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UV SPECTROPHOTOMETRIC METHOD DEVELOPMENT AND VALIDATION FOR ESTIMATION OF ETODOLAC IN BULK

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ABSTRACT

Sample, cost effective, accurate, precise and rapid UV Spectrophotometric method was developed for the estimation of etodolac in pure form. Absorbance maximum of etodolac was estimated at 279.5 nm in methanol and water (1:9V/V).The recovery studies ascertained the accuracy of the proposed method and the results were validated as per ICH guidelines. The drug exhibited the linearity in the concentration range of 10-60µg/ml with correlation coefficient of R^2 0.999.The % recovery of the drug for the proposed method was found to be 100.6% . The limit of detection (LOD) and limit of quantification (LOQ) were found to be 0.21µg/ml and 0.65µg/ml respectively. The apparent molar absorptivity and sandll's sensitivity were found to be 2.16 $mol^{-1}cm^{-1}$ and 0.143µg/cm^2 respectively.

INTRODUCTION:

Etodolac is chemically 2-{1,8-diethyl-1,h,3h,4h,9h-pyrano[3,4b]indol-1-yl}acetic acid. it is non steroidal anti inflammatory drug used for the management of mild to moderate pain, fever, and inflammation. it is licensed for the treatment of inflammation and pain caused by osteoarthritis and rheumatoid arthritis. etodolac blocks the cyclo-oxygenase enzyme (cox) which form prostanoids, it lower the concentration of prostaglandins. which results in inflammation , pain fever are reduced. etodolac is generally avoided during pregnancy and nursing methods. nasid's may cause adverse cardiovascular effects in foetus during pregnancy. . a survey of the literature reveals that there is no method available for the determination of etd in pure form and pharmaceutical formulations by oxidation-reduction reactions. a survey of the literature reveals that there are very few reported methods for the determination of etd in biological fluids, pharmaceutical formulations and in presence of its enantiomer. the aim of the present study was to develop a simple , accurate and validated uv spectrophotometric method for etodolac.

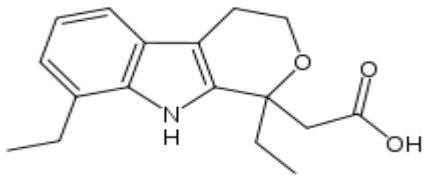

Fig 1: Structure of Etodolac

MATERIALS AND METHODS

Elico SL-159, UV-Visible spectrophotometer with matched cuvettes were used for the estimation. Ultrasonicator and electronic balance (Wensar ISO 9001-2000 certified) used for the experiment. Glass ware and filter paper (Whatmann No: 1) were used for the experiment.

Methodology:

Selection of solvent

The solubility of etodolac was determained in various solvent as pharmacopeia standard. Solubility test was carried out in different solvent like distilled water, methanol, chloroform, dimethyl sulfoxide and aqueous polyethylene glycol. From the solubility studies it was found that Etodolac was soluble in methanol and distilled water (1:9).

PREPARATION OF STANDARD SAMPLE:

The standard stock solution of etodolac was prepared by transferring accurately weighed 30mg of drug to 50ml volumetric flask and dissolving it with water and methanol (1:9) to get a concentration of 3000µg/ml. The solution was diluted accordingly to a concentration of 300µg/ml and was kept as the stock solution. The prepared stock solution was diluted with water and methanol to get working standard solutions of concentration 10-70µg/ml.

Determination of λ max:

The standard solution of etodolac (30µg/ml) was scanned in the wavelength region of 190 to 370 nm and the spectrum was recorded. Solvent methanol and water (1:9) was used as blank. It was observed that λ max was to be 279.5 nm by plotting a graph between absorbance vs. wavelength.

VALIDATION:

The objective of method validation is to demonstrate from the method is suitable for its intended purpose. The method was validated for linearity, precision, accuracy, LOD, LOQ, molar absorptivity and sandell's sensitivity as per ICH guidelines.

LINEARITY:

The sandared stock solution, the various dilutions in the conc. Of 10µg/ml, 20µg/ml, 30µg/ml, 40µg/ml, 50µg/ml &60µg/ml were prepared. The solution was scanned at 279.5nm and absorbance was recorded.

ACCURACY:

The accuracy of the proposed method was tested by recovery studies at 80%, 100% and 120% according to ICH guidelines by adding a known amount of pure drug to the pre-analysed formulation of concentration 15µg/ml. from above solution the mean was calculated according to the formula. From the mean, the standard deviation was calculated.

The Mean value = The sum of the absorbance/total absorbance

The Standard deviation Value =

$$s = \sqrt{\frac{1}{N-1}\sum_{i=1}^{N}(x_i - \overline{x})^2},$$

From standard deviation percent relative standard deviation was also calculated.

%RSD=SD/MEAN*100

The % recovery also calculated according to the below formula.

%Recovery=Amount formula/Amount found*100

PRECISION:

Precision was calculated by preparing six solutions of same concentration which is the middle concentration level among the linearity range mean, S.D was calculated for these 6 concentrations. The precision value was found to be 1.014%, which was found to be with in limits i.e < 2 .

Methanol and Water (1:9) was taken as blank and 6 readings were recorded at wavelength of 279.5nm then LOD, LOQ was calculated by following formula.

Limit of detection (LOD) = 3.3×SD\S
Limit of quantification (LOQ) = 10×SD\S
LOD of ETODOLAC = 0.212
LOQ of ETODOLAC = 0.657

SANDELL'S SENSIVITITY:

Determination of Sandell's Sensitivity Standard stock solution of etodolac: Sandell's Sensitivity was calculated from the linearity data for their respective absorbance values. From this observation table, the mean of observations was taken and Sandell's Sensitivity was calculated by the following formula:

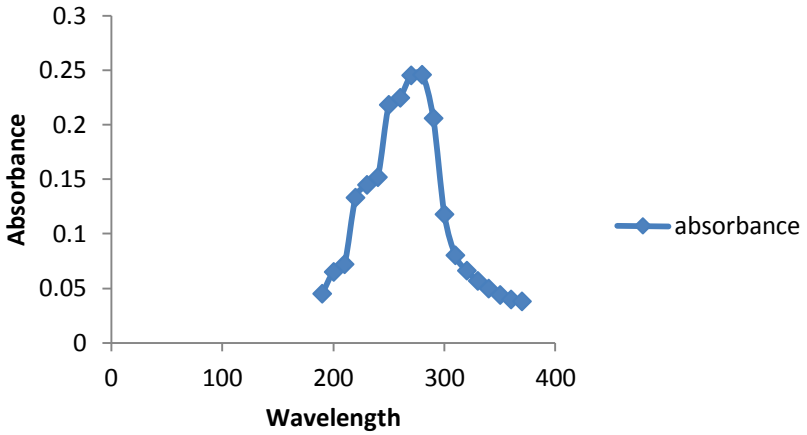

Fig 2: Absorption maximum of Etodolac by UV-Spectrophotometer

Table No: 1 Linearity data for Etodolac.

Concentration (µg/ml)	Absorbance(nm)
10	0.074
20	0.148
30	0.232
40	0.296
50	0.370
60	0.460

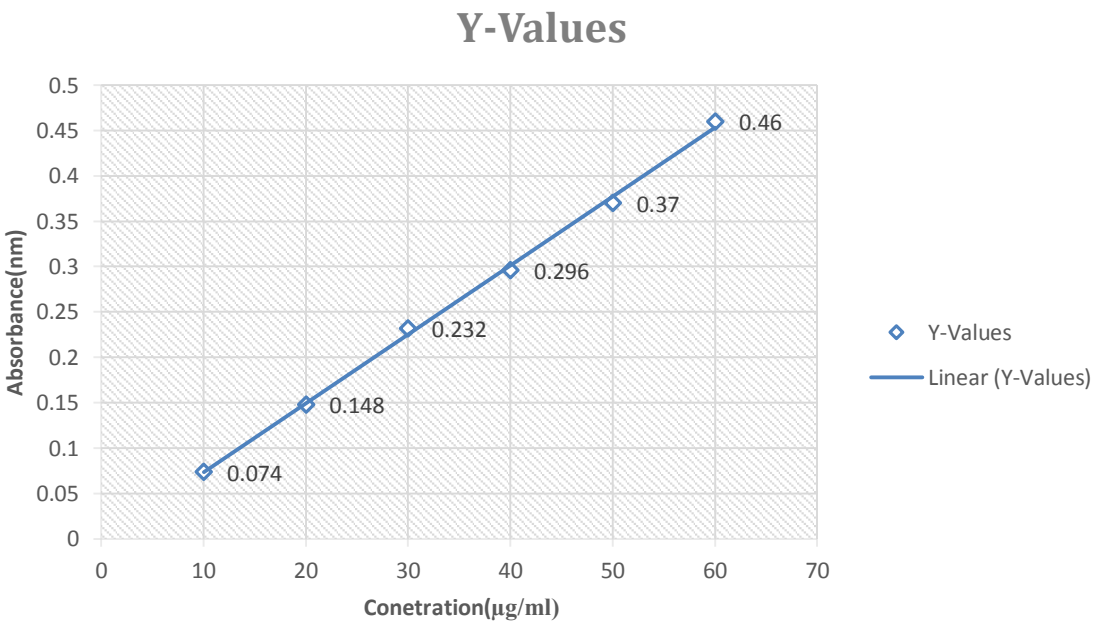

Fig 3: Linearity plot of Etodolac

Table no 2: ACCURACY DATA

Samples	%Recovery	Statistical Analysis
S1: 50% 50% 50%	101.78	Mean =0.114 SD =0.01 %RSD =0.877%
S2: 100% 100% 100%	98.26%	Mean =0.198 SD =0.001 %RSD =0.505%
S3: 120% 120% 120%	101.89%	Mean =0.3186 SD =0.0015 %RSD =0.470%

Table No 3: Precession Data

Concentration	Absorbance	Amount present	Statistical Analysis
30	0.135	10.0036	
30	0.137	10.0310	Mean=0.134667
30	0.135	10.0036	SD =0.001366
30	0.134	10.0310	%RSD=1.014%
30	0.134	10.0036	
30	0.133	10.0588	

Table No: 3 Sandell's Sensitivity Data

Sandell's Sensitivity (л) = Conc. (µg/100 ml) x 0.001/$D_{1.}$				
S.no	Concentration(µg/ml)	absorbance	Sensivity	Mean sensivity
1	10	0.074	0.144	
2	20	0.148	0.139	
3	30	0.232	0.131	0.143
4	40	0.296	0.137	
5	50	0.37	0.161	
6	60	0.46	0.146	

MOLAR ABSORPITIVITY:

Molar Absorpitivity was calculated by using the following formula.

$$slope / pat\square\ lengt\square\ ,$$

Where slope = log ($y_2\_y_1/x_2\_x_1$)

Slope/path length =2.16/1 =2.16 $mol^{-1}cm^{-1}$

Where slope is taken from the linearity plot, where X_1 =0.232, X_2 =0.37, Y_1 =30, Y_2 =50 and path length is taken as the cuvetts width i.e.. 1 cm.

RESULTS AND DISCUSSION:

The method developed and valied as per ICH guidelines. The method was validated in terms of linearity, precision, accuracy, LOD, LOQ, sandell's sensitivity and molar absorpitity. Detection wavelength was selectedat 279.5 nm linearity in response was observed in 10-60µg/ml having R^2=0.999. (R^2 not less than 0.996).The precision results show % RSD =1.014%(less than 2) each level clearly that the method is precision enough for the analysis of etodolac. The accuracy of the method was checked by recovery studies. The LOD =0.212 and LOQ=0.657 indicate

sensitivity of the method. The sandell's sensitivity for the developed method was found to be 0.143 and the moler asorpitivity was found to be 2.16 $mol^{-1}cm^{-1.}$

CONCLUSION:

A validation UV spectrophotometric method has been developed for the estimation of etodolac in bulk as well as pharmaceutical dosage form.In this proposed method the linearity was observed in the concentration range of 10-60µg/ml with correlation coefficient R^2 =0.99 for ETODOLAC at 279.5 nm. The developed was found to be simple, accurate, precise, specific, reprodouicible and linear over the concentration range studies. The proposed method can be used for routine analysis of etodolac. The method was validated as per ICH guidelines.

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