UV-Visible spectrophotometric method for the determination of ammonia nitrogen using potassium bromate as oxidant

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ABSTRACT

Ammonia nitrogen is an essential parameter for indicating water quality. Herein, a simple, sensitive, reproducible, and accurate method was developed to monitor ammonia nitrogen. The principle of the method is to use an oxidant to convert ammonia nitrogen to nitrite, with nitrite determined via Griess method. Potassium bromate in alkaline conditions was confirmed to be a suitable oxidant. Also, an optimization in the main procedure of the method was conducted to gain high sensitivity. Furthermore, under the optimal conditions, the absorbance was good linear with the concentration of ammonia nitrogen over the range of 0–448 $\mu g \cdot N \cdot L^{-1}$ with a correlation coefficient of 0.999. The limit of detection was 15.98 $ng \cdot N \cdot L^{-1}$. In addition, the proposed method was successfully applied to determine ammonia nitrogen in natural water samples from Ming Lake at Changzhou Institute of Technology. Recoveries of 96.13%–100.58% were obtained.

Keywords: Ammonia nitrogen; Nitrite; Griess reaction; Potassium bromate; Ammonia oxidation

1. Introduction

Ammonia nitrogen is an essential nutrient for the growth of aquatic organisms. However, excessive ammonia nitrogen can lead to eutrophication which not only disrupts the ecological balance but also pollutes the environment [1]. Additionally, ammonia nitrogen concentration is one of the factors used to assess self-purification of water [2]. Therefore, studies on the determination of ammonia nitrogen are necessary.

Several methods, such as ion selective electrodes [3,4], fluorescence [5], ion chromatography [6,7], and spectrophotometric or colorimetric assays [8,9], are used for the quantification of ammonia nitrogen in water. The merits of each method are numerous, but each also has certain disadvantages. For example, electrochemical methods that use ion selective electrodes for sensing ammonia nitrogen require few reagents, are highly accurate, and have a fast response time [10]. Nevertheless, due to their low sensitivity, electrochemical methods are not suitable for routine analyses of ammonia nitrogen [11]. Compared to electrochemical methods, fluorescence-based methods [12] are more sensitive [13], but their signals can be affected by air bubbles [11]. Ion chromatography is also highly sensitive and reproducible [14], but it is expensive and requires complex instruments [15]. In contrast, spectrophotometric methods, including Nessler's reagent method [16,17], the indophenol blue (IPB) method [18,19], and the hypobromite oxidation method, are more widely used due to their low costs, high sensitivity, and reliability [20,21]. Nevertheless, mercury(II) iodide, a component of Nessler's reagent, is extremely toxic. Ortho-chlorophenol, another product of the IPB method, is also toxic and highly volatile. By comparison, the hypobromite oxidation method has low toxic reagents and products, so it warrants further study.

The hypobromite oxidation method [22] is an indirect spectrophotometric method. In this method, ammonia nitrogen is converted to nitrite through the use of an

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oxidation agent [23], and nitrite is determined using the Griess method. According to Richards and Kletsch [24], ammonium is oxidized to nitrite by reaction with hypochlorite and potassium bromide. Moreover, many scholars [25,26] have greatly contributed to the application of the indirect method. However, a major drawback of this method is the instability of some of the reagents. For instance, sodium hypochlorite solution, an oxidation reagent in the hypobromite oxidation method, is stable for only 2 h. As a result, the oxidation reagent solution should be prepared fresh before use. This will increase labor intensity, but also reduce accuracy and reproducibility. In addition, most studies to determine ammonia concentrations were conducted on artificial samples, while rare studies were conducted on natural water.

This study investigated the efficacy of potassium bromate, a relatively stable and minimally toxic oxidant, in determining ammonia nitrogen concentration through oxidation methodology. Following the optimization of reaction conditions, the proposed method was employed to determine the ammonia nitrogen concentration in natural water samples. The results demonstrated that the proposed method was both sensitive and precise, effectively addressing the issue of oxidant instability.

2. Experimental set-up

2.1. Reagents

Chemicals, which include ammonium chloride (NH_4Cl), hydrochloric acid (HCl), potassium bromate ($KBrO_3$), sodium hydroxide (NaOH), sulfanilamide ($C_6H_8O_2N_2S$) and N-1-naphthylethylenediamine dihydrochloride ($C_{12}H_{15}ClN_2$), were obtained from Sinopharm Chemical Reagent Company Limited, (Shanghai, China). All reagents were of analytical grade. Deionized water was used to prepare all solutions.

A 1,000 $mg \cdot N \cdot L^{-1}$ NH_4^+ stock standard solution was prepared by dissolving 0.955 g of NH_4Cl in 250 mL deionized water.

A 14 $g \cdot L^{-1}$ $KBrO_3$ stock standard solution was prepared by dissolving 1.400 g of $KBrO_3$ in 100 mL deionized water.

A 400 $g \cdot L^{-1}$ NaOH stock standard solution was prepared by dissolving 20.000 g of NaOH in 50 mL deionized water. After becoming completely dissolved, the NaOH stock standard solution was transferred to a brown polyethylene bottle.

A 10 $g \cdot L^{-1}$ $C_6H_8O_2N_2S$ stock standard solution was prepared by dissolving 1.000 g of $C_6H_8O_2N_2S$ in 100 mL of HCl/H_2O solution with a volume ratio of 1:1.

A 1 $g \cdot L^{-1}$ $C_{12}H_{15}ClN_2$ stock standard solution was prepared by dissolving 0.500 g of $C_{12}H_{15}ClN_2$ in 500 mL deionized water.

Stock solutions were then stored at –7°C in brown bottles to prevent photo-degradation. When necessary, they were diluted with deionized water to give a series of working solutions. Finally, a series of duplicate calibration standards was prepared from stock solutions at concentrations ranging from 0 to 896 $\mu g \cdot N \cdot L^{-1}$.

2.2. Apparatus

A 1 cm quartz cuvette was used to hold the samples. A UV-Visible spectrometer (UV-5500, Shanghai Yuanxi Instrument Co., Ltd., Shanghai, China) was used to measure the absorbance at 543 nm of the post-reaction solution since the solution has an absorbance maximum at 543 nm.

2.3. Procedure

Each experiment was performed in a 10 mL colorimetric tube. The NaOH solution, the ammonium solution, and the $KBrO_3$ solution were added sequentially and thoroughly mixed after each addition. After that, the solution was allowed to stand for several minutes. Next, $C_6H_8O_2N_2S$ solution and $C_{12}H_{15}ClN_2$ solution were added successively, and the solution was uniformly mixed. Afterward, the volume was diluted with deionized water to 10 mL and allowed to stand for 15 min at 25°C to develop color. Finally, these solutions were measured by a UV-Visible spectrometer at 543 nm for their absorbance value (A). A blank sample was prepared as above, except no ammonium solution was added. UV-Visible spectrometer measurements were also made on the blank sample (A_0). Next, the calibration curve was created using several different ammonia nitrogen solutions, with ammonia nitrogen concentration as the horizontal axis and the absorbance value ($A–A_0$) as the vertical axis. Further, the ammonia nitrogen concentration of natural water was interpolated from the calibration curve.

3. Results and discussion

3.1. Spectral characteristics

In the first stage, ammonia nitrogen was oxidized to nitrite by $KBrO_3$, and then nitrite was used to form azoic dyes of red-violet color by reacting with $C_6H_8O_2N_2S$ and $C_{12}H_{15}ClN_2$. As expected, absorbance at 543 nm of the resulting colored solutions were linearly correlated with ammonia nitrogen concentration in the range of 0–448 $\mu g \cdot N \cdot L^{-1}$.

3.2. Optimization of reaction conditions

In order to develop a suitable system for ammonia nitrogen determination, the concentrations of NaOH, $KBrO_3$, $C_6H_8O_2N_2S$, and $C_{12}H_{15}ClN_2$ as well as the oxidation time were optimized. In addition, optimization experiments were performed with solutions containing 560 $\mu g \cdot N \cdot L^{-1}$ ammonia nitrogen. For each optimization experiment, three replicates were performed.

3.2.1. NaOH concentration

NaOH created an alkaline environment that not only converted the ammonium ion to ammonia [27], but also reduced the oxidative capacity of $KBrO_3$. Thus, the alkaline environment caused $KBrO_3$ to oxidize ammonia nitrogen into nitrite instead of nitrate. On the one hand, a high concentration of NaOH is necessary to maximize the absorbance signal to ensure the sensitivity of the method. On the other hand, since the red-violet colored azoic dye can only be synthesized under acid conditions, the concentration of NaOH in the solution should not be too high to make it neutral or alkaline. Therefore, the concentration of NaOH should be optimized. The results are summarized in Fig. 1.
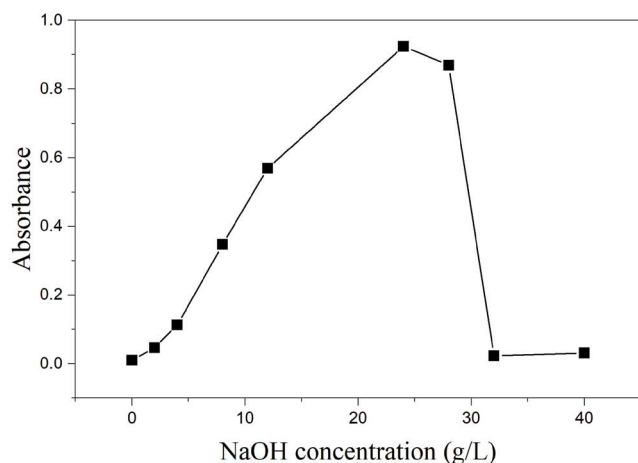
Fig. 1. Dependence of absorbance on NaOH concentration. Other reagents concentration: 560 µg·N·L⁻¹ ammonia nitrogen; 0.0112 g·L⁻¹ KBrO₃; 1 g·L⁻¹ C₆H₈O₂N₂S; 0.010 g·L⁻¹ C₁₂H₁₅ClN₂; oxidation time: 20 min.

As shown in Fig. 1, absorbance increased with increasing NaOH concentration and then peaked at 24 g·L⁻¹ NaOH. After that, as the NaOH concentration increased, absorbance declined and eventually became zero at 32 g·L⁻¹. Therefore, a NaOH concentration of 24 g·L⁻¹ was considered optimal.

3.2.2. KBrO₃ concentration

KBrO₃ acts as an oxidant in the whole reaction system. In an alkaline environment, KBrO₃ could oxidize ammonia nitrogen to nitrite. Thus, KBrO₃ must be present in sufficient concentration to convert all ammonia nitrogen to nitrite. However, the concentration of KBrO₃ should not be too high since a large excess of KBrO₃ could degrade C₆H₈O₂N₂S through oxidation. It is therefore necessary to optimize the concentration of KBrO₃. This is shown in Fig. 2.

In Fig. 2, as KBrO₃ concentration was increased, absorbance increased steeply at first, peaking at 0.0112 g·L⁻¹. After that, the absorbance of the resulting solutions decreased stepwise as KBrO₃ was increased. Therefore, a KBrO₃ concentration of 0.0112 g·L⁻¹ was considered optimal.

3.2.3. C₆H₈O₂N₂S concentration

Sulfanilamide (C₆H₈O₂N₂S) could react with nitrite to form diazonium compounds only in an acidic environment. Therefore, C₆H₈O₂N₂S solution was prepared by dissolving 1.000 g of C₆H₈O₂N₂S in 100 mL of HCl/H₂O solution with a volume ratio of 1:1. Therein, HCl neutralized the NaOH added in the previous stage, creating an acidic environment.

Fig. 3 shows that when C₆H₈O₂N₂S concentration was in the range of 0–0.6 g·L⁻¹, the absorbance of the resulting solutions was zero. It was because the result solutions were alkaline instead of acidic when C₆H₈O₂N₂S concentration was below 0.6 g·L⁻¹. In an alkaline environment, no diazonium compound could be formed. Therefore, the resulting solutions have no absorbance. In contrast, when the C₆H₈O₂N₂S concentration was greater than 0.6 g·L⁻¹, the
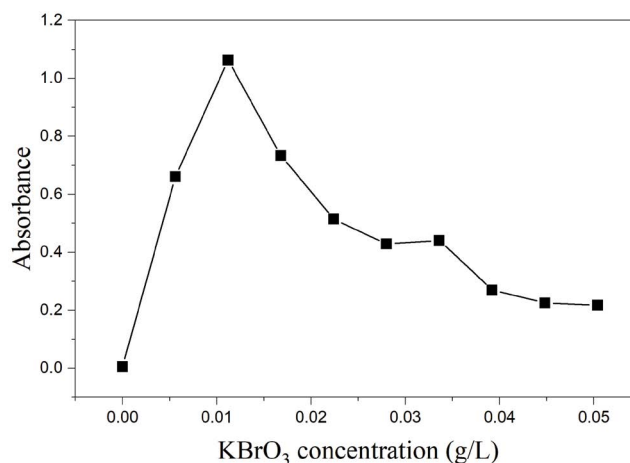
Fig. 2. Dependence of absorbance on KBrO₃ concentration. Other reagents concentration: 24 g·L⁻¹ NaOH; 560 µg·N·L⁻¹ ammonia nitrogen; 1 g·L⁻¹ C₆H₈O₂N₂S; 0.010 g·L⁻¹ C₁₂H₁₅ClN₂; oxidation time: 20 min.
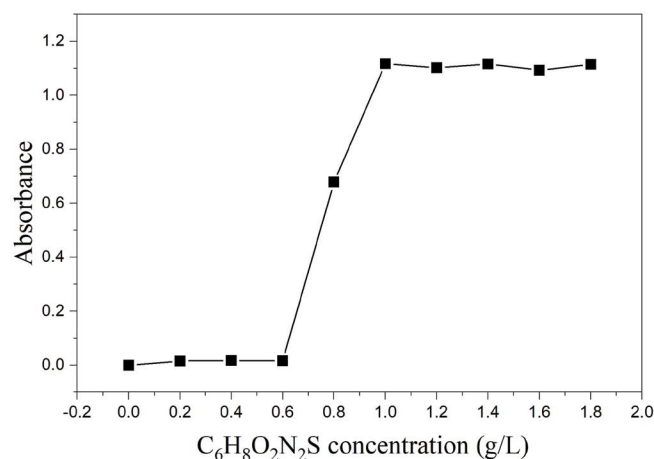
Fig. 3. Dependence of absorbance on C₆H₈O₂N₂S concentration. Other reagents concentration: 24 g·L⁻¹ NaOH; 560 µg·N·L⁻¹ ammonia nitrogen; 0.0112 g·L⁻¹ KBrO₃; 0.010 g·L⁻¹ C₁₂H₁₅ClN₂; oxidation time: 20 min.

resulting solutions became acidic. Therefore, the absorbance increased as C₆H₈O₂N₂S concentration increased, and peaked at 1.0 g·L⁻¹ after which the absorbance remained unchanged. Hence, 1.0 g·L⁻¹ of C₆H₈O₂N₂S was considered optimum.

3.2.4. C₁₂H₁₅ClN₂ concentration

C₁₂H₁₅ClN₂ reacts with the diazonium compound to form a red-violet azo dye with a characteristic absorption spectrum.

As shown in Fig. 4, the absorbance of the resulting solutions increased with increasing C₁₂H₁₅ClN₂ concentrations when C₁₂H₁₅ClN₂ concentrations were between 0–0.010 g·L⁻¹. Moreover, a maximum absorbance was also reached at 0.010 g·L⁻¹ of C₁₂H₁₅ClN₂. Furthermore, the absorbance did not change significantly when the concentration of C₁₂H₁₅ClN₂
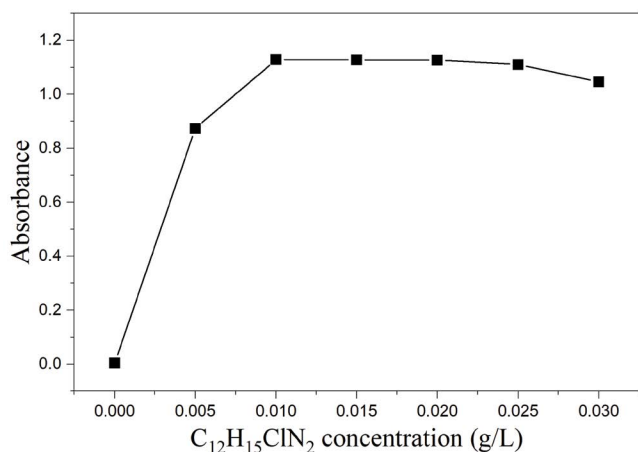
Fig. 4. Dependence of absorbance on $C_{12}H_{15}ClN_2$ concentration. Other reagents concentration: 24 $g·L^{-1}$ NaOH; 560 $\mu g·N·L^{-1}$ ammonia nitrogen; 0.0112 $g·L^{-1}$ $KBrO_3$; 1 $g·L^{-1}$ $C_6H_8O_2N_2S$; oxidation time: 20 min.

Fig. 5. Dependence of absorbance on oxidation time. Reagents concentration: 24 $g·L^{-1}$ NaOH; 560 $\mu g·N·L^{-1}$ ammonia nitrogen; 0.0112 $g·L^{-1}$ $KBrO_3$; 1 $g·L^{-1}$ $C_6H_8O_2N_2S$; 0.010 $g·L^{-1}$ $C_{12}H_{15}ClN_2$.

exceeded 0.010 $g·L^{-1}$. Thus, 0.010 $g·L^{-1}$ was chosen as the optimum concentration of $C_{12}H_{15}ClN_2$ for all experiments.

3.2.5. Oxidation time

For rapid detection of ammonia nitrogen, a short oxidation time is required, while a sufficient amount of time is needed for complete oxidation. Therefore, it is necessary to optimize oxidation time.

As can be seen in Fig. 5, no further changes occurred in absorbance after 20 min, indicating that the solution had completely developed its color. As a result, 20 min was considered optimal.

Based on the above observations, the following conditions were eventually chosen for further determination of ammonia nitrogen: 24 $g·L^{-1}$ of NaOH, 0.0112 $g·L^{-1}$ of $KBrO_3$, 1.0 $g·L^{-1}$ of $C_6H_8O_2N_2S$, 0.01 $g·L^{-1}$ of $C_{12}H_{15}ClN_2$ and 20 min of oxidation time.

3.2.6. Linearity

To detect ammonia nitrogen, an evaluation of the linearity, precision, and sensitivity of the method was conducted. Under the optimal conditions, the absorption spectra of the resulting solutions at different ammonia nitrogen concentrations are shown in Fig. 6.

A calibration curve, shown in Fig. 7, was plotted for ammonia nitrogen by using a series of standard solutions.

As shown in Fig. 7, a linear relationship between ammonia nitrogen concentration and absorbance was observed when the ammonia nitrogen concentration was between 0–448 $\mu g·N·L^{-1}$. There was a linear equation defined by $y = 0.00259x - 0.00201$, with $R^2 = 0.999$. When the ammonia nitrogen concentration was greater than 448 $\mu g·N·L^{-1}$, absorbance did not increase proportionally with increasing ammonia nitrogen concentration. The detection range of the method with $KBrO_3$ as oxidant was wider than that in the published literature [28]. The limit of detection for ammonia nitrogen, determined using the standard
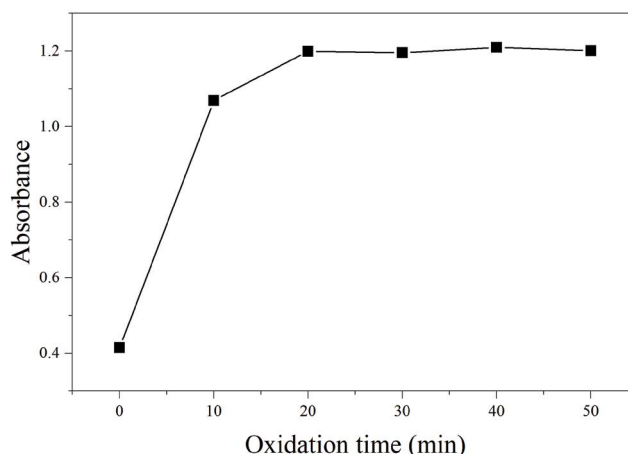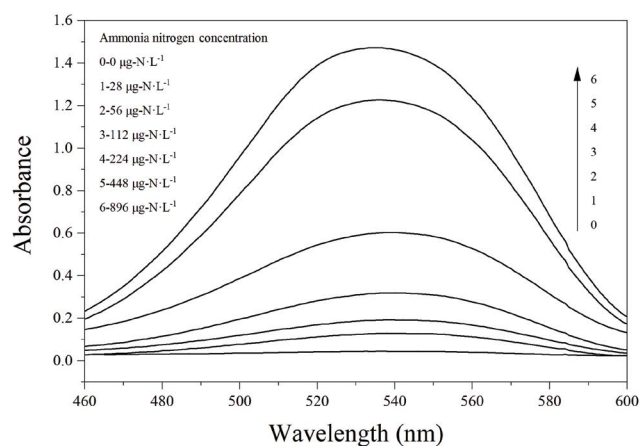

Fig. 6. Absorption spectra of resulting solutions under different ammonia nitrogen concentrations.

Table 1
Tolerance limits of interfering ions in the determination of 28 $\mu g·N·L^{-1}$ of ammonia nitrogen

Coexisting ion	Foreign ion to analyte ratio
Na^+, Ca^{2+}, K^+, Mg^{2+}, HPO_4^{2-}, $H_2PO_4^-$, SO_4^{2-}	2,000:1
NO_3^-	1,500:1
F^-	1,000:1
Cd^{2+}	200:1
Cu^{2+}	100:1

deviation of the response (σ) and the slope of the calibration curve (S) [29], was 15.98 $ng·N·L^{-1}$.

3.2.7. Selectivity

In order to verify the selectivity of the proposed method, it has been examined the influence of various substances

Table 2
Determination of ammonium nitrogen in natural water samples based on the proposed method

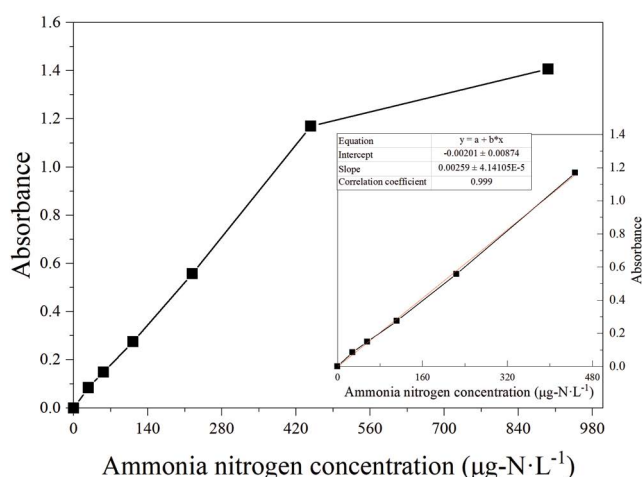| Sample	Added $C_{NH_4^+-N}$ ($\mu g \cdot N \cdot L^{-1}$)	Found $C_{NH_4^+-N}$ ($\mu g \cdot N \cdot L^{-1}$)	Relative standard deviation (%)	Recovery (%)
Natural water sample	56	54.3	0.4516	97.05
	84	80.7	0.3096	96.13
	224	222.8	0.1685	99.47
	280	281.6	0.0691	100.58

Fig. 7. Calibration curve of the ammonia nitrogen solution.

commonly present in water samples on the determination of 28 $\mu g \cdot N \cdot L^{-1}$ ammonia nitrogen using the developed method. An error of less than ±10% was defined as the maximum amount of interfering ion allowed. The results are given in Table 1. It can be seen that most of the examined ions did not interfere with ammonia nitrogen determination, indicating that the proposed method is highly selective.

3.2.8. Application

To explore the reliability of the proposed method, the method was successfully applied for the determination of ammonia nitrogen in water samples from Ming Lake at Changzhou Institute of Technology. The ammonia nitrogen concentration of Ming Lake water was 55.4 $\mu g \cdot N \cdot L^{-1}$. Furthermore, recovery tests were also carried out by the standard addition method. First, four levels of known ammonium standard solutions were added to a fixed amount of actual water samples, respectively. Next, each mixture was analyzed according to the proposed procedure. The results are given in Table 2.

It was noted that the relative standard deviation was less than 0.45%, suggesting this method was highly precise and accurate. Besides, the recovery rate ranged from 96.13% to 100.58%, which also indicated the good accuracy of the method.

4. Conclusions

A simple, sensitive, accurate and highly selective method for determining ammonia nitrogen in water has been established. In particular, $KBrO_3$ in an alkaline environment was used as the oxidant. Compared to hypobromite or hypochlorite, the new oxidant $KBrO_3$ is easily accessible and more stable. On the one hand, due to $KBrO_3$'s stability, the oxidizing solution can be prepared less frequently, thus reducing the overall workload. On the other hand, the reliability and reproducibility of the method were also promoted. Furthermore, the actual sample analysis showed that the proposed method was highly selective and sensitive to ammonia nitrogen. The proposed method was appropriate for the detection of ammonia nitrogen in natural water.

Author contributions

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Fuhua Jiang and Jiayi Tao. The first draft of the manuscript was written by Fuhua Jiang, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Acknowledgements

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