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Micelle-enhanced and terbium-sensitized spectrofluorimetric determination of gatifloxacin and its interaction mechanism

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ABSTRACT

A terbium-sensitized spectrofluorimetric method using an anionic surfactant, sodium dodecyl benzene sulfonate (SDBS), was developed for the determination of gatifloxacin (GFLX). A coordination complex system of GFLX–Tb^{3+}–SDBS was studied. It was found that SDBS significantly enhanced the fluorescence intensity of the complex (about 11-fold). Optimal experimental conditions were determined as follows: excitation and emission wavelengths of 331 and 547 nm, pH 7.0, 2.0×10^{-4} mol l^{-1} terbium (III), and 2.0×10^{-4} mol l^{-1} SDBS. The enhanced fluorescence intensity of the system (ΔI_f) showed a good linear relationship with the concentration of GFLX over the range of 5.0×10^{-10} to 5.0×10^{-8} mol l^{-1} with a correlation coefficient of 0.9996. The detection limit (3σ) was determined as 6.0×10^{-11} mol l^{-1}. This method has been successfully applied to the determination of GFLX in pharmaceuticals and human urine/serum samples. Compared with most of other methods reported, the rapid and simple procedure proposed in the text offers higher sensitivity, wider linear range, and better stability. The interaction mechanism of the system is also studied by the research of ultraviolet absorption spectra, surface tension, solution polarity and fluorescence polarization.

1. Introduction

Gatifloxacin[(±)-1-cyclopropyl-6-fluoro-1,4-dihydro-8-methoxy-7-(3-methyl-1-piperazinyl)-4-oxo-3-quinoline carboxylic acid, GFLX] (Fig. 1) is the fourth generation of a new class of synthetic antibacterial fluoroquinolone agents. It is a novel extended-spectrum fluoroquinolone with an improved Gram-positive and anaerobe coverage compared with older agents such as ciprofloxacin. GFLX acts intravenously by inhibiting topoisomerase II (DNA gyrase) or topoisomerase IV. It does not appear to exert phototoxic effects [1]. This fluoroquinolone is mainly excreted in urine (>75%) in unaltered form, and no pharmacokinetic differences have been observed between oral and i.v. administration [2].

Various analytical techniques have been employed for the determination of GFLX such as high-performance liquid chromatography (HPLC) [3–9], spectrophotometry [10–12], high performance thin-layer chromatography (HPTLC) [13,14], microbiological assay [15], voltammetry [16], and atomic absorption spectrometry [17]. Due to its high sensitivity and selectivity, the spectrofluorimetry has been used to estimate GFLX in pharmaceutical formulations and biological fluids [18–20]. Quinolones have suitable functional groups to form stable complexes with Tb^{3+} and Eu^{3+}. The presence of these ions in quinolone solutions leads to the formation of complexes that absorb energy at the characteristic wavelength of the organic ligand and emit radiation of the characteristic wavelength of Tb^{3+} or Eu^{3+}. These complexes show a large Stokes shift and narrow emission bands. This technique has been widely utilized for the determination of quinolones norfloxacin [21], enoxacin [22], garenoxacin [23], trovafloxacin [24], ciprofloxacin [25], etc. But the sensitivity is not very high as all the detection limits of these methods are over the range of 1.2×10^{-9} to 4.7×10^{-8} mol l^{-1}. However, the micelle-enhanced and terbium-sensitized spectrofluorimetry that the present paper describes could provide a much higher sensitivity. The detection limit (3σ) could attain 6.0×10^{-11} mol l^{-1} and the linear range is 5.0×10^{-10} to 5.0×10^{-8} mol l^{-1}, which is at a low concentration level. Furthermore, most of these references had not discussed the luminescence mechanism except that [21] and [22] merely studied it by the fluorescence spectra and ultraviolet absorption spectra. But in the present paper the interaction mechanism of the fluorescence system is studied through the research of surface tension, solution polarity and fluorescence polarization. Compared with most of other published procedures for the determination of GFLX, the proposed method offers advantages in higher sensitivity, wider linear range, and better stability. It has been successfully applied to the determination of gatifloxacin in

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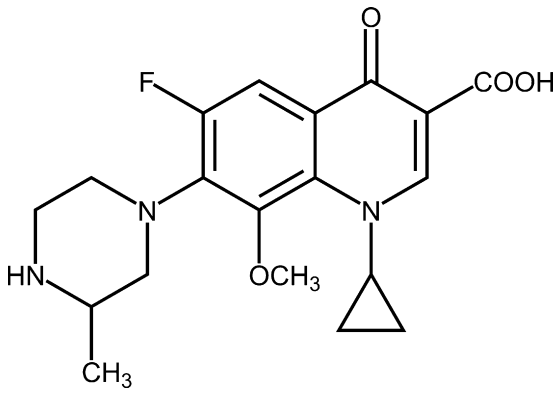

Fig. 1. Chemical structure of gatifloxacin.

pharmaceutical samples and human urine/serum samples, and the interaction mechanism of the system is also discussed. To the best of our knowledge, this is the first time that the terbium-sensitized spectrofluorimetric determination of GFLX has been reported.

2. Experimental

2.1. Apparatus

Normal fluorescence measurements were recorded with a Hitachi F-2500 Fluorescence Spectrophotometer (Japan), using a standard 10 mm pathlength quartz cell with 10 nm bandwidths for both the excitation and emission monochromators.

The fluorescence polarization spectra were obtained using an ISS K2 multifrequency phase and modulation spectrofluorometer (USA).

All absorption spectra were measured on a UV-2401PC spectrophotometer (Shimadzu, Japan) equipped with 10 mm pathlength quartz cells.

The surface tension was measured on a Processor Tensiometer-K12 (KRÜSS Corp., Germany) with the precise degree of the measurement 0.01 mN m^{-1} by the Wilhelmy plate.

The pH was measured using a Lei Ci pHs-3C pH-meter (Shanghai, China).

Serum samples were centrifuged with a Xiang Yi TDZ4A-WS brachytely desk centrifuge (Changsha, China).

2.2. Reagents

A stock standard solution (1.0×10^{-3} mol l^{-1}) of GFLX (96.0%, National Institute for the Control of Pharmaceutical and Biological Products) was prepared by dissolving 0.0391 g of GFLX in and diluting to 100 ml with water. The solution was kept at 4 °C in the refrigerator and protected from light. Working standard solutions were obtained by making appropriate dilutions of the stock standard solution with water.

Stock standard solution of Tb^{3+} (2.0×10^{-2} mol l^{-1}) was prepared by dissolving 0.3739 g Tb_4O_7 (purity, 99.99%) in 1:1 HCl and evaporating the solution to almost dryness before diluting to 100 ml with water. The stock standard solution was kept in the refrigerator at 4 °C. The working standard solutions were prepared by making appropriate dilutions with water.

An SDBS solution (2.0×10^{-3} mol l^{-1}) was prepared by dissolving 0.0774 g SDBS (90.0%) in and then diluting to 100 ml with water.

A Tris–HCl buffer solution (pH 7.0, 0.1 mol l^{-1}) was prepared by dissolving 3.0437 g Tris(trihydroxymethyl aminomethane) in 250 ml water and then adjusting pH to 7.0 with 1:1 HCl.

All reagents used were analytical grade unless otherwise indicated. Redistilled water made in-house was used throughout the study.

2.3. Methods

To a 10 ml volumetric flask, various solutions were added according to the following order: 1.0 ml of 2.0×10^{-3} mol l^{-1} Tb^{3+} solution, an aliquot of GFLX working standard solution, 1.0 ml of 0.1 mol l^{-1} Tris–HCl buffer solution, and 1.0 ml of 2.0×10^{-3} mol l^{-1} SDBS. The resultant mixture was diluted to the volume with water and mixed thoroughly by shaking, and then allowed to stand for 15 min at room temperature. Final gatifloxacin concentrations were in the range of 5.0×10^{-10} to 5.0×10^{-8} mol l^{-1}. The fluorescence intensity was measured in a 10 mm path-length quartz cell with excitation and emission wavelengths of 331 and 547 nm, respectively.

2.4. Serum and urine sample preparation

Each 1.0 ml of serum sample was deproteinized with 2.0 ml of acetonitrile, followed by vortexing for 5 min and centrifuging for 5 min at $1500 \times g$ [23]. No sample pretreatment was required for urine samples. Further dilutions with water were made to ensure that the concentrations of the drug in the sample solutions fall within the linear range of the method.

3. Results and discussion

3.1. Spectral characteristics

The excitation and emission spectra of GFLX (1, 1′), Tb^{3+} (2, 2′), GFLX–Tb^{3+} (3, 3′), Tb^{3+}–SDBS (4, 4′), and GFLX–Tb^{3+}–SDBS (4, 4′) system are shown in Fig. 2. The excitation spectrum of the GFLX–Tb^{3+}–SDBS system exhibited an excitation peak at 331 nm, which was selected as the excitation wavelength. The emission spectra obtained showed that neither GFLX aqueous solution (curve 1′) nor Tb^{3+} aqueous solution (curve 2′) gave a strong fluorescence peak at 547 nm. The characteristic fluorescence of Tb^{3+} cannot be observed. Curve 3′ was the emission spectrum of GFLX–Tb^{3+} system. It can be seen that the native fluorescence of GFLX at 445 nm decreased significantly in comparison with GFLX aqueous solution (curve 1′), while an emission peak was observed at 547 nm, which indicated that Tb^{3+} could interact with GFLX and resulted in the

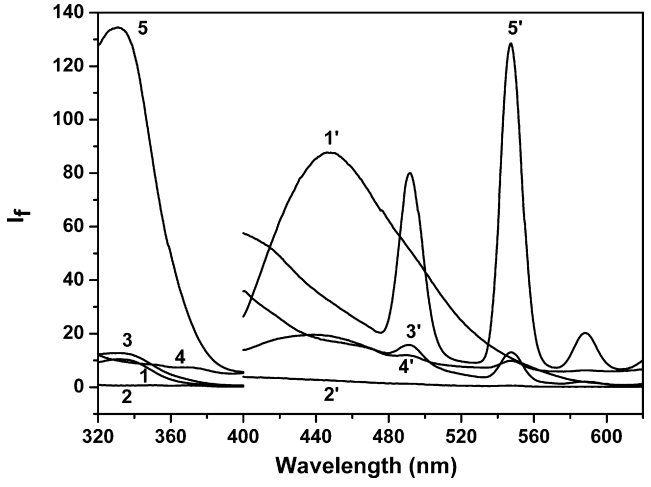

Fig. 2. Excitation spectra and emission spectra. 1, 1′ GFLX; 2, 2′ Tb^{3+}; 3, 3′ GFLX–Tb^{3+}; 4, 4′ Tb^{3+}–SDBS; 5, 5′ GFLX–Tb^{3+}–SDBS. Experimental conditions: GFLX, 1.0×10^{-6} mol l^{-1}; Tb^{3+}, 2.0×10^{-4} mol l^{-1}; SDBS, 2.0×10^{-4} mol l^{-1}; Tris–HCl, 0.01 mol l^{-1}, pH 7.0.

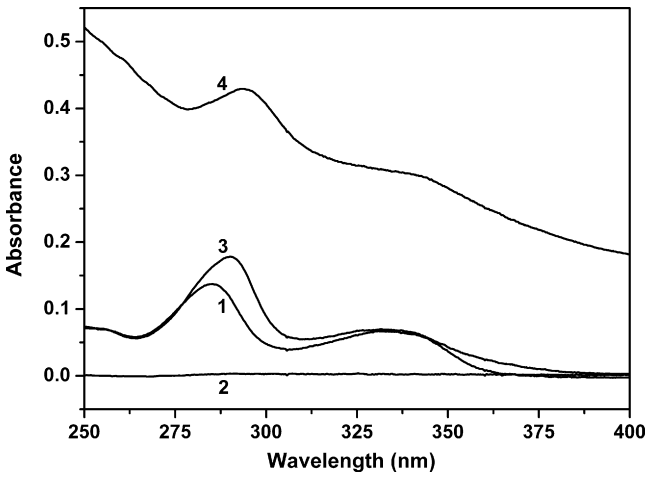

Fig. 3. Ultraviolet absorption spectra. Experimental conditions: GFLX, 1.0×10^{-5} mol l^{-1}; Tb^{3+}, 4.0×10^{-4} mol l^{-1}; SDBS, 2.0×10^{-4} mol l^{-1}; Tris–HCl, 0.01 mol l^{-1}, pH 7.0. (1) GFLX; (2) Tb^{3+}; (3) GFLX–Tb^{3+}; and (4) GFLX–Tb^{3+}–SDBS.

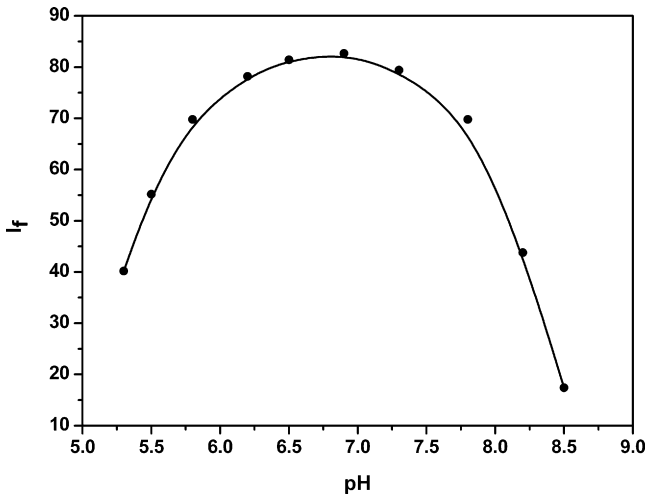

Fig. 4. Effect of pH. Experimental conditions: GFLX, 5.0×10^{-6} mol l^{-1}; Tb^{3+}, 2.0×10^{-4} mol l^{-1}; SDBS, 2.0×10^{-4} mol l^{-1}.

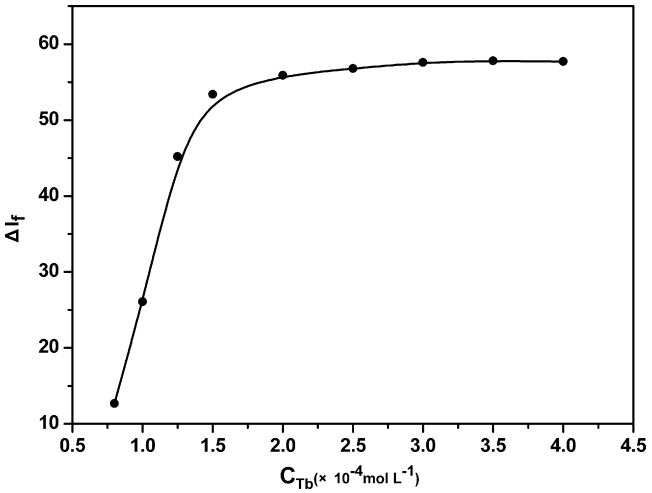

Fig. 5. Effect of Tb^{3+} concentration. Experimental conditions: GFLX, 5.0×10^{-6} mol l^{-1}; SDBS, 2.0×10^{-4} mol l^{-1}; Tris–HCl, 0.01 mol l^{-1}, pH 7.0.

energy transfer from the GFLX to Tb^{3+}. Weak characteristic fluorescence of Tb^{3+} was observed in the Tb^{3+}–SDBS system (curve 4′), indicating that SDBS could also interact with Tb^{3+} and emit the characteristic fluorescence of Tb^{3+} as a result of energy transfer from SDBS to Tb^{3+}. Curve 5′ was an emission spectrum of the GFLX–Tb^{3+}–SDBS system with an excitation wavelength of 331 nm. Strong emission peaks were observed at 491, 547, and 588 nm, which corresponded to the Tb^{3+} transitions of $^5D_4$–$^7F_6$, $^5D_4$–$^7F_5$ and $^5D_4$–$^7F_4$, respectively, while the native fluorescence peak of GFLX at 445 nm was absent. Among these emission peaks the 547 nm one displayed the strongest signal. Accordingly, 331 and 547 nm were selected as the excitation wavelength and the emission wavelength, respectively, for further study.

In order to study the interaction of GFLX, Tb^{3+}, and SDBS, several ultraviolet absorption spectra were recorded and are shown in Fig. 3. It could be seen in Fig. 3 that there were two absorption peaks of GFLX at 285 nm and 331 nm. With the addition of Tb^{3+}, the 285 nm absorption peak of the GFLX shifted to 290 nm while the fluorescence intensity increased. When both Tb^{3+} and SDBS solution was added, the absorbance of GFLX increased significantly, corresponding to a fluorescence enhancement of the fluorescence excitation spectrum of the GFLX–Tb^{3+}–SDBS system (see Fig. 2). A red shift of the maximum absorption wavelength from 290 to 294 nm indicated that a GFLX–Tb^{3+}–SDBS complex was formed [26].

3.2. Effect of pH and buffers

The effect of pH values on the fluorescence emission of a 5.0×10^{-6} mol l^{-1} GFLX solution is shown in Fig. 4. The data indicated that the signal exhibited a maximum around pH 7.0, which was chosen for further experiments. The effect of the following buffer solution media on the fluorescence intensity was then examined: NH_4Ac–HAc, NaAc–HAc, NH_4Cl–$NH_3 \cdot H_2O$, Tris–HCl, and KH_2PO_4–NaOH. It was found that 0.01 mol l^{-1} Tris–HCl offered the highest sensitivity. Therefore, a 0.01 mol l^{-1} Tris–HCl buffer solution of pH 7.0 was selected for the study.

3.3. Effect of terbium concentration

With a fixed GFLX concentration of 5.0×10^{-6} mol l^{-1}, the effect of Tb^{3+} concentration on the fluorescence intensity of the system was studied. The results are shown in Fig. 5. Fluorescence intensity reached a plateau with a Tb^{3+} concentration of 1.5×10^{-4} mol L^{-1}. Therefore, the Tb^{3+} concentration of 2.0×10^{-4} mol L^{-1} was selected for further experiments.

3.4. Effect of surfactants

The fluorimetric properties of GFLX were studied in a series of micellar media by preparing 5.0×10^{-6} mol L^{-1} GFLX in the presence of various concentrations of the surfactants CTAB (cationic), SDS (anionic), SDBS (anionic), GA (non-ionic), OP (non-ionic), and β-CD (non-ionic). The results are summarized in Table 1. It can be seen that no significant fluorescence enhancement was observed in

Table 1
Effect of different surfactants.

Surfactants and the optimal concentration	Fluorescence intensity
None	10.5
Cetrimonium bromide (CTAB), 3.0×10^{-5} mol l^{-1}	10.2
Arabic gum (GA), 1.5×10^{-3} g ml^{-1}	28.5
Polyoxyethylene nonylphenol ether (OP), 2.5% (v/v)	25.1
β-Cyclodextrin (β-CD), 5.0×10^{-4} mol l^{-1}	10.8
Sodium dodecyl sulfate (SDS), 5.0×10^{-4} mol l^{-1}	79.2
Sodium dodecylbenzene sulfonate (SDBS), 2.0×10^{-4} mol l^{-1}	111.4

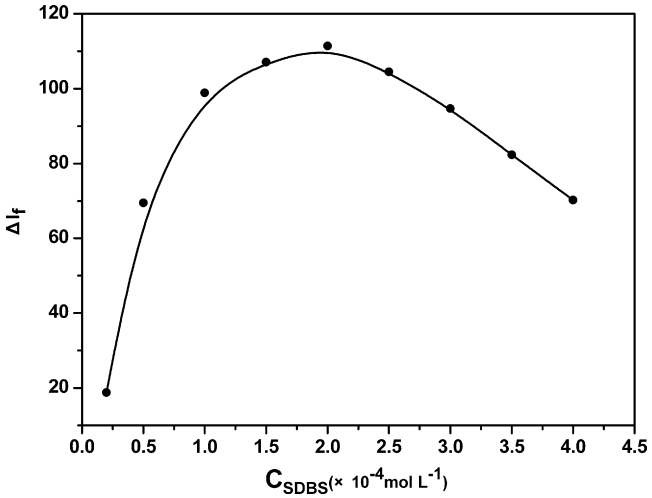

Fig. 6. Effect of SDBS concentration. Experimental conditions: GFLX, 5.0×10^{-6} mol l^{-1}; Tb^{3+}, 2.0×10^{-4} mol l^{-1}; Tris–HCl, 0.01 mol l^{-1}, pH 7.0.

the presence of either cationic or non-ionic surfactants. However, a significant increase in fluorescence was observed in the presence of anionic surfactants SDS and SDBS with the latter appeared to be more effective. The effect of the SDBS concentration on the fluorescence intensity was also determined (Fig. 6). Maximum fluorescence intensity was observed for an SDBS concentration of 2.0×10^{-4} mol l^{-1}, which was chosen for further experiments.

The mechanism of SDBS effect was studied by investigating the change of surface tension of the fluorescence system with an increase of the SDBS concentration, as shown in Fig. 7. It can be seen that the surface tension first decreased sharply, then an equilibrium was reached at the SDBS concentration of 5.0×10^{-5} mol l^{-1}, which may be considered as the apparent critical micelle concentration (CMC) of SDBS in this system. Since the SDBS concentration selected for the study (2.0×10^{-4} mol l^{-1}) was well above the CMC, it can be concluded that the formation of micelles had a great impact on the enhancement of the fluorescence intensity of the system.

3.5. Effect of reagent addition order and fluorescence stability

The effect of the order of reagent addition on the fluorescence intensity was studied. The result showed that an addition order of Tb^{3+}, GFLX, Tris–HCl, SDBS, offered the optimal performance, therefore, was chosen for further research. The experiments also showed that the fluorescence intensity of the system reached its maximum in 15 min after all the reagents had been added and remained stable for at least 3 h.

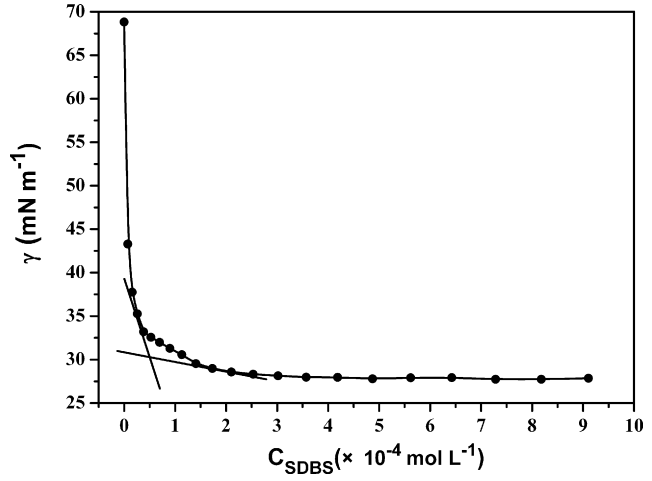

Fig. 7. Effect to the solution surface tension with the addition of surfactant SDBS. Experimental conditions: GFLX, 5.0×10^{-6} mol l^{-1}; Tb^{3+}, 2.0×10^{-4} mol l^{-1}; Tris–HCl, 0.01 mol l^{-1}, pH 7.0.

Table 2
Maximum permissible concentrations of interference species.

Interference species	Maximum permissible concentration (mol l^{-1})	Change of ΔI (%)
Cu^{2+}, SO_4^{2-}	4.0×10^{-7}	5.0
Vitamin C	7.0×10^{-7}	4.5
Fe^{3+}, Cl^-	1.5×10^{-6}	4.6
L-Methionine	7.0×10^{-6}	−4.5
Zn^{2+}, SO_4^{2-}	2.0×10^{-5}	−4.4
Al^{3+}, Cl^-	6.0×10^{-5}	3.8
Mg^{2+}, Cl^-	1.0×10^{-4}	−4.2
Ca^{2+}, Cl^-	1.0×10^{-4}	−4.7
L-Histidine	1.0×10^{-4}	−4.3
Na^+, Cl^-	1.5×10^{-4}	−4.2
L-Glutamine	2.0×10^{-4}	−5.0
K^+, Cl^-	4.0×10^{-4}	−4.9
L-Cysteine	5.0×10^{-4}	−4.5
Glycine	7.0×10^{-4}	−4.8
NH_4^+, Cl^-	1.0×10^{-3}	−4.7
Glucose	1.0×10^{-2}	−4.3

3.6. Effect of interfering species

In order to assess the applicability of the proposed method in biological samples, the effects of the potentially interfering species such as metal ions, sugars, and amino acids were examined. The results are summarized in Table 2. A 5% variation in intensity as a result of interference is considered acceptable. The results indicated that most species except for Cu^{2+}, Fe^{3+} and vitamin C have little effect on the fluorescence intensity of the system.

3.7. Analytical application

3.7.1. Calibration curve and detection limit

Under optimal conditions, the enhanced fluorescence intensity of the system (ΔI_f) showed an excellent linear relationship over the GFLX concentration range of 5.0×10^{-10} to 5.0×10^{-8} mol l^{-1}. The linear equation was $\Delta I_f = 6.35 + 1.97 \times 10^{10} C_{GFLX}$, with a correlation coefficient of 0.9996. The detection limit (3σ) was 6.0×10^{-11} mol l^{-1}.

In comparison with other methods reported, as shown in Table 3, the rapid and simple method proposed in this paper offers higher sensitivity and a wider linear range.

3.7.2. Recovery tests

GFLX is present in human serum and urine samples at the levels of approximately 2–5 and 20–100 mg l^{-1}, respectively [2] (viz. in serum: 5.3×10^{-6} to 1.3×10^{-5} mol l^{-1}; in urine: 5.3×10^{-5} to 2.7×10^{-4} mol l^{-1}). In order to make the analyte concentrations fall within the linear range of the method, urine samples were diluted 10,000-fold in water and then analyzed by standard addition method. Serum samples were deproteinized first and then diluted 1000-fold prior to analysis. The results of spiked urine and serum samples are shown in Table 4. All recoveries were in the range of 95.3–103.2%.

3.7.3. Pharmaceutical sample analysis

Three batches of gatifloxacin sodium chloride injection (Xi'an Wanlong Pharmaceutical Group Co. Ltd., China) were also analyzed using the proposed procedure. The results are shown in Table 5. The potencies of the products determined by the presented method were in good agreement with the label claims.

Table 3
Comparison of methods for the determination of GFLX.

Method	Detection limit (mol l^{-1})	Linear range (mol l^{-1})	Application	References
HPLC		2.7×10^{-7} to 2.7×10^{-5} (in human serum) 2.7×10^{-6} to 4.0×10^{-4} (in human urine)	Human serum, human urine	[3]
HPLC	3.2×10^{-7}	4.3×10^{-7} to 1.3×10^{-5}	Human serum	[4]
HPLC		2.7×10^{-7} to 2.7×10^{-5} (UV detection) 5.3×10^{-8} to 1.3×10^{-5} (fluorescence detection)	Human plasma	[5]
HPLC	3.5×10^{-7}	1.1×10^{-5} to 6.4×10^{-5}	Tablet	[6]
HPLC		2.7×10^{-7} to 1.6×10^{-5}	Human plasma	[7]
HPLC		1.1×10^{-5} to 3.7×10^{-5}	Tablet	[8]
HPLC/ESI-MS/MS	1.3×10^{-9}	2.7×10^{-8} to 2.7×10^{-6}	Human serum	[9]
UV	2.6×10^{-7}	2.7×10^{-6} to 3.8×10^{-5}	Tablet, injection, ophthalmic solution	[10]
UV	6.2×10^{-7}	5.3×10^{-6} to 4.3×10^{-5}	Tablet	[11]
UV		1.1×10^{-5} to 3.7×10^{-5}	Tablet	[12]
HP-TLC	2.7 ng spot^{-1}	400–1200 ng spot^{-1}	Bulk drug, polymeric nanoparticles	[13]
HP-TLC	40 ng spot^{-1}	100–500 ng spot^{-1}	Tablet	[14]
Microbiological assay		1.1×10^{-5} to 4.3×10^{-5}	Tablet	[15]
Voltammetric method	2.0×10^{-8}	8.0×10^{-9} to 4.0×10^{-7}	Tablet, human urine	[16]
Atomic absorption spectrometry	4.5×10^{-6}	2.7×10^{-5} to 4.0×10^{-4}	Tablet	[17]
Spectrofluorimetry	5.3×10^{-9}	5.3×10^{-8} to 1.2×10^{-6}	Human serum, human urine	[18]
Yttrium fluorescence probe spectrofluorimetry	9.0×10^{-9}	1.1×10^{-7} to 2.7×10^{-6}	Capsule, human serum, human urine	[19]
Europium-sensitized spectrofluorimetry	1.0×10^{-9}	1.0×10^{-8} to 8.0×10^{-7}	Injection, human serum, human urine	[20]
Terbium-sensitized spectrofluorimetry	6.0×10^{-11}	5.0×10^{-10} to 5.0×10^{-8}	Injection, human serum, human urine	Present paper

Table 4
Recovery of GFLX in spiked human urine and serum samples ($n = 5$).

Sample	Added (mol l^{-1})	Found (mol l^{-1})	Recovery ± R.S.D. (%)
Urine 1	1.00×10^{-9}	0.953×10^{-9}	95.3 ± 2.5
Urine 1	1.00×10^{-8}	1.03×10^{-8}	103.2 ± 1.8
Serum 1	1.00×10^{-9}	0.980×10^{-9}	98.0 ± 2.1
Serum 2	1.00×10^{-8}	1.01×10^{-8}	100.6 ± 2.6
Serum 3	3.00×10^{-8}	2.93×10^{-8}	97.8 ± 1.9

Table 5
Pharmaceutical sample analysis ($n = 5$).

Batch no.	Mark concentration (mg/100 ml)	Found (mg/100 ml)	Average (mg/100 ml)	R.S.D. (%)
070603-3	200.0	196.3, 198.6, 198.4, 200.8, 201.7	199.2	2.1
070604-3	200.0	197.4, 195.1, 197.9, 199.2, 201.3	198.2	2.3
070605-1	200.0	202.3, 203.2, 202.1, 198.9, 200.4	201.4	1.7

3.8. Interaction mechanism

3.8.1. Luminescence of Tb^{3+}

In the GFLX–Tb^{3+}–SDBS system, GFLX was excited to its excited singlet state after absorbing light energy, and then changed to its triplet state via an inter-system crossing. Subsequently, GFLX can transfer its energy to the level $^5D_4$ of Tb (III) via intra-molecular energy transfer, resulting in the observation of the characteristic fluorescence of Tb (III). This luminescence mechanism was also called the Antenna Effect [27].

3.8.2. The fluorescence enhancement of GFLX–Tb^{3+}–SDBS system

Fig. 2 clearly shows that SDBS can greatly enhance the fluorescence intensity of GFLX–Tb^{3+} complexes. Furthermore, it can be seen that the absorption peak of GFLX shifted to longer wavelength, as a result of the change in microenvironment of the system. This change can also be demonstrated by the variation in polarity and fluorescence polarization of the system as shown in Table 6.

The ratio of the first to the third fluorescence bands of pyrene monomer (I_1/I_3) is a well-established parameter which reflects the

Table 6
Comparison of the polarity and fluorescence polarization of the systems.

System	GFLX–Tb^{3+}	GFLX–Tb^{3+}–SDBS
I_1/I_3	1.721	1.327
P	0.163	0.332

polarity variation of a system experienced by the pyrene probe with a lower value as an indication of a lower polar environment [28,29].

The microviscosity of the microenvironment can be estimated using the fluorescence polarization of fluorescence probe according to Perrin equation [30]. A higher value of polarization reflects a higher microviscosity.

Table 6 illustrates that, with the addition of SDBS to the GFLX–Tb^{3+} system, the value of I_1/I_3 decreased and the value of P increased. Therefore, it can be concluded that SDBS medium could provide an optimal hydrophobic environment with low polarity and high viscosity for GFLX–Tb^{3+} complexes and contribute to the fluorescence enhancement of the complexes. In addition, the hydrophobic environment provided by SDBS can also prevent the collision of the complex and water and reduce the energy loss of the GFLX–Tb^{3+}–SDBS system. Consequently, the fluorescence quantum yield was improved, resulting in the significant enhancement of the fluorescence intensity.

4. Conclusion

This paper describes a new fluorimetric method for the determination of GFLX. Under optimal conditions, the enhanced fluorescence intensity was in proportion to the GFLX concentration over the range of 5.0×10^{-10} to 5.0×10^{-8} mol l^{-1}. The detection limit (3σ) was 6.0×10^{-11} mol l^{-1}. In comparison with most of other methods reported, the proposed method is rapid and simple, and has higher sensitivity, a wider linear range, and better reproducibility. The interaction mechanism is also discussed. It is demonstrated that SDBS could provide a hydrophobic environment with low polarity and high viscosity, resulting in the fluorescence enhancement of the GFLX–Tb^{3+} complex. The proposed method has been applied to the determination of GFLX in pharmaceuticals and human urine/serum samples with satisfactory results. Moreover, due to its ultrahigh sensitivity, this method would also be useful for the trace analysis of GFLX in food or environment.

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