Development and Validation of Visible Spectrophotometric methods for the Estimation of Mesalamine in Pharmaceutical Dosage Forms

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ABSTRACT

Three simple and sensitive spectrophotometric methods (A, B and C) have been developed for the quantitative determination of Mesalamine in bulk drug and pharmaceutical preparations. Method A, B are based on the condensation of Mesalamine with p-dimethyl aminobenzaldehyde and p-dimethyl amino cinnamaldehyde to form schiff’s bases, to yield a yellow, red colored chromogen and exhibits absorption maxima at 440, 523.5 nm. Method C is based on the reaction of Mesalamine with Folin-Ciocalteu (phenol’s) reagent under alkaline conditions forming a blue colored chromogen and exhibits absorption maxima at 616 nm. Beer’s law was obeyed in the concentration range of 50-250, 20-100 and 10-50 µg/ml for method A, B and C respectively. These methods were extended to pharmaceutical formulations and there was no interference from any common excipients. The results of analysis have been validated statistically and by recovery methods.

Key words: Spectrophotometry, p-dimethyl amino benzaldehyde, p-dimethyl amino cinnamaldehyde, FC reagent, Schiffs base.

INTRODUCTION

Mesalamine is chemically known as 5 – amino – 2 – hydroxy benzoic acid[1-4], is an anti-inflammatory drug used to treat inflammation of the digestive tract (crohn’s disease) and mild to moderate ulcerative colitis. It is a bowel-specific amino salicylate drug that is metabolized in the gut and has its predominant actions there, thereby having fewer systemic side effects [5-6]. The literature survey reveals that few analytical methods for this drug are reported, which include chromatographic[7], and spectrophotometric methods[8-9]. The present investigation has been undertaken to develop three simple and accurate spectrophotometric methods using p-dimethyl amino benzaldehyde, p-dimethyl amino cinnamaldehyde and Folin-Ciocalteu reagent.
Which are essential for routine quality control analysis of pharmaceutical products containing Mesalamine as active constituent.

MATERIALS AND METHODS

Instrument
All spectral measurements were made on Shimadzu 1800 UV-Visible spectrophotometer with 1 cm matched quartz cell were used.

Materials
Pure drug of Mesalamine was obtained from Cosmopharmaceutical Pvt Ltd, Goa and commercial formulations were procured from local market. All the chemicals used were of analytical grade.

Reagents
Alcoholic solution of P-dimethyl amino benzaldehyde (0.5 %w/v)
Alcoholic solution of P-dimethyl amino cinamaldehyde (0.5 %w/v)
Folin-Ciocalteu reagent (1N) was diluted to 2N with distilled water
Aqueous solution of Sodium Hydroxide (1N)
Aqueous solution of Hydrochloric acid (0.1N)

Preparation of Standard solution:
Weigh accurately 100 mg of Mesalamine and transferred into 100 ml volumetric flask and dissolve in 100 ml of 0.1N HCl to obtain a concentration of 1 mg/ml. From this suitable dilutions were made to obtain the working standard concentration of 100 µg/ml.

Preparation of sample solution:
Two brands of commercially available tablets were taken, twenty tablets each weighing 400mg were weighed and powered. A tablet powder equivalent to 100 mg was weighed accurately and transferred into 100 ml volumetric flask containing 50 ml of 0.1N HCl, the flask was sonicated for 5 min, the volume was made up to mark with 0.1N HCl, and the solution was filtered through whatmann filter paper 41, from the above stock solution, working standard solution of 100mg/ml were prepared by further dilution with 0.1N HCl, the above procedure was applied for analysis.

Assay Procedure:
Method A
Aliquots of standard drug solution ranging from 0.5 to 2.5 ml (1ml=1000µg/ml) were transferred in to a series of 10 ml volumetric flasks. To each flask 1 ml of P-dimethyl amino benzaldehyde, 1ml of concentrated HCl were added and kept for 20 min heating at 60°C and the volume was made up to the mark with water. The absorbance of yellow colored chromogen was measured at 440 nm (fig 1) against a reagent blank. The amount of drug present in the sample was computed from its calibration curve (fig 2).

Method B
Aliquots of standard drug solution ranging from 0.2 to 1.0 ml (1ml=1000µg/ml) were transferred in to a series of 10 ml volumetric flasks. To each flask 1 ml of P-dimethyl amino cinamaldehyde,
1ml of concentrated HCl were added and kept for 20min heating at 40°C and the volume was made up to the mark with water. The absorbance of red colored chromogen was measured at 523.5 nm (fig 3) against a reagent blank. The amount of drug present in the sample was computed from its calibration curve (fig 4).

**Method C**

Aliquots of standard drug solution ranging from 1 to 5 ml (1ml=100µg/ml) were transferred in to aseries of 10 ml volumetric flasks. To each flask 1.0 ml of Folin-Ciocalteu reagent and1.0 ml of 1N sodium hydroxide were added, kept for 10 min to develop the color and the volume was made up to themark with distilled water. The absorbance of blue colored chromogen was measured at 616 nm(fig 5) against a reagent blank. The amount of drug present in the sample was computed from its calibrationcurve (fig 6).

**RESULTS AND DISCUSSION**

The optical characteristics such as Beer’s law limits, Molar absorptivity, and relative standard deviation were calculated and the results are summarized in Table 1. Regression characteristics like slope, intercept and correlation coefficient were calculated and are presented in Table 1. Commercial tablets of Mesalamine were successfully analyzed by the proposed methods and the results are presented in Table 2. Comparison of the results obtained with the proposed and UV methods for dosage forms (Table 2) confirms the suitability of these methods for Pharmaceutical dosage forms. To evaluate validity and reproducibility of the methods recovery experiments were conducted and the results are summarized in Table 2. The other active ingredients and excipients usually present in pharmaceutical dosage forms did not interfere.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Method A</th>
<th>Method B</th>
<th>Method C</th>
</tr>
</thead>
<tbody>
<tr>
<td>λmax (nm)</td>
<td>440</td>
<td>523.5</td>
<td>616</td>
</tr>
<tr>
<td>Beer’s law limits (µg/ml)</td>
<td>50-250</td>
<td>20-100</td>
<td>10-50</td>
</tr>
<tr>
<td>Molar absorptivity (L mol⁻¹ cm⁻¹)</td>
<td>5.1454 X 10²</td>
<td>1.3093 X 10³</td>
<td>2.4042 X 10³</td>
</tr>
<tr>
<td>Regression equation (Y = a+bc)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Slope (b)</td>
<td>0.0033</td>
<td>0.0084</td>
<td>0.0197</td>
</tr>
<tr>
<td>Intercept (a)</td>
<td>0.0218</td>
<td>0.0044</td>
<td>0.0001</td>
</tr>
<tr>
<td>% R S D</td>
<td>0.1992</td>
<td>0.1953</td>
<td>0.2025</td>
</tr>
<tr>
<td>Correlation coefficient (r)</td>
<td>0.9998</td>
<td>0.9999</td>
<td>0.9999</td>
</tr>
<tr>
<td>Limit of Quintitation (LOQ)</td>
<td>0.3350</td>
<td>0.0084</td>
<td>0.6067</td>
</tr>
<tr>
<td>Limit of Detection (LOD)</td>
<td>0.1105</td>
<td>0.1848</td>
<td>0.2002</td>
</tr>
<tr>
<td>Range of error**</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Confidence limit with 0.05 level</td>
<td>1.0573 X 10⁻³</td>
<td>1.0576 X 10⁻³</td>
<td>4.8960 X 10⁻⁴</td>
</tr>
<tr>
<td>Confidence limit with 0.01 level</td>
<td>1.5648 X 10⁻³</td>
<td>1.5648 X 10⁻³</td>
<td>7.2436 X 10⁻⁴</td>
</tr>
</tbody>
</table>

Y=bC+a where C is the concentration of Mesalamine in µg/ml and Y is absorbance unit

**for eight measurements**

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Table-2 Evaluation of Mesalamine in Tablet Dosage formulations

<table>
<thead>
<tr>
<th>Label Claim (mg)</th>
<th>Amount of drug obtained by proposed methods (mg)</th>
<th>Reference method UV</th>
<th>% Recovery*</th>
<th>% Recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A</td>
<td>B</td>
<td>C</td>
<td>UV A</td>
</tr>
<tr>
<td>M_{1} 400</td>
<td>398.14</td>
<td>398.64</td>
<td>399.92</td>
<td>398.64</td>
</tr>
<tr>
<td>M_{2} 400</td>
<td>399.92</td>
<td>399.58</td>
<td>398.74</td>
<td>398.74</td>
</tr>
</tbody>
</table>

*mean of six determinations, M_{1} = Mesacol (UnipharmaPvt Ltd), M_{2} = Walasa(Wallace PvtLtd)
CONCLUSION

The proposed visible spectrophotometric methods for the estimation of Mesalamine are simple, sensitive, accurate and can be used for the routine quality control of the drug in bulk as well as in pharmaceutical formulations.

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REFERENCES