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# DEVELOPMENT OF VALIDATED UV SPECTROPHOTOMETRIC METHODS FOR THE SIMULTANEOUS ESTIMATION OF FEBUXOSTAT AND DICLOFENAC POTASSIUM IN TABLET DOSAGE FORM UTILISING SIMULTANEOUS EQUATION AND ABSORBANCE RATIO METHOD

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## ABSTRACT

Two validated uv spectrophotometric methods for the simultaneous estimation of Febuxostat and Diclofenac in a multicomponent dosage form have been developed, utilising simultaneous equation and absorbance ratio method. The method is based on the measurement of absorbance of Febuxostat and Diclofenac at their respective wavelengths of 315 nm and 282nm and at the iso absorptive wavelength of 288 nm in methanol. Febuxostat and Diclofenac at their respective $\lambda_{max}$315 nm and 282nm obeyed Beer's law in the concentration range 2-10µg/ml and 4-14µg/ml respectively with correlation coefficient 0.9993 for Febuxostat and 0.9992 for Diclofenac. The results have been validated statistically as per ICH guidelines.

**KEYWORDS: Febuxostat; Diclofenac; Simultaneous estimation; absorbance ratio, Validation.**

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# INTRODUCTION

Febuxostat (FEB) $O^3$-(2-methoxyethyl) $O^5$-[(*E*)-3-phenylprop-2-enyl] 2,6-dimethyl-4-(3-nitrophenyl)-1,4-dihydropyridine- 3,5-dicarboxylate is a novel non-purine selective inhibitor of xanthine oxidase, is a potential alternative to allopurinol for patients with hyperuricemia and gout.[1]

Diclofenac is used to treat pain, inflammatory disorders, and dysmenorrhoea. Inflammatory disorder may include musculoskeletal complaints, especially arthritis, rheumatoid arthritis, polymyositis, dermatomyositis, osteoarthritis, dental pain, TMJ, spondylarthritis, ankylosing spondylitis, gout attacks,[2].Chemically it is 2-(2-(2,6-dichlorophenylamino)phenyl)acetic acid. The combination is recommended to alleviate thesigns and symptoms associated with gouty arthritis

Literature review revealed only very few methods of estimation for the drug Febuxostat have been carried out which include an HPLC[3] method and an uv spectrophotometric method[4].

For the simultaneous estimation of Febuxostat and Diclofenac in two component dosage forms, no method has been reported so far. Hence it was proposed to develop economical, rapid and simple uv spectrophotometric methods for the simultaneous estimation of these drugs in tablet dosage forms.

### *Materials*

All the chemicals and reagents used were of analytical grade. Febuxostat was obtained as gift sample from MSN Organics Pvt.Ltd.,Nalgonda. The combined dosage form was purchased from local market. Methanol HPLC was procured from SD Fine-Chem limited, Mumbai.

### *Equipment*

The Jasco double beam uv-vis spectrophotometer with spectral band width 2.0 nm wavelength accuracy 0.5nm and matched quartz cells of 1 cm path length were used for all spectral and absorbance measurements. Class A volumetric glass wares were used.

### *Experimental Methods*

### *Standard solutions and Calibration curves*

Stock solutions for spectrophotometric measurements were prepared by dissolving FEB and DFC in methanol to obtain concentration of 1mg/ml for each compound. For calibration, series of above solutions were prepared containing FEB 2.0,4.0,6.0,8.0,10.0 µg/ml and DFC 4.0,6.0,8.0,10.0,12.0,14.0 µg/ml by diluting the stock standard solution with methanol in standard volumetric flasks(10ml).The solutions were scanned in the range of 220-350nm.

### *Selection of wavelengths*

### *Method I. Simultaneous equation method*

Overlain spectra for both the drugs are shown in Fig.1. Two wavelengths selected for the use of simultaneous equation were 315 and 282nm. The absorbance was recorded at the selected wavelengths and the absorptivity values were determined for Febuxostat and Diclofenac. Statistical parameters like slope, intercept, coefficient of correlation and SD were determined. (Table 1).

### *Method II. Absorbance ratio/Q value method*

From the overlain spectra of the two drugs, the isoabsorptive wavelength of 288nm and the λ max of Febuxostat at 315 nm were selected for this method. The absorptivity values were calculated at 288nm and 315nm. Statistical parameters like slope, intercept, coefficient of correlation and SD were determined. (Table 1).

### *Derivation of Equations*

### *Method I*

### *Simultaneous equation method*

The absorbance and the absorptivity values at the particular wavelength were calculated and substituted in the following equation, to obtain the concentration of these two drugs in combination in their pharmaceutical formulations.

$Cx = (A_1ax_2 – A_2ax_1) / (ax_2ay_1 – ax_1ay_2)$.

$Cy = (A_2ay_1 – A_1ay_2) / (ax_2ay_1 – ax_1ay_2)$.

Where,

Cx = Concentration of FEB

Cy = Concentration of DFC
$A_1$&$A_2$ absorbance of sample at 315 nm and 282 nm respectively
$ax_1$&$ax_2$ absorptivity of FEB at 315 nm and 282 nm respectively
$ay_1$&$ay_2$ absorptivity of DFC at 315 nm and 282 nm respectively.

***Method II***

***Absorbance ratio/Q value method***

In Q analysis method, absorbances were measured at selected wavelength i.e.288 nm (isoabsorptive point) and at 315 nm (λ max of FEB).The absorptivity coefficient of each drug at both the wavelengths were determined. The concentration of each drug in the tablet formulation were determined by substituting the absorbances and absorptivity coefficients in the equation

$Cx = (Qm - Qy)/(Qx - Qy) \times A1/ax_1$

$Cy = (Qm - Qx)/(Qy - Qx) \times A1/ay1$

where,

Qm = Absorbance of sample at 315 nm / Absorbance of the sample at 288 nm
Qx = Absorptivity of FEB at 315 nm / Absorptivity of FEB at 288 nm
Qy = Absorptivity of DFC at 315 nm / Absorptivity of DFC at 288 nm
A1 = Absorbance of the sample of sample at 288nm( isoabsorptive point)
$ax_1$= Absorptivity of FEB at 288 nm
$ay_1$ = Absorptivity of DFC at 288 nm

***ANALYSIS OF FORMULATION***

Twenty tablets of brand XANFEB DSR (Indoco Remedies Ltd.) containing 40 mg of FEB and 100 mg of DFC were weighed, average weight determined and finely powdered. Appropriate quantity of powder equivalent to 4 mg of FEB and 10 mg DFC was accurately weighed, transferred to a 100 ml volumetric flask and volume was made up to 100 ml with methanol and shaken vigorously for 15 minutes. The solution was then sonicated for 5 minutes and filtered through the Whatman filter paper no.41. Necessary dilutions of filtrate were made with methanol to get final concentration 4 µg/ml of FEB and 10 µg/ml of DFC. Absorbance of this solution was measured at 315 nm (λ max of FEB) 282nm ((λ max of DFC), and 288 nm (Isoabsorptive Point), The values obtained were substituted in the respective formulae of Method 1 & 2 to obtain concentrations of FEB and DFC. The results are shown in Table 3.

***Method Validation[5].***

***Calibration curve (linearity of the method)***

Calibration curves were constructed by plotting absorbance vs. concentrations of FEB and DFC,at their respective λmax and the regression equations were calculated. The calibration curves were plotted over different concentrations in the range 2-10 µg/mL and 4-14 µg/mL for FEB And DFC, respectively.(Fig.2 and Fig.3) The optical parameters and statistical parameters are depicted in Table 1.

***Accuracy (% Recovery)***

The accuracy of the method was determined by calculating recoveries of FEB and DFC by the standard addition method. Known amount of standard of FEB and DFC (80%, 100%, and 120%) were added to the sample solutions of tablet dosage forms. The amounts of FEB and DFC were estimated by method I and II. The results are shown in Table 2. The values prove that the method is accurate.

***Method Precision***

The reproducibility of the method was determined by performing the assay on the same day (intra-day precision)and three different days(inter day precision). Precision studies were carried out by preparing nine concentrations (3 replicates of three different concentrations covering the entire linear range). The RSD values were found to be below 2% which indicate that the proposed methods are precise. (Table 2).

**Figure 1**
***Overlain Absorption spectra of Febuxostat and Diclofenac***

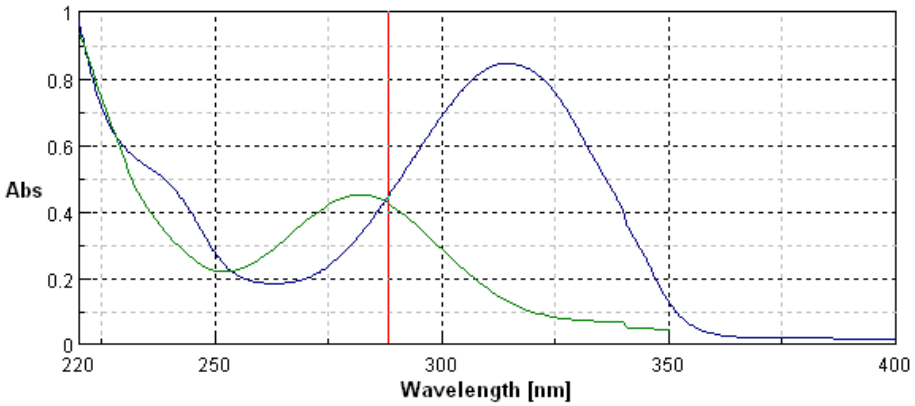


**Figure 2**
***Calibration plot of fundamental spectra of FEB***

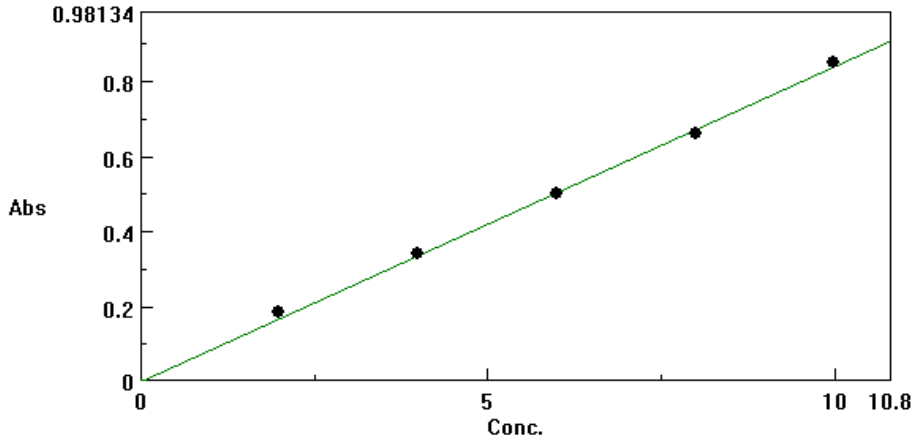


**Figure 3**
***Calibration plot of fundamental spectra of DFC***

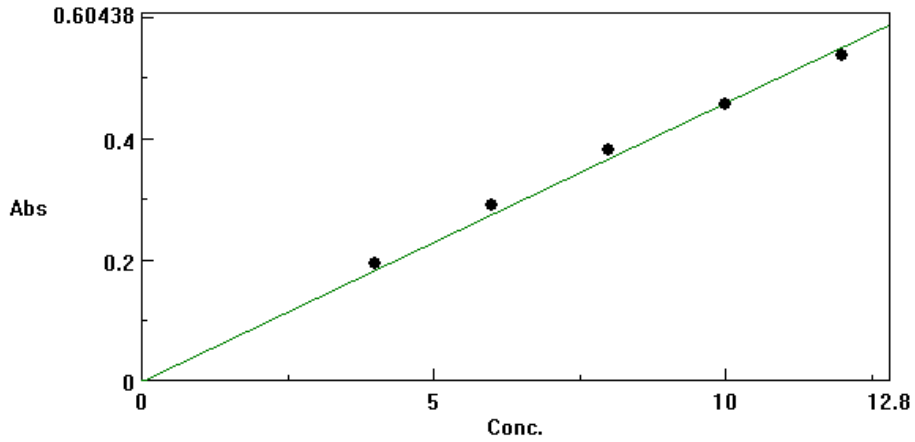

**Table 1**
***Optical characteristics Data***

| Parameters | FEB | DFC |
|---|---|---|
| Working λmax | 315nm | 282 nm |
| Beer's law limit | 2-10µg/ml | 4-14µg/ml |
| Correlation coefficient | 0.9993 | 0.9992 |
| Intercept | 0.0112 | 0.0914 |
| Slope | 0.0825 | 0.0366 |
| Regression equation | y=0.0825x+0.0112 | y=0.0366x+0.0914 |

**Table 2**
***Summary of validation parameters for the proposed methods***

| Parameter | FEB | | DFC | |
|---|---|---|---|---|
| | Method I | Method II | Method I | Method II |
| Accuracy % | 99.72 | 98.94 | 100.10 | 98.95 |
| Precision (RSD, %) | | | | |
| Repeatability(n=3) | 0.18 | 0.28 | 0.22 | 0.32 |
| Intraday(n=3) | 0.20-0.25 | 0.21-0.39 | 0.22-0.84 | 0.35-0.78 |
| Interday(n=3) | 0.68-1.22 | 0.48-0.88 | 0.62-1.12 | 0.68-0.94 |

**Table 3**
***Compilation of results of commercial formulation***

| Brand name | Company | Formulation | Label Claim | Amount found (mg) | |
|---|---|---|---|---|---|
| | | | | Method I | Method II |
| XANFEB | Indoco Remedies Ltd. | Tablets | FEB 40mg | 39.4±0.10 | 38.9±0.12 |
| DSR | | | DFC 100mg | 99 .89±0.34 | 98.77±1.56 |

# CONCLUSION

The methods discussed in the present work provide a convenient and accurate way for simultaneous estimation of FEB and DFC in combined oral dosage forms. In simultaneous equation method, wavelengths selected for analysis were 315 nm (λmax of FEB) and 282 nm (λmax of DFC).In Q-analysis method wavelengths selected were 291 nm (isoabsorptive point) and 282 nm (λmax of DFC). In both the methods linearity for detector response was observed in the concentration range of 2-10 µg/ml (for FEB) and 4-14 µg/ml (for DFC). Absorptivity coefficient were calculated for both the drugs at selected wavelengths and substituted in equations for determining concentration of FEB and DFC in its tablet dosage form. Percent label claim for FEB and DFC in tablets was determined by simultaneous equation method and by Q-analysis method. Accuracy of proposed methods was ascertained by recovery studies. Hence the proposed methods can be employed for routine quality control of Febuxostat and Diclofenac in its combined dose formulations.

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