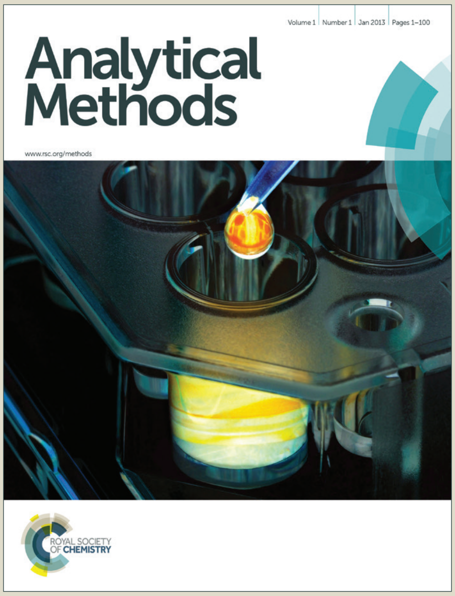
# Analytical Methods



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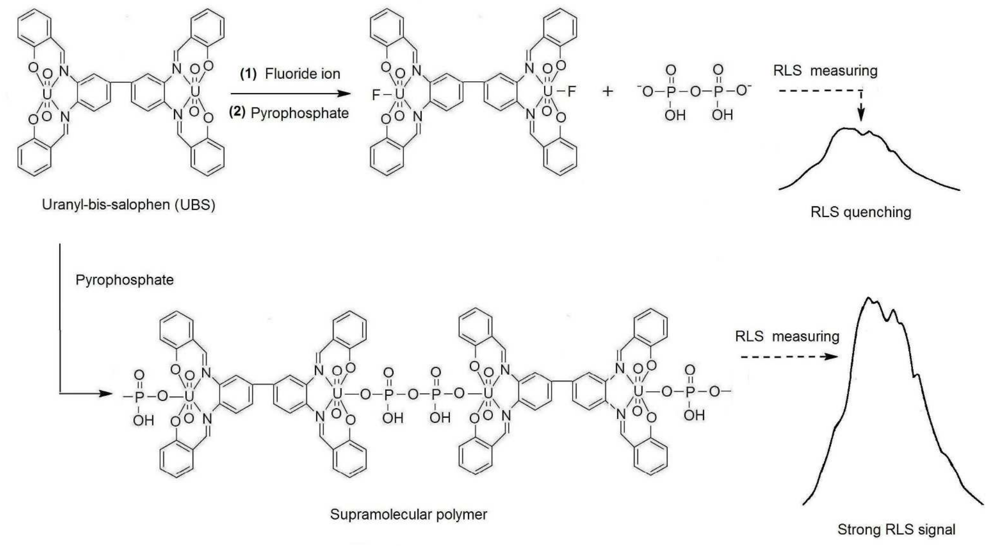

Illustrated procedures of detecting fluoride ion by RLS method
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ARTICLE TYPE

# Resonance light scattering detecting fluoride ion based on the formation of uranyl coordination supramolecular polymer

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A resonance light scattering (RLS) method for the detection of fluoride ion in aqueous solution is reported in this paper. Uranyl-bis-salophen (UBS), a binuclear uranyl complex prepared through the reaction of uranyl with a ditopic tetradentate Schiff base ligand bis-salophen, was used for the experiment. It is found that after UBS self-assembled with pyrophosphate to form uranyl coordination supramolecular polymer (SP), the system produced a strong RLS signal. When fluoride ion coexisted with UBS, The formation of SP was inhibited due to that the fluoride ion occupied the coordination sites in UBS. This resulted in a RLS quenching. Based on these findings, we established a RLS method for the detection of fluoride ion in aqueous solution. Under optimal conditions, a linear range was found to be 0.005 to 1.5 nmol/mL with a detection limit of 0.0028 nmol/mL. The method was applied to determine fluoride ion in environmental water samples with the relative standard deviations of 1.3 to 3.1% and the recoveries of 98.7–101.3%.

## Introduction

Supramolecular polymers (SP) have gained considerable contemporary research interest because of their various and tunable properties. The applications of SP have been involved in many fields. An important application field needed to be developed is using SP for the recognition and detection of target analytes.[1]

Resonance light scattering (RLS) technique is an efficient analytical technique with the advantages of simplicity, rapidness and high sensitivity.[2] It is based on the particle volume enlargement caused by the assembly or aggregation of the particles. RLS technique has been used for the detection of target analytes based on various assembly and aggregation reactions, such as the detection of ions and molecules based on the self-assembly of cations with anions to form ion association particles,[3] the detection of ions and biological molecules based on the aggregation of gold nanoparticles or magnetic nanoparticles,[4] the detection of antibodies and antigens based on their combination to form immune complexes,[5] and the detection of medicines and drugs based on their binding with proteins and DNA.[6] The self-assembly of monomers to form SP is also a typical procedure of particle volume enlargement and can cause an enhanced RLS signal. Up to date, this type of enhanced RLS signal has hardly been used to detect analytes. Therefore, it is necessary to exploit a new type of RLS technique for the detection of target analytes by utilizing the formation of SP.

Fluoride is the only existing form of fluorine in nature. It occurs as ion in natural waters with various concentrations. Low concentration of fluoride ion in drinking water is beneficial as it prevents tooth decay owing to its cariostatic activity. However, a large amount of ingested fluoride will result in a wide series of toxic effects to the body.[7] For example, excessive uptake of fluoride may adversely affect children's intelligence.[8] According to the World Health Organization guidelines, the permissible limit for fluoride in drinking water is 1.5 mg/L.[9] Therefore, the monitoring of fluoride levels in water and food products is important to ensure good water and food quality.

A set of analytical methods have been developed for the determination of fluoride in various sample matrices, including spectrophotometry with various chromogenic reagent,[10] potentiometry with fluoride ion selective electrode,[11] chromatography,[12] fluorometry,[13] capillary electrophoresis[14] and polarography.[15] However, until now, RLS technique has hardly been used for the detection of fluoride.

Previous studies demonstrated that the complexes of uranyl with tetradentate ligands have strong ability to recognize fluoride and phosphate group with high affinity and high selectivity.[16] Considering these, in this work we intended to establish a RLS method for the detection of trace fluoride ion in aqueous solution by utilizing the self-assembly of a binuclear uranyl complex with pyrophosphate to form uranyl coordination SP. The used binuclear uranyl complex is uranyl-bis-salophen (UBS). It is a complex of two uranyl with a ditopic tetradentate ligand bis-salophen. We found that after UBS self-assembled with pyrophosphate, the system produced a strong RLS signal due to the enlargement of the molecular volume through the formation of SP. Furthermore, we found that when fluoride ion was present, the RLS was quenched due to that fluoride occupied the coordination sites in UBS and then inhibited the formation of SP. Based on these findings, we established the RLS method for the detection of trace fluoride ion in aqueous solution. The proposed method has been successfully applied for the detection of fluoride ion in real water samples with satisfactory results.

## Experimental

### Chemicals and apparatus

Salicylaldehyde and 3,3',4,4'-tetraminobiphenyl were purchased from Aladding (Shanghai, China). Uranyl nitrate hexahydrate was obtained from Hubei Chushengwei Chemical Co., Ltd. (Wuhan, China). Potassium fluoride, sodium pyrophosphate and other chemicals were purchased from Merck and Aldrich. All chemicals used were of analytical grade.

Bis-salophen (*N*,*N'*,*N''*,*N'''*-tetrasalicylidene-3,3'-diaminobenzidine), the ditopic tetradentate Schiff base ligand, was prepared according to the literature procedures by utilizing the Schiff base condensation reaction of salicylaldehyde with 3,3',4,4'-tetraminobiphenyl.[17] The binuclear uranyl complex UBS was prepared through the chelating reaction of uranyl with bis-salophen.

RLS spectra were recorded on a Hitachi 4500 spectrofluorometer.

### Procedure of fluoride detection

An appropriate amount of fluoride ion calibration solution or sample solution, 1.0 mL of pH 7.5 Tris-HCl buffer and 1.0 mL of 15.0 nmol/mL UBS solution were added successively into a 10 mL volumetric flask. The solution was mixed thoroughly and was incubated for 15 min. Then 1.0 mL of 15.0 nmol/mL pyrophosphate solution was added and the total volume of the solution was fixed at 10 mL with water. The solution was mixed thoroughly and was incubated again for 15 min. After that the RLS spectra of the system were recorded on the spectrofluorometer. A calibration curve of the RLS intensity versus fluoride ion concentration was plotted. The procedure of fluoride ion detection is illustrated in Fig. 1.

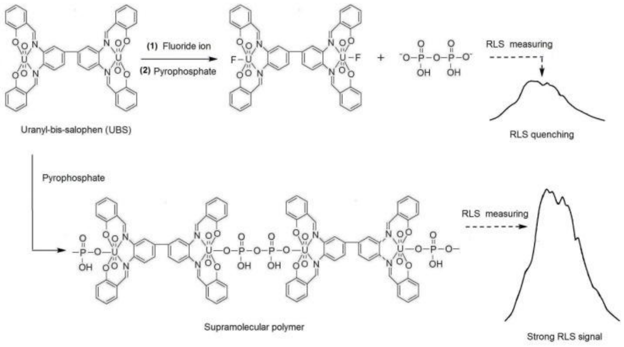

**Fig. 1** Illustrated procedures of detecting fluoride ion by RLS method.

## Results and discussion

### RLS spectral characteristics

Typical RLS spectra of UBS-pyrophosphate system with different concentrations of fluoride ion are shown in Fig. 2. It can be seen that the RLS intensity of UBS-pyrophosphate system is strong in the absence of fluoride. The reason is that under this condition UBS has self-assembled with pyrophosphate to form SF with large molecular volume. When fluoride ion is present, the RLS intensity is decreased with the increase of fluoride ion concentration. The reason is that in the presence of fluoride, coordination sites in UBS are occupied by fluoride ion, and the self-assembly reaction is inhibited. This results in a RLS quenching. The maximum scattering wavelength is at 405 nm. Under a certain condition there is a linear relationship between the RLS intensity and the concentration of fluoride ion in a certain concentration range. Therefore, a RLS method for the determination of fluoride ion can be established by utilizing the inhibition effect of fluoride ion on the self-assembling reaction. In this work the RLS intensity at 405 nm was selected for the detection of fluoride ion.

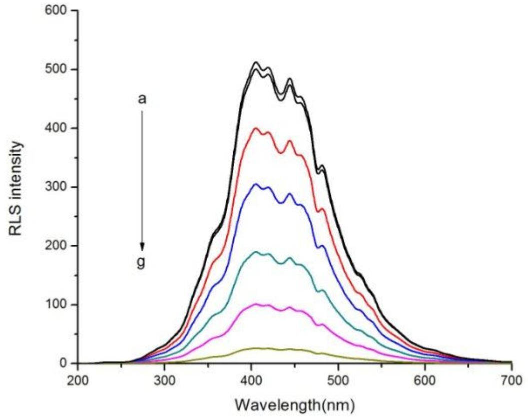

**Fig. 2** RLS spectra of the system. The concentrations of both UBS and pyrophosphate are 1.5 nmol/mL. The concentrations of fluoride ion are (a) 0.0, (b) 0.005, (c) 0.3, (d) 0.6, (e) 0.9, (f) 1.2, and (g) 1.5 nmol/mL.

### Effect of UBS and pyrophosphate concentrations

The effect of UBS and pyrophosphate concentrations on the RLS intensity and the inhibition effect of fluoride ion were investigated. The results show that under fixed pyrophosphate concentration, the RLS intensity increases with the increase of UBS and reaches the maximum when UBS concentration is equal to or higher than pyrophosphate concentration. Similarly, under fixed UBS concentration, the RLS intensity also reaches the maximum when pyrophosphate concentration is equal to or higher than UBS concentration. Therefore, in this work, the concentration of UBS and that of pyrophosphate were selected to be equal. As shown in Fig. 3, the RLS intensity increases with the increase of equal concentrations of UBS and pyrophosphate. When UBS and pyrophosphate concentrations are higher than 1.5 nmol/mL, the RLS intensity still increases with the increase of UBS and pyrophosphate concentrations. However, the inhibition effect of fluoride ion is reduced. Therefore, in this work, 1.5 nmol/mL of UBS and 1.5 nmol/mL of pyrophosphate were selected for the detecting experiment.

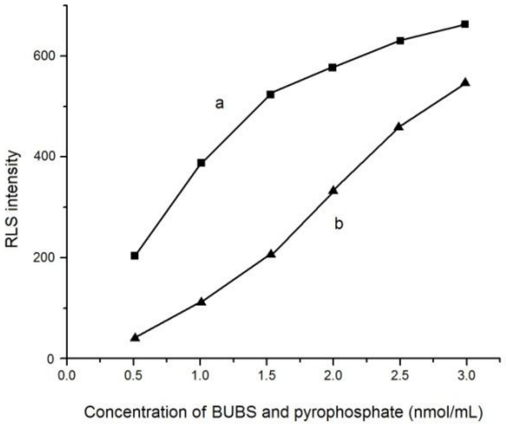

**Fig. 3** Effect of UBS and pyrophosphate concentrations (a) fluoride ion is absent. (b) 1.0 nmol/mL fluoride ion is present.

### Influence of medium acidity

The influence of medium acidity to the inhibition effect of fluoride ion was investigated and the results are shown in Fig. 4. When fluoride ion is absent, the RLS intensity was found to be nearly a constant in the pH range of 5.5 to 9.0. When the pH is higher than 9.0, the RLS intensity decreases with the increase of pH. This is due to that under strong basic condition the self-assembly reaction is inhibited by hydroxyl ion. When fluoride ion is present, the RLS intensity decreases at first, then reaches a minimum in the pH range of 7.0 to 10.0 and the intensity is nearly a constant in this pH range. These results demonstrate that in the pH range of 7.0 to 9.0, fluoride ion has the strongest inhibition effect on the self-assembly reaction, and that the reaction needs to be carried out in this pH range. These also result from that under acidic condition the inhibition effect of fluoride ion is reduced due to its combination with hydrogen ion. Under strong basic condition, the inhibition effect of fluoride ion is also reduced due to that hydroxyl ion competes with fluoride to occupy the coordination sites in UBS. In this work, the medium acidity was controlled at pH 7.5 by using Tris-HCl as buffer for the reaction.

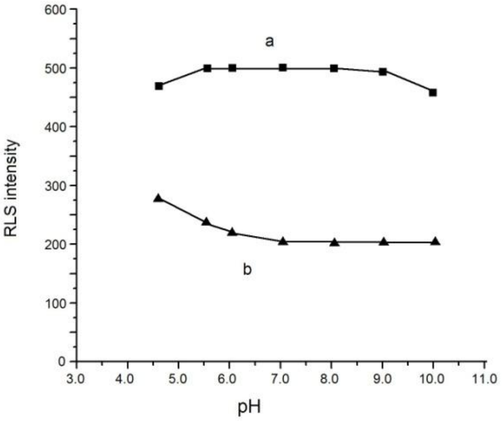

**Fig. 4** Effect of medium acidity. (a) fluoride ion is absent. (b) 1.0 nmol/mL fluoride ion is present.

### Effect of addition sequence of the reagents and reaction times

The effect of the addition sequence of the reagents was tested. The results show that the linear relationship between the RLS intensity and fluoride ion concentration with the addition sequence of first adding UBS and then adding pyrophosphate is better than that with the addition sequence of first adding pyrophosphate and then adding UBS. The reason is that when UBS is added first, fluoride ion can react completely with UBS, resulting in a good linear relationship between the RLS intensity and fluoride ion concentration. Otherwise, when pyrophosphate is added first, it competes with fluoride ion to occupy the coordination sites in UBS. Thus, the reaction of fluoride ion further substituting the pyrophosphate in the polymer is too slow, resulting in a departure from the linear relationship. Therefore, in this work, the selected addition sequence was adding UBS first and then adding pyrophosphate.

The effects of the reaction time of UBS with fluoride ion before adding pyrophosphate and the reaction time after adding pyrophosphate were also investigated. The results show that when both of the reaction times are more than 10 min, the linear relationship is good. Therefore, in this work, the selected reaction time for the reaction of UBS with fluoride ion was 15 min. The selected reaction time after adding pyrophosphate was also 15 min.

### Calibration curve, detection limit and precision

Under optimal conditions, the calibration curve for the determination of fluoride ion was tested with different amounts of fluoride ion. The curve shown in Fig. 5 demonstrates that there is a linear relationship between the RLS intensity ($I_{\mathrm{RLS}}$) and fluoride ion concentration ($c$) in the range of 0.005 to 1.5 nmol/mL. The linear regression equation is $I_{\mathrm{RLS}} = -328.3\ c$ (nmol/mL) + 502.0 with a correlation coefficient of $r = 0.9978$. The detection limit of the method was determined to be 0.0028 nmol/mL calculated from three times the standard deviation through 11 parallel blank tests. The relative standard deviation (RSD) of six parallel determinations for 0.6 nmol/mL fluoride was 1.05%.

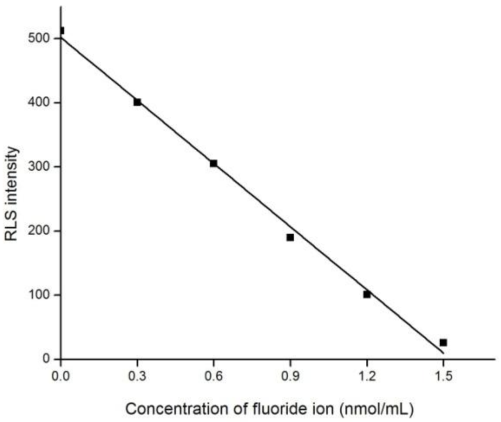

**Fig. 5** Calibration curve for the determination of fluoride ion

### Influences of coexistent substances and their elimination

The influence of several coexistent ions to the determination of 1.0 nmol/mL fluoride ion was investigated. The influence was examined individually by adding a relatively high concentration of these ions. An error of less than 5% was considered to be no interference to the determination. The results shown in Table 1 demonstrate that 100 times of $SO_4^{2-}$, $NO_3^-$, $Cl^-$, $Br^-$, $Cu^{2+}$, $Zn^{2+}$, $Cd^{2+}$, $Co^{2+}$ and $Ni^{2+}$ do not interfere with the determination， but 10 times of $Al^{3+}$, $Fe^{3+}$, $Mn^{2+}$ and $La^{3+}$, and 2 times of $PO_4^{3-}$ have interference. These interferences can be eliminated by the following operation procedure. First, $PO_4^{3-}$ can be precipitated by adding cupric sulphate. Then the metal ions can be precipitated by changing the solution acidity to pH10 with NaOH.

**Table 1** Influence of coexistent ions to the determination of 1.0 nmol/mL fluoride ion

| Coexistent ion | Concentration (nmol/mL) | Found of fluoride ion (nmol/mL) | Error (%) |
|---|---|---|---|
| $SO_4^{2-}$ | 100 | 1.01 | +1.0 |
| $NO_3^-$ | 100 | 1.03 | +2.5 |
| $Cl^-$ | 100 | 1.03 | +3.2 |
| $Br^-$ | 100 | 1.01 | +1.3 |
| $PO_4^{3-}$ | 2 | 1.15 | +14.6 |
| $Cu^{2+}$ | 100 | 1.01 | +0.5 |
| $Zn^{2+}$ | 100 | 1.01 | +1.3 |
| $Cd^{2+}$ | 100 | 0.97 | −3.1 |
| $Co^{2+}$ | 100 | 1.01 | +1.1 |
| $Ni^{2+}$ | 100 | 1.03 | +2.6 |
| $Al^{3+}$ | 10 | 0.94 | −6.4 |
| $Fe^{3+}$ | 10 | 0.92 | −8.0 |
| $Mn^{2+}$ | 10 | 0.93 | −7.2 |

| | | | |
|---|---|---|---|
| $La^{3+}$ | 10 | 0.91 | −8.5 |

### Analysis of real samples

In order to test the validation of the RLS method for real samples, five water samples were analyzed by this method. The water samples were pre-disposed by the following operation. 1.0 mL of 0.1 mmol/L cupric sulphate was added into 100 mL of water sample. Then the acidity of the solution was changed to pH 10 by adding 0.1 mol/L NaOH. After filtrated, the acidity of the solution was changed to near pH 7 by adding 0.1 mol/L HCl. The pre-disposed water samples were analyzed according to the experimental procedure of fluoride ion detection. The recovery experiment of added standard fluoride ion concentrations was carried out to evaluate the applicability and reliability of this developed method. These samples were also analyzed with a control experiment carried out by a reported fluorescence method for the comparison.[18] All results were summarized in Table 2. It can be observed that the fluoride ion concentration values obtained from the current method show good agreement with that obtained from the fluorescence method, which demonstrate that the new RLS method based on the inhibition effect of fluoride ion on the self-assembly reaction can be successfully applied to real samples with good recoveries.

**Table 2** Analytical results of fluoride ion in water samples (*n* = 6)

| Sample | Found (nmol/mL) | RSD (%) | Added (nmol/mL) | Total Found (nmol/mL) | Recovery (%) |
|---|---|---|---|---|---|
| Potable Water | 0.40<br>(0.43) | 3.1<br>(2.6) | 0.40 | 0.81 | 101.3 |
| Well Water | 0.37<br>(0.39) | 2.2<br>(3.0) | 0.40 | 0.76 | 98.7 |
| Surface Water | 0.56<br>(0.55) | 1.5<br>(2.4) | 0.50 | 1.05 | 99.0 |
| Rain Water | 0.48<br>(0.47) | 1.9<br>(2.5) | 0.50 | 0.98 | 100.0 |
| Tap Water | 0.37<br>(0.40) | 1.3<br>(1.9) | 0.40 | 0.47 | 100.2 |

Note: The data in the brackets were obtained from the control experiment.

## Conclusions

In this study, we established a RLS method for the determination of trace fluoride ion in aqueous solution. The method is based on the fact that the self-assembly reaction of UBS with pyrophosphate to form SP can produce a strong RLS signal and fluoride ion can inhibit the self-assembly reaction to quench the RLS. The method is simple, sensitive and convenient in operation. Although only applied to determine fluoride ion as described in this paper, the RLS method based on the formation of SP may open new strategy for the detection of other targets.

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## Notes and references

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