Spectrophotometric determination of flucloxacillin sodium using Folin-Ciocalteu reagent

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ABSTRACT

A simple accurate reproducible spectrophotometric method was established for the estimation of Flucloxacillin sodium based on oxidative coupling reaction. The method is based upon formation of light green colored product formed, when drug reacts with Folin-ciocalteu reagent (F.C). The coloured species has absorption maximum at 912 nm and obeys Beer's law in the concentration range of 0.05-0.25mg/ml. The optimum experimental parameters have been studied. Statistical analysis of the results has been carried out revealing high accuracy and good precision. The proposed method was successfully applied for the determination of Flucloxacillin in pharmaceutical formulations.

Keywords: Flucloxacillin sodium, Spectrophotometry, FC Reagent.

INTRODUCTION

Flucloxacillin chemically is (6R)-6-[3-(2chloro-6-fluorophenyl)-5-methylisoxazole-4carboxamido] penicillanic acid¹⁻². Flucloxacillin is an isoxazolyl pencillin used primarily for the treatment of infections due to Staphylococci resistant to benzyl pencillin. These include bone & joint infections, endocarditis, pneumonia, skin infections and toxic shock syndrome. Like other β -lactam antibiotics, flucloxacillin acts by inhibiting the synthesis of bacterial cell walls. It inhibits cross-linkage between the linear peptidoglycon polymer chains that make up a major component of the cell wall of Grampositive bacteria. It is a β -lactamase resistant pencillin used as first choice antibiotic for grampositive cocci. The therapeutic importance of this drug has prompted the development of many methods for its assay. This drug is official in BP and European Pharmacopoeia³⁻⁴. In Literature, Spectrophotometric⁵⁻¹¹, HPLC¹²⁻¹⁴, Polarography¹⁵,

Micro analysis¹⁶, LC¹⁷⁻¹⁸ methods have been developed for the determination of flucloxacillin. The aim of the present work was to develop a simple, validated, and rapid visible spetrophotometric method for routine analysis of Flucloxacillin sodium in pure drug and in pharmaceutical dosage form. The chemical structure of Flucloxacillin was reported in Fig. 1.

EXPERIMENTAL

Materials and Methods

All spectral and absorbance measurements were made on an ELICO-SL164 UV/ VIS double beam spectrophotometer with 10mm matched quartz cells.

All chemicals and reagents were of analytical grade. A 1mg/ml stock solution was prepared by dissolving 100 mg of Flucloxacillin sodium in 100 ml of methanol, 1N FC was prepared by diluting 50 ml of FC reagent to 100 ml with distilled water. 0.1N NaOH was prepared by dissolving 420 mg of NaOH in 100 ml of distilled water

Aliquot samples of Flucloxacillin sodium solution ranging from 0.5 to 2.5 ml ($1ml = 1000\mu g$) were transferred into the series of 10 ml volumetric flask. 2 ml of NaOH (0.1N) was added and mixed well. Then 0.5 ml of F.C reagent (1N) was added to each flask, mixed well and appropriate volume of distilled water was added to each flask to bring the volume to 10ml. The absorbance was measured at 912 nm against reagent blank. The amount of Flucloxacillin sodium present was computed and its calibration graph was reported in Fig. 2.

RESULTS AND DISCUSSION

Validation of the method

The optimum condition of the color was established by varying the concentration of FC, NaOH and observing the effect produced on the absorbance of the colored species

The optical characteristics such as Beer's law limits, molar absorptivity and sandell's sensitivity were given in Table 1. The precision of the method was found by measuring the absorbance of 6 separate samples containing known amounts of drugs and the results obtained are incorporated in Table 1. Regression analysis using the method of



Fig. 1: Chemical structure of Flucloxacillin



Fig. 2: Beer's laws plot of Flucloxacillin sodium with FC reagent

Table 1: Optical and regression characteristics of the proposed method for Flucloxacillin sodium

Parameters			
λmax,nm	912		
Beer's law Limit(mg/ml)	0.05-0.25		
Molar absorptivity,L/mol.cm	4.779		
Sandell's sensitivity (µg/cm²/0.001Absorbance unit)	15x10 ⁻²		
Regression equation Y=a+bc			
Slope(b)	0.457311		
Intercept(a)	0.001008		
Correlation coefficient	0.999841		

Table 2: Analysis of tablet formulation

Label claim	Amount found	%R.S.D	%Recovery[n=5]
500mg	499.98	0.228	99.96
500mg	498.99	0.230	99.79

least squares was made to evaluate the slope (b), intercept (a) and correlation coefficient (r).

The accuracy of the method was ascertained by comparing the results by proposed results of studied methods and those of reference. The similarity of the results is obvious evidence that during the application of these methods the excipients present in the formulation do not interfere in the assay of proposed method. As an additional check of accuracy of the proposed method recovery studies were carried out. The recovery of the added amounts of standard drug was studied at 3 different levels. Each level was repeated 6 times. From the amount of drug found, the percentage recovery was calculated.

CONCLUSION

The proposed visible spectrophotometric method is simple and sensitive with reasonable precision, accuracy and constitute better alternative to the existing ones for the routine determination of Flucloxacillin sodium in bulk and pharmaceutical formulation

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