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## CHEMICAL INVESTIGATION OF GERMINATED PEGANUM HARMALA SEEDS

KARIMULLAH A. ZIRVI and ASLAM BUTT \*

PCSIR Laboratories, Peshawar

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Germination embodies the mobilization of the food reserves stored in the seeds for the building up of raw material, and thus provides an important and interesting means to study the metabolic processes going on in plants. This is why several workers<sup>1-6</sup> have studied the biogenetic transformations in various plants by germinating their seeds.

The seeds of *Peganum harmala*, an alkaloidal plant indigenous to Pakistan, have been reported? to contain vasicine (peganine), a nonindole alkaloid, alongwith three indole alkaloids, namely, harmaline, harmine, and harmalol.

In the present work, we have germinated the seeds of *Peganum harmala*, and isolated an oil from the seedlings. Besides, two pure crystalline compounds having m.ps 137°C and 210–12°C have also been isolated. These have been characterized by the preparation of their derivatives and study of their elemental and spectral analysis, as β-sitosterol and vasicine respectively. We were unable to isolate any alkaloid of the harmine series.

## Experimental

Germination of the Seeds and Extraction.—The seeds were soaked in water for 24 hr and then spread in the folds of jute matting which was kept wet. The room temperature was kept between 35-37°C. while the seeds were exposed to diffused sunlight. After one week the seeds started sprouting. When the seedlings had acquired a length of  $1\frac{1}{2}$ " these were separated from the seed coverings and dropped in rectified spirit. Three percolations were effected, each after every 24 hr. On removal of the solvent under reduced pressure, the combined percolate gave 21 g of a reddish viscous matter as residue (19% by wt of the seedlings). It was digested with warm distilled water and the decanted solution was charcoaled and filtered. The clear reddish yellow solution was extracted with ether. The remaining aqueous solution was basified (IN KOH) and extracted with ethyl acetate, till the aqueous portion gave a negative alkaloid test.

Vasicine.—The ethyl acetate extract was dried (Na<sub>2</sub>SO<sub>4</sub>). On evaporation of the solvent

vasicine was obtained which was recrystallized several times from chloroform, m.p. and mixed m.p. 212° with decomposition. Its IR spectrum was superimposable with that of an authentic sample. (Found: C, 70.3; H, 6.5; N, 14.85; O, 8.5%. Calc. for C<sub>11</sub>H<sub>12</sub>N<sub>2</sub>O:C, 70.19; H, 6.43; N, 14.88; O, 8.5%.) Its picrate melted at 199°, and did not depress the m.p. of an authentic sample of vasicine picrate. (Found: C, 48.8; H, 3.5; N, 16.70%. Calc. for C<sub>11</sub>H<sub>12</sub>N<sub>2</sub>O.C<sub>6</sub>H<sub>3</sub>N<sub>3</sub>O<sub>7</sub>: C, 48.9; H, 3.6; N, 16.8%.)

 $\beta$ -Sitosterol and an Oil.—The ether extract, dried over Na<sub>2</sub>SO<sub>4</sub>, on evaporation gave a golden yellowish oily material. This was chromatographed on a column of neutral alumina, and eluted with light petroleum, light petroleum-benzene, benzene, and benzene-chloroform successively. The light petroleum eluate on removal of the solvent and purification by chromatography and repeated crystallization, gave pure β-sitosterol, m.p. and mixed m.p. 136–37°C. Acetylation with acetic anhydride and pyridine at room temperature yielded β-sitosterol acetate, which when crystallized from methanol, melted at 129–30°C. The IR spectra of the sterol and its acetate were identical with those of the authentic β-sitosterol and its acetate.

The light petroleum-benzene eluate gave an oil, which was found to have the following physicochemical characteristics: acid value=11.70; iodine value=12.20; hydroxyl value=12.50; acetyl value=12.28; saponification value=204.60.

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<sup>\*</sup>PCSIR Laboratories, Karachi.