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# Analysis of tryptophan at nmol $l^{-1}$ level based on the fluorescence enhancement of terbium–gadolinium–tryptophan–sodium dodecyl benzene sulfonate system

Shufang Liu [a], Jinghe Yang [a,*], Xia Wu [a], Benyu Su [b], Changxia Sun [a], Feng Wang [c]

[a] *Key Laboratory of Colloid and Interface Chemistry, School of Chemistry and Chemical Engineering, Shandong University, Ministry of Education, Jinan 250100, PR China*
[b] *Shandong Supervision and Inspection Institute for Product Quality, Jinan 250100, PR China*
[c] *Department of Chemistry, Zaozhuang Teacher's College, Zaozhuang 277160, PR China*

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**Abstract**

It is found that $Tb^{3+}$ can react with tryptophan (Trp) and sodium dodecyl benzene sulfonate (SDBS), and emits the intrinsic fluoresence of $Tb^{3+}$. The fluorescence intensity can be enhanced by $La^{3+}$, $Gd^{3+}$, $Lu^{3+}$, $Sc^{3+}$ and $Y^{3+}$, among which $Gd^{3+}$ has the greatest enhancement. This is a new co-luminescence system. The studies indicate that in the Tb–Gd–Trp–SDBS system, there is both Tb–Trp–SDBS and Gd–Trp–SDBS complexes, and they aggregate together and form a large congeries. The fluorescence enhancement of the Tb–Gd–Trp–SDBS system is considered to originate from intramolecular and intermolecular energy transfers, and the energy-insulating sheath effect of Gd–Trp–SDBS complex.

Under the optimum conditions, the enhanced intensity of fluorescence is in proportion to the concentration of Trp in the range from $4 \times 10^{-8}$ to $4 \times 10^{-5}$ mol $l^{-1}$. The detection limit is $10^{-9}$ mol $l^{-1}$. The proposed method is one of the most sensitive fluoremetries of Trp.


*Keywords:* Energy transfers; Fluorescence enhancement; Tryptophan; Determination; Rare earth ion

## 1. Introduction

Tryptophan (Trp) is among the most reactive amino acids which forms proteins in life systems and it is one of the important factors for the evaluation of food nourishment. So, to find a simple, quick and sensitive method of Trp is becoming more and more important. Many methods have been reported to determine Trp such as chemiluminescence [1], capillary electrophoresis [2], fluorescence [3,4], high-performance liquid chromatography with fluorescence detection [5] and so on. But the sensitivity of most of these methods is not high.

In present paper, a new co-luminescence system of Tb–Gd–Trp–sodium dodecyl benzene sulfonate (SDBS) is found and studied. The experiments indicate that the fluorescence intensity is in proportion to the concentration of Trp, the detection limits is $10^{-9}$ mol $l^{-1}$. Compared with most fluorescence methods reported, this method is more sensitive and stable. The reaction mechanism has been investigated.

## 2. Experimental

### 2.1. Chemicals

Stock standard solutions (0.01 mol $l^{-1}$) of rare earth ions were prepared by dissolving the corresponding oxides (Yuelong Chemical Co., Shanghai, 99.9%) in hydrochloric acid and diluting with water.

SDBS solution ($1.00 \times 10^{-2}$ mol $l^{-1}$) is prepared by dissolving 0.8712 g SDBS (Chemical Co. of China, Shanghai) in 250 ml volumetric flask with water.

The Tris–HCl buffer was made by dissolving 12.5 g of Tris in 500 ml water. The pH was adjusted to 10.10 with 6 mol $l^{-1}$ HCl.

* Corresponding author. Tel.: +86-531-8365459; fax: +86-531-8564464.
*E-mail address:* yjh@sdu.edu.cn (J. Yang).

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The Trp solution ($1.00 \times 10^{-3}$ mol l$^{-1}$) was prepared by dissolving 0.0204 g Trp (Bio Basic Inc., Canada) in 100 ml volumetric flask with water. This solution needed to be stored at 0–4 °C.

All the chemicals used are of analytical grade and doubly distilled water was used throughout.

### 2.2. Apparatus

All fluorescence measurements were made with FL-4500 spectrofluorimeter (Hitachi). All absorption spectra were recorded with an UV-240 spectrophotometer (Shimadzu). All pH measurements were made with a Delta 320-S pH meter (Mettler Toledo). The $^1$H NMR spectra are recorded with a Bruker AVANCE-600 spectrometer (600 MHz) with a ultra low temperature probe in $D_2O$ as solvent.

### 2.3. Procedure

To a 25 ml test tube, solutions were added in the following order: $Tb^{3+}$, $Gd^{3+}$, Trp, SDBS, and Tris–HCl buffer. The mixture was diluted to 10 ml with distilled water. The fluorescence intensity was measured in a 1 cm quartz cell, the excitation and emission slits were both 10 nm.

## 3. Result and discussion

### 3.1. Fluorescence spectra

Emission spectra of Tb (1), Tb–Trp (2), Tb–SDBS (3), Tb–Trp–SDBS (4) and Tb–Gd–Trp–SDBS (5) systems at excitation wavelength of 290 nm are shown in Fig. 1. From this figure, it can be seen that the systems of (2)–(5) have the same emission peaks at 490 and 545 nm of $Tb^{3+}$, corresponding to the $^5D_4$–$^7F_6$ and $^5D_4$–$^7F_5$ transitions of $Tb^{3+}$, respectively. The fluorescence intensity at 545 nm is the strongest. The fluorescence spectrum of Tb–Trp–SDBS system is similar to that of Tb–Gd–Trp–SDBS system, but the fluorescence intensity of the system is much enhanced when $Gd^{3+}$ is added to the Tb–Trp–SDBS system, this is a newly found co-luminescence system. So, we choose 545 nm as the emission peak in this research.

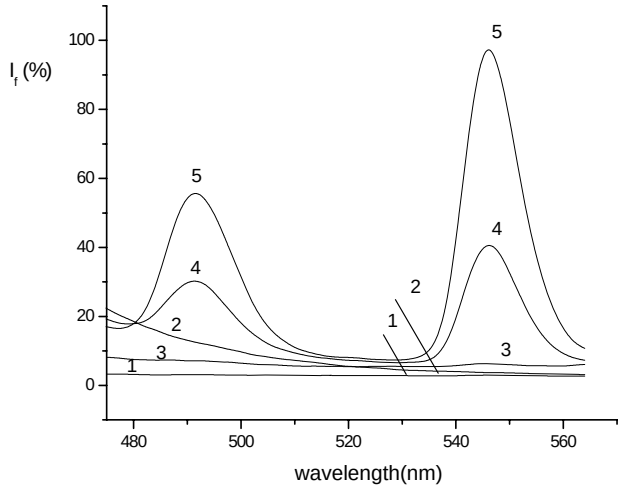



Fig. 1. Emission spectra: (1) Tb; (2) Tb–Trp; (3) Tb–SDBS, (4) Tb–SDBS–Trp; (5) Tb–Gd–SDBS–Trp. Conditions: $Tb^{3+}$: $1.3 \times 10^{-4}$ mol l$^{-1}$; $Gd^{3+}$: $1.25 \times 10^{-4}$ mol l$^{-1}$; SDBS: $3 \times 10^{-4}$ mol l$^{-1}$; Trp: $1.0 \times 10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

Table 1
Choice of enhancing ions for the co-luminescence

| | Gd | La | Lu | Sc | Y | Sm | Ho | Nd | Pr | Eu | Dy | Tm |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| $\Delta I_f$ (%) | 100 | 93 | 23 | 90 | 92 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

### 3.2. Choice of enhancing rare earth ions

The effects of other rare earth ions ($1.25 \times 10^{-4}$ mol l$^{-1}$) on the fluorescence intensity of Tb–SDBS–Trp system are tested and shown in Table 1. The results show that $La^{3+}$, $Gd^{3+}$, $Lu^{3+}$, $Sc^{3+}$ and $Y^{3+}$ can increase the fluorescence intensity of the system, among which $Gd^{3+}$ has the greatest enhancement. So, $Gd^{3+}$ is chosen as the enhancing ion for further research.

### 3.3. Effect of pH

The effect of pH on the fluorescence intensity of this system is shown in Fig. 2. It can be seen that the fluorescence intensity reaches the maximum at pH 10.10. Different buffers are also tested, such as HMTA, Tris–HCl, $Na_2B_4O_7$–HCl, $NH_4Cl$–$NH_3$, NaAc–HAc, $Na_2B_4O_7$–$H_3BO_3$. The results indicate that 2.0 ml of Tris–HCl is the most suitable buffer.

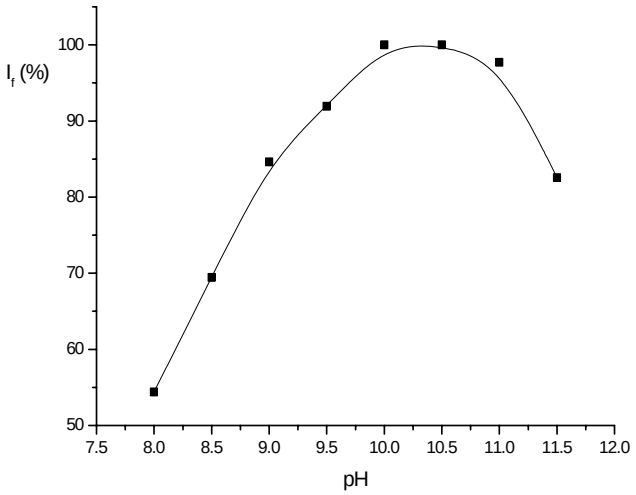



Fig. 2. Effect of pH. Conditions: $Tb^{3+}$: $1.3 \times 10^{-4}$ mol l$^{-1}$; $Gd^{3+}$: $1.25 \times 10^{-4}$ mol l$^{-1}$; SDBS: $3 \times 10^{-4}$ mol l$^{-1}$; Trp: $10^{-5}$ mol l$^{-1}$.

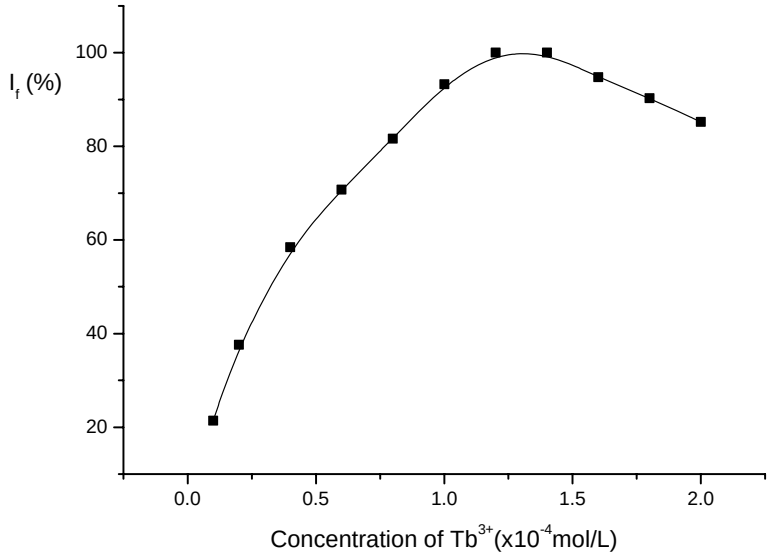



Fig. 3. Effect of the concentration of $Tb^{3+}$. Conditions: $Gd^{3+}$: $1.25 \times 10^{-4}$ mol l$^{-1}$; SDBS: $3 \times 10^{-4}$ mol l$^{-1}$; Trp: $1.0 \times 10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

### 3.4. Effect of $Tb^{3+}$ concentration

The effect of the concentration of $Tb^{3+}$ was tested as shown in Fig. 3. The fluorescence intensity of the system reached maximum and remained constant when the concentration of $Tb^{3+}$ is $1.3 \times 10^{-4}$ mol l$^{-1}$. So, $1.3 \times 10^{-4}$ mol l$^{-1}$ was chosen in the research.

### 3.5. Effect of $Gd^{3+}$ concentration

With the change in the concentration of $Gd^{3+}$, the corresponding change of fluorescence intensity of this system is shown in Fig. 4. It can be seen that the concentration of $Tb^{3+}$ is in the range of $3.0 \times 10^{-5}$ to $1.3 \times 10^{-4}$ mol l$^{-1}$, the tendency of the effect of $Gd^{3+}$ concentration on the fluorescence intensity of the system is the same with a maximum at $1.25 \times 10^{-4}$ mol l$^{-1}$. This indicates that there is no constant molar ratio between $Tb^{3+}$ and $Gd^{3+}$. So, in this paper, the concentration of $Gd^{3+}$ for study is chosen as $1.25 \times 10^{-4}$ mol l$^{-1}$.

### 3.6. Effect of SDBS concentration

The effects of different surfactants were studied and shown in Table 2. It indicated that the fluorescence intensity was the strongest when using SDBS as the surfactant.

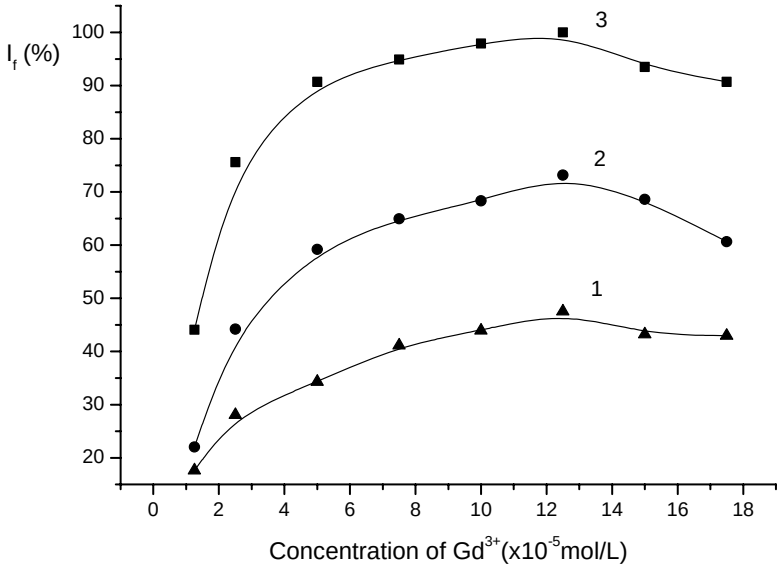



Fig. 4. Effect of the concentration of $Gd^{3+}$. Conditions: $Tb^{3+}$: $3 \times 10^{-5}$ mol l$^{-1}$ (1); $6 \times 10^{-5}$ mol l$^{-1}$ (2); $1.3 \times 10^{-4}$ mol l$^{-1}$ (3); SDBS: $3 \times 10^{-4}$ mol l$^{-1}$; Trp: $1.0 \times 10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

Table 2
Effects of different surfactants

| | SDS | SDBS | CPB | Trixon 100 | SLS | CTMAB |
|---|---|---|---|---|---|---|
| $\Delta I_f$ (%) | 37.28 | 100 | 4.12 | 6.85 | 68.85 | 6.11 |

The effect of SDBS concentration was studied and shown in Fig. 5. It is found that the fluorescence intensity is the strongest when SDBS concentration is $3.0 \times 10^{-4}$ mol l$^{-1}$. So, this concentration is chosen for this research.

From Fig. 6, it can be seen that the surface tension of this system first decrease sharply with the increase of SDBS concentration, soon gets to a minimum and then remains constant. The concentration $9 \times 10^{-4}$ mol l$^{-1}$ may be regarded as the apparent critical micelle concentration (CMC)

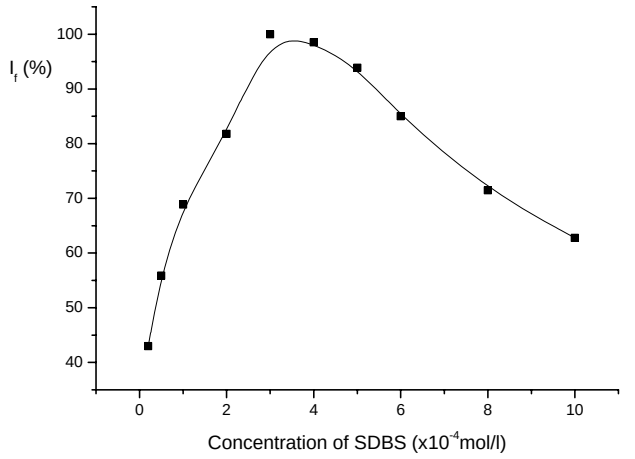



Fig. 5. Effect of SDBS concentration. Conditions: $Tb^{3+}$: $1.3 \times 10^{-4}$ mol l$^{-1}$; $Gd^{3+}$: $1.25 \times 10^{-4}$ mol l$^{-1}$; Trp: $1.0 \times 10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

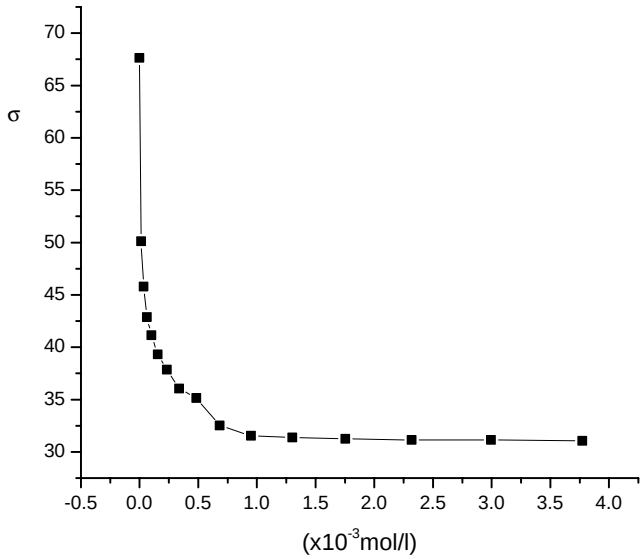



Fig. 6. Surface tension of the Tb–Trp–SDBS–Gd system. Conditions: $Tb^{3+}$: $1.3 \times 10^{-4}$ mol l$^{-1}$; $Gd^{3+}$: $1.25 \times 10^{-4}$ mol l$^{-1}$; Trp: $1.0 \times 10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

of SDBS in this system. Thus, it is possible that SDBS exists in the pre-micella or single molecule in the studied system.

### 3.7. The addition order and stability of this system

In this system, we also investigated the effect of the adding order. The result indicates that the order of $Tb^{3+}$, $Gd^{3+}$, Trp, SDBS and Tris is the best.

Under the optimum condition, the effect of time on the fluorescence intensity was studied. The result showed that the fluorescence intensity immediately reached a maximum after all the reagents had been added and remained stable for over 12 h. Therefore, this system exhibits good stability.

### 3.8. Effect of foreign substances

The interference of foreign substances was tested and shown in Table 3. It was found that most of amino acids, proteins and metal ions except $Al^{3+}$, $Mg^{2+}$, $Mn^{2+}$, $Fe^{3+}$ had little effect on the determination of Trp, under the permission of ±5% relative error.

Table 3
Effect of foreign substances

| Foreign substance | Concentration coexing ($\times 10^{-5}$ mol l$^{-1}$) | Change of $I_f$ (%) |
|---|---|---|
| KCl | 40 | −4.2 |
| $Al(NO_3)_3$ | 0.5 | −4.8 |
| $MgSO_4$ | 0.5 | −4.3 |
| $Na_2CO_3$ | 3 | −3.9 |
| $BaCl_2$ | 20 | −4.8 |
| $MnSO_4$ | 0.5 | −5.0 |
| $ZnSO_4$ | 16 | −5.0 |
| $FeCl_3$ | 0.4 | −5.0 |
| NaCl | 40 | −5.0 |
| $Fe_2(SO_4)_3$ | 0.2 | −5.0 |
| $AlCl_3$ | 6 | −3.8 |
| $NH_4Cl$ | 30 | −5.0 |
| $Na_2SO_4$ | 40 | −4.8 |
| $CaCl_2$ | 75 | −5.0 |
| DL-Thr | 15 | −4.7 |
| DL-Tyr | 8 | −5.0 |
| L-His | 32 | −4.5 |
| Lys | 25 | −5.0 |
| L-Lin | 4 | −5.0 |
| DL-Lys–HCl | 30 | −5.0 |
| L-Phe | 40.0 | −5.0 |
| L-Ala | 3 | −5.0 |
| L-Asp | 40 | −3.3 |
| Cys | 15 | −5.0 |
| L-Arg–HCl | 18 | −3.9 |
| y-RNA | 2.4 | −3.9 |
| Ct-DNA | 20 | −5.0 |
| Fs-DNA | 5 | 5.0 |
| BSA | 30 | −5.0 |
| HSA | 4 | −5.0 |
| ADP | 2 | −5.0 |
| UTP | 1 | −3.7 |
| GMP | 2 | −4.1 |

(Note: a. $1 \times 10^{-7}$ g ml$^{-1}$) Conditions: $Tb^{3+}$: $1.3 \times 10^{-4}$ mol l$^{-1}$; $Gd^{3+}$: $1.25 \times 10^{-4}$ mol l$^{-1}$; SDBS: $3 \times 10^{-4}$; Trp: $1.0 \times 10^{-6}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

Table 4
Analytical parameters of this method

| Amino acid | Linear range (mol l$^{-1}$) | Linear regression equation (mol l$^{-1}$) | $r$ | LOD (mol l$^{-1}$) |
|---|---|---|---|---|
| Trp | $4 \times 10^{-8}$ to $4.0 \times 10^{-5}$ | $I_f = 40.81512 + 3.49701 \times 10^{-7}\ C$ | 0.99975 | $1 \times 10^{-9}$ |

Table 5
Determination of Trp in actual sample

| Trp | Standard value ($\times 10^{-4}$ g ml$^{-1}$) | Determination value ($\times 10^{-4}$ g ml$^{-1}$) | Average ($\times 10^{-4}$ g ml$^{-1}$) | R.S.D. (%) |
|---|---|---|---|---|
| Proposed method | 9.0 | 8.80, 8.96, 9.22, 9.15, 9.06 | 9.01 | 0.18 |
| UV-spectrophotometric method | 9.0 | 9.30, 9.06, 9.06, 9.40, 9.22 | 9.21 | 0.15 |

## 4. Analytical applications

### 4.1. The calibration graph and detection limit

Under the optimum condition defined, the calibration graphs for Trp are obtained and showed in Table 4. It can be seen that there is a linear relationship between the fluorescence intensity of the system and the concentration of Trp in the range of $4.0 \times 10^{-8}$ to $4.0 \times 10^{-5}$ mol l$^{-1}$, and the detection limit is $10^{-9}$ mol l$^{-1}$.

### 4.2. Determination of actual sample

The standard addition method is used for the determination of Trp in compound amino acid injection (Sichuang Kelunda Pharmaceutical Ltd. Co.) and compared with the UV spectrophotometric method (Table 5). The results are shown in Table 4; it can be seen that the accuracy and precision of the method are satisfactory.

From Table 6, it can be seen that the proposed method is preferable to most of other traditional fluorimetric methods for the determination of Trp, in both sensitivity and linear range. But some of traditional fluorimetric methods can allow simultaneous determination of more metabolites in real matrices because of the use of HPLC [5,7,8,11] or capillary electrophoresis [12] and second-order derivative techniques [10]. So, the proposed method is adapt to the pure analytical research.

## 5. Interaction mechanism of the system

### 5.1. Formation of Tb–SDBS–Trp complex

It is well known that the isoelectric point of Trp is 5.98; when pH of the solution is 10.10, Trp is negatively charged. Thus, $Tb^{3+}$ can react with anionic surfactant SDBS and Trp through electrostatic force and an association complex of Tb–Trp–SDBS is formed. This can be seen from the fluorescence spectra in Fig. 1.

From Fig. 6, it can be seen that the apparent critical concentration (CMC) of SDBS in this system is $9.0 \times 10^{-4}$ mol l$^{-1}$, while the optimum SDBS concentration ($3.0 \times 10^{-4}$ mol l$^{-1}$) in this experiment is lower than its CMC. So, SDBS exists as the pre-micellar or single molecule.

The NMR measurement was introduced to investigate the interaction between SDBS and Trp and the chemical shifts of aromatic ring were shown in Table 7, it can be seen that after the addition of SDBS into Trp, the peaks corresponding to the aromatic ring of Trp moved to the upfield weakly. We consider that there is weak stacking interaction between aromatic rings of both SDBS and Trp [13–17].

The resonance light scattering (RLS) spectra of Trp(1), SDBS(2), Tb–Trp(3), Tb–SDBS(4), Tb–SDBS–Trp(5) and Tb–SDBS–Trp–Gd(6) systems are shown in Fig. 7. When $Tb^{3+}$ was added into Trp(1) and SDBS(2) systems, the resonance light scattering spectra of the two systems

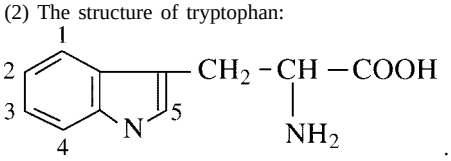
Table 6
Compared with other luminescent methods in sensitivity

| Luminescent methods (fluorigenic reagent) | LOD ($\times 10^{-9}$ mol l$^{-1}$) | Linear range (mol l$^{-1}$) | Reference |
|---|---|---|---|
| Spectrofluorimetric method (phenylglyoxal) | 20 (in HCl), 34 (in $H_3PO_4$) | $2 \times 10^{-7}$ to $1 \times 10^{-4}$ | [6] |
| HPLC-fluorimetric detection (phenylglyoxal) | 72 | – | [7] |
| Spectrofluorimetric method (fluorescamine) | 30 | 0 to $1 \times 10^{-6}$ | [5] |
| HPLC-fluorimetric detection | 100 | $4.9 \times 10^{-6}$ to $4.9 \times 10^{-4}$ | [8] |
| Electrogenerated chemiluminescence | 100 | $1 \times 10^{-7}$ to $8.4 \times 10^{-5}$ | [9] |
| Second-order derivative fluorimetric method | 11.26 | $1.96 \times 10^{-8}$ to $9.8 \times 10^{-7}$ | [10] |
| HPLC-fluorimetric detection | 42.60 | $2.45 \times 10^{-7}$ to $7.83 \times 10^{-6}$ | [11] |
| UV-pulsed laser-induced fluorescence detection | 0.15 | $2 \times 10^{-8}$ to $2.3 \times 10^{-5}$ | [12] |
| Spectrofluorimetric method (nitrous acid) | 49 | $9.8 \times 10^{-7}$ to $7.8 \times 10^{-6}$ | [3] |

Table 7
Upfield shifts for aromatic ring of tryptophan stacking

| H | H-1 | H-2 | H-3 | H-4 | H-5 |
|---|---|---|---|---|---|
| $\delta$ | 7.693665 | 7.24182 | 7.158903 | 7.49646 | 7.27545 |
| $\delta'$ | 7.683785 | 7.21948 | 7.138503 | 7.47515 | 7.25002 |
| $\Delta\delta$ | 0.00988 | 0.02234 | 0.0204 | 0.02131 | 0.02543 |

*Notes*: (1) $\delta$ and $\delta'$ are the chemical shifts of H corresponded to the aromatic ring of Trp without and with SDBS, respectively; $\Delta\delta = \delta - \delta'$. (2) The structure of tryptophan:




enhanced. This indicated that Tb–Trp and Tb–SDBS complexes formed. The resonance light scattering intensity of Tb–SDBS–Trp(5) system was higher than that of Tb–Trp(3) and Tb–SDBS(4) systems. This proved that $Tb^{3+}$ can react with Trp and SDBS, and form Tb–SDBS–Trp complex in this system.

From Fig. 7, it can be seen that the RLS intensity of the Tb–SDBS–Trp–Gd(6) system is larger than that of the Tb–SDBS–Trp(5) system. This indicates that there exists a large congeries in Tb–SDBS–Trp–Gd system. In the light of similar chemical properties of all lanthanide elements, we consider that both Tb–SDBS–Trp and Gd–SDBS–Trp ternary complexes are formed in Tb–Gd–SDBS–Trp system. Because SDBS possesses the tendency to self-assemble as pre-micella or micella, so the two complexes are in close proximity in the system.

### 5.2. The energy transfer of the systems

When $Gd^{3+}$ as enhancing rare earth ion is added to Tb–Trp–SDBS system, the intrinsic emission peaks of $Tb^{3+}$ are much enhanced. The reasons of this fluorescence enhancing effect are the following:

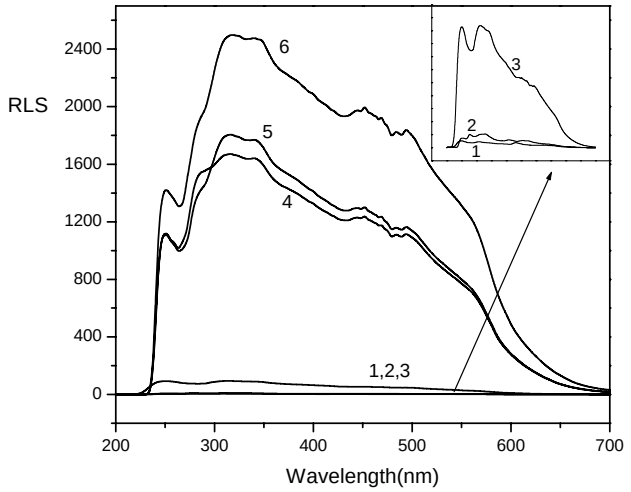



Fig. 7. The resonance light scattering (RLS) spectra: (1) Trp; (2) SDBS, (3) Tb–Trp; (4) Tb–SDBS; (5) Tb–Trp–SDBS; (6) Tb–Gd–Trp–SDBS. Conditions: $Tb^{3+}$: $1.3\times10^{-4}$ mol l$^{-1}$; $Gd^{3+}$: $1.25\times10^{-4}$ mol l$^{-1}$; SDBS: $1.5\times10^{-5}$ mol l$^{-1}$; Trp: $1.0\times10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

(1) *Intramolecular energy transfer*: It is well known that both Trp and SDBS can absorb light energy, which is transferred to $Tb^{3+}$. From Fig. 1, it can be seen that both Tb–SDBS and Tb–Trp systems have very weak fluorescence, which indicates that the energy transfer efficiencies from SDBS and Trp to $Tb^{3+}$ are very low. But after Trp is added to the Tb–SDBS system, the fluorescence intensity of the system is much higher than that of both Tb–Trp system and Tb–SDBS systems. On the basis of the results mentioned above, we consider that after the Trp–SDBS complex absorbs light energy, $Tb^{3+}$ can be excited to the $^5D_4$ levels by intramolecular energy transfer from Trp–SDBS to the $Tb^{3+}$. Emission then takes place by transitions from the $^5D_4$ level of $Tb^{3+}$ to the $^7F_6$ and $^7F_5$ levels, corresponding the emission peaks of 490 and 544 nm, respectively. In addition, it is

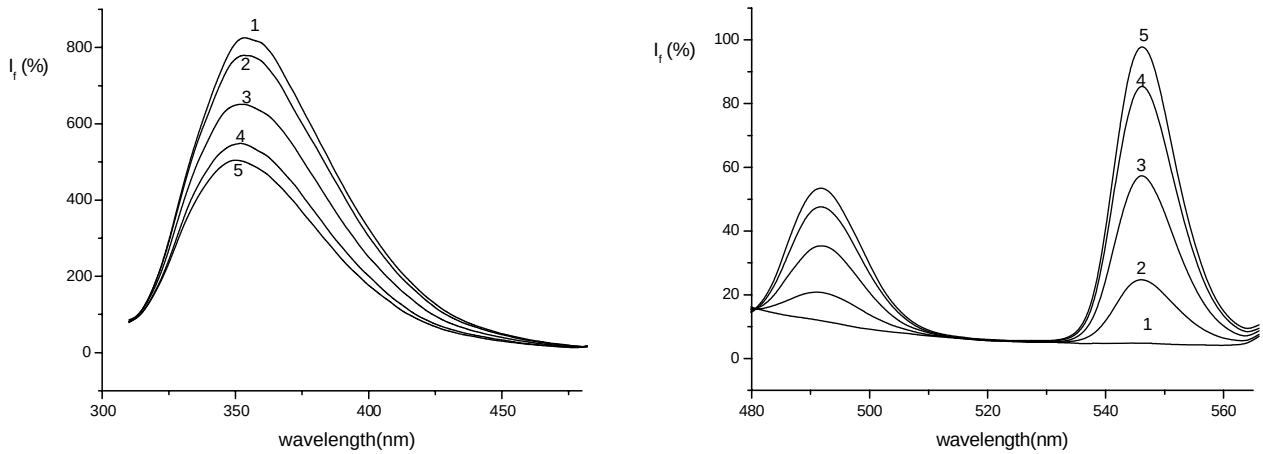



Fig. 8. Effect of concentration of $Tb^{3+}$ on the fluorescence intensity of Tb–Gd–Trp–SDBS system: (1) 0.0 mol l$^{-1}$ $Tb^{3+}$; (2) $1.0\times10^{-5}$ mol l$^{-1}$ $Tb^{3+}$; (3) $4.0\times10^{-5}$ mol l$^{-1}$ $Tb^{3+}$; (4) $8.0\times10^{-5}$ mol l$^{-1}$ $Tb^{3+}$; (5) $1.3\times10^{-4}$ mol l$^{-1}$. Conditions: $Gd^{3+}$: $1.25\times10^{-4}$ mol l$^{-1}$; SDBS: $1.5\times10^{-5}$ mol l$^{-1}$; Trp: $1.0\times10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

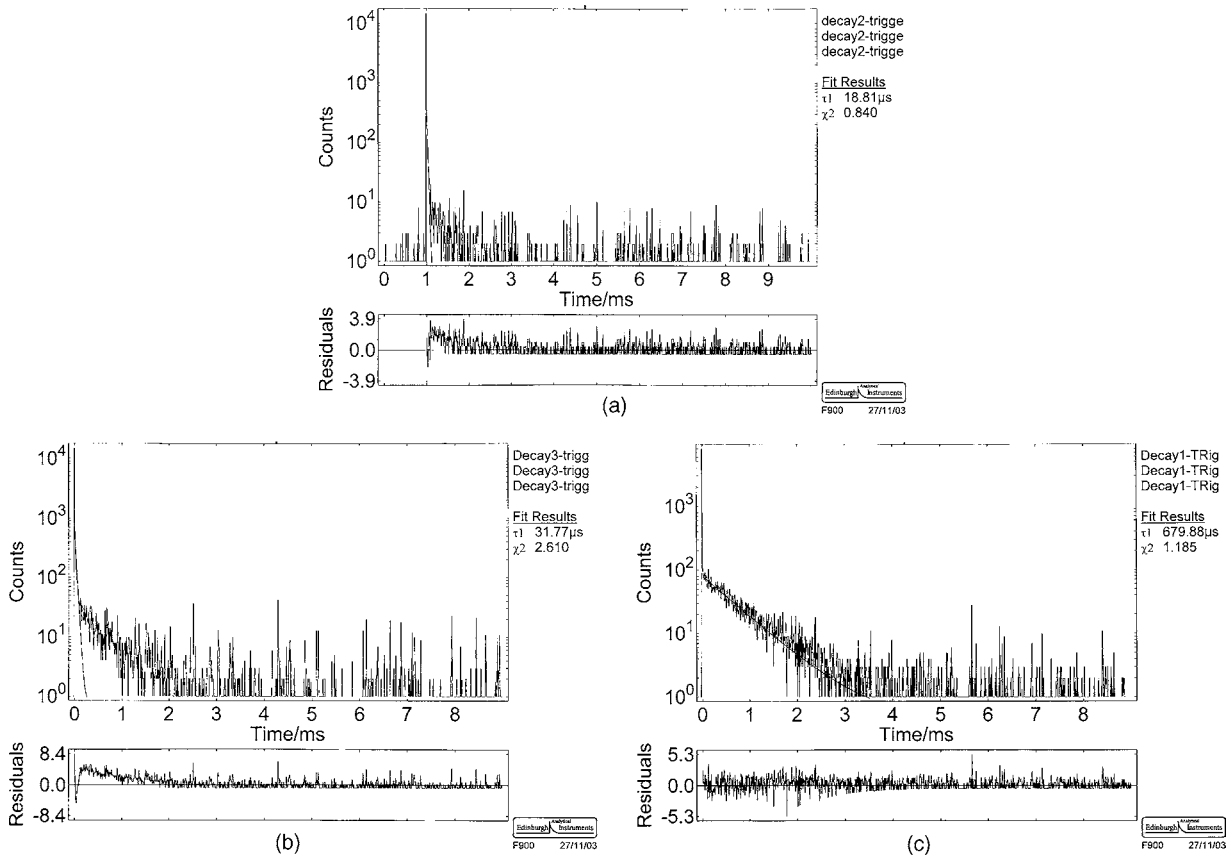



Fig. 9. The fluorescence life-time change of $Tb^{3+}$ in different systems: (a) Tb–Trp and Tb–SDBS, (b) Tb–SDBS–Trp, (c) Tb–SDBS–Trp–Gd. Conditions: $Tb^{3+}$: $1.3 \times 10^{-4}$ mol l$^{-1}$; $Gd^{3+}$: $1.25 \times 10^{-4}$ mol l$^{-1}$; SDBS: $3 \times 10^{-4}$ mol l$^{-1}$; Trp: $1.0 \times 10^{-5}$ mol l$^{-1}$; Tris–HCl buffer (pH = 10.10).

possible that the stacking interaction between aromatic rings of both Trp and SDBS can benefit the intramolecular energy transfer in Tb–Trp–SDBS system, which makes the fluorescence intensity of this system enhance.

(2) *Intermolecular energy transfer*: From Fig. 8, it can be seen that Gd–Trp–SDBS complex emits ultraviolet fluorescence at 360 nm. After continuously adding $Tb^{3+}$ to Gd–Trp–SDBS complex, the fluorescence of Gd complex is gradually weaken and that of $Tb^{3+}$ is gradually enhanced. This indicates that there has the intermolecular energy transfer between Tb and Gd complexes, the energy absorbed by Gd–Trp–SDBS complex can be transferred to $^5D_4$ of $Tb^{3+}$ in Tb–Trp–SDBS, and then enhances the fluorescent intensity of $Tb^{3+}$.

(3) *The energy-insulating sheath*: Fluorescence enhancement also originate from the energy-insulating sheath as well as the intramolecular and intermolecular energy transfers in this system. This can be proved from the fluorescence life-time change of $Tb^{3+}$ in different systems shown in Fig. 9. When Trp is added to the Tb–SDBS system, the fluorescence life-time of $Tb^{3+}$ can be delayed from 18.81 to 31.77 μs. This shows that Trp replace water molecule coordinated in $Tb^{3+}$, resulting in both the decrease of energy loss on $Tb^{3+}$ and the increase of the fluorescence life-time of $Tb^{3+}$. While $Gd^{3+}$ is added to Tb–SDBS–Trp system, the fluorescence life-time of $Tb^{3+}$ is greatly prolonged from 31.77 to 679.88 μs. This is due to the fact that in Tb–Gd–SDBS–Trp system, each terbium complex is surrounded by many gadolinium complexes, the latter can act as an energy-insulting sheath, which can prevent collision with water molecules and decrease the energy loss of $Tb^{3+}$. Therefore, the fluorescence life-time of $Tb^{3+}$ is greatly increased.

## 6. Conclusion

In this paper, a new co-luminescence system is found. In this system, the fluorescence intensity of Tb–Trp–SDBS can be enhanced by $La^{3+}$, $Gd^{3+}$, $Lu^{3+}$, $Sc^{3+}$ and $Y^{3+}$, among which $Gd^{3+}$ has the greatest enhancement. This method can be applied for the determination of Trp. The detection limits are $10^{-9}$ mol l$^{-1}$. In comparison with most fluorescence methods reported, this method has good sensitivity and stability.

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