of aegelin reported earlier.

Aegelin, isolated by Chatterjee and Bose [1] from the leaves of *Aegle marmelos* Correa, was believed by them to be a steroid, $C_{18}H_{18}O_4$. In preliminary communications,[2] however, we showed that it was nitrogenous ($C_{18}H_{19}O_3N$) and proved its structure as the (±)-*trans*-form of compound (I) by carrying out the reactions shown in the scheme and by synthesis of the four compounds shown there.




Chatterjee and Chaudhuri [3] recently adopted our formula and structure and reported alternative syntheses, but all without reference to our prior publications. We therefore record here details of our work.

### Experimental

*Modified Method for Isolation of Aegelin.*—Air-dried powdered leaves (14 kg.) of *Aegle marmelos* were extracted repeatedly with cold 90% ethanol. The extract was mixed with filter-paper pulp, dried at 80°, and extracted with ether (Soxhlet) for about 80 hr. The extract was concentrated to *ca.* 1·5 l. and kept at 0° for a week, and the separated product collected, washed with ether, crystallised from ethyl acetate and then from ethanol–acetone, passed in ethanol through aluminium oxide, and finally crystallised from ethanol, giving aegelin as plates, m. p. 178—179° [Found: C, 72·7; H, 6·3; N, 4·45; OMe, 9·6; active H, 0·75%; *M* (Rast), 291. Calc. for $C_{18}H_{19}O_3N$: C, 72·7; H, 6·4; N, 4·7; 1OMe, 10·4; 2 active H, 0·67%; *M*, 297·3].

From mature leaves (November to March) the yield of aegelin is very low, but from young leaves (May to August) the yield is 0·15—0·2%. It has an absorption maximum at 275 mμ (log ε 4·42), as reported by Chatterjee and Bose,[1] but also maxima at 212 and 219 mμ (log ε 4·36, 4·36). *trans-N*-Methylcinnamamide has similar maxima at 218 and 270 mμ (log ε 4·18, 4·39). Aegelin evolves little hydrogen selenide when heated with selenium according to Chatterjee and Bose's method.[1]

*Aegelin Esters.*—Aegelin (0·1 g.), acetic anhydride (2 c.c.), and pyridine (2 drops) were heated on a water-bath for 4 hr. Alternatively, aegelin (0·6 g.), acetic anhydride (2 c.c.), and sodium acetate (0·6 g.) were heated under reflux for 20 min. The *acetate*, crystallised from ethyl acetate and then aqueous methanol, had m. p. 123—125° (Found: C, 71·1; H, 6·5; Ac, 13·5. $C_{20}H_{21}O_4N$ requires C, 70·8; H, 6·2; 1Ac, 12·7%). Chatterjee and Bose's product,[1] of m. p. 159—160°, appears to have been a mixture.

The *benzoate* was prepared from benzoyl chloride (1·5 c.c.) and aegelin (0·6 g.) in pyridine (12 c.c.) at room temperature overnight and crystallised from ethanol in needles, m. p. 147—148° [Found: C, 75·0; H, 5·4; N, 3·1; OMe, 5·2; Bz, 28·1%; *M* (Rast), 419. $C_{25}H_{23}O_4N$ requires C, 74·8; H, 5·8; N, 3·5; 1OMe, 7·7; 1Bz, 26·2%; *M*, 401·44].

*Aegelone.*—Chromic acid (0·2 g.) was added in small lots with shaking during 4 hr. to a solution of aegelin (0·5 g.) in glacial acetic acid (25 c.c.) at the room temperature. Acetic acid was distilled off under reduced pressure, and the residue diluted with water. The separated solid was filtered off and repeatedly crystallised from ethanol, giving *aegelone* as colourless

[1] Chatterjee and Bose, *J. Indian Chem. Soc.*, 1952, **29**, 425.

[2] Chakravarti and Dasgupta, *Chem. and Ind.*, 1955, 1632; *Bull. Calcutta School Trop. Med.*, 1956, **4**, 69, 123, 167; Chakravarti, Ann. Rept. Calcutta School Trop. Med., 1955—56, 70; Dasgupta, D.Phil. (Science) Thesis, Calcutta, 1956.

[3] Chatterjee and Chaudhuri, *Science and Culture*, 1957, **23**, 155.

needles, m. p. 159—160° (0·35 g.) (Found: C, 73·6; H, 5·8; N, 4·25. $C_{18}H_{17}O_3N$ requires C, 73·2; H, 5·8; N, 4·7%).

Aegelone does not colour Schiff's reagent nor does it respond to the Angeli–Rimini test for aldehydes. When heated on a water-bath it reduces ammoniacal silver nitrate solution within a minute. The *semicarbazone* is obtained in needles, m. p. 234—235° (decomp.) (Found: C, 64·3; H, 5·6; N, 15·2. $C_{19}H_{20}O_3N_4$ requires C, 64·8; H, 5·7; N, 15·9%). The ultraviolet absorption spectrum of aegelone (max. at 280 mμ; log ε 4·67) resembles that of ω-amino-*p*-methoxyacetophenone hydrogen sulphate (max. at 282 mμ; log ε 4·27).

*Dihydroaegelin.*—Aegelin (0·5 g.) was shaken in acetic acid (25 c.c.) solution with Adams catalyst (50 mg.) under hydrogen [absorption, complete in 15 min.: 42 c.c. (N.T.P.); 1 mol. = 37·7 c.c.]. After filtration acetic acid was distilled off under reduced pressure, and the residue diluted with water. The separated *dihydroaegelin* crystallised from ethanol in leaflets, m. p. 140—141° (0·33 g.) (Found: C, 72·4; H, 7·15; N, 4·4. $C_{18}H_{21}O_3N$ requires C, 72·2; H, 7·1; N, 4·7%). Dihydroaegelin, like aegelin, does not reduce ammoniacal silver nitrate solution when heated on a water-bath for several minutes.

*Dihydroaegelone.*—This was prepared, in the same way as aegelone, by chromic acid oxidation of dihydroaegelin. *Dihydroaegelone* crystallised from ethanol in needles, m. p. 126—127° (Found: C, 72·9; H, 6·5; N, 4·5. $C_{18}H_{19}O_3N$ requires C, 72·7; H, 6·4; N, 4·7%). It reduces ammoniacal silver nitrate solution on a water-bath in a few minutes, and gives a *semicarbazone* (needles), m. p. 138—140° (Found: C, 64·0; H, 6·4; N, 15·0. $C_{19}H_{22}O_3N_4$ requires C, 64·4; H, 6·3; N, 15·8%).

Dihydroaegelone was also prepared by partial hydrogenation of aegelone (0·5 g.) in acetic acid (20 c.c.) over Adams catalyst (50 mg.). Hydrogenation was stopped when the rate of absorption became slow [absorption, 45 c.c. (N.T.P.); 1 mol. = 37·9 c.c.]. The product had m. p. 126—127°.

*Catalytic Hydrogenation of Dihydroaegelone.*—A solution of dihydroaegelone (0·5 g.) in glacial acetic acid (25 c.c.) was shaken with Adams catalyst (50 mg.) under hydrogen [absorption, complete in 30 min.: 40 c.c. (N.T.P.); 1 mol. = 37·7 c.c.]. Dihydroaegelin obtained in this way and crystallised from ethanol had m. p. 140—141°.

*Alkaline Permanganate Oxidation of Aegelin.*—Potassium permanganate (4 g.) and potassium hydroxide (0·5 g.) in water (100 c.c.) were gradually added to a boiling aqueous suspension of aegelin (1 g.) in water (10 c.c.). The excess of permanganate was then decomposed with methanol, the whole filtered, and the filtrate concentrated to 20 c.c., cooled, and acidified with hydrochloric acid. A white precipitate (0·4 g.) was obtained. It was separated into two crops by repeated fractional crystallisations from hot water.

The less soluble crop yielded needles (from hot water), m. p. 183—184° [Found: C, 63·0; H, 5·4; OMe, 19·8%; *M* (Rast), 150. Calc. for $C_8H_8O_3$: C, 63·15; H, 5·3; OMe, 20·4%; *M*, 152·14]. It was identified as *p*-anisic acid (mixed m. p.).

The more soluble crop yielded glistening plates (from water), m. p. 120—121° [Found: C, 68·6; H, 5·1%; *M* (Rast), 125. Calc. for $C_7H_6O_2$: C, 68·8; H, 4·95%; *M*, 122]. It was identified as benzoic acid (mixed m. p.).

In a second experiment the oxidation was carried with distillation. The clear aqueous distillate yielded benzaldehyde 2 : 4-dinitrophenylhydrazone (60 mg.), m. p. and mixed m. p. 235°.

*Hydrolysis of Aegelin with Hydrochloric acid.*—Aegelin (2 g.) was refluxed for 8 hr. with 90% ethanol (72 c.c.) and concentrated hydrochloric acid (12 c.c.), and then distilled in steam. The oily globules from the second fraction of the distillate were taken up in ether, washed with sodium hydrogen carbonate solution, then water, and dried. The oil (0·427 g.) obtained on evaporation of the ether was warmed with aqueous semicarbazide hydrochloride and sodium acetate containing a little ethanol for 0·5 hr. Overnight oily globules with a few crystals separated. The aqueous solution was removed and the remaining product washed with water, drained, and treated with ether to dissolve the adhering oil. The crystalline product (45 mg.) obtained was purified by crystallisation from ethanol and identified as *p*-anisaldehyde semicarbazone (plates), m. p. and mixed m. p. 203—204° (Found: C, 56·2; H, 5·5; N, 20·9; OMe, 15·25. Calc. for $C_9H_{11}O_2N_3$: C, 55·95; H, 5·7; N, 21·75; OMe, 16·1%). The ether washings from the semicarbazone on evaporation gave an oil (smell of ester) which was hydrolysed with 10% ethanolic potassium hydroxide, freed from ethanol, and acidified with hydrochloric acid; *trans*-cinnamic acid (0·19 g.) separated and crystallised from hot water as plates, m. p. and mixed m. p. 133—134°.

The residual liquid obtained after removal of volatile matter by steam was extracted with ether (extract A). The aqueous layer, on being made alkaline, became turbid, with evolution of ammonia. On extraction with ether and removal of the solvent a minute amount of basic crystals was obtained which in acid solution gave a creamy precipitate with Meyer's reagent.

In one experiment the acid solution after steam-distillation and extraction with ether was evaporated to dryness and the dry salt obtained was heated with alkali. The gases evolved were absorbed in ethanol and allowed to react with 1-chloro-2 : 4-dinitrobenzene: 2 : 4-dinitroaniline was obtained, and had m. p. and mixed m. p. 175—176°, confirming the presence of ammonia in the products of hydrolysis of aegelin.

The ethereal extract (A) was washed with sodium hydrogen carbonate solution. The alkaline extract on acidification with hydrochloric acid yielded *trans*-cinnamic acid (0·32 g.) which crystallised from hot water in plates, m. p. and mixed m. p. 133—134° [Found: C, 72·8; H, 5·25%; *M* (Rast), 147; equiv., 146·9. Calc. for $C_9H_8O_2$: C, 73·0; H, 5·4%; *M*, equiv., 148].

ω-*Amino*-p-*methoxyacetophenone Hydrogen Sulphate*.—This was prepared by a modification of Kindler and Peschke's method.[4] *p*-Methoxybenzoyl cyanide (1·3 g.) in acetic acid (15 c.c.) was hydrogenated over Adams catalyst (0·1 g.) at atmospheric pressure and room temperature. The hydrogenation was stopped after absorption of 2 mols. of hydrogen (20 min.). After filtration the dark brown acetic acid solution was treated with concentrated sulphuric acid (0·46 c.c.) and diluted with ether; ω-amino-*p*-methoxyacetophenone hydrogen sulphate (1·3 g.) separated. Crystallised from acetic acid, it had m. p. 167—168°.

ω-*Cinnamoylamino*-p-*methoxyacetophenone*.—An aqueous solution of the preceding sulphate (0·3 g.) was treated with *trans*-cinnamoyl chloride (0·3 c.c.) at room temperature. 10% Sodium hydroxide solution was added gradually with shaking till the mixture gave an ammoniacal smell and became pink. Then more cinnamoyl chloride (0·2 c.c.) was added and the whole shaken till the pink colour disappeared. The mixture was again made alkaline. The separated solid was collected and crystallised from ethanol, giving ω-cinnamoylamino-*p*-methoxyacetophenone as needles (0·2 g.), m. p. 159—160° (Lister and Robinson [5] give m. p. 153—154°) undepressed on admixture with aegelone (Found: C, 73·4; H, 5·9%). It gave a semicarbazone, m. p. and mixed m. p. 234—235° (decomp.).

As described by Lister and Robinson,[5] under the dehydrating action of concentrated sulphuric acid, aegelone also gives 5-*p*-methoxyphenyl-2-styryloxazole which in solution exhibits intense blue fluorescence.

p-*Methoxy*-ω-β-*phenylpropionamidoacetophenone*.—β-Phenylpropionyl chloride (0·3 c.c.) was added to an aqueous solution of the sulphate (0·3 g.), followed by 10% sodium hydroxide solution with shaking and cooling. The initial red colour disappeared on shaking and a pasty mass separated which gradually disintegrated into small lumps. When distinctly alkaline, the separated brick-red solid crystallised from ethanol, giving p-*methoxy*-ω-β-*phenylpropionamidoacetophenone* as needles (0·2 g.), m. p. and mixed m. p. with dihydroaegelone, 126—127° (Found: C, 72·7; H, 6·5; N, 4·5%). The semicarbazone crystallised from aqueous ethanol in needles, m. p. and mixed m. p. 138—140°.

*Catalytic Hydrogenation of the Hydrogen Sulphate of* ω-*Amino*-p-*methoxyacetophenone*.—The sulphate (0·7 g.) was shaken in glacial acetic acid (15 c.c.) and water (2 c.c.) with Adams catalyst (50 mg.) over hydrogen at atmospheric pressure. After absorption of about 0·66 mol. of hydrogen (*ca.* 45 min.), the rate of absorption became very slow. Fresh platinum catalyst (50 mg.) was introduced and the hydrogenation continued: it stopped after absorption of the remaining 0·34 mol. of hydrogen (*ca.* 20 min.). After filtration the clear colourless liquid was distilled under reduced pressure to remove acetic acid, and the residue treated with water and divided in two parts.

One part was condensed with *trans*-cinnamoyl chloride (0·3 c.c.) as above and the pasty mass obtained was crystallised from ethanol, giving *aegelin* as plates, m. p. and mixed m. p. 178—179° (0·15 g.) (Found: C, 72·85; H, 6·4; N, 4·5%).

The other part of the solution was condensed with β-phenylpropionyl chloride (0·3 c.c.) as before, and the solid product obtained was crystallised from ethanol, giving *dihydroaegelin* as leaflets, m. p. and mixed m. p. 140—141° (0·16 g.) (Found: C, 71·9; H, 7·1%).

Department of Chemistry, School of Tropical Medicine,
Calcutta-12, India.

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[4] Kindler and Peschke, *Arch. Pharm.*, 1931, **269**, 581.
[5] Lister and Robinson, *J.*, 1912, **101**, 1297.