Spectrophotometric determination of frusemide in some pharmaceuticals via oxidative coupling reaction

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الخلاصة

طورت طريقة طيفية بسيطة وذات دقة عالية لتقدير الفروسامايد في عدد من المستحضرات الدوائية باستخدام تفاعل الاقتران التأكسدي و تعتمد هذه الطريقة على تفاعل الفروسامايد مع البروميثازين في الوسط المائي الحامضي وبوجود أيون الهايبوكلورات بوصفه عاملا مؤكسدا وقيس الناتج ذو اللون الأزرق عند (625)نانوميتر والذي يصل ذروته حالا ويبقى مستقرا مدة لاتقل عن 24 ساعة.وقد لوحظ إن قانون بير يسرى على الكميات التي تتراوح بين(2-20)جزء بالمليون بامتصاصية مولارية 0.625×10^4 لتر.مول $^{-1}$.سم $^{-1}$ وان الانحراف القياسي النسبي للطريقة أقل من 2% ودقة الطريقة (معدل نسبة الاسترجاعية) 100.15. واستخدمت الطريقة بنجاح لتقدير الفروسامايد في حبوب وأمبول لازكس . واختبر نجاح الطريقة بمقارنتها بالطريقة القياسية الدستورية المعتمدة باستخدام اختبار (t) عند حدود ثقة 95% مما يدل على صلاحية التطبيق التحليلي للطريقة.

Abstract

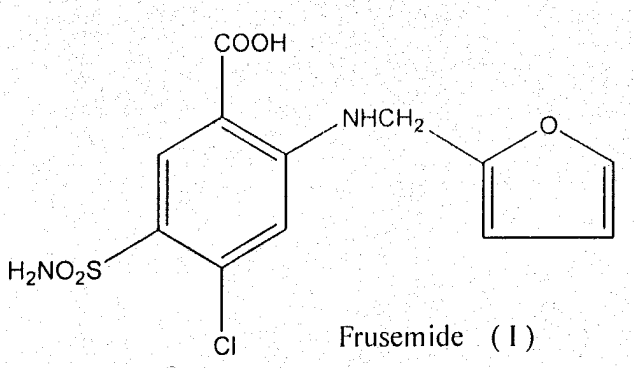
A simple and accurate spectrophotometric method has been described for the determination of frusemide in pharmaceutical preparation.The method is based on oxidative coupling reaction of frusemide with promethazine in aqueous acidic solution in the presence of hypochlorite ion and the absorbance of the blue product was measured at 625 nm against a reagent blank. The blue color formed immediately and stable for at least 24h.Calibration graph was rectinlinear for 2-20 ppm.The molar absorptivity is 0.625×10^4 l.mol $^{-1}$.cm $^{-1}$, relative standard

deviation of the method was less than 2% , and accuracy (average recovery %) was 100.15. The proposed method can satisfactorily be applied to the analysis of frusemid in pharmaceutical preparations. The present method compared statistically with official method using (t) value at 95% confidence level, the results indicated that there is no systematic error and the present method has good validity.

Keywords: frusemide,spectrophotometric,oxidative coupling.

Introduction:

Frusemide [4-chloro-2-furfurylamino-5-sulphamoyl benzoic acid] (I) is a loop diuretic which reduces the resorption of electrolytes by the proximal and distal renal tubules and by the loop of henele.Excretion of sodium,potassium and chloride ions is increased and as a consequence water exceretion is enhanced [1]. Frusemide is indicated in the treatment of edema associted with congestive heart failure, cirrhosis of the liver,renal disease and hypertension [2].



Frusemide (I)

Few methods for the determination of frusemide have been described in the literature, among these the high performance liquid chromatography [3-6] . Very few spectrophotometric methods have been reported for the determination of frusemide including the official method [7] .Another methods involves diazotization [8] , flow-injection spectrophotometric [9] , complexation with palladium(II) [10] and first derivative spectrophotometric methods [11-12] .However these methods lack of simplicity needed for routine analysis.This paper describe simple,rapid and accurate analytical method for determination of frusemide in aqueous solution without extraction by an organic solvent.

The proposed method was applied successfully for the assay of frusemide in pharmaceutical preparations .

Experimental:

Apparatus:

Spectrophotometer: A Genway 6405 Uv/Vis was used.

Reagents:

All reagent used should be of analytical reagent grade . The water was always twice distilled .

Frusemide .HCl solution: 100 PPM ($3x10^{-4}$ M)

Dissolve 0.1 g of frusemide .HCl in 1L ethanol.

Promethazine.HCl solution: ($6x10^{-4}$ M)

Dissolve 0.192 g of reagent in 1L distilled water.

Acetic acid: 3.5 %

Sodium hypochlorite: 0.1 %

Recommended procedure:

A known volume of sample containing 50-500μg of frusemide was transferred into a 25ml calibrated flask followed by 5ml of promethazine solution, 0.5 ml of 3.5 % acetic acid solution and 3ml of hypochlorite solution. The mixture was diluted to volume with distilled water, and allows standing for 5 min. The absorbance was measured at 625 nm against a reagent blank.

Procedures for pharmaceutical preparations:

Tablets: weigh and finally powder 10 tablets .Shake a quantity of powder containing 0.1g of frusemide with 150 ml of 0.1M NaOH for 10 min. Filter and dilute to 1L with distilled water. Treat 3ml of this solution as mentioned under recommended procedure.

Injection: To a volume containing the equivalent of 10 mg of frusemide add sufficient distilled water to produce 100 ml. Treat 3ml of this solution, as described under recommended procedure.

Results and Discussion:

The absorption spectra of the reagent and its frusemide complex are shown in Fig.1. The maximum absorbance of the blue product is at 625 nm.

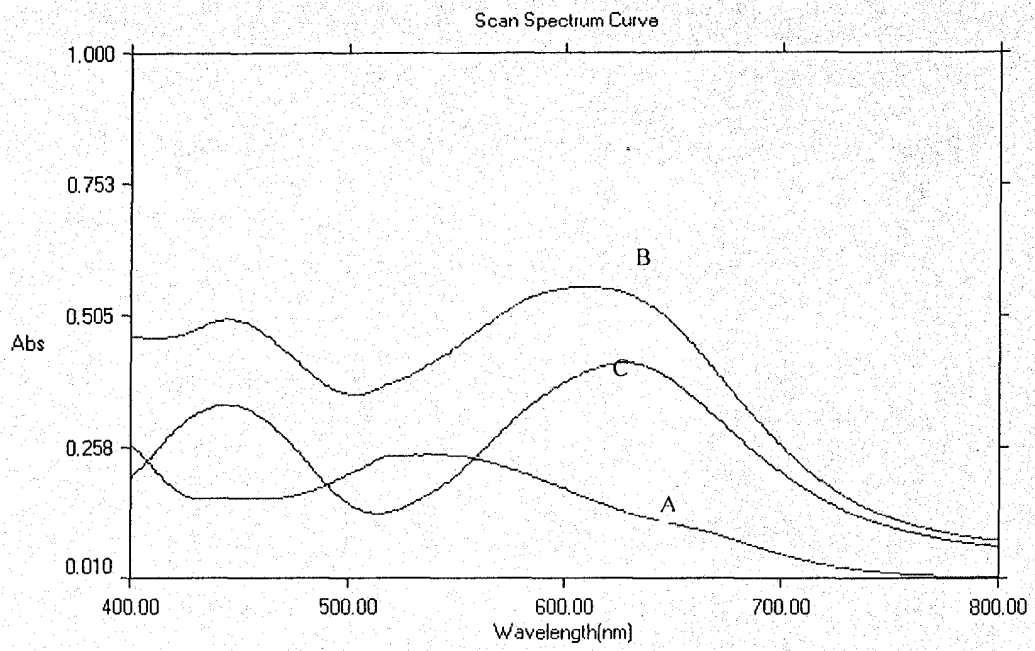


Fig. (1): Absorption spectra of 300µg/25 ml frusemide treated as discribeed under the recommended procedure conditions.
A-blank against water.
B-promethazine-frusemide complex against water.
C-promethazine-frusemide complex against blank.

Effect of pH:

The effect of pH 2-14 on the sensitivity of the product was examined .Only pH 2.7 (3.5 % acetic acid) was found to be optimum.The effect of the amount of (3.5 % acetic acid) used was also investigated and 0.5 ml was found the optimal.

Effect of reagent concentration:

The amount of promethazine solution ($6x10^{-4}$ M) for maximal color intensity was examined. The maximum constant intensity was reached at 3ml and remained constant up to 7ml. However 5ml of the reagent solution was selected for the subsequent work.

Effect of oxidant concentration:

The amount of hypochlorite solution (0.1%) for maximal color intensity was examined. The maximum constant intensity was reached at 2ml and remained constant up to 5ml .However; 3ml of the hypochlorite solution was selected for the subsequent work.

Effect of reaction time:

The maximum time for complete color development of the complex was found to be 5min, at room temperature and the absorbance was stable for at least 24 h.

Order of the addition of reagents:

To test the effect of order of the addition of the reagents on absorbance, different orders were tested. The selected order was sample solution, promethazine, acetic acid followed by the oxidant solution which was gave high absorbance value.

Beers Law:

Under the recommended procedure described above a linear calibration graph for frusemide with concentration range of 50-500 µg /25 ml and the molar absorptivity 0.625x 10^{4} $l.mol^{-1}.cm^{-1}$, with accuracy(average recovery. %) was 100.15 and the relative standard deviation of less than 2%was obtained .

The optimum reaction conditions are given in table (1).

Table (1): Optimum reaction conditions.

λmax	Temp.	PH	Reagent conc.	Oxid.conc	Time	Beer'Law	Accuracy (Average recovery %)	Precision RSD(%)
625 nm	R.T	2.7	$6x10^{-4}$M 5ml	0.1 % (3ml)	5 min	50-500 µg/25 ml	100.15	< 2

Stoichiometry of reaction:

The stoichiometry of the reactants was investigated by Job's method of continuos variations [13] .Standard solution of frusemide ($3x10^{-4}$ M) and promethazine ($3x10^{-4}$ M) were prepared. A series of standard solutions of frusemide and promethazine in different proportions totaling 10 ml (from 0+10 to 10+0 inclusive) were prepared in 25 ml calibrated flasks and diluted to the mark with distilled water. After 5 mints, the absorbance was measured at 625 nm.

The results indicated the existence of 1:1 frusemide: promethazine ratio at 625 nm. Fig (2).

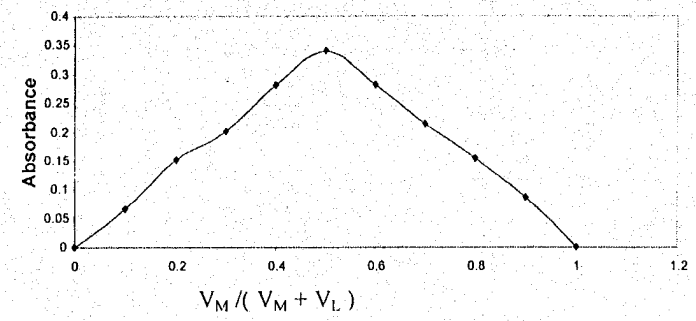
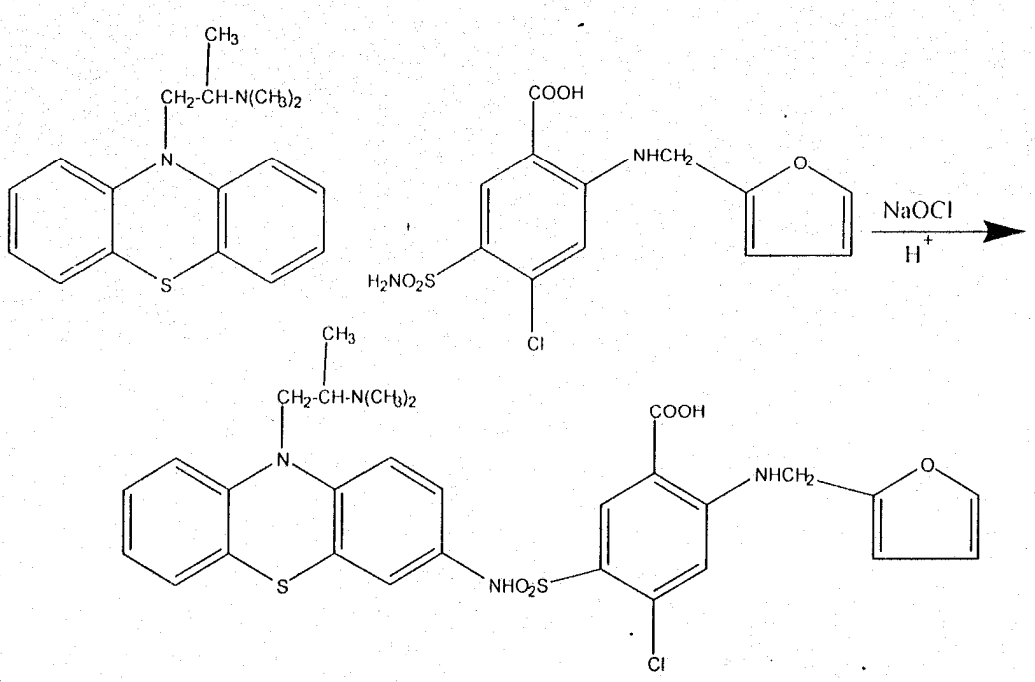


Fig. (2): Continuos variation plot for frusemide (v_m) with promethazine (v_L).

Therefore the formation of the product probably occurs as follows:



Apparent stability constant of the product:

The conditional stability constant of the product can be estimated by using the following equation [14].

$$K_n = \frac{a - (\Delta A / \epsilon)}{n^{n+1} (\Delta A / \epsilon)}$$

Where: a = Frusemide total concentration.

Δ A = sample absorbance in reagent excess minus the sample Absorbance at Stoichiometric reagent amount

ϵ = molar absorptivity at the measured wavelength.
And n = number of ligands.

The stability constant (mean of five values) is found to be 6.55×10^{8} l. mol^{-1} indicating that the product is very stable.

Applications:

The recommended procedure was applied for the determination of frusemide in tablets and injection preparations .The results are given in table (2) from these results it may be seen that the amount of frusemide found in pharmaceutical preparations agrees closely with the amount of frusemide on the label and the amount of frusemide analyzed through the official method [7].

Table (2): Assay of frusemide in pharmaceutical preparations.

Pharmaceutical preparations supplied by NDI	Amount of frusemid *		
	Present method	B.P method	Labeled value
Lasimex tablets	40.06 mg / tab	40.09 mg / tab	40 mg / tab
Lasimex injection	2.01 mg / amp	2.0 mg / amp	2.0 mg / amp

*Mean of five determinations.

Validity of the method:

The same batch of lasimex ampoules were analyzed simultaneously using the British pharmacopoeia method and the proposed method. Ten replicate analyses were prefomed by each method. The t-exp is 1.98 , this value less than 2.262 (t - test form table) at 95 % confidence level for nine degrees of freedom indicated that there is no

systematic error and there is no significant difference between the two methods .Therefor , the present method seems to have good validity .

Conclusion

A simple, rapid, precise and sensitive spectrophotometric method has been developed for the determination of trace amounts of frusemide in aqueous solution based on its oxidative coupling reaction with promethazine.HCl and sodium hypochlorite in the presence of acetic acid. The proposed method does not require temperature control or the solvent extraction step; the ingredients with frusemide often formulated have been shown not to interfere. The method was applied successfully on pharmaceutical samples.

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