

## VALIDATED SPECTROPHOTOMETRIC METHODS FOR ESTIMATION OF LOPERAMIDE HYDROCHLORIDE FROM TABLET DOSAGE FORM

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**Abstract:** Two simple, economical, precise and reproducible visible spectrophotometric methods have been developed for the estimation of loperamide hydrochloride in tablet formulation. The developed methods are based on the formation of chloroform extractable complex of loperamide hydrochloride with bromo phenol blue and thymol blue in double distilled water. The complex with bromo phenol blue (method I) shows absorbance maxima at 421.8 nm and linearity in the concentration range of 5-40 µg/ml. The extracted complex with thymol blue (Method II) shows absorbance maxima at 437.8 nm and the linearity in the concentration range of 10-100 µg/ml. Results of analysis for both the methods were validated statistically and by recovery studies.

**Keywords:** Loperamide hydrochloride, visible spectrophotometry, quantitative estimation, bromo phenol blue, thymol blue.

### INTRODUCTION

Loperamide hydrochloride, chemically 4-[4-(4-chlorophenyl)-4-hydroxypiperidin-1-yl]-N, N-dimethyl-2, 2-di(phenyl)butanamide, is an opioid-receptor agonist and acts on the  $\mu$ -opioid receptors in the myenteric plexus large intestines; by itself it does not affect the central nervous system like other opioids. It works by decreasing the activity of the myenteric plexus, which, like morphine, decreases the tone of the longitudinal smooth muscles but increases tone of circular smooth muscles (anal sphincter) of the intestinal wall. This increases the amount of time substances stay in the intestine, allowing for more water to be absorbed out of the fecal matter [1]. Loperamide also decreases colonic mass movements and suppresses the gastrocolic reflex. It is official in BP [2], USP24NF19 [3] and Merck Index [4]. This describes potentiometric [2], high performance liquid chromatographic [4] method for its quantitation. Literature survey reveals one RP-HPLC methods [5]. The objective of the present investigation was to develop simple, accurate and economical spectrophotometric methods for quantitation of Loperamide Hydrochloride in tablet formulation. Shimadzu UV 1700, UV/Vis double beam spectrophotometer with spectral band width of 1 nm, wavelength accuracy of  $\pm 0.3$  nm and 1.0 cm matched quartz cells was used for analytical method development.

### MATERIALS AND METHODS

All the chemicals and reagent used were of analytical grade. Bromo phenol blue (Merck) reagent and thymol blue (Loba Chemie) reagent were prepared in double distilled water. Both the reagents were extracted several times with chloroform so as to remove chloroform soluble impurities. Tablet formulations of loperamide hydrochloride [Lomin Tab. (Intra Labs), Starlop Tab. (Cadila)] were procured from local pharmacy. Standard solution of loperamide hydrochloride was prepared by dissolving 10 mg in 100 ml mixture of double distilled water + Methanol (Ratio-20:1) to give stock solution of concentration 100 µg/ml of drug.

#### Procedure for preparation of calibration curve

For method I, in a series of 10 ml volumetric flask, aliquots of standard drug solution (100 µg/ml) in double distilled water + Methanol (Ratio-20:1) were transferred and diluted with same so as to give several dilutions in concentration range of 5-40 µg/ml of loperamide hydrochloride. To 5 ml of each dilution taken in a separating funnel, 5 ml of bromo phenol blue (0.2 % w/v) reagent and 5 ml of chloroform was added. Reaction mixture was shaken gently for 5 min and allowed to stand so as to separate two layers. The chloroform layer was separated out and absorbance maxima measured at 421.8 nm (Figure 1) against a reagent blank. Calibration curve was plotted between concentration of loperamide hydrochloride and measured absorbance (Figure 2). Spectral characteristics of loperamide hydrochloride are given in Table 1.

For method II, in a series of 10 ml volumetric flask, aliquots of standard drug solution (100 µg/ml) in double distilled water + Methanol (Ratio-20:1) were transferred and diluted with same so as to give several dilutions in concentration range of 10-100 µg/ml of

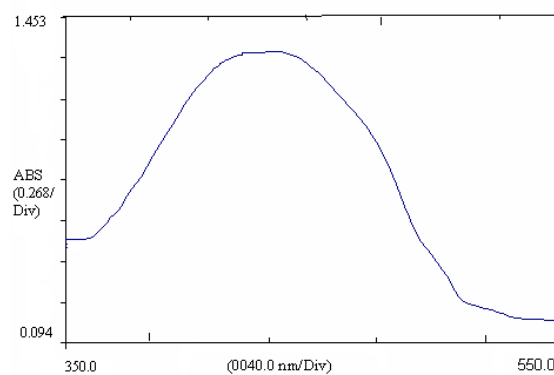


Figure 1. Spectra of loperamide hydrochloride using method I

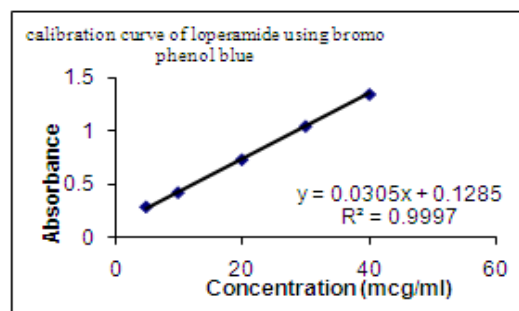


Figure 2. Calibration curve of loperamide hydrochloride using method I

loperamide hydrochloride. To 5 ml of each dilution taken in a separating funnel, 5 ml of thymol blue reagent (0.25 % w/v) and 5 ml of chloroform was added. Reaction mixture was shaken gently for 5 min and allowed to stand so as to separate two layers. The chloroform layer was separated out and absorbance maxima measured at 437.8 nm (Figure 3) against a reagent blank. Calibration curve was plotted between concentration of loperamide hydrochloride and measured absorbance (Figure 4). Spectral characteristics of Loperamide Hydrochloride are given in Table 1.

#### Procedure for analysis of tablet formulation

For analysis of tablet formulation 20 tablets (2 mg) of loperamide hydrochloride were weighed accurately and finely powdered. An accurately weighed powdered sample equivalent to 10 mg of loperamide hydrochloride was taken in a 100 ml volumetric flask containing 40 ml of double distilled water + Methanol (Ratio-20:1), sonicated for 10 min. The resultant solution was filtered through Whatman filter paper no. 41 into another 100 ml volumetric flask. The filter paper was washed several times with double distilled water

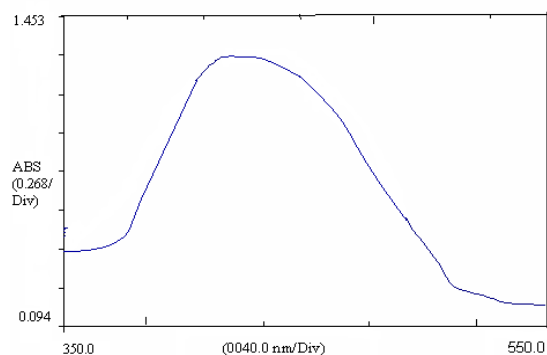


Figure 3. Spectra of loperamide hydrochloride using method II

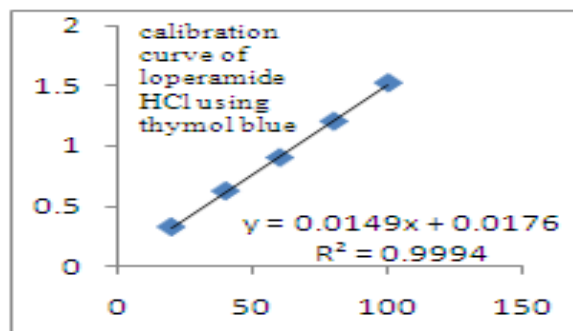


Figure 4. Calibration curve of loperamide hydrochloride using method II

Table 1. Spectral characteristics of loperamide hydrochloride

Parameters	Method I	Method II
$\lambda$ max	421.8 nm	437.8 nm
Beer's law limit ( $\mu\text{g/ml}$ )	5-40 $\mu\text{g/ml}$	20-100 $\mu\text{g/ml}$
Regression equation*	$y = 0.1285 + 0.0305x$	$y = 0.0176 + 0.0149x$
Slope (b)	0.030	0.014
Intercept (a)	0.128	0.017
Correlation coefficient ( $r^2$ )	0.9997	0.9994

\*  $y = a + bx$ , where  $c$  is the concentration in  $\mu\text{g/ml}$  and  $y$  is the absorbance unit of five replicate samples.

+ Methanol (Ratio-20:1). The washings were added to the filtrate and final volume was made up to the mark with double distilled water + Methanol (Ratio-20:1).

For method I, 2 ml of filtrate of the sample solution was diluted to 10 ml with double distilled water + Methanol (Ratio-20:1). These were treated

as per the procedure for preparation of calibration curve and amount of the drug present in sample was computed from respective calibration curve.

For method II, 5 ml of filtrate of the sample solution was diluted to 10 ml with double distilled water + Methanol (Ratio-20:1). These were treated as per the procedure for preparation of calibration curve and amount of the drug present in sample was computed from respective calibration curve.

The procedure of analysis from tablet formulations for both the methods was repeated five times with two different tablet formulations and results are reported in Table 2.

## RESULTS AND DISCUSSION

The proposed methods are simple, rapid, accurate, precise and reproducible. They do not suffer from any interference due to common excipients of tablet. Beer's law is obeyed in the concentration range of 5-40  $\mu\text{g/ml}$  and 20-100  $\mu\text{g/ml}$  for Method I and Method II respectively. Both the methods were validated in terms of accuracy, reproducibility, and the results are recorded in (Table - 2). The accuracy of the methods was proved by performing recovery studies in the commercially available formulations. Values near or greater than 99.0% indicate that the proposed methods are accurate for the analysis of drug. The optical characteristics and linearity are recorded in (Table 1). The regression analysis using the method of last sequence was made for the slope (b), intercept (a) and correlation coefficient (r) obtained from different concentrations. The results are summarized in (Table 1). Rigorous analysis of the results shows that the presence of excipients in tablet formulation did not interfere with the final determination of the active component

Loperamide hydrochloride solubility problem are resolved by using cost effective mixture of distilled water + Methanol (20:1).

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Table 2. Results of analysis and recovery studies of commercial formulations of loperamide hydrochloride

Method	Formulation	Label claim (mg/tab)	% Label claim*	% recovery**	SD
I	Lomin tab.	2	99.63	99.39	$\pm 0.459$
	Starlop tab.	2	99.32	98.96	$\pm 0.541$
II	Lomin tab.	2	99.66	98.98	$\pm 0.648$
	Starlop tab.	2	99.31	99.71	$\pm 0.446$