

# Rapid Determination of Trace Potassium in Drinks and Serum by Rayleigh Light Scattering Technique

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## ABSTRACT

In this paper, a sensitive, rapid and selective method based on Rayleigh light scattering technique (RLS) was proposed for the determination of potassium using a conventional spectrofluorometer. Potassium was transformed to  $\text{KB}(\text{C}_6\text{H}_5)_4$  particles, which displayed intense light scattering in aqueous solutions. Effects of factors such as wavelength, acidity, ionic strength and coexistent interferents on the RLS of  $\text{B}(\text{C}_6\text{H}_5)_4\text{-K}$  were investigated. The RLS intensity of the  $\text{KB}(\text{C}_6\text{H}_5)_4$  suspension was obtained in NaAc solution (0.50 M, pH = 8.0) and the addition of 0.5 mL of sodium dodecyl benzene sulfonate (5.05  $\mu\text{M}$ ) and 1.0 mL of tetraphenylboron sodium (1.0%). The Rayleigh scattering light intensity at the maximum peak of 567 nm was proportional to the concentration of potassium ion in the range of 0.20-1.60  $\mu\text{g mL}^{-1}$  with a detection limit of 0.10  $\mu\text{g mL}^{-1}$ . To determine the feasibility of the proposed method, samples of water, drinks and serum were analyzed. The attained results were in agreement with the ion-selective electrode method. Good recovery was also obtained in a range of 95.10-107.02%. The sensitivity and selectivity of the RLS method are high enough to determine trace amounts of potassium ion without any significant interference from high levels of other components such as common anions and cations.

Key words: Rayleigh light scattering, tetraphenylboron sodium, potassium, drinks and serum, determination

## INTRODUCTION

Rayleigh light scattering (RLS), an elastic scattering, occurs when the incident beam is close to an absorption band. RLS is a sensitive and selective technique for monitoring molecular assemblies. With the advantages of speed, convenience and sensitivity, RLS is an appropriate technique for detecting and characterizing the extended aggregates of chromophores. Since the RLS technique was established by Pasternack *et al.* to study the biological macromolecules with a common fluorescence spectrometer<sup>(1-3)</sup>, great attention was paid to study and determine the pharmaceutical<sup>(4)</sup> and biological macromolecules such as nucleic acids<sup>(5-11)</sup>, proteins<sup>(12-15)</sup> and nanoparticle<sup>(16)</sup>. However, the study and determination of the life important ion  $\text{K}^+$  by RLS technique was not yet reported.

Potassium ion is a mineral ion that helps the kidneys function normally. It also plays a key role in cardiac, skeletal, and smooth muscle contraction, making it an important nutrient for normal heart, digestive, and muscular function. A diet high in potassium from fruits, vegetables, and legumes is generally recommended for optimum heart health. For most people, a healthy diet rich in vegetables and fruits provides all the potassium needed.

Many analytical methods for  $\text{K}^+$  have been developed

so far, such as atomic absorption spectrometry<sup>(7)</sup>, ion-selective electrodes (ISE)<sup>(18)</sup>, spectrophotometry<sup>(19)</sup>, thermal decomposition method<sup>(20)</sup>, atomic emission spectrometry<sup>(21)</sup>, ion chromatography<sup>(22)</sup>, turbidimetry<sup>(23)</sup>, enzymatic method<sup>(24)</sup> and capillary zone electrophoresis<sup>(25)</sup> etc..

Among these methods, the atomic emission and absorption spectrometry method were popular and reliable to determine  $[\text{K}^+]$ . In turbidimetry, tetraphenylboron sodium (Figure 1) was used to form  $\text{KB}(\text{C}_6\text{H}_5)_4$  deposition. The determination of potassium is more troublesome than that of most biologically important cations; at the same time there are many conditions involving variations of the electrolyte content of body fluids accurate estimations of potassium are desirable. It wants a try to devise an analytical method for potassium, which should be simpler than those in use. We now report the development of a sensitive and selec-

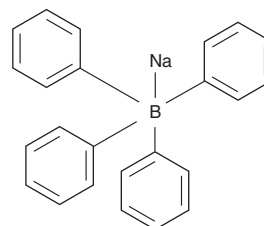


Figure 1. The structure of tetraphenylboron sodium.

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tive method for the determination of potassium with  $\text{NaB}(\text{C}_6\text{H}_5)_4$  as a probe by RLS technique. We compare the attained results with that of ISE method to evaluate the RLS technique.

## MATERIALS AND METHODS

### I. Apparatus

RLS and absorption spectra were obtained using a JASCO FP-6500 spectrofluorometer (Tokyo, Japan), and a JASCO V-550 spectrophotometer, respectively. A Rapidchem 744 electrolyte analyzer (Bayer, UK) was used in the ISE determination. A galvanothermy water bath (Shanghai, China) was used for the temperature control. The pH measurements were carried out on a PHS-3C pH meter (Shanghai, China).

### II. Reagents

The stock solution of  $\text{K}^+$  was prepared by dissolving KCl (>99.99%, Sigma, USA) in water. The working solution of the  $\text{K}^+$  was  $10.0 \mu\text{g mL}^{-1}$ . A working solution of 1.0% tetraphenylboron sodium (>99.8%, Shanghai Chemical Reagent Company, China) was prepared in methanol-water solution (20:80, v/v). A working solution of  $5.05 \mu\text{M}$  sodium dodecyl benzene sulfonate (SDBS, >99.5%, Shanghai Chemical Reagent Company, China) was prepared with water and used as a stabilizer. Sodium acetate (0.50 M, pH = 8.0) was used to control the pH, while 0.1 M NaCl (>99.8%, Shanghai Chemical Reagent Company, China) was used to adjust the ionic strength of the solutions. Doubly distilled water was used throughout.

### III. Standard Procedure

An appropriate aliquot of  $\text{K}^+$  working solution was added to a mixture of 1.0-mL tetraphenylboron sodium solution (1.0%), 1.0-mL NaAc solution (0.50 M, pH = 8.0) and 0.5-mL SDBS solution ( $5.05 \mu\text{M}$ ) and then diluted to 10 mL with water. Five minutes later, the absorption and RLS measurements were obtained against the blank treated in the same way without  $\text{K}^+$ . The RLS spectrum was obtained by scanning simultaneously the excitation and emission monochromator in the wavelength range of 250~750 nm with bandwidths 3.0 nm. RLS intensity was measured at 454.0, 567.0 and 606.0 nm. The enhanced RLS intensity of  $\text{B}(\text{C}_6\text{H}_5)_4\text{-K}$  system was represented as  $\Delta I = I - I_0$  ( $I$  and  $I_0$  were the RLS intensities of the system with and without  $\text{K}^+$ ).

### IV. Sample

The samples of mineral water and pure water were filtered through cellulose membrane filters ( $0.45 \mu\text{m}$ ,

Whatman, NJ, USA) if precipitate is possible. Serum was separated from the fresh blood. The serum samples were diluted 100-fold with water. A 2.0 mL aliquot of the prepared sample solutions was diluted to a 10 mL in a volumetric flask and RLS was measured as described above.

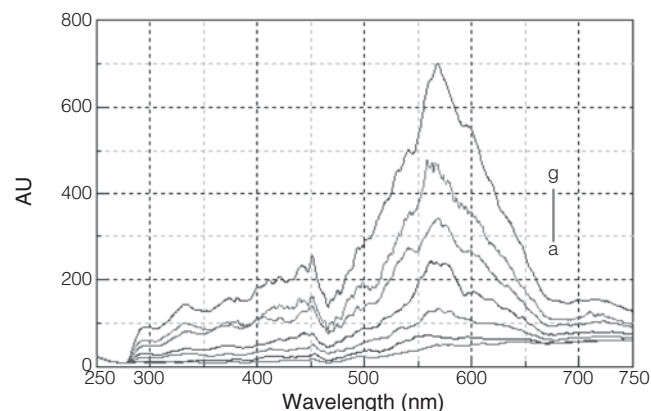
## RESULTS AND DISCUSSION

### I. Characteristics of the RLS Spectra

The RLS spectra of  $\text{B}(\text{C}_6\text{H}_5)_4\text{-Na}$  and  $\text{B}(\text{C}_6\text{H}_5)_4\text{-K}$  system were shown in Figure 2. It is shown that the RLS intensity of the  $\text{K}^+$  (Figure 2A) and  $\text{B}(\text{C}_6\text{H}_5)_4\text{-Na}$  (Figure 2B) is weak for the whole scanning wavelength region. However, the enhanced RLS intensity can be observed with three peaks at 454, 567 and 606 nm when a trace amount of  $\text{K}^+$  was added to. Moreover, the enhanced RLS intensity increases with the increasing of  $[\text{K}^+]$ . The maximum RLS peak is located at 567 nm. Therefore, 567 nm was selected as the analytical wavelength. As shown in Figure 2, the RLS intensities of  $\text{K}^+$  and  $\text{B}(\text{C}_6\text{H}_5)_4\text{-K}$  system are quite weak. It can be concluded that  $\text{B}(\text{C}_6\text{H}_5)_4\text{-Na}$  reacted with  $\text{K}^+$  and produced a new-formed compound whose RLS intensity was much higher than those of  $\text{K}^+$  and  $\text{B}(\text{C}_6\text{H}_5)_4\text{-Na}$ .

### II. Effects of pH Values in Medium

In order to test the electrostatic attraction, we adjusted the pH of the medium in the range of 4.0-11.0. Owing to the negatively charged  $\text{B}(\text{C}_6\text{H}_5)_4^-$ , it is necessary to consider that adjusting pH-value of the medium should induce the change of charges on  $\text{B}(\text{C}_6\text{H}_5)_4^-$  ion. As shown in Figure 3, with the variation of pH, the RLS intensities of  $\text{NaB}(\text{C}_6\text{H}_5)_4$  solution didn't change, where-



**Figure 2.** RLS spectra of (a)  $1.0 \mu\text{g mL}^{-1} \text{K}^+$  and (b-g)  $\text{B}(\text{C}_6\text{H}_5)_4\text{-K}$  systems. Conditions: 1.0 mL of  $\text{NaB}(\text{C}_6\text{H}_5)_4$  solution (1.0%), 1.0 mL HAc-NaAc solution (0.50 M, pH = 8.0) and 0.5 mL SDBS solution ( $5.05 \mu\text{M}$ ) were added to a 10-mL volumetric flask. b-g: 0, 0.2, 0.4, 0.6, 0.8,  $1.0 \mu\text{g mL}^{-1}$  of  $\text{K}^+$ .

as those of the  $B(C_6H_5)_4-K$  system presented different traits. That is, the RLS intensities of the system increased from pH 3.0 to pH 8.0, and decreased after pH 8.0. The intensity reached a maximum in the region of pH 7.0-9.0 so we selected pH 8.0 as the optimum value.

### III. Effect of Ionic Strength

NaCl solution (0.1 M) was used to adjust the ionic strength of the system. It could be seen in Figure 4 that RLS intensity of  $B(C_6H_5)_4-K$  system changed little with ionic strength. Therefore, the system can be allowed at high ionic strength.

### IV. Addition Orders

The effect of addition order on the RLS intensity was listed in Table 1 ( $1.0 \mu\text{g mL}^{-1}$  of  $K^+$ ). It was found that the addition order of reagents exerts little effect on the RLS intensity.

**Table 1.** Effect of adding order

1	2	3	$\Delta I$ (%)
$NaB(C_6H_5)_4$	$K^+$	HAc-NaAc	100
$NaB(C_6H_5)_4$	HAc-NaAc	$K^+$	97.81
HAc-NaAc	$K^+$	$NaB(C_6H_5)_4$	98.67

### V. Stability

The formation of  $B(C_6H_5)_4-K$  particle includes three steps: nucleation, crystal growth, and aggregation, which will affect the particle size directly. Because the particle size is one important factor on RLS intensity, stabilizer must be used to control the size of  $B(C_6H_5)_4-K$  particles, prevent the rapid sedimentation of the particles, and to improve the reproducibility of RLS intensity. In this study, SDBS was used to stabilize the  $B(C_6H_5)_4-K$  system. The average deviation of RLS signal was found to be lower than 4.30%.

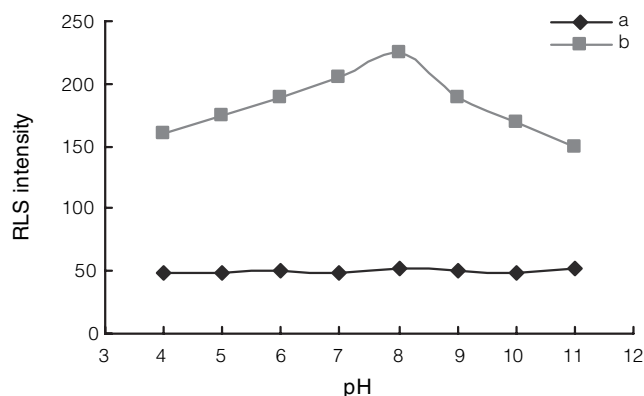
### VI. Tolerance of Foreign Substances

Effects of cationic and anionic species in water on RLS intensity were studied by the addition of foreign substances. Their concentration relative to  $[K^+]$  and the corresponding influence to the determinations were displayed in Table 2. Few coexisting ions affect the determination of  $[K^+]$ . Common ions such as  $Na^+$ ,  $Ca^{2+}$ ,  $Ba^{2+}$ ,  $Mg^{2+}$ ,  $Zn^{2+}$ ,  $Co^{2+}$ ,  $Cu^{2+}$ ,  $Al^{3+}$ ,  $Pb^{2+}$  and  $Fe^{2+}$  can be tolerated at high concentrations because they did not combine with  $B(C_6H_5)_4^-$ . However, some ions such as  $NH_4^+$  can only be tolerated at very low concentration ( $2 \mu\text{g mL}^{-1}$ ). In the studied species,  $NH_4^+$  was interfered seriously due to similar ionic radius. However,  $NH_4^+$  is unstable and nearly absent in the sample, so it seldom interferes the determination. The most abundant  $Na^+$  would interfere at the concentration of up to 1000 times that of  $K^+$ . Because  $Na^+$  was studied by adding NaAc and the molecular weight of  $Ac^-$  is larger than that of  $Na^+$ , the tolerance level of  $Ac^-$  is larger. Other ions have nearly no effects on the determination when their concentration was the same or more than potassium ion. Therefore, the tolerant level of interference in the samples was high. Due to the good selectivity of this method, assays can be performed without removing other coexisting ions.

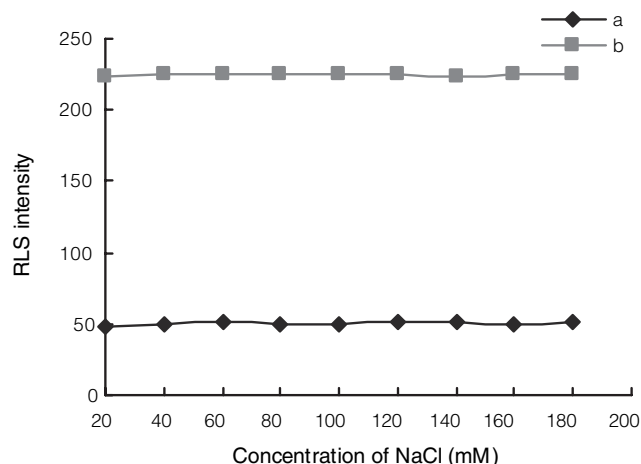
## RESULTS AND DISCUSSION

### I. Detection and Quantification Limits

The detection limit was calculated as  $s_b + 3s$ , where  $s_b$  is the average signal of ten blank solutions and  $s$  the standard deviation. On the other hand, the quantification limit was calculated as  $s_b + 10s$ . With detection wave-



**Figure 3.** Effect of pH on the RLS intensity. (a) the blank solution, (b) the  $B(C_6H_5)_4-K$  suspension with  $0.60 \mu\text{g mL}^{-1}$  of  $K^+$ .



**Figure 4.** Effect of NaCl as ionic strength regulator on the RLS intensity. (a) the blank solution, (b) the  $B(C_6H_5)_4-K$  suspension with  $0.60 \mu\text{g mL}^{-1}$  of  $K^+$ .

**Table 2.** Effect of interfering ions on the  $K^+$  determination<sup>a</sup>

Foreign cationic ions	Ratio of the conc. (foreign ions/[ $K^+$ ])	Change in RLS (%)	Foreign anionic ions	Ratio of the conc. (foreign ions/[ $K^+$ ])	Change in RLS (%)
$Na^+$	1000	1.20	$Cl^-$	1000	2.62
$Ca^{2+}$	100	2.19	$Ac^-$	1000	2.34
$Ba^{2+}$	100	2.65	$SO_4^{2-}$	1000	2.47
$Mg^{2+}$	100	-2.59	$NO_3^-$	1000	3.94
$Zn^{2+}$	100	3.42	$CO_3^{2-}$	1000	3.01
$Co^{2+}$	100	-2.46	$SO_3^{2-}$	1000	2.77
$Cu^{2+}$	100	1.85	$PO_4^{3-}$	1000	-0.79
$Al^{3+}$	100	4.21			
$Fe^{2+}$	100	3.42			
$NH_4^+$	5	5.42			
$Pb^{2+}$	10	6.86			

<sup>a</sup>The cationic ions were added in the form of chloride, and the anionic ions were added in the form of sodium.

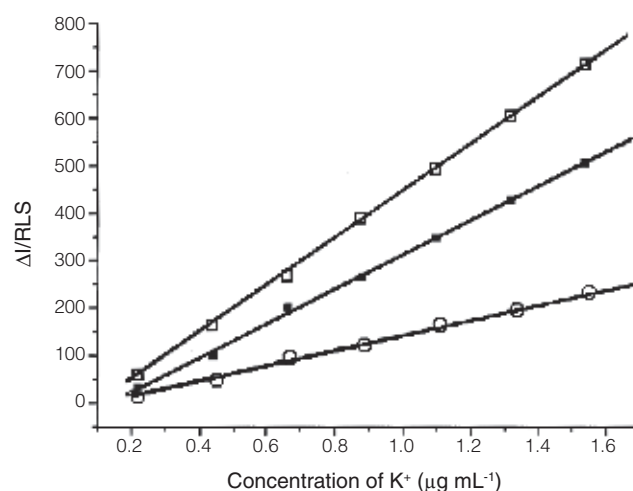
length of 567 nm, the detection limit and quantification limit were  $0.10 \mu\text{g mL}^{-1}$  and  $0.20 \mu\text{g mL}^{-1}$ , respectively.

## II. Detected Wavelength and Calibration Curves

According to the above standard procedure, the calibration curves were obtained by plotting seven different concentrations of  $K^+$  against the intensity of RLS spectrum at 454, 567 and 609 nm under the optimum conditions (Figure 5). Each calibration sample was detected in triplicate. To maximum RLS peak of 567 nm, the linear regression equation using the least square method was  $\Delta I = -53.81 + 490.79C_{K^+}$  ( $\mu\text{g mL}^{-1}$ ) ( $r^2 = 0.9983$ ) in the range of  $0.20$ – $1.60 \mu\text{g mL}^{-1}$  of  $K^+$ . The parameters and correlation coefficients of the calibration plots with three wavelengths were indicated in the Figure 5. The regression analysis were then used to back-calculate the concentration from the  $\Delta I$ , and the appropriate summary statistics [mean, standard deviation (SD), and relative SD (RSD)] were calculated. The lowest detection limit and quantification limit took place at 567 nm which is the maximum RLS peak. For trace  $K^+$  in drinks, and serum, the 567 nm was used for highest sensitivity.

## III. Precision

The precision was comprised of repeatability and reproducibility. These were developed in five different samples (mineral water and serum) which contained low, medium and high  $K^+$  levels. The repeatability was established by analyzing the samples five times. The reproducibility was determined by analyzing each sample on 3 different days over about 1 month. The RSDs of the repeatability and of the reproducibility were lower than 3.36% and 4.67%, respectively. These results indicate that the present method can be used for quantitative analyses of  $K^+$ .



**Figure 5.** Relationships between RLS intensities of  $B(C_6H_5)_4-K$  system and the concentration of  $K^+$  at various wavelengths. The linear regression equations for (a) 567, (b) 609, and (c) 454 nm were  $y = -53.81 + 490.79x$  ( $r = 0.9983$ ),  $y = -48.50 + 375.94x$  ( $r = 0.9958$ ), and  $y = -18.68 + 158.94x$  ( $r = 0.9913$ ), respectively ( $y$  is  $\Delta I$ ;  $x$  is the  $K^+$  concentrations ( $\mu\text{g mL}^{-1}$ )). The linear range was  $0.20$ – $1.60 \mu\text{g mL}^{-1}$ .

## IV. Recovery

To establish the accuracy of the method, this procedure was performed by adding  $K^+$  to samples. The recoveries of  $K^+$  by this method were shown in Table 3 and good recoveries were observed in a range of 95.10–107.02%.

## V. Assay of Samples

The proposed method was carried to determine  $[K^+]$  in drinks and serum and the results were compared with the ISE method. As shown in Table 4, the results were in

**Table 3.** Recovery tests of some samples<sup>a</sup>

Sample	K <sup>+</sup> in samples (μg)	Added K <sup>+</sup> (μg)	Found (μg)	Recovery range (%)
Mineral water	2.37	1.00, 2.00, 3.00	3.43, 4.51, 5.46	106.00, 107.00, 103.00
Mineral water	12.34	10.00, 12.00, 15.00	21.85, 23.96, 27.13	95.10, 96.83, 98.60
Serum	141.49	100.00, 150.00, 200.00	238.74, 300.59, 345.67	97.25, 106.07, 102.09
Serum	115.72	80.00, 100.00, 120.00	201.34, 221.49, 238.49	107.02, 105.77, 102.31
Serum	180.75	150.00, 180.00, 200.00	325.39, 358.71, 382.04	96.43, 98.87, 100.65

<sup>a</sup>1 or 3 mL of samples was used, and 100 μg of the work solution was added to each sample.

agreement with the ISE method, which indicated that the assay of trace potassium is practical. The samples will be diluted if necessary.

As we know, ISE method involves electrochemistry analysis, in which the variation of temperature, fluctuation of voltage and treatment of electrode may bring on significant influence on the determination though it features high selectivity. However, the RLS method hardly suffers from this drawback.

## VI. Mechanism Discussion

Light scattering is caused by the presence of fine particles. Because the dimension of B(C<sub>6</sub>H<sub>5</sub>)<sub>4</sub>-K particles is much less than the wavelength of incident light, it should obey the Rayleigh light scattering formula, which is shown as follows<sup>(26-27)</sup>:

$$R(\theta) = \frac{9\pi^2}{2\lambda^4} \left( \frac{n_1^2 - n_0^2}{n_1^2 + 2n_0^2} \right)^2 N_0 v^2 (1 + \cos^2\theta) \quad (1)$$

where  $R(\theta)$  is the Rayleigh light ratio at the scattering angle  $\theta$ , which is equal to the ratio of the scattering intensity of incident light  $I(\theta)$  at the angle  $\theta$  to the intensity of incident light  $I_0$ ;  $n_1$  and  $n_0$  are the refractive indices of solute and medium, respectively;  $N_0$  is the number of particles per unit volume;  $v$  is the volume of the particle;  $\lambda$  is the wavelength of incident light in the medium.

If  $c$  is the concentration of B(C<sub>6</sub>H<sub>5</sub>)<sub>4</sub>-K solution and  $\rho$  is the density of each particle.  $N_0 v$  is equal to  $c/\rho$ . The formula above can be expressed in eq 2.

$$R(\theta) = \frac{9\pi^2}{2\lambda^4} \left( \frac{n_1^2 - n_0^2}{n_1^2 + 2n_0^2} \right)^2 \frac{c}{\rho} v (1 + \cos^2\theta) \quad (2)$$

In the experiment,  $\theta$  is 90° and  $v$  remains nearly constant because the experiment conditions such as acidity and the volume of stabilizer and other reagents added were kept constant as identical as possible to obtain the same size particles;  $n_1$ ,  $n_0$ ,  $\lambda$  and  $\rho$  were all constant. According to eq 2, RLS intensity is proportional to the concentration of B(C<sub>6</sub>H<sub>5</sub>)<sub>4</sub>-K suspension ( $c$ ) or the number of particles in the unit volume ( $N_0$ ). Therefore, K<sup>+</sup> can be determined based on this theory.

## CONCLUSIONS

Potassium ion can interact with B(C<sub>6</sub>H<sub>5</sub>)<sub>4</sub><sup>-</sup> anion

**Table 4.** Determination results of potassium in waters, drinks, and seruma

Samples	RLS (μg mL <sup>-1</sup> )	RSD (%)	ISE (μg mL <sup>-1</sup> )	RSD (%)
Mineral water				
Nongfu spring	0.35	4.31	0.31	3.46
Laoshan	ND		ND	
Nestlé	2.71	2.43	2.56	1.54
Robust	0.58	3.24	0.49	2.23
Watsons	6.51	3.46	6.68	4.22
Masterkong	12.34	1.03	12.08	1.68
Pure water				
Wahaha	ND		ND	
Nongfu spring	ND		ND	
Green tea	5.79	1.96	5.86	1.64
Serum				
1	134.54	1.30	142.17	1.65
2	166.36	0.98	161.49	1.29
3	180.75	0.75	179.64	1.01
4	141.49	1.02	138.71	0.99
5	154.30	0.88	155.07	1.21
6	121.97	1.14	123.45	1.32
7	128.63	1.05	124.31	1.19
8	147.80	0.76	145.27	0.89
9	159.43	0.91	158.74	1.08
10	115.72	1.38	119.56	1.42

<sup>a</sup>ND means "not found".

in aqueous solution, giving rise to B(C<sub>6</sub>H<sub>5</sub>)<sub>4</sub>-K particle, which produced intense Rayleigh scattering light. Stable and intense Rayleigh scattering light of B(C<sub>6</sub>H<sub>5</sub>)<sub>4</sub>-K suspension was obtained in NaAc solution (0.50 M, pH = 8.0) in the presence of SDBS as a stabilizer. The Rayleigh scattering light intensity at the maximum peak of 567 nm was proportional to the concentration of K<sup>+</sup> in the range of 0.20-1.60 μg mL<sup>-1</sup> with a detection limit of 0.10 μg mL<sup>-1</sup>. The method was used to determine the trace amounts of K<sup>+</sup> in serum, showing high sensitivity and accuracy compared with the clinically used ISE method.



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