

## PYROGALLOL AS A REAGENT FOR SPECTROPHOTOMETRIC DETERMINATION OF AMMONIUM-NITROGEN ( $\text{NH}_4^+\text{-N}$ ) IN AQUEOUS SOLUTION

P O Okolo\* and A Jideonwo

Chemistry Department, University of Benin, Benin City, Nigeria

(Received 6 December 2001; accepted 22 May 2002)

Pyrogallol (1,2,3-trihydroxybenzene) is proposed as a reagent for the spectrophotometric determination of ammonium-nitrogen ( $\text{NH}_4^+\text{-N}$ ) in aqueous solution. Pyrogallol forms instantaneous pale-yellow complex with aqueous  $\text{NH}_4^+\text{-N}$ . The pyrogallol/ammonium-nitrogen (P/ $\text{NH}_4^+\text{-N}$ ) complex has absorption maximum ( $\lambda_{\text{max}}$ ) at 400nm and molar absorptivity ( $\epsilon$ ) of  $9.706 \times 10^2 \text{ l mol}^{-1} \text{ cm}^{-1}$ . Beer's law is obeyed up to  $5 \mu\text{g NH}_4^+\text{-N ml}^{-1}$ . The correlation coefficient ( $r$ ) is 0.9983. The optimum pyrogallol concentration and optimum pH are found to be  $0.06\%(\text{mv}^{-1})$  and 7.00, respectively. The method sensitivity is  $0.063 \mu\text{g NH}_4^+\text{-N ml}^{-1}$ . The extent of ionic interference in the use of the proposed method has also been quantitatively evaluated. The most tolerated ions were  $\text{K}^+$  and  $\text{Cl}^-$  ( $7.90 \times 10^{-3} \text{ mol l}^{-1}$  maximum) and the least tolerated ion was  $\text{Al}^{3+}$  ( $3.12 \times 10^{-7} \text{ mol l}^{-1}$  maximum). Recovery of  $\text{NH}_4^+\text{-N}$  from spiked water samples shows good result (98.77% to 101.94%). The proposed method has been applied to the determination of  $\text{NH}_4^+\text{-N}$  in some potable waters. Results are in good agreement with those obtained by a standard spectrophotometric method (Nessler's method).

**Key words:** Ammonium-nitrogen determination, Aqueous solution, Spectrophotometry, Pyrogallol.

### Introduction

Different methods have been developed for the determination of ammonium-nitrogen ( $\text{NH}_4^+\text{-N}$ ) in aqueous solutions. Some of them (e.g. Kjeldahl, distillation and titration methods), published in Standard Methods by APHA, AWWA, WPCF (1989) and also by Wagner (1940), are insensitive below  $5 \mu\text{g NH}_4^+\text{-N ml}^{-1}$  and inapplicable to the quantitative analysis of  $\text{NH}_4^+\text{-N}$  in drinking waters and clean surface waters which normally contain low  $\text{NH}_4^+\text{-N}$  concentrations. Others, e.g. ammonia selective electrode, phenate, and Nessler's methods, (Vogel 1961; Rossum and Villarruz 1963; Weather 1967; APHA, AWWA, WPCF 1989) are applicable to lower  $\text{NH}_4^+\text{-N}$  concentrations ( $> 5.0 \mu\text{g NH}_4^+\text{-N ml}^{-1}$ ) but often are either cumbersome or expensive.

Pyrogallol, which has not been previously reported as a reagent, is proposed here as a reagent for simple and accurate spectrophotometric determination of  $\text{NH}_4^+\text{-N}$  in aqueous solution.

The method was extended to other polyphenols, e.g. quinol (1,4-dihydroxybenzene) and the results will be published elsewhere.

### Experimental

**Apparatus.** A Unicam SP500 series ultraviolet and visible spectrophotometer with matched 1-cm silica cells was used for absorbance measurements. A model 3020-pH meter (Jenway) was employed for pH measurements.

**Reagents.** All chemicals were of analytical-reagent grades from BDH.

**Stock ammonium-nitrogen solution.** ( $0.06\text{M}$  or  $848.505 \mu\text{g ml}^{-1}$  in  $\text{NH}_4^+\text{-N}$ ). Dissolve 1.0g di-ammonium hydrogen orthophosphate,  $(\text{NH}_4)_2\text{HPO}_4$ , in distilled water and dilute to 250ml.

**Calibration solution.** (0.0, 0.5, 1.0, 2.0, 3.0, 4.0, 5.0, and 6.0  $\mu\text{g NH}_4^+\text{-N ml}^{-1}$ ). Dilute 0.00, 0.01, 0.03, 0.06, 0.09, 0.12, 0.15 and 0.18ml of stock ammonium-nitrogen solution in 25ml calibration flasks with distilled water.

**Pyrogallol solution ( $1.0\% \text{ mv}^{-1}$  or  $0.0794\text{M}$ ).** Dissolve 1.0g pyrogallol (M W 126.11) in water and dilute to 100ml with distilled water.

1M HCl or 1M NaOH was used for pH adjustments of the reaction mixtures.

**Recommended procedures.** Pipette out 0.03, 0.06, 0.09, 0.12, 0.15, 0.18, 0.21, 0.24, 0.27 and 0.30ml of the stock  $\text{NH}_4^+\text{-N}$  solution into 25-ml calibrated flasks. Then add 1.50ml of the pyrogallol solution. Make up to mark with distilled water. These are the reaction mixtures for calibration curve. Also prepare a reagent blank. Allow both reaction mixture and reagent blank to stand for 35 min. Adjust the pH of both to 7.00 using 1M HCl or 1M NaOH solutions. Take the absorbance reading of each reaction mixture versus reagent blank at 400nm in matched 1 cm silica cells. All processes take place at ambient temperatures ( $25\text{-}30^\circ\text{C}$ ).

\*Author for correspondence

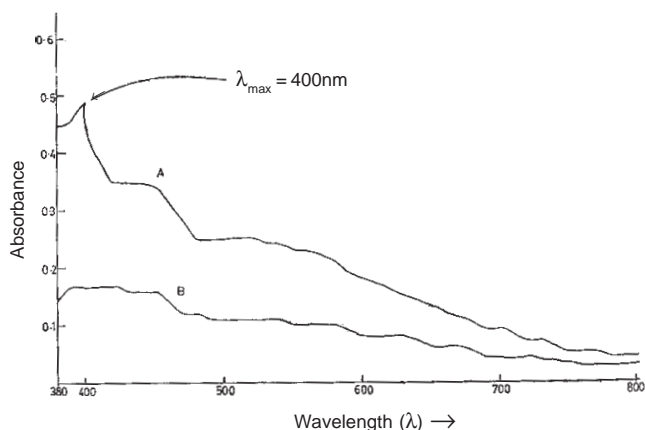
**Sample (test) solution.** Pipette 5-20ml of sample solution, containing 25-125  $\mu\text{g NH}_4^+\text{-N}$ , into a 25-ml calibrated flask. Add 1.5ml of the pyrogallol solution and make this volume up to mark with distilled water. Procedure for color development, pH adjustment, ambient temperatures and absorbance readings are as stated under calibration curve.

## Results and Discussion

**Absorption spectra.** The absorption spectra (Kunihiro *et al* 1995; Demetrius *et al* 1995) of the pyrogallol/  $\text{NH}_4^+\text{-N}$  complex as well as the reagent blank are shown in Fig1. The maximum absorbance of the reaction mixture vs reagent blank (A) was far larger than that for reagent blank vs distilled water (B). The wavelength of maximum absorbance,  $\lambda_{\text{max}}$ , for the former was located at 400nm.

**Effect of pyrogallol concentration.** Fig 2 shows that at pH 7.0 and when  $\text{NH}_4^+\text{-N}$  concentration is 5.0  $\mu\text{g ml}^{-1}$  and pyrogallol concentration is varied from 0.005 to 0.7%  $\text{m v}^{-1}$ , the absorbance at 400nm increased from 0.28 (for 0.005%  $\text{m v}^{-1}$  pyrogallol) to a peak of 0.43 (for 0.06%  $\text{m v}^{-1}$  pyrogallol) and then decreased to almost a constant value of 0.27 (from 0.20 – 0.70 %  $\text{m v}^{-1}$  pyrogallol). Pyrogallol concentration of 0.06%  $\text{m v}^{-1}$  (or 1.50 ml of 1%  $\text{m v}^{-1}$  pyrogallol solution in 25ml final volume) is therefore chosen as the optimum pyrogallol concentration. This is a molar excess of 10.1 of pyrogallol over  $\text{NH}_4^+\text{-N}$ .

**Effect of pH.** The effect of pH on absorbance is investigated when  $\text{NH}_4^+\text{-N}$  and pyrogallol concentrations are 5.0  $\mu\text{g ml}^{-1}$  and 0.06%  $\text{m v}^{-1}$ , respectively. Absorbance reading are taken at 400nm in matched 1cm silica cells. From Fig 3, absorbance values are almost steady at about 0.29 for pH values of

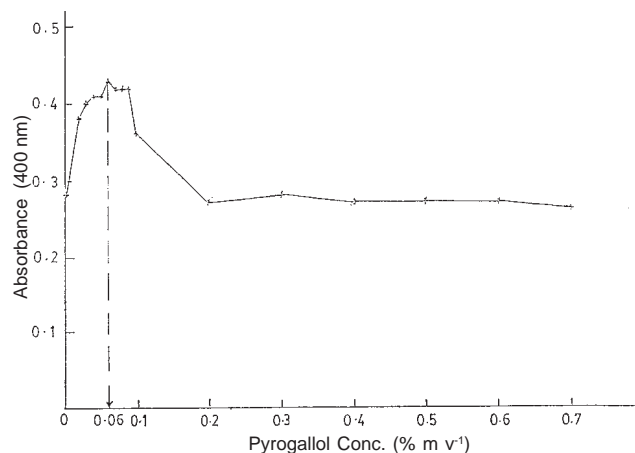


**Fig 1.** Absorption spectra for pyrogallol/ $\text{NH}_4^+\text{-N}$  Complex. Reaction mixture (curve A):  $\text{NH}_4^+\text{-N}$ , 5.0  $\mu\text{g ml}^{-1}$ ; pyrogallol, 0.06%  $\text{m v}^{-1}$ ; pH 7.0; Final volume, 25 ml; Reagent blank (curve B): same as in reaction mixture except for  $\text{NH}_4^+\text{-N}$ .

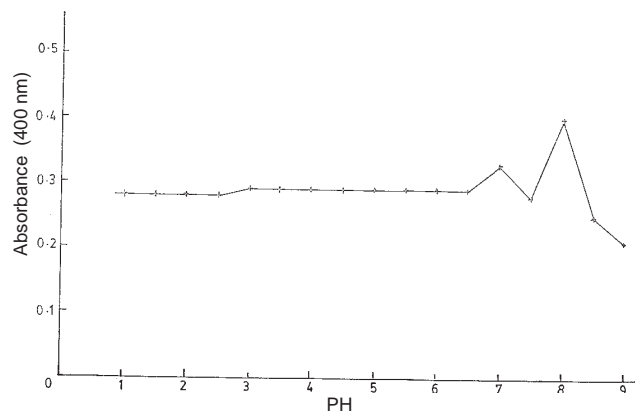
1.0 – 6.5. Two absorbance peaks were outstanding at 0.33 and 0.40 corresponding to pH of 7.0 and 8.0 respectively. Further investigations revealed that at pH 8.0 the absorbance of the reagent blank was large (absorbance of the reagent blank vs water = 0.7 at 400nm) and both reaction mixture and reagent blank were unstable (they lasted for about 20 min). While at pH of 7.0, the reagent blank had minimal absorbance at 400nm (absorbance of reagent blank vs water = 0.17 at 400nm), and both reaction mixture and the reagent blank were more stable (they lasted for 2 h). Therefore, the optimum pH for this method is 7.0.

**Effect of reaction time.** Fig 4 shows that, when the concentrations of pyrogallol and  $\text{NH}_4^+\text{-N}$  are 0.06%  $\text{m v}^{-1}$  and 5.0  $\mu\text{g ml}^{-1}$  respectively and pH is fixed at 7.0, the color developed after 5 min and reached a maximum after 35 min. Therefore duration of 35 min is chosen as the maximum for color development.

Further investigations show that, after attaining the maximum absorbance, the reaction mixture and the reagent blank remained stable for 2h.



**Fig 2.** Effect of Pyrogallol conc. on absorbance.



**Fig 3.** Effect of pH on absorbance.

**Order of addition of reagents.** The preferred order of reagent addition is as follows: first, pipette 0.15ml of stock ammonium-nitrogen solution into a 25 ml calibrated flask. Then add 1.50 ml of the pyrogallol solution and finally dilute to 25 ml mark with water. Allow 35 min for maximum color development and adjust pH to 7.00 with 1M HCl or 1M NaOH.

**Figures of merit.** A series of standard ammonium-nitrogen solutions were prepared. When the pyrogallol concentration was 0.06% m v<sup>-1</sup> and the total final volume and pH were 25ml and 7.00 respectively, Beer's Law was obeyed up to 5.0 µg ml<sup>-1</sup> NH<sub>4</sub><sup>+</sup>-N (Fig 5). The stoichiometry of P/NH<sub>4</sub><sup>+</sup>-N complex is 1:2, that is P(NH<sub>4</sub>)<sub>2</sub> where P is pyrogallol.

The apparent molar absorptivity (ε) (Robert *et al* 1974; Donald *et al* 1979) of the P/ NH<sub>4</sub><sup>+</sup>-N complex was 9.706 x 10<sup>2</sup> l mol<sup>-1</sup> cm<sup>-1</sup>. The correlation coefficient (r) (Kealey 1986) was found to be 0.9983 which revealed a high degree of correlation

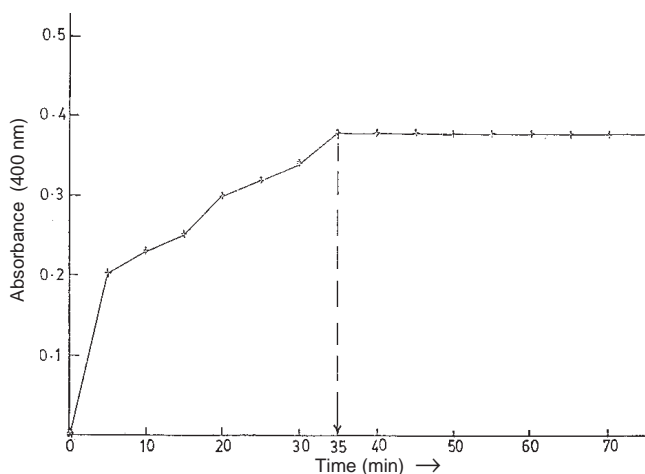


Fig 4. Effect of reaction time on absorbance.

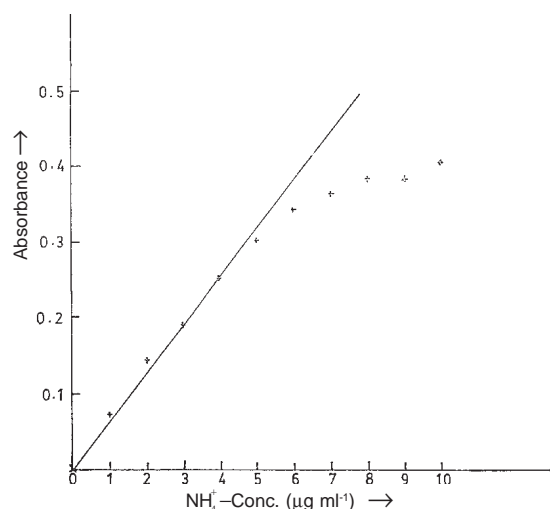


Fig 5. Adherence to Beer's law.

**Table 1**  
Recovery test

Recovery of ammonium - nitrogen	1	2	
NH <sub>4</sub> <sup>+</sup> -N	Added (µg ml <sup>-1</sup> ):	1.2728	5.0900
	Originally present (µg ml <sup>-1</sup> ):	0.0000	0.0000
	Found (µg ml <sup>-1</sup> ):	1.2975	5.0275
% Recovery:	101.94	98.77	
Std. Deviation*, S (µg ml <sup>-1</sup> ):	± 0.10	± 0.10	
Rel. std deviation, Sr (%):	8.5	1.99	
95% confidence limit (µg ml <sup>-1</sup> ):	1.2975±0.1350	5.0275±0.1243	

\*repeated 5 times

**Table 2**

Effect of foreign substances on the absorbance of an NH<sub>4</sub><sup>+</sup>-N of 5.0 µg ml<sup>-1</sup> solution

S.no.	Species	Added as	Tolerated concentration (mol l <sup>-1</sup> )	Relative <sup>+</sup> deviation(%)
1	NO <sub>3</sub> <sup>-</sup>	KNO <sub>3</sub>	2.66 x 10 <sup>-4</sup>	4.35
2	Al <sup>3+</sup>	Alk(SO <sub>4</sub> ) <sub>2</sub> .12H <sub>2</sub> O	3.12 x 10 <sup>-7</sup>	2.30
3	NO <sub>2</sub> <sup>-</sup>	NaNO <sub>2</sub>	3.00 x 10 <sup>-3</sup>	-2.30
4	V <sup>+5</sup>	V <sub>2</sub> O <sub>5</sub>	2.00 x 10 <sup>-6</sup>	-4.60
5	Na <sup>+</sup> , ClO <sub>4</sub> <sup>-</sup>	NaClO <sub>4</sub> .H <sub>2</sub> O	3.76 x 10 <sup>-4</sup>	4.36
6	As <sup>v</sup>	Na <sub>2</sub> HAsO <sub>4</sub> .7H <sub>2</sub> O	4.50 x 10 <sup>-5</sup>	-2.30
7	W <sup>vi</sup>	Na <sub>2</sub> WO <sub>4</sub> .2H <sub>2</sub> O	2.60 x 10 <sup>-6</sup>	-2.30
8	PO <sub>4</sub> <sup>3-</sup>	NaH <sub>2</sub> PO <sub>4</sub> .2H <sub>2</sub> O	5.13 x 10 <sup>-5</sup>	4.70
9	K <sup>+</sup> , Cl <sup>-</sup>	KCl	7.90 x 10 <sup>-3</sup>	4.70
10	CO <sub>3</sub> <sup>-</sup>	Na <sub>2</sub> CO <sub>3</sub> (anhy)	3.60 x 10 <sup>-5</sup>	-2.30
11	Fe <sup>3+</sup>	Fe <sub>2</sub> (SO <sub>4</sub> ) <sub>3</sub>	3.70 x 10 <sup>-3</sup>	-2.30
12	Ca <sup>2+</sup> , SO <sub>4</sub> <sup>2-</sup>	CaSO <sub>4</sub>	1.47 x 10 <sup>-4</sup>	-2.50
13	Mg <sup>2+</sup>	MgSO <sub>4</sub> .7H <sub>2</sub> O	1.62 x 10 <sup>-4</sup>	-2.17

$$^+ \text{Relative deviation (\%)} = \frac{A \text{ without interference} - A \text{ with interference}}{A \text{ with interference}} \times 100$$

**Table 3**  
Determination of NH<sub>4</sub><sup>+</sup>-N in water samples

Sample	Proposed method	Standard method
Tap water	ND <sup>++</sup>	ND <sup>++</sup>
River water	2.65	2.71
Well water	ND <sup>++</sup>	ND <sup>++</sup>

<sup>++</sup> Not detected; concentrations (µg NH<sub>4</sub><sup>+</sup>-µg ml<sup>-1</sup>) given are the average of three replicates.

(linearity) between the absorbance and analyte (NH<sub>4</sub><sup>+</sup>-N) concentration. The regression equation (Kealey 1986) for the curve is given by y = 0.054x + 0.028 where 'y' is the measured

absorbance at 400nm and 'x' the concentration of  $\text{NH}_4^+\text{-N}$  in  $\mu\text{g ml}^{-1}$ . The method sensitivity (ACS 1980; APHA, AWWA, WPCF 1989; Andres *et al* 1995; Valentinus *et al* 1995) was found to be  $0.063 \mu\text{g ml}^{-1}$ .

**Recovery test.** The accuracy and precision of the proposed method were studied by spiking water samples (deionised, well and tap waters) with  $\text{NH}_4^+\text{-N}$ , and analyzing five replicates of each, using the recommended procedures. The percent recovery (%), standard deviation (S), relative standard deviation (Sr) and confidence limits (95%) for various spiked samples are given in Table 1.

**Interference studies.** The effect of foreign ions (Sallnas *et al* 1995; Saurina and Hernandez 1995; Susanto *et al* 1995) on the absorbance of  $\text{NH}_4^+\text{-N}$  solution ( $5.0 \mu\text{g ml}^{-1}$ ) is shown in Table 2. Some of the ions chosen (e.g.  $\text{Na}^+$ ,  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ ,  $\text{Cl}^-$  or  $\text{SO}_4^{2-}$ ) are the main components of natural waters, while others, which can form ion - associates may be present as pollutants (e.g.  $\text{As}^v$ ,  $\text{V}^v$ , or  $\text{P}^v$ ). The criterion for interference was fixed at  $\pm 5.0\%$  (Saurina and Hernandez 1995), thus concentrations of foreign ions which cause relative deviations equal to or lower than 5.0% were tolerated. Most of the ions tested were tolerated in a wide range of concentrations. In all cases, the concentrations of the possible interfering species in natural waters are far lower than those found as the maximum tolerated in Table 2.

**Applications.** The proposed method was applied to determination of ammonium - nitrogen ( $\text{NH}_4^+\text{-N}$ ) in some natural and portable waters. Results were compared with those obtained by a standard spectrophotometric method - Nessler's method. As shown in Table 3, both methods are in good agreement.

## Conclusion

The proposed method provides an alternative approach to the determination of ammonium-nitrogen ( $\text{NH}_4^+\text{-N}$ ) upto  $5.0 \mu\text{g ml}^{-1}$  in aqueous solutions. The advantages include:

- The method is cheap and affordable to laboratories in under - developed countries.
- No sophisticated equipments are needed. A pH meter and a UV/visible spectrophotometer are required.
- No hazardous chemicals like mercuric chloride (used in Nessler's method) are involved.
- It is faster than distillation/titration method.

## References

ACS 1980 Committee on Environmental Improvement. *Anal Chem* **52** 2241 – 2249.  
 Andres S M P, Marina M L, Vera S 1995 Spectrophotometric determination of copper(II), nickel (II) and cobalt (II) as

complexes with sodium diethyldithiocarbamate in the Anionic Micellar Media of dodecylsulfate salts. *Analyst* **120** 256.  
 A P H A, A W W A, W P C F 1989 *Standards Methods for the Examination of Water and Waste Water*. 17th ed. pp 1-4 to 1-5; 1-18 to 1-20; 4-40 to 4-120.  
 Demetrius G T, Goerge A Z, Stratis J A 1995 Selective spectrophotometric determination of cobalt (III) using 2, 2' -dipyridyl-2-pyridylhydrazone and a flow injection manifold. *Analyst* **120** 1597.  
 Donald L P, Gary M L, George S K 1979 *Introduction to Spectroscopy*. Saunders College Publishing. West Washington Square, Philadelphia, P A 19105, p 186.  
 Kealey D 1986 *Experiments in Modern Analytical Chemistry*. Chapman and Hall, New York, pp 4 - 18.  
 Kunihiro K, Misako T, Kiyosti T, Motohiro N 1995 Spectrophotometric determination of sodium chondroitin sulphate in eye drops after derivatization with 4-amino-3-hydrazino-5-mercapto 1,2,4-triazole. *Analyst* **120** 2756.  
 Robert M S, Clayton G B, Terence C M 1974 *Spectrophotometric Identification of Organic Compounds*, 3rd ed. Wiley Int Ed, New York pp 232 – 236.  
 Rossum J R, Villarruz P A 1963 Determination of ammonia by the indophenol method. *J Amer Water Works Ass* **55** 57.  
 Sallnas F, Espinosa A, Lopez-de-Alba P L 1995 Extraction - spectrophotometric determination of silver in ores. electronics flowsoldiers and white metals with 2-carboxybenzaldehyde thiosemicarbazone. *Analyst* **120** 2859.  
 Saurina J, Hernandez-Cassou S 1995 Flow injection spectrophotometric determination of silicate based on the formation of the ion associated between molybdosilicate and malachite green. *Analyst* **120** 2603.  
 Susanto J P, Oshima M, Motomizu S, Mikasa H, Hori Y 1995 Determination of micro amounts of phosphorus with malachite green using a filtration dissolution preconcentration method and flow injection - spectrophotometric detection. *Analyst* **120** 191.  
 Valentinus P B, Danielle C E, Josette A 1995 Validation of modified spectrophotometric method for the determination of nitrate in dry milk using 2-sec-butylphenol. *Analyst* **120** 2747.  
 Vogel A I 1961 *Quantitative Inorganic Analysis*, 3rd ed. Longman Group Ltd, London, pp 51, 133, 490, 503, 783 - 784, 1121 - 1128.  
 Wagner E C 1940 Titration of ammonia in the presence of boric acid. *Ind Eng Chem Anal Ed* **12** 711.  
 Weather Burn M W 1967 Phenolhypochlorite reaction for determination of ammonia. *Anal Chem* **39** 71.