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ANALYSIS OF INDAPAMIDE DRUG BY CHROMATOSPECTROPHOTOMETRIC METHOD

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Annotation

Diuretics are drugs that increase the excretion of pus from the body. Drugs reduce the amount of fluid in tissues and body cavities, so they are used for certain heart, kidney and liver diseases, in which water accumulates in the body, causing swelling, as well as lowering blood pressure. Drugs should only be used as directed by a doctor, as there are several side effects of these drugs (hypo and hyperkalemia, hyperuricemia, alkalosis, acidosis, cardiac arrhythmias, gynecomastia, allergic reactions, nephrotoxicity, etc.).

Key words: *diuretic, indapamide, thin layer chromatography, chromatospectrophotometry, solvent system, reagents.*

INTRODUCTION:

Diuretic drugs, including the toxicity of indapamide, are associated with their pharmacological effects, reducing fluid volume and contributing to electrolyte loss; these include dehydration, hypokalemia (or hyperkalemia with spironolactone and Triamterene), hypomagnesemia, hyponatremia, and hypochloremic alkalosis[4]. Electrolyte imbalances can lead to cardiac arrhythmias and increase digital toxicity. Diuretics are considered to have pharmacological mechanisms that affect solution and water loss [2]. In case of overdose, arterial hypotension, bradycardia and drowsiness appear. The natriuretic, kaliuretic and vascular dilation effect of indapamide is enhanced by an overdose, which leads to diuresis, hypokalemia and arterial hypotension. Intracellular potassium metabolism leads to hypokalemia [3]. Experts assure that elderly patients with a high risk of self-poisoning should be recognized and avoid the use of drugs with a high dose of toxic effects. In order to reduce cases of acute poisoning and death, it will be important to limit the use of dangerous drugs (for overdose). [5,7].

MATERIALS AND METHODS

In forensic chemical practice, the method of chromatospectrophotometry is widely used to determine the amount of toxic substances isolated from biological objects. In this case, a thin layer of toxic substances extracted from biological objects is isolated by the chromatography method, and then the amount of the substance is determined by the spectrophotometry method. For this reason, the aim of the study was to determine the amount of indapamide drug in a chromatospectrophotmetric way [8].

The richness of separations from biological materials, which are forensic chemical and chemical-toxicological objects, the presence of other substances in its composition and the presence of a verifiable substance in low quantities require the development of sensitive, highly effective and private methods for analysis. Among the methods that meet the same requirements, the role of chromatography is incomparable. The method has not lost its practical significance to this day due to its simplicity, breadth of application area, sensitivity[10]. On the contrary, it is known that the method is used in forensic chemistry, ecology and chemical-toxicological analyzes, on the one hand as a method for determining the quality of matter, and on the other hand, for cleaning deductions from biological objects. This plays an important role in these analyzes.

The method of UB spectrophotometry is widely used in daily analyzes, considering that experiments are carried out quickly and easily, and almost all laboratories are provided with this instrument, it is considered appropriate to study the UB spectra of indapamide. The sensitivity of this method allows it to analyze extracts extracted from the composition of biological objects, after certain cleaning. Taking this into account, in forensic chemical practice it was found that the amount of indapamide isolated from the composition of biological objects and biological fluids should be determined by the method of UB spectrophotometry [6].

RESULTS AND DISCUSSIONS:

Selection of solvent system in thin layer chromatography analysis.

In thin-layer chromatography, it is important to choose the right system of solvents when distributing and separating substances from each other. After all, a properly selected mixture of organic solvents is a quality indicator (in terms of Rf value) for this substance, but also plays an important role in cleaning from foreign substances[1]. Also, when analyzing substances in the method, it is necessary that their Rf values are in the range of 0.4 - 0.8, and this is achieved by using organic solvents in different proportions. Before choosing a mixture of organic solvents, the solubility of indapamide in various solvents was experimentally checked. The results of the experiment are presented in Table 1

1-table

№	Solvent	Solubility
1	Water	Does not melt
2	0.1H NaOH	Low melting
3	Sulfuric acid	Soluble
4	0.1 H hydrochloric acid	Soluble
5	Acetone	Soluble
6	Benzal	Soluble
7	Hexane	Soluble
8	Ethylacetate	Soluble
9	Chloroform	Soluble
10	Xidmətlər	Soluble
11	Ethyl alcohol	Soluble

To carry out the analysis in the method, it was dripped from 0.1 ml of the standard solution of indapamide in ethyl alcohol, which stored 1 mg/ml on the start line of the plates "Silufol UB-254", and dried at room temperature. Then, dropping plates into chromatographic chambers with a mixture of several organic solvents, the mixture of solvents rose to a height of 10 cm, and when it reached the finish line, the plates were taken and dried at room temperature. When conducting the experiment, various chemical reagents were used in order to determine the place of accumulation of the substance on the chromatographic plate. Of these: the formation of colored spots was observed using UB-rays (pumpkin-colored), a 50% solution of nitric acid (pink). The results are presented in Table 2.

Results of the selection of a mixture of organic solvents used in the analysis of indapamide by the method of thin-layer chromatography

2-table

№	Mixture of organic solvents	Rf value of indapamide
1	Benzal: acetone (80:20)	0,43-0,45
2	Benzene : dioxane: ammonia (60:35: 5)	0,67-0,69
3	Chloroform: acetone (18: 1)	0,60-0,62
4	Diaksan: talual (10: 3)	0,80-0,82
5	Prapanal: benzal: talual (10: 5:2,5)	0,88-0,90
6	Benzene: dichloroethane:propanol (10: 7: 3)	0,85-0,87
7	Dichloroethane : dioxane: ammonia (8:4: 1)	0,36-0,38
8	Propanol : dioxane: ammonia (7:4: 0.5)	0,77-0,79
9	Chloroform: dichloroethane: ammonia (8:7:1)	0,44-0,46
10	Benzene: propanol: ammonia (7:7:0,5)	0,88-0,90
11	Chloroform: benzene: ammonia (6:5:0,5)	0,65-0,67
12	Toluol : dioksan: ammiak (10:4:1)	0,55-0,57
13	Benzene: dichloroethane (7:2,5)	0,20-0,22
14	Toluene : dichloroethane (10:3)	0,01-0,03
15	Dichloroethane: toluene: benzene (7:3:1)	0,02-0,04
16	Benzene: ammonia (15:0,5)	0,01-0,03

Determination of reagent sensitivity in thin layer chromatographic analysis of indapamide

In chromatography, it is important to determine the sensitivity of the substance-detecting reagents for this substance. In this regard, the sensitivity of the following proposed methods of analysis for this substance was studied. For this, a number of working standard solutions were prepared, the concentration of which decreases from the solution of the standard sample of indapamide. Samples of these solutions were dripped into the start line of the chromatographic plate using microshprits in a circle 0.4-0.5 cm wide at a distance of 2cm from each other, and chromatographed in a mixture of organic solvents stored in chloroform : benzene : ammonia (6:5:0.5), and then sprayed with the recommended chemical reagents. The results of the analysis performed are presented in Table 3.

Results of determining the sensitivity of stain-forming reagents by the method of Chromatography of a thin layer of indapamide.

3-table

Reaction sensitivity,mkg	Stain-forming reagents					
	UB-light	HNO_3 50%	Marki reactant	Erdman reactant	Mandelin reactant	5% $FeCL_3$ 20%HCl 50% HNO_3
10	+	+	+	+	+	+
9	+	+	+	+	+	+
8	+	+	+	+	+	+
7	+	+	+	+	+	+
6	+	+	+	+	+	+
5	+	+	+	+	+	+
4	+	+	-	+	+	+
3	+	+	-	+	+	+
2	+	+	-	-	-	+
1	+	+	-	-	-	-
0,5	+	+	-	-	-	-

Based on the results presented in Tables 2 and 3, a mixture of solvents in the ratio chloroform : benzene : ammonia (6:5:0.5) from the system of organic solvents used to perform chromatographic analysis of indapamide was found to be appropriate. In it, the approval of indapamide in the Rf_value range of 0.65-0.67 was achieved. When lightening the stain, the most sensitive was considered to be UV-light peeling and a 50% solution of nitric acid. Their sensitivity to the substance was 0.5 mcg.

Selection of sorbents in thin-layer chromatographic analysis of indapamide. At the next stage of the experiment, the effect of sorbents on the analysis of YUQX in determining the chinity of indapamide was studied. For this," Silufol 254 "was used" Siligagel", chromotographic plates that kept the sorbent. Chromatographic plates, in which silicagel (13% kept plaster) stored sorbent, were prepared in laboratory conditions. Chromatographic plates were dried at room temperature and activated on a drying shelf for 30 minutes at 105Ċ. Ready-made silicagel plates were stored in special containers until analysis – an exciter. To perform the analysis, the plates were dripped 0.1 ml of the standard solution of indapamide 1 mg/ml into the start line, dried at room temperature, and the analysis was carried out under the conditions described above. The results of the analysis performed are presented in Table 4.

Results of the study of the effect of sorbents in the analysis of indapamide by the method of thin-layer chromatography.

4-table

Selected systems	Applied plates		
	"Silufol UV-254"	Aluminum oxide	Siligagel
Benzol:dioksan:ammiak (60:35:5)	0,63-0,65	0,67-0,69	0,67-0,69
Xloroform :atseton (18:1)	0,40-0,42	0,66-0,68	0,60-0,62
Xloform:benzol :ammiak (6:5:0,5)	0,21-0,32	0,80-0,82	0,65-0,67

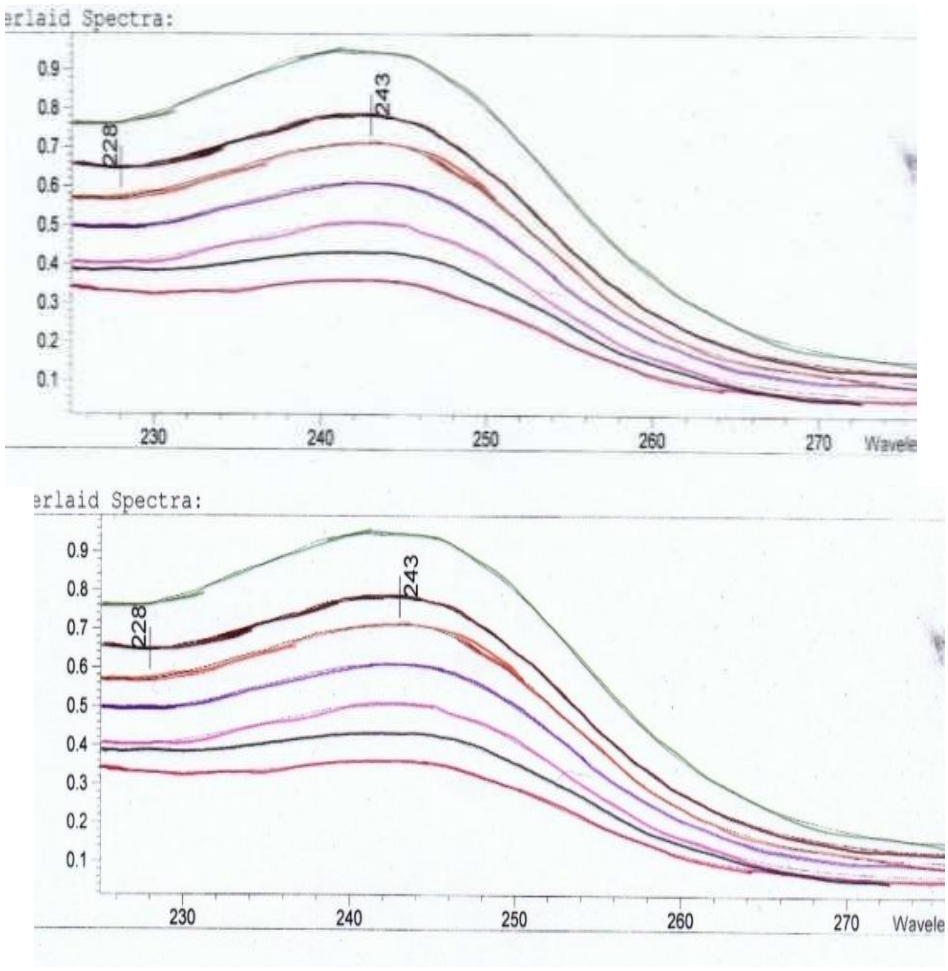
The results of the analysis showed that the use of chromatographic plates containing" aluminum oxide "and the sorbent" Siligagel " prepared in laboratory conditions was recognized as expedient.

Conducting qualitative analysis of indapamide by spectrophotometric method. The analysis of the indapamide drug in the UB-spectrophotometric method was carried out on a spectrophotometer branded 8453e Spectroscopy System from the firm "Agilent Technologies".

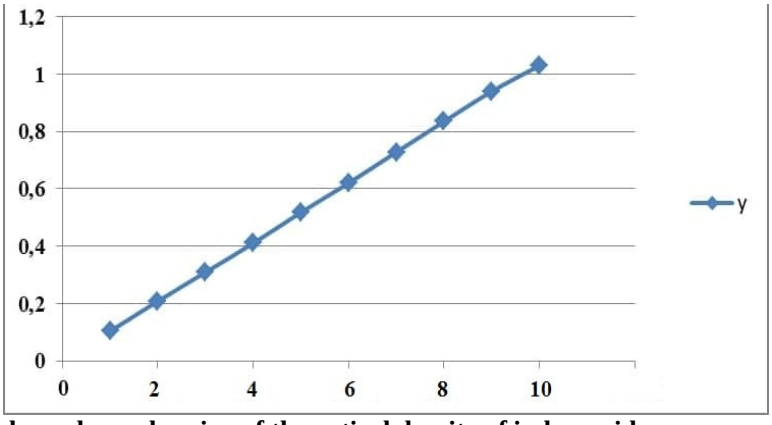
When the substance was completely dissolved, it was delivered with 96% ethyl alcohol up to the size mark (a solution). A was taken 1ml from the solution and placed in a measuring flask with a volume of 100mL, and the volume was delivered with 96% ethyl alcohol (solution B). In cuvette with a layer thickness of 10 mm from this solution, an analysis of the wavelength from 200 to 400 nm was carried out. 96% ethyl alcohol was used as a comparable solution. It has been achieved to confirm that the 96% ethyl alcohol solution of indapamide has a high beam absorption rate at a wavelength of 243 nm.

Conducting quantitative analysis of indapamide by spectrophotometric method.

UB-quantitative analysis of indapamide in the spectrophotometric method was calculated through a calibrated drawing. To do this, from the V solution prepared above, working standard solutions of indapamide were prepared, which kept 1-10 mcg/ml in the composition, and an analysis was carried out in cuvette with a layer thickness of 10 mm, at a wavelength of 243 nm. 96% ethyl alcohol was used as a comparable solution. The results of the analysis are presented in Figure 2 and Figure 3.



2-fig Beam absorption indicators of indapamide working standard solutions



3-fig. Concentration dependence drawing of the optical density of indapamide

Based on the results obtained, the comparative and molar light absorption indicator values of indapamide were calculated. The results of the analysis performed are presented in Table 5.

Comparative and molar light absorption indicators of indapamide
Determination results (n=5)

5-table

Amount of substance, mkg/ml	Optical density (D)	Specific light absorption indicator (E)	Molar light absorption indicators (ε)
1	0,103	103	3765
2	0,208	104	3801
3	0,309	103	3764
4	0,410	100	3655
5	0,518	104	3787
6	0,620	103	3776
7	0,728	104	3801
8	0,835	104	3801
9	0,940	104	3816
10	1,030	103	3765
Average		103	3774

According to him, the specific and molar light absorption indicators of indapamide were 103 and 3774 values, respectively.

A quantitative analysis of indapamide was carried out with the aim of checking the accuracy and repeatability of the analysis conditions developed by the spectrophotometric method. For this, 5 samples were prepared from 7 μg/ml of indapamide solution, and the optical density of solutions was determined at a wavelength of 243 nm on a spectrophotometer. Based on the compiled calibration drawing, the amount of indapamide was determined and the Metrological report was calculated according to the DF XI edition. The results of the analysis obtained are presented in Table 6 [9].

Results of UB-spectrophotometric analysis of the amount of indapamide

6-table

Preparat miqdori mkg/ml	Topilgan miqdori		Metrologik tavsifi
	mkg/ml	%	
7,0	7,02	100,3	$X_{ўр}$=100,04 S^2=0,037 S =0,336 S_x=0,150 ΔX=1,963 $ΔX_{ўр}$= 0,877 E=1,962% $E_{ўр}$=0,877
7,0	6,98	99,8	
7,0	7,02	100,4	
7,0	6,97	99,6	
7,0	7,01	100,1	

As can be seen from Table 6, as a result of spectrophotometric analysis of indapamide, an average determination of 100.04% was achieved. The average relative error in this was 0.877%. The results obtained indicate that the developed method can be used in determining the amount of indapamide extracted from a biological object.

Determination of the quality and quantity of indapamide drug by chromotospectrophotometric method.

1. To carry out this analysis, the indapamide, which stored 1 mg/ml on the start line of 5 “silica gel” plates, was dripped from 0.1 ml of standard solutions and dried at room temperature, dropping plates into chromatographic chambers with a mixture of organic solvents, which stored chloroform : benzene : ammonia (6:5:0.5), the solvent mixture rose to a height of Then the sorbents glued on the surface of the plates were dissolved using 96% ethyl alcohol, eluating the indapamide contained in them. By filtering solutions, the volume was increased to 10 ml and carried out in the UV spectrophotometer of the Agilent Technologies firm 8453 e Spectroscopy System brand, in cuvette with a layer thickness of 10 mm, at a wavelength of 243 Nm. The Metrological report of the quantitative analysis of indapamide, determined by the chromotospectrophotometric method, was calculated according to the DF XI edition. The results of the analysis are presented in Table 7.

The average relative error of 97.6% as a result of chromatospectrophotometric analysis of the amount of indapamide was 2.72 values. The results presented in the table also indicate that the developed method can be used to determine the amount of indapamide extracted from biological objects to determine the amount.

Results of quantitative analysis of indapamide by chromotospectrophotometric method 7- table

Amount of substance, mg/ml	The amount of substance found as a result of the analysis		Results of Metrological analysis
	mg	%	
1,0 1,0 1,0 1,0 1,0	0,98 0,96 0,96 0,98 0,98	98,8 96,5 96,7 98,2 98,2	X=97,6 S=1,01 S_x=0,45 ΔX=0,0304 ΔX=0,0136 E=6.08 E_a=2.72

CONCLUSION:

1. Analysis conditions for indapamide by the method of thin-layer chromatography have been developed. A mixture of solvents in a ratio of chloroform : benzene : ammonia (6:5:0.5) from the system of organic solvents used to carry out the analysis was found to be appropriate. When lightening the stain, the most sensitive was considered to be UB-light peeling and a 50% solution of nitric acid. Their sensitivity to the substance was 0.5 mcg.

2. The conditions for the analysis of indapamide by the quantitative method of UB spectrophotometry were studied. The average values of salishtima and molar light absorption indicators at high beam absorption wavelengths in the UB-light field were 103 and 3774 values on average, respectively. Under the conditions of analysis, the linearity range of indapamide was 1-10 mcg/ml.

3. As a result of chromatospectrophotometric analysis of the amount of indapamide, the average was X=97.6 %, and the average relative error was E=2.72 %.

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