

Base (VII) amounted to 10% of the total and was isolated in the form of the perchlorate with mp 216-217°C; it is an oily compound giving a hydrochloride with mp 196-197°C and a hydriodide with mp 231-233°C.

Bases (VIII) and (IX) with mp 143-144°C and 174-175°C, mol. wt. 317 (homo derivatives with mp 187-188°C and 147-148°C, respectively) were identical with piptanthine and piptamine [1]. The piptanthine amounted to 10% of the total and the piptamine to 0.07%.

The last two alkaloids have been isolated previously from this plant [3], thus, in the separation of the combined alkaloids we isolated five known and four new bases.

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ALKALOIDS OF *Thalictrum strictum*. III

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Continuing a study of the alkaloids of *Th. strictum* [1], we have separated the mixture of bases from the leaves, seeds, roots, and rhizomes. A preparative chromatographic investigation showed that the combined alkaloids of the leaves, of the seeds, and of the epigeal part have similar qualitative compositions. By chromatographing the combined bases from the leaves on a column of alumina followed by preparative separation on plates with a fixed layer (silica gel-gypsum) we isolated argemonine (I), thalicmine (II), and 2,3,7-trimethoxy-N-methyl-8,9-methylenedioxypavinan (II) [2]. Similarly, from the seeds we obtained bases (I) and (III). From the ether-soluble alkaloids of the roots and rhizomes we isolated (II), thalicsimidine, an unidentified base A (IV) [1], and base (V). Alkaloid (IV) is inactive, mp 117-118°C (CH₃OH), and gives a hydrochloride with mp 202-203°C. The UV spectrum of (IV) ($\lambda_{\max}$ 264, 285, 313, 327, 348, 366 nm) is characteristic for the ethylaminophenanthrene bases. The mass spectrum of (IV) shows the peak of the molecular ion with m/e 353 and strong peaks of ions with m/e 295, 251, 209, 58 (100%). The NMR spectrum (CDCl₃, δ scale) has signals at 2.34 ppm (singlet, 3H, NCH₃), 2.5-3.35 ppm (multiplet, 4H, -CH₂-CH₂-), 3.85 and 4.02 ppm (two singlets, 3H each, 2OCH₃), 6.04 ppm (singlet, 2H, CH₂O₂), 7.12 ppm (singlet, 2H, Ar-H), and 7.47 and 7.75 ppm (two one-proton doublets, J = 10 Hz). These facts have permitted the conclusion that the base (IV) is thalichthuberine, isolated previously only from *Th. thunbergii* [3]. A direct comparison of the R_f values and IR and NMR spectra of (IV) and of thalichthuberine* confirmed their identity.

Base (V) was isolated in the form of a viscous oil. UV spectrum of (V): $\lambda_{\max}$ 280, 305, 315 nm. A bathochromic shift was observed in an alkaline medium. The mass spectrum showed peaks of ions with m/e 371 (M)⁺, 370, 356, 340, 328. In the NMR spectrum there were signals at 2.49 ppm (singlet, 3H, NCH₃), 3.80, 3.85, and 3.89 ppm (12H, 4OCH₃), and 6.76 and 7.91 ppm (two one-proton singlets, Ar-H). The spectral characteristics and a comparison of them with literature data permitted the assumption that (V) is preocotene [4]. To confirm this, we prepared the O-methyl ether of (V) and showed its identity with an authentic sample from

*The sample of thalichthuberine and its IR and NMR spectra were kindly given to us by Prof. Tomimatsu (Japan).

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its R_f values and mass and NMR spectra. From the fourth fraction we obtained magnoflorine and berberine iodides.

Thus, from *Th. strictum*, which has not previously been studied chemically, we have isolated ten bases: thalicminine, O-methylcassyfiline [1], thalicmine, argemonine, 2,3,7-trimethoxy-N-methyl-8,9-methylenedioxypavinan, thalictuberine, preocoteine, thalicsimidine, magnoflorine, and berberine. Thalichminine has been isolated from the roots of *Th. minus*, *Th. simplex*, and *Th. isopyroides* [5] but in the plant under investigation thalicminine was found in the seeds, leaves, and epigeal part but it was not detected even chromatographically in the roots. 2,3,7-Trimethoxy-N-methyl-8,9-methylenedioxypavinan is a new base which we first isolated from the epigeal part of *Th. strictum* [1, 2].

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ISOLATION OF AMINOACYL-tRNA SYNTHETASES AND THEIR USE IN THE AMINOACYLATION OF tRNA

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In the present communication we give the results of the isolation of aminoacyl-tRNA synthetases (ARSases) from the seeds of the cotton plant of variety 108-F and their use in the aminoacylation of the tRNA from the same source.

The ARSases were obtained by the method of Merrick and Dure [1] with some modifications. A dry defatted powder of cotton seeds was extracted with homogenization in the cold in 0.1 M tris-HCl buffer, pH 8.0, containing 0.01 M $MgCl_2$, 0.005 M 2-mercaptoethanol, and 0.001 M EDTA. The ratio of seeds to buffer was 1:6. The homogenate was centrifuged at 27,000g. The precipitate was discarded, and the supernatant was treated with 2% protamine sulfate to eliminate nucleic acids. After centrifuging, the supernatant liquid was fractionated with $(NH_4)_2SO_4$, and the fraction between 30% and 60% saturation was collected. It was dissolved in the minimum volume of the extraction buffer and stored at $-20^\circ C$. The protein was determined by Lowry's method [2].

In aminoacylation, we used the tRNA isolated from cotton seeds by the method of Holley et al. [3] with some modifications. The incubation mixture with a volume of 0.25 ml contained (μ mole): tris-HCl buffer pH 7.0-9.0, 25; EDTA, 0.25; 2-mercaptoethanol, 0.25; Mg^{2+} , 2.5; ATP, 0.25-2.5; NH_4Cl , 2.5; [^{14}C](amino acid)s, 2 μCi ; tRNA, 2-3 o.u.; enzyme, 3 mg of protein. The time of incubation was varied.

Samples with a volume of 50 μl each were deposited on paper disks, these were washed several times with 5% TCA, ethanol, and ether, and were dried in the air. The radioactivities of the disks were counted in a LS-100C liquid scintillation counter with an efficiency of 65% for ^{14}C .

In a study of the influence of ATP and Mg^{2+} on the catalytic activity of the enzyme it was found that the largest amount of valyl-tRNA was synthesized when 2.5 μ mole of Mg^{2+} and

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