DEVELOPMENT OF FIRST ORDER AND SECOND ORDER DERIVATIVE FLUOROMETRIC METHODS FOR CARVEDILOL IN BULK AND FORMULATIONS

CM Jamkhandi1*, PS Kumbhar1, JI Disouza1, As Sherikar1 and DA Bhagwat2
1Tatyasaheb Kore College of Pharmacy. Warananagar, Kolhapur, Maharashtra, India.
2Bharati Vidyapeeth College of Pharmacy, Kolhapur, Maharashtra, India.

ABSTRACT
Objective: A simple, accurate, precise, easily accessible fluorometric estimation method has been developed for Carvedilol in 0.1 M Methanolic sodium hydroxide. Method: Fluorescent properties of Carvedilol have been studied in 0.1 M Methanolic sodium hydroxide showed linearity in the range of 1 to 10µg/ml. First derivative and second derivative method of applied and found to be sensitive to small changes in concentration. Results: Zero order readings are used for statistical calculation of validation parameters. Thus obtained readings were used calculate First and second order graphs to study the sensitivity of the drug with change concentration of the drug. On validation of the method shows correlation coefficient of 0.996556, Standard deviation of 32.97, Intercept 8.5333, slope was 9.0121, and Relative standard deviation was 0.03512. Conclusion: The fluorometric properties of Carvedilol were studied in various solvents and fluorometric property in methanolic sodium hydroxide was used to develop fluorometric method for estimation. Analytical method was validated for parameters like accuracy, precision, specificity, ruggedness, robustness and percentage recovery and all parameters were found within the specified range. Keywords: Fluorometry, Carvedilol, Methanolic sodium hydroxide, Derivative spectroscopy.

INTRODUCTION
Carvedilol is β-blocker used to treat high blood pressure and cardiac failure. Chemically Carvedilol is 2-Propanol, 1-(9H-carbazol-4-yloxy)-3-[[2-(2-methoxy phenoxy) ethyl amino]-, (+)-; (+)-1-(Carbazol-4-yloxy)-3-[2-(o-methoxy phenoxy) ethyl] amino]-2-propanol. There are various methods of estimation Carvedilol have been described such as UV-Visible spectrophotometry [1-8], RP–HPLC[9-10] and HPLC methods[11-13]. Fluorescent properties of Carvedilol and fluorometric estimation methods[14-16]. Under the present study we have studied fluorometric properties of Carvedilol in Methanolic sodium hydroxide. Literature survey revealed that Fluorometric property of Carvedilol in Methanolic sodium hydroxide has not been reported and it was observed that the fluorescent property of Carvedilol increases in methanolic sodium hydroxide.

MATERIALS AND METHODS
All reagents and solvents used in the experimental work are analytical grade. Elico Fluorometer model CL-53 was used. The fluorescence intensity of test and reference solutions was recorded in 3 ml borosilicate cells. The Relative Intensity or % Transmission was measured with filters of excitation wavelength of 366 nm and emission wavelength of 475 nm.

Preparation of 0.1 M Methanolic NaOH: 4.2 gm NaOH dissolved in 5 mL distilled water. Sufficient aldehyde free methanol added to produce 1000 ml. The solution is allowed to stand in tightly stopper bottle.

Procedure for estimation: Stock solution of Carvedilol was prepared with 100 mg of Carvedilol dissolved in 100 ml of 0.1 M Methanolic sodium hydroxide solution and various concentrations in the range of 1µg/ml to 10µg/ml were prepared for recording. Highest concentration was used set 100% intensity of Fluorometer. The 0.1 M Methanolic sodium hydroxide solution was as blank. Sample readings were recorded using commercial brands of Carvedilol. Standard dilutions in the range of 1-10µg/ml prepared for linearity measurement.

Estimation of commercial tablets: 20 tablets were weighed and ground to fine powder. The tablet powder equivalent to 100 mg of
carvedilol was weighed and transferred to 100 ml volumetric flask; 50 ml of 0.1 M Methanolic sodium hydroxide was added, filtered and final volume was adjusted to 100 ml. Dilution containing highest concentration of 10μg/ml was used to set 100% intensity and readings were recorded against 0.1 M methanolic sodium hydroxide as blank.

RESULTS AND DISCUSSION
Fluorometric method is sensitive which can measure the sample in low concentration. Carvedilol is fluorescent substance and this property of fluorescence is increased in 0.1M Methanolic sodium hydroxide solution. The influence of other solvents on fluorescence property of carvedilol was also studied (Fig 2). Pure drug and marketed formulations were used to get the results. Linearity range of carvedilol in 0.1M Methanolic sodium hydroxide solution was 1µg/ml to 10µg/ml. The validation parameters such as precision, accuracy, coefficient correlation, standard deviation and relative standard deviation were also calculated. All results obtained were within the range of standard values (table 1). The results of zero order were used for first and second order calculations, graphs were plotted accordingly. First and second order graphical representation showed that the percentage of Transmission is optimum at different concentrations. In the graph of a zero-order derivative change in %Transmission and concentration produces a straight line. The expected shapes of the curves for change in %Transmission and concentration with concentration for a first-order derivative. A plot of squares of change in %Transmission and concentration with concentration then the reaction is second order (Fig 1, 3 and 4).

![Fig. 1: Linear relationship between % relative intensities and Concentration](image1)

![Fig. 2: % Transmission with carvedilol in different solvents](image2)
Table 1: Statistical calculations of validated parameters

<table>
<thead>
<tr>
<th>Validation Parameter</th>
<th>Calculated Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Range and Linearity</td>
<td>1 µg/ml to 10 µg/ml</td>
</tr>
<tr>
<td>Relative Standard Deviation</td>
<td>0.03512015</td>
</tr>
<tr>
<td>Coefficient Correlation</td>
<td>0.996556</td>
</tr>
<tr>
<td>Accuracy</td>
<td>91% to 95%</td>
</tr>
<tr>
<td>Slope</td>
<td>9.012121</td>
</tr>
<tr>
<td>Intercept</td>
<td>8.533333</td>
</tr>
</tbody>
</table>

Fig. 3: Graphical representation of First Order derivative

Fig. 4: Graphical representation of Second Order derivative

CONCLUSION
The developed method is easily accessible, precise, and accurate and showed linearity in range of 1µg/ml to 10µg/ml. Thus developed can be successfully applied for estimation of Carvedilol in pure and bulk formulations.

ACKNOWLEDGEMENTS
Authors are thankful to Hon’ble Dr. Vanayji V Kore (Savkar) President of Shree Warana Vibhag Shikshan Mandal Warananagar for continuous encouragement and support.

REFERENCES
2. A Mali, V Kekan, R Dongare, S Gholve, R Bathe. Simultaneous UV Spectrophotometric Methods for...


