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Indirect Visible Spectrophotometric Method For the Determination of Framycetin with Periodate, p-N,N-Dimethylphenylenediamine and Sulphanilamide

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ABSTRACT
Framycetinis beingoxidized with sodium metaperiodate. The iodate so formed is determined at pH 3.0 with p-N,N-dimethylphenylenediamine (DMPD) and sulphanilamide masking the excess periodate with sodium molybdate. The absorbance of the p-N,N-dimehylbenzoquinonemonoimine – sulphanilamide charge-transfer complex is measured at 520nm. This proposed method is simple, rapid and sensitive with reasonable precision and accuracy. The precision of the method was found by analyzing a set of eight solutions, each containing a final concentration value approximately in the middle of the Beer's law range. The percent relative standard deviation in this method is presented in table-5.5. The accuracy of the method was determined by taking different known amounts (with in Beer's law limits) of the drug and analyzing them by proposed method. The results are given in table-5.4. In the determination of Framycetin the excipients usually present in formulations (glucose, starch, sodium hexa phosphate) and the other antibiotics did not interfere.
Keywords: pectrophotometer, Framycetin, DMPD, sodium metaperiodate, sulphanilamide, sodium molybdate, Buffer pH 3.0.

INTRODUCTION

Aqueous solution of Sodium metaperiodate at pH 4.0 or below is the most suitable one as the oxidant [1].Periodic acid oxidation is applicable to compounds having two hydroxyl groups or a hydroxyl and an amino group attached to adjacent carbon atoms and is characterized by the cleavage of the carbon-carbon bond, as illustrated by the following equations [2-11]-

$$RCH(OH)CH(OH)R^1 + HIO_4 \rightarrow RCHO + R^1CHO + H_2O + HIO_3$$
$$RCH(OH)CH(NH_2))R^1 + HIO_4 \rightarrow RCHO + R^1CHO + NH_3 + HIO_3$$

The methods of analysis which are used to determine polyhydroxy compounds by oxidation with periodate can be divided in to eight main groups.

i. Determination of the amount of periodate consumed
ii. Determination of iodate formed
iii. Determination of the formic acid produced by cleavage of α, β, γ –triols and related structures. Other acids are also occasionally determined.
iv. Determination of formaldehyde produced from the terminal primary alcohol groups of compounds containing the 1, 2 – diol structure.
v. Determination of acetaldehyde
vi. Determination of ammonia and amines produced by periodate oxidation of α-aminoalcohols.
vii. Determination of carbon-dioxide
viii. Methods based on combinations of two or more of the above groups

The author describes the Indirect visible spectrophotometric determination of sodium metaperiodate consumed during oxidation of the Framycetin. The iodate, so formed is determined at pH 2.8 ± 0.2 with DMPD and sulphanilamide masking the excess periodate with sodium molybdate. The absorbance of the purple redcoloured p-N-N-dimethylbenzoquinonemonoimine – sulphanilamide charge-transfer complex is measured at 520nm.

Chloramphenicol (after reduction), Dihdrostreptomycin, and Framycetin were estimated by indirect visible spectrophotometer by using periodateand metol[12]. DMPD (p-N-N-Dimethylaminophenol as sulphate) and potassium iodatehas been found to be valuable reagent for the determination of primary aromatic amines[13]**.**

Table-1: Optimum Conditions of the Method

	Optimum Range	Conditions in Procedure
Sodium metaperiodate	0.8 – 1.1 ml	1.0 ml
Temperature for oxidation	25 – 35 °C	28±2 °C
Time for oxidation	1 – 5 min	2 min
pH Range	6.5 – 7.5	7.0
Sodium molybdate	1.5 – 2.5 ml	1.5 ml
DMPD	1.25 – 1.75ml	1.5 ml
Sulphanilamide	1.25 – 1.75 ml	1.5 ml
Temperature and time necessary for maximum colour development		70°C for 2min

Table-2: Determination of Iodate with DMPD and Sulphanilamide

Iodate Taken (µg)	Iodate obtained (µg)	Iodate + Masked periodate (µg)
200	201.0	201.6
300	301.8	302.2
400	398.8	399.7
500	502.0	502.9
2,000 (µg)		

MATERIALS AND METHODS

Preparation of reagents

DMPD Solution (0.05%): It was freshly prepared by dissolving 50 mg of the Analytical grade substance in 100 ml of water.

Sodium meta periodate Solution (0.2%): It was prepared by dissolving 200 mg of the analytical grade reagent in 100ml water.

Sodium Iodate solution ($9.3X10^{-3}$M): It was prepared by dissolving 185 mg of sodium Iodate in 100 ml of water.

Sodium molybdate solution($4.9X10^{-2}$M): It was prepared by dissolving 1.0 gm of sodium molybdate in 100 ml of water.

Sulphanilamide solution($1.2X10^{-2}$M): It was prepared by dissolving 200 mg of recrystallised sulphanilamide in 100 ml of water.

Buffer solution (pH3.0): It was prepared by mixing 50 ml potassium acid phthalate (0.2M) and 40.8 ml of hydrochloric acid (0.1M) and 109.2ml of water.

Framycetin: USP grade framycetin was prepared by dissolving 100mg of the substance in 100ml of water. Working solutions were prepared by appropriate dilution of stock solution to 200µg/ml.

All the other chemical reagents were of analytical grade.

Instrumentation
Spectral and absorbance measurements were made on Shimadzu double beam spectrophotometer UV – 140 with 1 cm quartz cells pH measurements were carried out using Systronics pH meter 335.

Absorbance curve
The absorbance curve of iodate in the presence of appropriate reagents was scanned on a spectrophotometer in the range 400-600 nm against the reagent blank. However, the maximum characteristic absorption is obtained at 520nm. The results were graphically represented in Fig.-1. Whereas the periodate in the presence of molybdate, DMPD and sulphanilamide have practically no absorption in this region. Prepared the calibration graph similarly (Fig.-2).

Optical characteristics- adherence to beer's law:
In order to know the Beer's law limits of the proposed method, the absorbances of a series of solutions containing varying amounts of Framycetin and specified concentrations of the remaining as given in the procedure in a total volume of 25ml were measured at 520nm against a reagent blank. The linearity of plot between absorbance and the concentration of Framycetin shows that the system obeys Beer's Law (Fig.-3).
The Beer's law limits, regression equation, correlation coefficient, molar absorptivity, sandell'ssensitivity, optimum photometric range were calculated and recorded in Table-3.

Table-3

Concentration range (µg/ml) (C)	4-24
Regression equation	A=0.001+0.0125C
Correlation co-efficient	0.9999
Molar absorptivity (1 $mole^{-1}$ cm^{-1})	$1.1x10^4$
Sandell's sensitivity (ug/cm^2/0.001 absorbance unit)	0.0800
Optimum photometric range (µg/ml)	11.5 – 22.5

Table-4: Accuracy of the Method

Antibiotic	Amount of antibiotic (µg)		
	Taken	Found	%error
Framycetin sulphate	500	497.6	0.48

Procedure
To a 25ml standard flasks containing 0.2 – 2.0 ml of Framycetin solutions 1.0ml of sodium metaperiodate solution was added and maintained the pH in the range 7.0 – 7.5. Then 1.5ml of sodium molybdate solution was added. After 10 min 15ml of pH 3.0 buffer and 1.5ml of DMPD solution were added. After 2 min 1.5ml of sulphanilamide solution was added and heated for 2 min at 70°C. Cooled, madeup to the mark with distilled water, and measured the absorbance at 520 nm against a reagent blank. Prepared the calibration graph similarly (Fig.-2).

For dosage forms
A known quantity of sample equivalent to 100mg of the Framycetin was taken, dissolved it in 100ml of water, filtered and diluted accurately to 250ml. Diluted the solution as necessary and determined the Framycetin content by recommended procedure.

Accuracy of the method
The accuracy of the method was determined by taking aliquots containing known quantities of Framycetinand estimated them by proposed method and the results were tabulated in Table-4.

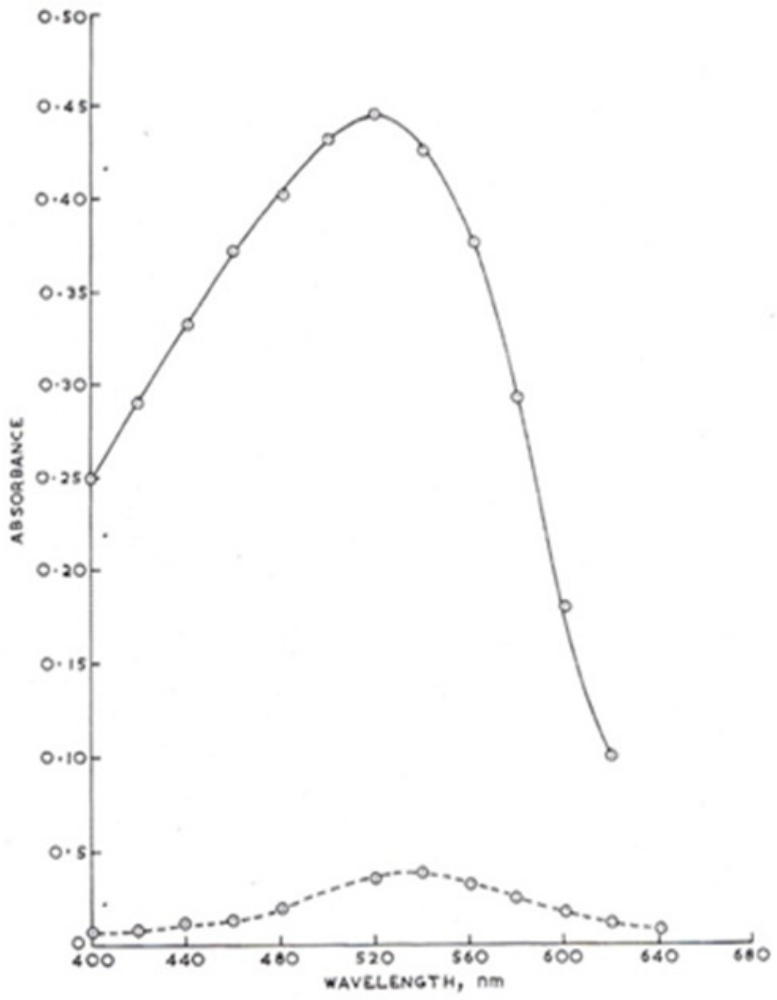


Fig.-1: Absorption Spectra of Iodate –DMPD-Suphalilamide System

Table-5: Precision of the Method

Antibiotic	%RSD	Percent of error confidence limit	
		0.05 level	0.01 level
Framycetin sulphate	1.52	± 1.86	± 2.26

RESULTS AND DISCUSSION

Interference studies

In the determination of Framycetinthe excipients usually present in formulations (glucose, starch, sodium hexa phosphate) and the other antibiotics did not interfere.

Analysis of pharmaceutical preparations and recovery experiments

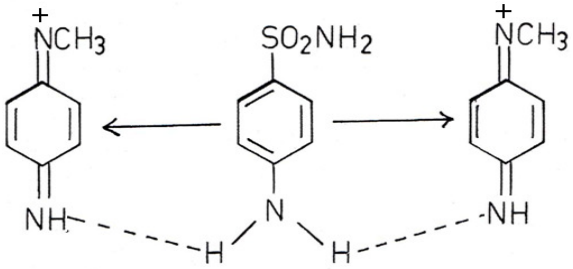
In order to find out the suitability of the proposed method, the pharmaceutical preparations were analyzed by the proposed and reported methods. Further, recovery experiments were conducted by adding a known amount of the drug to previously analyzed formulation and the total content of the drug was determined by the proposed method. The results were tabulated in Table-6.

Aqueous solution of Sodium metaperiodate at pH 4.0 or below is the most suitable one as the oxidant. Periodic acid oxidation is applicable to compounds having two hydroxyl groups or a hydroxyl and an amino group attached to adjacent carbon atoms and is characterized by the cleavage of the carbon-carbon bond.
Comparison of the results incorporated in Table-3 to 6 reveal that the proposed method is rapid, sensitive and simple with reasonable precision and accuracy. Sensitivity of the method is better than many of the methods.

Table-6: Assay of Formulation and % Recovery Data

SAMPLE	Labelled Amount (mg)	Amount found (mg) in method		%recovery (Proposed method)
		Proposed	Reported	
Framycetin sulphate powder	100	99.3	98.2	99.3

Chemistry involved
Sodium metaperiodate can oxidize the Framycetin. The iodate so formed can further oxidize DMPD into p-N-N-dimethylbenzoquinonemonoimine. Which can form a purple red p-N-N-dimethylbenzoquinonemonoimine – sulphanilamide charge-transfer complex with sulphanilamide at pH 3.0. Which can be measured at 520nm.



Conclusion
The proposed method is simple, rapid and sensitive with reasonable precision and accuracy and it is useful for the determination of Framycetin in bulk samples and pharmaceutical preparations. Though HPLC method is more accurate and rapid, it is expensive.

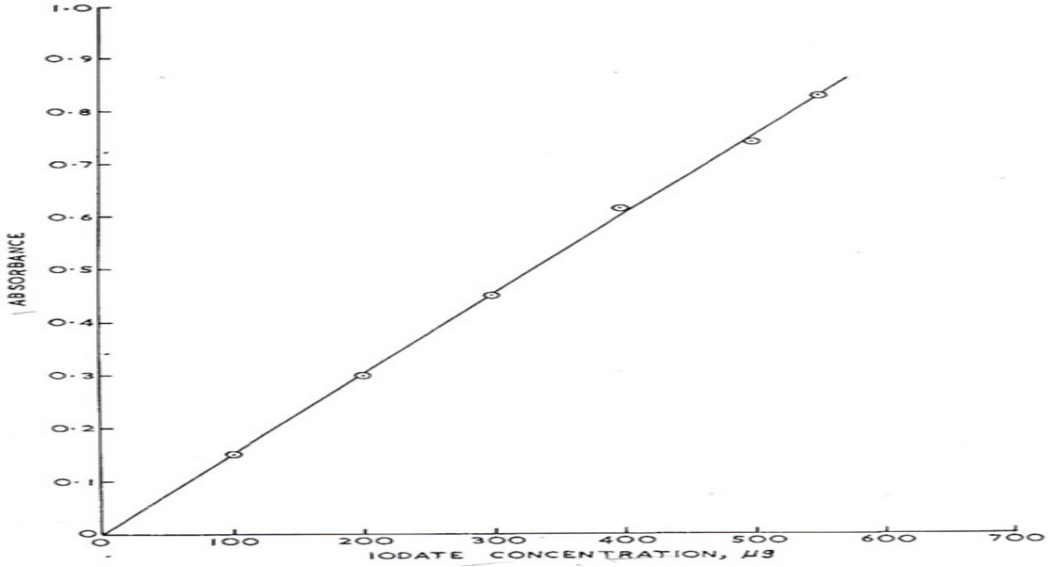


Fig.-2: Adherence to Beer's law of Iodate –DMPD-Suphalilamide System

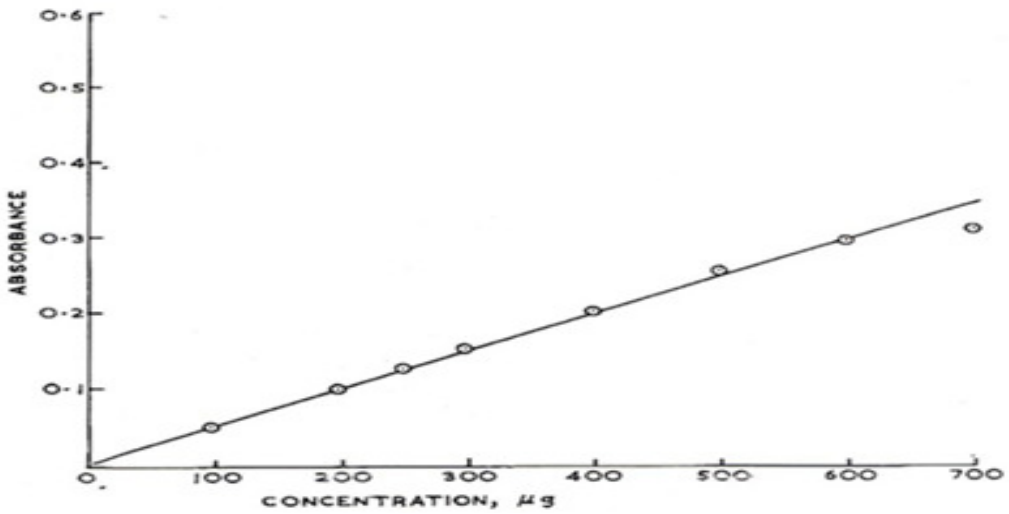


Fig.-3: Beer's Law plot of Framycetin

REFERENCES

1. Jones J.H. and Heckman N., J.Am. Chem. Soc. **969**(1947) 536.
2. Dryhurst G. , Periodate oxidation of diol and other functional groups, Ed.R.Belcher and DMW.Anderson, Pergamon press, London.(1970)
3. Skalarz , Quarterly Reviews, **21** (1967) 3.
4. Jackson E.L., Organic Reactions, **2**(1944) 341.
5. Bobbit J.M., Adv.Carbohydrate Chemistry, **11** (1956) 1.
6. Hough L., Methods in Carbohydrate Chemistry, **77** (1965) 370.
7. Barker S.A.and Somers P.J., Carbohydrates Research, **8** (1966) 220.
8. Dyer J.R., Methods of biochemical analysis, Ed by D.Glick, Interscience publishers, New York **3** (1956) 131.
9. Hay G.H., Lewis B.A., Smith F.and Unrau A.M., Methods in carbohydrate Chemistry **5**(1965) 251.
10. Belcher R.,Submicro methods of organic analysis, pergamon press, London(1967).
11. Speck J.C., Methods in Carbohydrate Chemistry, **1**(1962) 441.
12. Sastry C.S.P., Divakar T.E., Talanta **33** (1986) 164.
13. Ramakrishna R., Siraj P. and Sastry C.S.P., Acta Ciencia Indica, **6**(1980) 140

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