Research Journal of Pharmaceutical, Biological and Chemical Sciences

Validated HPTLC Method for the Simultaneous Determination of Diclofenac Potassium and Tizanidine Hydrochloride in Tablet Dosage Form

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ABSTRACT

A simple, precise, accurate and rapid reverse phase high performance thin layer chromatographic method was developed for the simultaneous estimation of Diclofenac Potassium and Tizanidine Hydrochloride in tablet dosage forms. A Precoated silica gel Plate 60F 254 (Merck) stationary phase with mobile phase Toluene: Isopropyl alcohol: Ammonia (30:20:2.5) was used. The effluents were monitored at 280 nm. The R_f value were found to be between 0.72 - 0.79 and 0.85-0.92 for Diclofenac Potassium and Tizanidine Hydrochloride respectively .The Linearity for Diclofenac Potassium and Tizanidine HCl were in the range of 3000 to 7000 g/ml for Diclofenac Potassium and 120 to 260 g/ml respectively. Results of the analytical method were validated statically, and by recovery studies. The proposed method can be successfully used to deter mine the drug contents of marketed formulation.

Keywords: HPTLC, Diclofenac Potassium, Tizanidine HCl

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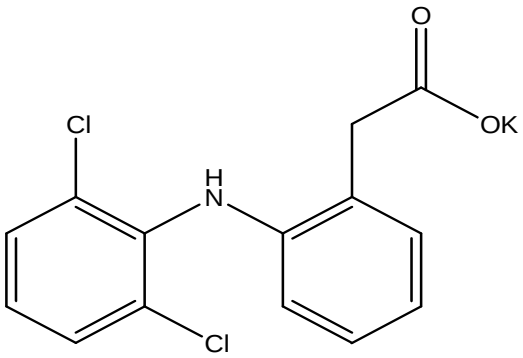
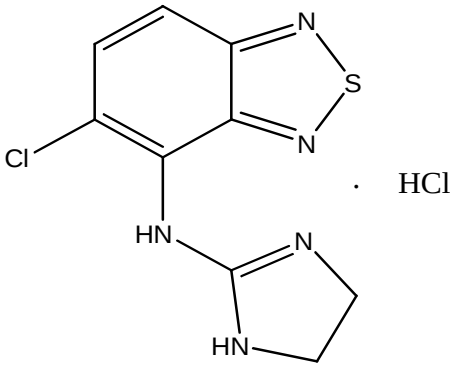
INTRODUCTION

Diclofenac Potassium and Tizanidine HCl combination is used clinically for its NSAID properties. Diclofenac K is chemically 2-[(2,6-dichlorophenyl)amino] benzeneaceticacid, monopotassium salt and Tizanidine HCl is 5-chloro-4-(2-imidazoline-2-ylamino)- 2,1,3-benzothiodiazole hydrochloride.



Diclofenac Potassium



· HCl

Tizanidine Hydrochloride

The mechanism of action of Diclofenac Potassium is not completely under-stood but may be related to prostaglandin synthetase inhibition. Tizanidine is an agonist at α2-adrenergic receptor sites and presumably reduces spasticity by increasing presynaptic inhibition of motor neurons. On detailed literature survey, it was found that these drugs have been estimated individually and in combinations by various methods [1-11]. Besides, UV method for simultaneous estimation of this combination was reported [1]. In this communication we report a new HPTLC method for simultaneous estimation of Diclofenac Potassium and Tizanidine HCl from Tablet dosage form, which is simple, rapid and precise.

MARERIALS AND METHODS

Parameters for HPTLC analysis

The stationary phase used was pre-coated Silica Gel Plate 254 (Merck) with the mobile phase as Toluene: Isopropyl alcohol: Ammonia (30:20:2.5). The plate size is 20 x 15 cm with 15 mm distance between two bands. The densitogram were developed in CAMAG twin through chamber by applying 5 µl volume of the sample. The eluents were monitored at 280nm. CAMAG Linomat IV was the sample applicator and CAMAG TLC Scanner III and version 4.01 was the scanner mode.

Reagents and Chemicals

Methanol Isopropyl alcohol (HPLC Grade), Toluene (AR Grade), Ammonia (AR Grade), Diclofenac Potassium (working standard), Tizanidine Hydrochloride (Working standard).

Preparation of standard stock solution

An accurately weighed quantity of 20 mg of Tizanidine Hydrochloride and 500 mg of Diclofenac Potassium was dissolved in methanol. Make up to 10 ml to obtain a stock solution of 2000 mcg/ml of Tizanidine Hydrochloride and 50000 mcg/ml of Diclofenac Potassium.

Linearity and Calibration

To evaluate the linearity range of Tizanidine Hydrochloride and Diclofenac Potassium standard stock solution was diluted with methanol to give a minimum of 5 concentrations in the range of 120-260 mcg/ml Tizanidine Hydrochloride and 3000-7000 mcg/ml of Diclofenac Potassium respectively.

Sample preparation

Twenty tablets were weighed and powdered. The powder equivalent to 20 mg of Tizanidine Hydrochloride and 50 mg of Diclofenac Potassium was weighed and transferred into a volumetric flask. Made up to 10 ml with methanol, shaked for 15 min. mixed all the contents with the aid of ultrasonicator for 1 min and filtered through Whatman filter paper 42.

Estimation method

The sample solution was spotted on the chromplate with the help of Linomat IV spotting system. The chromplate was developed in a twin trough chamber containing the Mobile Phase. The chromatograms were recorded and R_f values were determined for Tizanidine Hydrochloride and Diclofenac Potassium. The amount of drug present was calculated by comparing the peak area values of standard with that of sample as follows:

$$\text{Amount of Drug in Each tablet} = \frac{\text{Peak area of test}}{\text{Peak area of standard}} \times \frac{\text{Standard dilution factor}}{\text{sample dilution factor}} \times \text{Average weight of tablet}$$

Recovery studies

To ensure the reliability of the method, mixing a known quantity of standard drug with the pre analyzed sample formulation carried out recovery studies and the contents were analyzed by the proposed method. The Densitogram showing the recovery behavior of the method.

Statistical validation

The precision or reproducibility of the analytical method was determined by repeating the analysis three times for each brand of product and the statistical parameters were calculated.

RESULTS

Assay

The solvent system consisting of Toluene: Isopropyl alcohol: Ammonia (30:20:2.5) was found to be ideal mobile phase for the separation of Tizanidine Hydrochloride and Diclofenac Potassium. The well resolved bands of the drugs were scanned for R_f values in the scanner at the wave length 280nm. The R_f value was found to be between 0.72 - 0.79 and 0.85-0.92 for Diclofenac Potassium and Tizanidine Hydrochloride respectively.

Assay

	PEAK AREA OF STANDARD		PEAK AREA OF SAMPLE	
	Diclofenac potassium	Tizanidine HCl	Diclofenac potassium	Tizanidine HCl
	50340.1	4198.1	51025	4105.1
	50127.1	4187.2	50304.1	4157.9
	49987.1	4175.1	51021.1	4102.1
AVG	50151.4	4186.8	50783.4	4127.7
STD DEV	177.75357	11.50522	415.09056	31.38598
RSD	0.354433688	0.274797368	0.817374489	0.761481528

Linearity

Linearity was evaluated by plotting peak area as a function of analyte concentration for. Diclofenac Potassium and Tizanidine Hydrochloride. From the linearity studies the specified range determined was 3000 to 7000 g/ml for Diclofenac Potassium and 120 to 260 g/ml.

Linearity and Calibration

DICLOFENAC POTASSIUM		TIZANIDINE HCl	
CONCENTRATION (µg/ml)	PEAK AREA	CONCENTRATION (µg/ml)	PEAK AREA
0	0	0	0
3000	30123.1	120	2510.1
4000	39789.2	160	3097.1
5000	49987.1	200	4198.1
6000	60024.2	240	5031.2
7000	71435	260	5467.5

Repeatability

A system repeatability test was applied to a representative Densitogram to check the repeatability of the proposed method. The proposed method was used to the quantitative estimation of the Diclofenac Potassium and Tizanidine Hydrochloride tablet dosage form.

Recovery data

The validity of the proposed method was verified by the recover y studies. The recovery studies were carried out and the results expressed as percentage recovery (%). The percentage recovery studies are quite optimum for the method developed.

Recovery data

Sample	mg of std drug added	mg of drug* recovered	% of recovery
Diclofenac Potassium	0.2	0.19	99.54
Tizanidine Hydrochloride	5	4.63	99.33

* Mean of 5 values

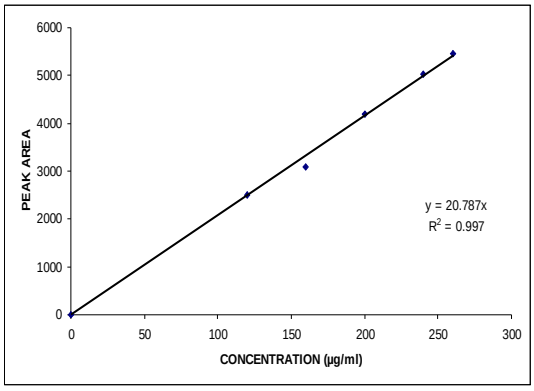
Statistical validation

Drug	Lable claim (mg/tablet)	Amount estimated (mg/tablet)	Std dev	%RSD	Std Error
Diclofenac Potassium	50	9.85	0.08	0.80	± 0.04
Tizanidine Hydrochloride	2	1.99	0.03	0.30	± 0.01

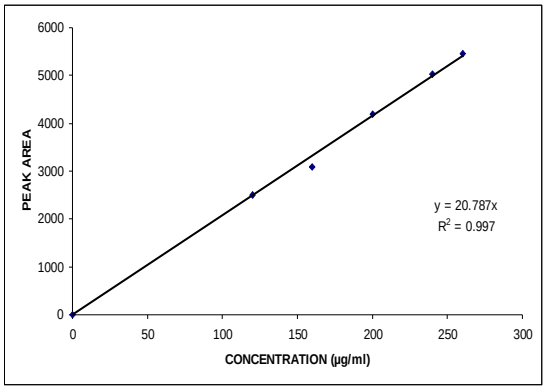
System suitability parameters

Parameter	TIZANIDINE HCL	DICLOFENAC K
Tailing Factor	O.5	1.07
Resolution Factor	8.87	.....

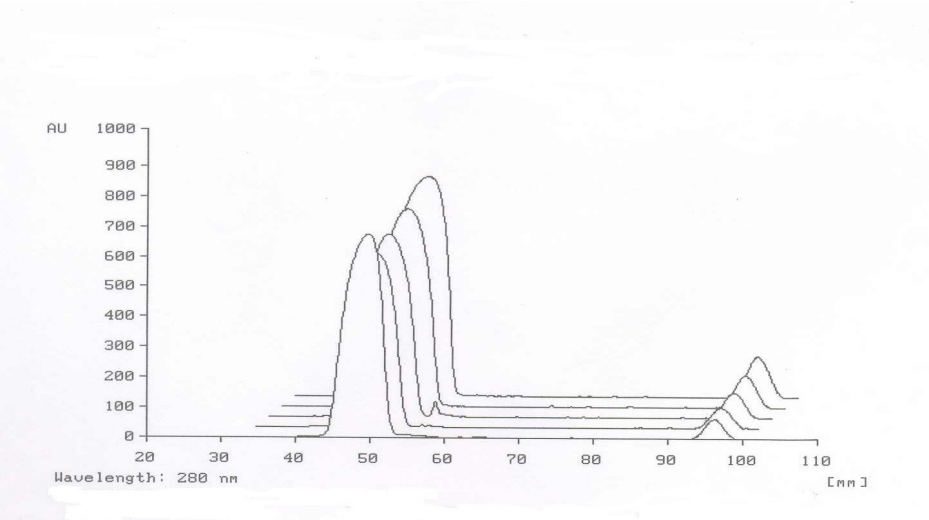
Linearity of DICLOFENAC POTASSIUM



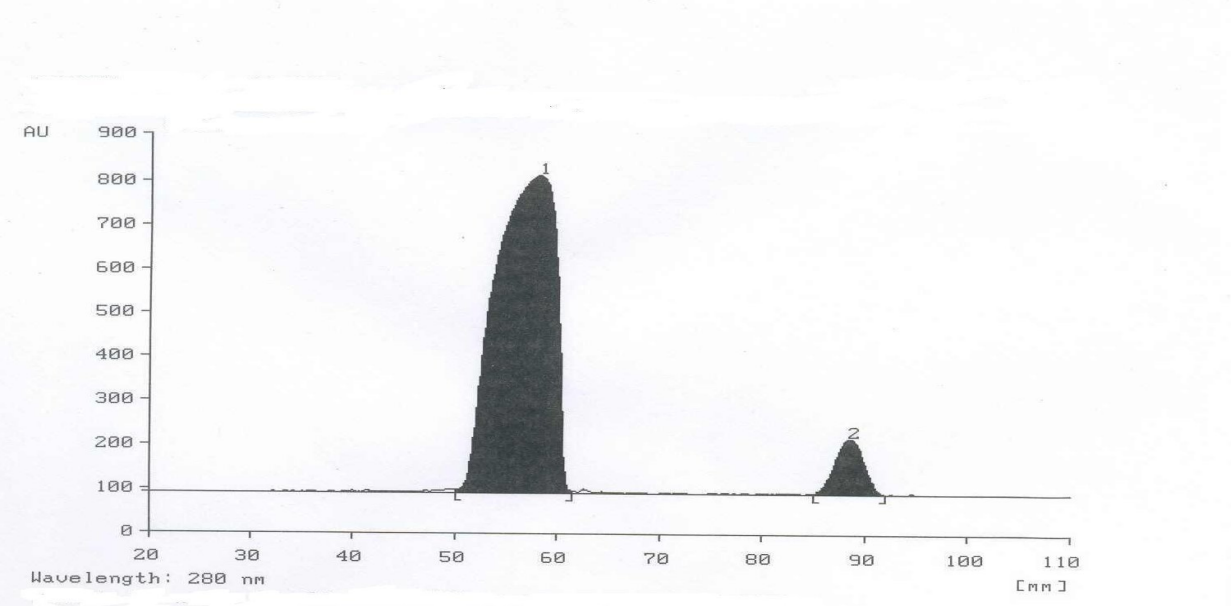
Linearity of TIZANIDINE HCl



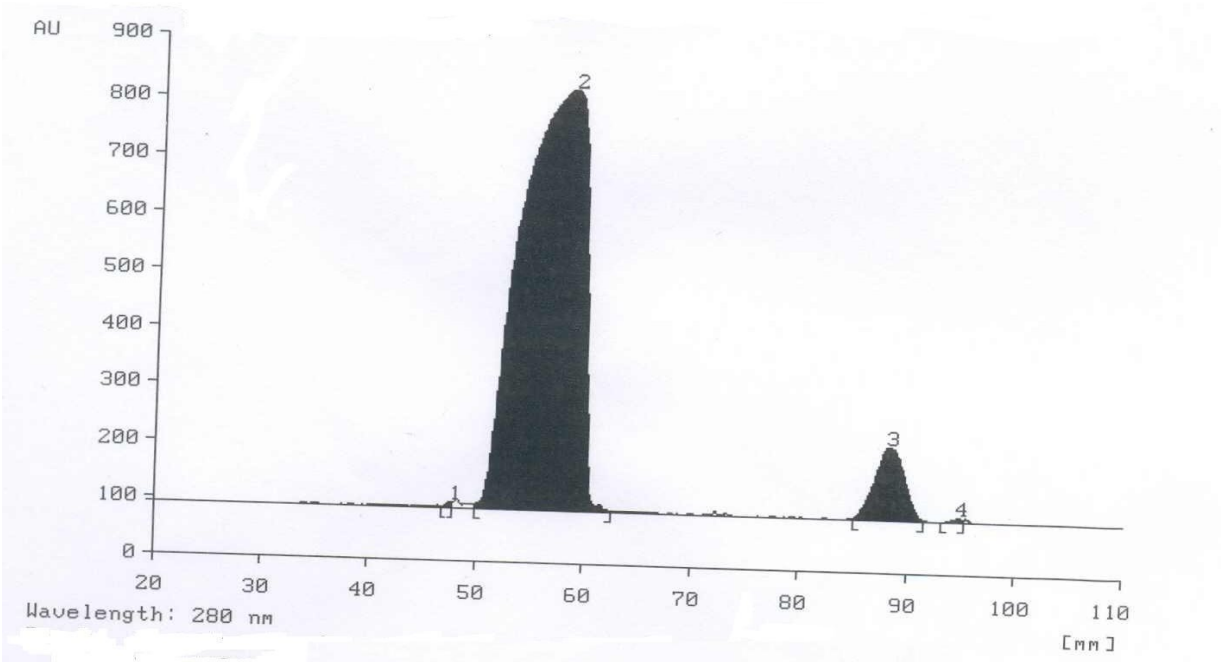
Overlain densitogram for the mixed standard



Densitogram for the mixed standard



Densitogram for the assy of combined tablet dosage form



CONCLUSION

In this present study an attempt has been made to develop an analytical method for the simultaneous estimation of Diclofenac Potassium and Tizanidine Hydrochloride in combined tablet dosage for m.The present combination of Diclofenac Potassium and Tizanidine Hydrochloride was marketed as one formulation (TIZARAN).

Diclofenac Potassium – 50mg /tab Tizanidine Hydrochloride – 2mg/tab

The fixed dose combination tablet of Diclofenac Potassium and Tizanidine Hydrochloride was subjected to simultaneous estimation by HPTLC method. Highly reliable and cost efficient HPTLC method was developed for the quantitative estimation of Diclofenac Potassium and Tizanidine Hydrochloride in combined tablet dosage form.

The results obtained were reproducible and reliable. The validity and precision of the methods were evident from the statistical and analytical parameters obtained. From the forgoing it is concluded that the method developed is simple, rapid, selective and precise hence suitable for application in routine analysis of pharmaceutical preparations.

REFERENCES

[1] Ashok Kumar, Anroop B, Kshif Nazim. The Indian Pharmacist 2005;4:81-84.

[2] Ciapina EG, Santini AO, Weinert PL, Gotardo MA, Pezza H R Pezza L. Chromatographia 2003; 65:315.

[3] Subramanian G, Musmade P, Agarwal S, Udupa N. J Pharm Biomed Anal 2004; 66: 5.

[4] Kubala.T, Gambhir B, BORST S.I, Chromatographia, 2007, Sep; 66 (1):87 – 91.

[5] Lei Wang, Mei-Ling Qi, Chromatographia 2006; 20(15):2286-2292

[6] Qi, Peng Wang and Lei Wang, Shenyang Pharmtech Institute of Pharmaceuticals 2002; 32:13-17

[7] MaríaS . Aurora-Prado, Martin Steppe, Marina FM. Tavares, Erika RM, J Pharm Biomed Anal 2005;3: 107-110.

[8] Mei- Ling Qi, Peng Wang and Lei Wang, Shenyang Pharmtech Institute of Pharmaceuticals, 2002; 32: 39-41

[9] Ramakrishna.V. S,Nirogi , Vishwottam N. Kandikere, Manoj Shukla, Koteshwara Mudigonda, Santosh Maurya, Asian J Chem 2006; 18 (4):3123 – 3125.

[10] Mukherjee B, Mahapatra S, Asian J Chem 2003; 65:73-75.

[11] Nagoji.K.E.V, Vijayasrinivas.S, Kiran Kumar.M, Mathivanan.N, Satish Kumar.M, Rao M.E.B. Asian J Chem 1993;19:76-79