

The optimum conditions for the formation of a complex of berberine bisulfate with eosin have been determined. A procedure has been developed for the photometric determination of berberine bisulfate.

Berberine is an alkaloid isolated from the roots and leaves of Berberis vulgaris. It is used in medical practice in the form of the bisulfate as a cholagogue [1].

Various methods have been proposed for the quantitative determination of berberine bisulfate: acid-base titration [2], extraction-spectrophotometric [3], phototurbidimetric [4, 5], and UV-spectrometric and UV-spectrophotometric [6-8] determinations. The low sensitivity of the titrimetric procedure, the lengthiness of the extraction-spectrophotometric method, and the inadequate accuracy of this phototurbidimetric method of determination are the main disadvantages of the procedures described.

Our aim was to develop a highly sensitive procedure simple in performance for the quantitative determination of berberine bisulfate. It has been found that berberine bisulfate interacts with the chromogenic organic reagent with an acid nature eosin (2,4,5,7-tetrabromofluorescein) with the formation of a complex compound. The colorations and optical densities of solutions of berberine with eosin depend on the pH of the solutions, the concentration on eosin and of berberine, and other factors.

In this work we used aqueous solutions of eosin (0.07%), of polyvinyl alcohol (0.1%), and of hydrochloric acid (0.05 N). The optical densities of the solutions were measured on a SF-16 spectrophotometer and a KFK-2 MP photoelectric colorimeter in cells with a layer thickness of 1 cm, and the pH values of the solutions were monitored with the aid of a EV-74 universal pH-meter. The optimum conditions for the photometric determination were investigated and selected: pH 2.4-3.3. The necessary concentration of hydrogen ions was maintained by the addition of 1 ml of a 0.05 N solution of hydrochloric acid. The stability of the coloration of solutions of the complex increases in the presence of polyvinyl alcohol. The optical densities of the solutions of the complex reached their maximum value in the course of 3-5 min and then remained constant for not less than 3 h. In view of the considerable overlapping of the electronic spectra of eosin (λ_{\max} 515-520 nm) and the complex (λ_{\max} 530-540 nm), it is recommended to perform the measurements of the optical densities of the solutions in the spectral region corresponding to the maximum difference between the light-absorption of the complex and of eosin (540-545 nm).

The calculated value of the molar absorption coefficient ($7.2 \cdot 10^4$) shows the high sensitivity of the photometric reaction of berberine with eosin. The light-absorption of solutions of the complex obey Beer's law in the interval of berberine bisulfate concentrations of 0.3-8.0 $\mu\text{g/ml}$.

A procedure has been developed for the photometric determination of berberine in sulfate in 0.005-g tablets. It was established by preliminary experiments that the fillers and auxiliary substances present in the tablets do not interfere with the photometric determination of berberine bisulfate by the reaction with eosin.

Below we give the results of the determination of berberine bisulfate in 0.005-g tablets:

Weight of the tablet mass, g	Amount of berberine found in one tablet, %	Metrological characteristics
0,212	101,42	$\bar{x} = 100,23\%$
0,2085	100,17	$S = 1,79$
0,1954	98,84	$S_{\bar{x}} = 0,80$
0,2104	98,20	$E_{0,95} = 2,22$
0,1970	102,54	$\Delta_{rel} = 2,21$

The procedure developed is highly sensitive, simple in use, and does not require the use of harmful reagents and organic solvents. The determination of berberine bisulfate is possible both on a spectrometer and on a photoelectric colorimeter, which makes the method more accessible. It can be used in the analysis of medicinal forms containing berberine bisulfate.

EXPERIMENTAL

Procedure for Determining Berberine Bisulfate in 0.005-g Tablets. About 0.2 g of a powder of ground tablets is placed in a 100-ml measuring flask, 50 ml of distilled water is added and, after 3 minutes' shaking, the volume of the solution is made up with water to the mark and it is stirred and filtered. Of this filtrate, 1 ml is added to a 25-ml measuring flask together with 2 ml of a 0.1% solution of polyvinyl alcohol, 1.5 ml of a 0.7% solution of eosin, and 1.5 ml of a 0.05 N solution of hydrochloric acid, and the volume of the solution is made up to the mark with water. Solutions for constructing a calibration graph containing 10-200 μg of berberine bisulfate corresponding to the requirements of GFX [State Pharmacopeia, Xth edition] are prepared similarly. The optical densities of the solutions are measured in a spectrophotometer at 545 nm or in a photoelectric colorimeter in cells with a layer thickness of 1 cm relative to a comparison solution containing all the ingredients apart from the berberine bisulfate. The amount of berberine bisulfate in the tablets is calculated from the calibration graph with allowance for the dilution performed.

SUMMARY

1. The optimum conditions for the formation and the spectral characteristics of a complex of berberine bisulfate with eosin has been determined.

2. A procedure has been developed for the quantitative determination of berberine bisulfate in tablets.

LITERATURE CITED

1. M. D. Mashkovskii, Drugs [in Russian], Meditsina, Moscow, Vol. 1 (1968), p. 513.
2. FS [Pharmaceutical Standard] 42-727-73.
3. T. Sakai and N. Ohno, Chem. Lett., No. 1, 107 (1982).
4. N. V. Cherepanova, É. K. Titova, E. V. Vaganova, and N. A. Reneva, The Investigation of Drugs and the Provision of the Population with Them [in Russian], Sverdlovsk (1983), p. 68.
5. A. I. Popov, L. M. Bazhina, G. I. Oleshko, and G. I. Kudymov, Farmatsiya, 26, No. 2, 78 (1977).
6. A. I. Tses'ko and E. Ya. Ladygina, Farmatsiya, 20, No. 2, 28 (1971).
7. A. I. Tses'ko and E. Ya. Ladygina, Farmatsiya, 23, No. 5, 27 (1974).
8. V. M. Musyanovich, Khim.-farm. Zh., 11, No. 10, 134 (1977).