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# Spectrophotometric studies and application of imipramine–eriochrome cyanine R system for determination of imipramine in pharmaceuticals

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**Abstract**

Eriochrome cyanine R (ECR) has been tested as reagent for the determination of imipramine. It reacts in neutral medium with imipramine forming reddish compound, which can be quantitatively extracted into n-butanol. This property has been successfully used for the extractive-spectrophotometric determination of imipramine. Beer's law is obeyed in concentration range of 10–80 μg $ml^{-1}$ of imipramine. The method was applied to the determination of imipramine in its pharmaceutical. © 2000 Elsevier Science B.V. All rights reserved.

*Keywords:* Spectrophotometry; Imipramine–eriochrome cyanine R system; Imipramine; Pharmaceuticals

## 1. Introduction

Imipramine, a dibenzazepine derivative, is widely used for the treatment of depression. Due to its structure, it is often referred to a tricyclic antidepressant. Studies have indicated that the efficacy of this drug in alleviating depression at night is due to the enhancement of noradrenergic activity through the blockage of norepinephine reuptake in blockage peripheral and central noradrenergic system. In vieco of the importance of imipramine, considerable work has been done for their detection and quantification [1]. There are several methods of determination of imipramine. Chromatographic methods with UV or coulometric [2,3] detection are very often applied for this purpose. In scientific literature are also described [4,5] procedures utilised for determination of imipramine through various voltammetric [6] methods. The spectrophotometric method seems to be most often employed for the determination of imipramine in bulk and pharmaceuticals due to their simplicity and common access to instrumentation. Those methods are based on chemical properties of imipramine. Some spectrophotometric methods are based on the formation of coloured compounds with a number of organic substances, e.g. methyl orange [7], bromothymol and methylthymol blue and chromazurol S [8,9], arylosulphonic acids [10,11], picric acid [12], picric chloride [13], erythrosine B [14].

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As a continuation of our previous studies on the determination of tricyclic antidepressants [15–17] this paper deals with the application of eriochrome cyanine R (ECR) to the extractive-spectrophotometric determination of imipramine. The application is based on the ability of ECR to form with imipramine coloured compound quantitatively extracted with n-butanol.

The composition of this compound has been established and some of its physicochemical properties have been examined.

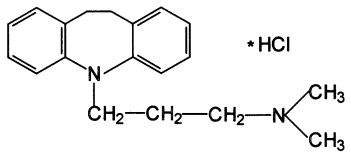
## 2. Experimental

### 2.1. Apparatus

Spekol-11 spectrophotometer (Carl Zeiss Jena, Germany), Hewlett-Packard spectrophotometer, Model 8452 A, Nicolet spectrometer ST-IR, Magna 550.

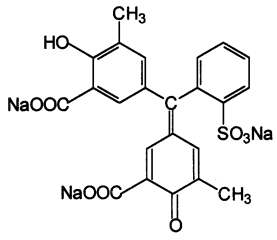
### 2.2. Reagents

Imipramine (10,11-dihydro-*N*,*N*-dimethyl-5H-dibenz [b,f]-azepin-5-propanamine) hydrochloride (IMP)




from Sigma Chemical Co. The standard $1 \times 10^{-2}$ mol $l^{-1}$ solution was prepared by dissolving 0.3162 g of IMP · HCl in distilled water.

Eriochrome cyanine R (ECR) from Lachema, Chemapol Praque. The substance was crystallised twice from acetone.




### 2.3. Procedure

Transfer a portion of 0.5–4 ml of $6.31 \times 10^{-4}$ mol $l^{-1}$ solution of imipramine solution into a 25 ml separatory funnel. Add 0.6–4.8 ml of $3.15 \times 10^{-3}$ mol $l^{-1}$ ECR solution and dilute with distilled water to the volume of 10 ml. The mixture, shake well with two 5-ml portions of n-butanol. Dilute the combined extracts to volume in a 10-ml standard flask with n-butanol. Measure the absorbance of coloured extracts at 520 nm in 1-cm cells against a reagent blank prepared in the same way but without imipramine.

### 2.4. Analysis

In order to confirm the applicability of extractive-spectrophotometric method, imipramine hydrochloride was determined in injections IMIPRAMIN (from Polfa). The declared concentration was 25 mg in 2 ml of solution and 20 different samples were analysed. The appropriate volume of sample was transferred into a 100-ml calibrated flask and made up to the mark with distilled water. Imipramine hydrochloride content in the diluted solution was determined as described above using extractive-spectrophotometric method. The results obtained were compared with those given by the pharmacopoeial method [13].

## 3. Results and discussion

Imipramine reacts with ECR in neutral medium to form reddish compound, which can be quantitatively extracted into n-butanol. This solvent does not extract ECR. Variation in the shaking time from 1 to 5 min revealed that the extraction is quantitative after 1 min at room temperature. The extracts were stable for about 1 h.

The effect of acids HCl, $H_2SO_4$, $HNO_3$, $H_3PO_4$, and boranic buffer on the formation and extraction of the complex into n-butanol was examined. It was found that the addition of the acids or boranic buffer (pH 6.7) decreases the absorbance of the extracts.

The influence of ECR concentration in relation to imipramine in aqueous phase on the absorb-

ance of the extracted compound was examined. The maximum absorbance was achieved for a 3–9 excess of ECR with respect to imipramine.

Chemical composition of the compound of imipramine with ECR was established by Job's method and spectrophotometric titration; the stoichiometry was found to be 1:1.

Studies of the spectroscopic properties of reagents and the compound formed in the IMP–ECR system was carried out in the UV–VIS and IR regions. It was found that the compound exhibits two main absorption peaks in ultraviolet region at 220 and 252 nm and one peak at 520 nm in visible region. The spectrum was compared with UV–VIS spectra of imipramine hydrochloride and ECR. In the visible range, the band of ECR ($\lambda = 520$ nm) is preserved in the compound, while in the UV range, two imipramine hydrochloride absorption bands ($\lambda = 210$ and $\lambda =$ 250 nm) are also preserved. Furthermore, no new absorption bands were observed in the spectrum of the compound examined. This data supports thesis that the formation of the compound by electrostatic interaction between cations of imipramine hydrochloride and anion of ECR. This type of compound can be classified as an ion-association complex.

The ion-association character of the examined compound was confirmed by Infrared (IR) spectroscopy. IR spectra of the compound (KBr discs) was recorded in the region 400–4000 $cm^{-1}$ with a Nicolet spectrometer ST-IR, Magna 550-series. The spectrum of the compound in the region 400–1600 $cm^{-1}$ is the sum of the spectra of the reagents. Significant changes in the spectrum of the compound are observed in the region 2300–2600 $cm^{-1}$. A wide band, appearing in the imipramine spectrum in the region 2300–3000 $cm^{-1}$ and characteristic vibrations of the $NH^+$ group did not appear in the spectrum of compound. This suggests that compound is formed with participation of a nitrogen atom from the tertiary amine group in the aliphatic chain of the imipramine hydrochloride.

From the obtained results, it can be concluded that the compound formed between IMP hydrochloride and ECR has an ion-association character.

This compound is precipitated from neutral aqueous solutions in the form of coloured sediment, sparingly soluble in water, which could be quantitatively extracted into n-butanol. Taking advantage of these properties a new extractive-spectrophotometric method for the imipramine hydrochloride determination was developed. Under the described optimal experimental conditions, the linear relationship between absorbance and concentration of imipramine hydrochloride was tested. The obtained results and the statistical evaluation of precision are given in Table 1. The results show that the Beer's law is obeyed at the concentration range of 10–80 μg $ml^{-1}$ of imipramine hydrochloride with correlation coefficient equal 0.9975. The coefficient of molar absorptivity is $4.8 \times 10^3$ l $mol^{-1}$ $cm^{-1}$ at 520 nm.

The elaborated method was applied successfully to the determination of imipramine hydrochloride in injections fluid IMIPRAMIN containing 25 mg

Table 1
The results of the extractive-spectrophotometric determination of imipramine hydrochloride using ECR

| Amount of IMP taken (μg $ml^{-1}$) | Absorbance $A$ | S.D. | R.S.D. (%) | $\pm t_{0.95} \cdot \bar{S}$ |
|---|---|---|---|---|
| 10 | 0.214 | 0.01 | 4 | 0.02 |
| 20 | 0.323 | 0.008 | 2 | 0.02 |
| 30 | 0.511 | 0.02 | 4 | 0.05 |
| 40 | 0.656 | 0.01 | 1 | 0.02 |
| 50 | 0.798 | 0.006 | 0.7 | 0.01 |
| 60 | 0.923 | 0.005 | 0.5 | 0.01 |
| 70 | 1.092 | 0.01 | 0.9 | 0.02 |
| 80 | 1.252 | 0.002 | 0.1 | 0.005 |

Table 2
The results of determination of imipramine hydrochloride in injection IMIPRAMIN

| IMP · HCl taken | IMP · HCl found using method | | Error (%)[a] | S.D. | R.S.D. (%) |
|---|---|---|---|---|---|
| | Proposed (mg) | Pharmacopoeial [18] (mg) | | | |
| Ampoule ≈25 mg | 25.15 | | 0.2 | | |
| | 25.2 | 25.1 | 0.4 | 0.029 | 0.11 |
| | 25.15 | | 0.2 | | |

[a] Versus pharmacopoeial method.

imipramine hydrochloride. The measurements were carried out according to the procedure described in the Section 2. The results of the proposed method show good agreement with those obtained by the pharmacopoeial method [18] (Table 2). Some excipients usually present in pharmaceuticals [18] like sodium sulphite, sodium pyrosulphite, ascorbic acid and sodium chloride do not disturb the determination of active compound. The elaborated method is simple, rapid and useful for the determination of imipramine hydrochloride content in injections IMIPRAMIN.

## References

[1] B.A. Scoggins, K.P. Maquire, T.R. Norman, G.D. Burrows, Clin. Chem. (Winston-Salem, N.C.) 26 (1980) 5.
[2] S.D. Yoo, J.W. Holladay, T.K. Fincher, M.J. Dewey, J. Chromatogr. B 668 (1995) 338.
[3] J.P. Foglia, D. Sorisio, J.M. Perel, J. Chromatogr. 57 (1991) 247.
[4] H. Hopkała, G. Misztal, Pharmazie 51 (1996) 96.
[5] H. Hopkała, M. Toborek, S. Zaręba, Acta Polon. Pharm. 53 (1996) 181.
[6] E. Bishop, W. Hussein, Analyst 109 (1984) 73.
[7] T. Perez-Ruitz, C. Martinez-Lozano, C. Alonso, Talanta 41 (1994) 1523.
[8] W.N. French, F. Matsui, J.F. Truelove, J. Can. Pharm. Sci. 3 (1968) 33.
[9] B.A. Person, S. Eksborg, Acta Pharm. Sci. 7 (1970) 353.
[10] M. Gajewska, J. Iliaszenko, M. Sokołowska-Muszyńska, Acta Polon. Pharm. 49 (1992) 17.
[11] M. Gajewska, J. Iliaszenko, M. Sokołowska-Muszyńska, Acta Polon. Pharm. 45 (1988) 125.
[12] M.E. Elnemma, F.M. El Zawawy, S.S.M. Hassan, Mikrochim. Acta 110 (1993) 79.
[13] S.A. Hussein, A.M.I. Mohamed, H.Y. Hassan, Talanta 36 (1989) 1147.
[14] T. Perez-Ruiz, C. Martinez-Lozano, V. Tomas, C. Sidrach, Analyst 120 (1995) 1103.
[15] B. Starczewska, J. Trace Microprobe Tech. 16 (1998) 151.
[16] B. Starczewska, J. Karpińska, J. Anal. Lett. 29 (1996) 2475.
[17] B. Starczewska, H. Puzanowska-Tarasiewicz, Farm. Pol. 49 (1993) 34.
[18] Polish Pharmacopeia, V, Warsaw, 1993.