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# MONITORING OF THE PURITY OF ANABASINE HYDROCHLORIDE

### BY THIN-LAYER CHROMATOGRAPHY

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Anabasine hydrochloride has been recommended by the Pharmacological Committee of the Ministry of Public Health of the USSR for manufacture and wide medicinal use. The starting material for the production of anabasine hydrochloride is N-nitrosoanabasine, isolated from technical anabasine sulfate by nitrosation [1]. The possibility of the presence of N-nitrosoanabasine in a pharmacopoeial preparation of anabasine hydrochloride must be avoided. The chromatographic analysis of N-nitroanabasine in a pharmacopoeial preparation of anabasine hydrochloride in a nonfixed layer of alumina has been described only in a provisional pharmacopoeial paper [2]. The main disadvantage of this method is the inconvenience of its performance, which is connected with the free-flowing nature of the alumina and its low sensitivity for the determination of N-nitrosoanabasine (500  $\mu$ g).

There have been no publications devoted to the thin-layer chromatography of anabasine hydrochloride in a fixed layer of silica gel. Consequently, we have made a choice of the optimum system of solvents and of the amount of deposited substance.

For thin-layer chromatography we used type KSK silica gel prepared by the method of the State Pharmacopoeia [3]. Silica gel (2 g) and gypsum (0.1 g) were carefully mixed with distilled water (8 ml), and a uniform layer was deposited on a glass plate with dimensions of  $50\times200$  mm. The prepared plate was activated by drying at room temperature for 24 h. On the starting line, 125 µg of the preparation in the form of a solution in methanol was deposited as a spot. Chromatography was carried out by the ascending method in a cylindrical chamber  $(80\times200 \text{ mm})$ . The plate was treated with the Dragendorff reagent. The following solvent systems were used: 1) chloroform methanol (2:1), 2) chloroform methanol—acetone (25:7:3), 3) ether methanol (50:1), and 4) benzene methanol—acetone (10:5:2). The best separation was achieved in system 2. The selected solvent system permits the separation of anabasine hydrochloride ( $R_f$  0.35) from N-nitrosoanabasine ( $R_f$  0.92). The minimum detectable amount of anabasine hydrochloride is 1 µg and of N-nitroanabasine 1.2 µg.

The chromatographic method in a thin layer of sorbent that we have developed is recommended for introduction into the draft Pharmacopoeia paper on anabasine hydrochloride and the Provisional Pharmacopoeial paper on anabasine hydrochloride — a standard as a test for the purity of the latter.

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The purity of five laboratory and five industrial batches of the pharmacopoeial preparation of anabasine hydrochloride and of five laboratory batches of standard anabasine hydrochloride was checked by the above-described method.

No N-nitrosoanabasine was detected in any of the batches, which shows the high purity of the pharmacopoeial preparation and of the standard anabasine hydrochloride.

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ALKALOIDS OF Veratrum nigrum. II

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From the combined alkaloids extracted by ethers from the roots and rhizomes of Veratrum nigrum L. [1], by chromatography on a column of cellulose [2] we have isolated five alkaloids with  $R_f$  0.1 (I), 0.22 (II), 0.6 (III) (chloroform saturated with formamide; paper impregnated with formamide), 0.47 (IV), and 0.7 (V) (chloroform-benzene (7:3)).

The compounds have been identified by the methods used for the alkaloids of *Veratrum lobelianum* Bernh. [1, 3-7]. The results of analysis showed that (I) was dideacetylprotoveratrine A [3], (II) was veramarine [4], (III) was germidine [1], (IV) was protoveratrine A, and (V) was verazine [4]. This is the first time that the alkaloids (I-V) have been found in *Veratrum nigrum*.

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