of additional peak was observed. Repeatability and reproducibility studies demonstrated the precision of the method. Repeatability studies were done by consequently injecting the standard solution at five different concentrations. These solutions were prepared in duplicate and injected as per assay procedure. The % RSD was found to be 0.42 and 1.2, respectively for two brands.

The system suitability parameters like peak asymmetry factor, capacity factor, peak area or height of their repetitive injection were carried out as specified in USP. Although USP requires only two of these criteria for method validation, *parameters like column efficiency* (10,156), *capacity factor* (1.05) and peak asymmetry factor (1.25) were calculated in the present study. Tailing factor was found to be 1.225. The LOD and LOQ of the developed method were determined by injecting progressively low concentration of the standard solution into HPLC system under optimum condition. The LOD and LOQ of the drug valdecoxib were found to be 0.5 ng/ml and 200 ng/ml respectively. In the present study the mobile phase, standard solution and sample solution were subjected to 12 h at room temperature and under refrigeration. The stability of the solution were studied by performing the experiment and looking for change in Rt, Rs and tailing of the peak compared to the pattern of the chromatogram of freshly prepared solution. The solutions stored under room temperature as well as refrigeration were stable up to 1 h. The result obtained by the method was precise and reproducible for the drug valdecoxib. The high percentage recovery and low percentage deviation were satisfactory and it shows the accuracy, reliability and suitability of the method. Hence the method developed can be used for routine analysis of valdecoxib in tablet formulations.

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Analysis of Gatifloxacin in tablet dosage form

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Two simple methods, spectrofluorimetric method and spectrophotometric method for the determination of gatifloxacin in pharmaceutical formulation are described. For spectrofluorimetric method, the excitation and emission wavelengths were found to be 365 nm and 492 nm respectively. In the case of spectrophotometric method gatifloxacin in alkaline medium on treatment with ceric

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ammonium sulphate and 3-methyl-2-benzothia-zolinone hydrazone gave a green colored chromogen (λ_{max}=452 nm). For the spectrofluorimetric and spectrophotometric methods, the linearity was found to be in the range of 0.2 to 1 µg/ml and 10 to 30 µg/ml respectively.

Gatifloxacin[1] chemically known as ±1-cyclopropyl-6-fluoro-1,4-dihydro-8-methoxy-7-[3-methyl-1-piperazinyl]-4-oxo-3-quinoline carboxylic acid is an antibacterial agent against acute bacterial exacerbation of chronic bronchitis. Liquid chromatographic methods[2-4] were reported for determination of gatifloxacin in plasma and urine. Literatures were also available on photodegradation studies[5-7] of gatifloxacin. Neither spectrofluorimetric nor spectrophotometric methods have been reported for estimation of gatifloxacin from formulation. Hence, it is essential to develop a simple and accurate method which can be routinely employed. The present communication reports two simple, precise and accurate spectrofluorimetric and spectrophotometric methods for the estimation of gatifloxacin from tablet dosage form.

Gatifloxacin was obtained as gift sample from Unichem Pharmaceuticals Ltd., Roha and 200 mg tablet, Gatilox, manufactured by Sun Pharmaceuticals Ltd., Vadodara was procured from the local market. Sodium hydroxide pellets, ceric ammonium sulphate (CAS), 3-methyl-2-benzothiazolinone hydrazone hydrochloride (MBTH), ethanol and methanol were procured from S. D. Fine Chemicals Ltd., Mumbai and Qualigens Fine Chemicals Ltd., Mumbai. All chemicals used in this study were of analytical grade. All solutions were prepared in distilled water.

A Jasco FP–750 spectrofluorimeter was used for fluorescence measurements and Jasco V–530 UV/Vis spectrophotometer was employed for absorbance measurements.

Fluorescence occurs because of transition from first excited singlet state to ground state by emission of light[8,9]. Gatifloxacin consists of a heterocyclic fused ring structure, which is responsible for fluorescent behavior. Fluorophore in citric acid and ethanol showed an excitation wavelength at 365 nm and emission wavelength at 492 nm.

For spectrofluorimetric estimation, aliquots of standard gatifloxacin solutions ranging from 0.2-1 ml (from 10 µg/ ml solution) were pipetted into a series of 10 ml volumetric flasks. Then 6 ml of 5% citric acid was added to each flask, contents of the flasks were shaken well and finally the volume was made up to 10 ml with ethanol. The fluorescence intensity of the resulting solution was measured at emission wavelength of 492 nm keeping excitation wavelength as 365 nm. The calibration curve was prepared by plotting concentration of gatifloxacin Vs fluorescence intensity of respective solutions. Twenty tablets of gatifloxacin were weighed and average weight was calculated. Quantity of powder equivalent to 5 mg was weighed accurately and transferred to a 100 ml volumetric flask. The active ingredient was extracted with methanol and volume was made up with ethanol and filtered. The filtered solution was further diluted to get requisite concentrations and analyzed as described under the procedure for pure sample. The concentration of gatifloxacin in tablet formulation was calculated from standard graph. Results are given Table 1.

The second method is spectrophotometric method which is based on the following probable mechanism. In the presence of an oxidant like ceric ammonium sulphate (CAS), 3-methyl-2-benzothiazolinone hydrazone hydrochloride (MBTH) forms an electrophilic intermediate compound, which reacts to most nucleophilic site of aromatic ring or moieties (NH_2, OH) to give a colored oxidative coupling product[10,11]. Here by the reaction of CAS and MBTH with gatifloxacin (fig. 1), a green colored

TABLE 1: ASSAY OF GATIFLOXACIN IN TABLET DOSAGE FORM

Drug	Labeled amount (mg/tablet)	Amount found * by proposed method (mg/tablet)		% Recovery*	
		Spectrofluorimetric method	Spectroscopic method	Spectrofluorimetric method	Spectroscopic method
Gatiflox	200	203.1±0.17	213.0±0.09	99.5±0.07	100.1±0.05

*All values are Mean ± standard deviation of six determinations

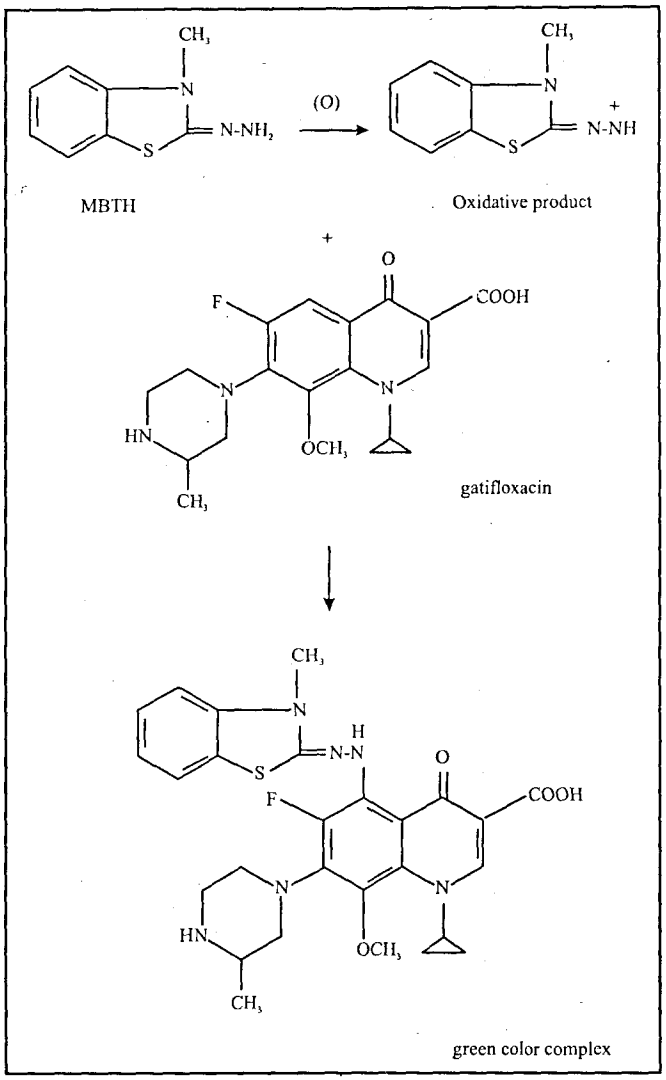

Fig. 1: Probable mechanism of color development

chromogen was obtained whose absorbance was noted at 452 nm.

For spectrophotometric estimation, aliquots of standard gatifloxacin solutions ranging from 1-3 ml (from 100 µg/ml solution) were pipetted into a series of 10 ml volumetric flasks. Then 3 ml of 0.7% w/v CAS and 2 ml of 0.2% MBTH solutions were added. The contents of the flasks were shaken well, allowed to stand for 10 minutes and finally the volume was made up to 10 ml with 0.1N NaOH. The absorbances of these solutions were noted at 452 nm. The calibration curve was prepared by plotting concentration of gatifloxacin Vs absorbance of respective solutions. Twenty tablets of gatifloxacin were weighed and average weight was calculated. Quantity of powder equivalent to 10 mg was weighed accurately, dissolved and volume was made up to 100 ml with 0.1N NaOH. This solution was filtered and further diluted to get requisite concentrations and analyzed as described under procedure for pure samples. The concentration of gatifloxacin from formulation was calculated from standard graph. The results are given in Table 1.

The developed methods were validated for accuracy by carrying out the recovery studies. It was carried out by mixing a known quantity of standard gatifloxacin with the analyzed tablet formulation and the contents were reanalyzed by the proposed method (Table 1). Parameters such as Beers law range, molar extinction coefficient, correlation coefficient, slope and intercept are given in Table2.

The spectrofluorimetric method shows linearity in the concentration range of 0.2-1 µg/ml, which is established by good correlation coefficient value (0.9996). The UV spectroscopic method shows linearity in the concentration range of 10-30 µg/ml. The recovery values close to 100% indicate the reproducibility and accuracy of the methods. Low standard deviation values establish the reproducibility of the developed methods. The proposed methods are simple and sensitive with good precision and accuracy and can be used for routine analysis of gatifloxacin in tablets dosage forms.

TABLE 2: VALIDATION PARAMETERS

Parameter	Spectrofluorimetric method	Spectroscopic method
Linearity Range (µg/ml)	0.2-1	10-30
Molar Absorptivity (l/mole cm)	$7.2x10^7$	$0.65x10^4$
Regression Equation (Y=a + bc) Slope(b)	159.6141	0.0179
Intercept (a)	13.6380	-0.0110
Correlation Coefficient (r)	0.9996	0.9988

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A Validated High Performance Thin Layer Chromatographic Determination of Fenofibrate

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The present work describes a validated high performance thin layer chromatographic method for estimation of fenofibrate in capsules. Aluminum plates precoated with Silica gel 60 F_{254} was used as stationary phase and toluene:chloroform (7:3 v/v) as mobile phase. Quantification was carried out by the use of densitometric absorbance mode at 296 nm. The amount of fenofibrate estimated as percentage of labeled claimed was found to be 101.43/100.68 (as per the peak height and peak area) in capsules. The proposed high performance thin layer chromatographic method was quantitatively evaluated in terms of stability, precision, repeatability and accuracy. The calibration correlation proving, it's utility for routine analysis of fenofibrate capsule.

Fenofibrate, chemically is 2-[4-(4-chlorobenzoyl) phenoxy]-2-methyl propionic acid,1-methyl ethyl ester. It is a lipid-lowering agent. Fenofibrate is official in BP[1]. Literature survey revealed HPLC and GC-MS methods for the estimation of fenofibrate aquatic environmental samples[2,3] and in human plasma[4-6]. No methods have been reported in the literature for its estimation in pharmaceutical dosage form. The present study describes the development and validation of a simple, specific, accurate and precise HPTLC method for determination of fenofibrate in capsules.

Fenofibrate working standard was a gift sample from US Vitamin India limited, Mumbai. Silica Gel 60 F_{254} TLC plates (10x10 cm, E. Merck) were used as stationary phase. Twenty fenofibrate capsules (Lipicard-200 mg, USV Ltd.) were purchased from a local pharmacy. Chloroform and toluene of GR grade (E. Merck) purity were procured from local supplier. A Camag HPTLC system (Switzerland) comprising of Camag Linomat IV semiautomatic sample applicator, Camag TLC scanner 3, Camag twin trough chamber (10x10 cm), Camag Cats 4 Software, Hamilton syringe (100 μl) were used during the study.

An accurately weighed quantity of fenofibrate (50 mg) was transferred in to a 50 ml volumetric flask. It was dissolved and diluted up to the mark with methanol to give a standard stock solution of 1 mg/ml. Five milliliters of this stock solution was further diluted to 25 ml with methanol to give a working standard solution of 200 μg/ml.

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