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Determination of bromate in bread additives and flours by flow injection analysis

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Abstract

A new, simple, rapid and sensitive spectrophotometric flow injection analysis (FIA) method was developed for the determination of bromate based on its reaction with 3,5-dibromo-PADAP and thiocyanate in a strongly acidic medium. This produced an unstable violet product with a maximum absorption at 602 nm. The calibration curve was linear in the range of 2.00×10^{-6}–2.10×10^{-5} mol/l and the detection limit was 8.00×10^{-7} mol/l. The sampling frequency was 90 h^{-1}. The method has been successfully applied to the determination of bromate in commercial bread additives and flours. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Bread; Flours; Bromate; Flow injection analysis

1. Introduction

The breadmaking quality of freshly milled flour tends to improve with storage for a period of up to two months but this process may be accelerated by addition of chemical substances called improvers. Among other substances, the Standard of Using Food Additives of China (Ling, Wang & Tang, 1989) permits the use of potassium bromate up to a maximum level in bread of 50 mg/kg of flour mass. In 1996, The Association of Official Analytical Chemists (AOAC) committee (Warner, 1996) recommended continued study of bromates in baked products because of its suspected carcinogenic potential. So, it is necessary for the bakers to be able to analyze bread additive and flour rapidly in order to determine bromate content.

Many analytical methods such as the spectrophotometric method (Boschserrat, 1997; Roman Ceba, Jimenez Sanchez & Galeano Diaz, 1983), spectrofluorometric method (Gahr, Huber & Niessner, 1998; Gong, Jia, Qu & Wang, 1993), capillary electrophoresis (Doble, Macha & Haddad, 1998) and ion chromatography (Cox, Harrison, Jandik & Jones, 1985; Heitkemper, Kaine, Jackson & Wolnik, 1994; Lu, Mou, Tong & Riviello, 1998; Qi, Qu, Liu & Mou, 1998) have been developed for the determination of bromate. However, the methods mentioned above suffer from more or less time-consuming procedures and complicated instrumentation. Flow injection analysis (FIA) has been widely applied to determine many cations and anions. However, a few papers reported on the determination of bromate by FIA. Brian G. Osborne (Osborne, 1987) determined bromate in flour based on its reaction with acidified potassium iodine and starch. Chen Xingguo et al. (Xingguo, Xingwei, Zhide & Zongyan, 1990) proposed a procedure for determining this anion in chemical reagent by FIA.

FIA has undergone extensive development since 1975. It offers various advantages that led to a rapid growth in popularity. One of the important trends observed in FIA is the growing interest in the exploitation of unstable chemical reactions for quantitative assay using FIA techniques (Fang, Sun & Xu, 1992). The precise timing characteristic of FIA makes it ideally suited for such proposes.

In this paper, we report a new FIA method for the direct determination of bromate, that based on an unstable violet product with a maximum absorption at 602 nm produced by the reaction of bromate, thiocyanate and 2-(3,5-dibromo-2-pyridylazo)-5-diethylaminol (3,5-dibromo-PADAP) in a strongly acid medium. And

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it has been applied to the determination of bromate in commercial bread additives and flours with good precision, accuracy, selectivity and simple procedures.

2. Experimental

2.1. Apparatus

The FIA system used in this work was a Tecator FIAstar 5020 analyzer with a Tecator Chemifold II, as shown schematically in Fig. 1. The detector was a 5023 FIAstar spectrophotometer consisting of the 5032 Detector Controller and 5023-011 spectrophotometer unit. Teflon tubes (i.d. 0.5 mm) and connectors were used to build the manifold. Sample solutions were aspirated into 100 μl sample loop and then injected into a carrier stream. The gain factor was set at 1.00. The change in the absorbance was monitored with a 1-cm path flow cell (inner volume 18 μl) at 602 nm.

A model 721 spectrophotometer (Shanghai Third Analytical Instrument Factory, China) was used in the measurement of the absorbance as a function of time.

2.2. Reagents and chemicals

All reagents used were of analytical grade. Distilled water was used throughout. 2-(3,5-dibromo-2-pyridylazo)-5-diethylaminol (3,5-dibromo-PADAP, The Institute of Tianjin Chemical Reagents, Tianjin, China), 4.67×10^{-4} mol/l. Prepared by dissolving 0.0200 g of the reagent in 100 ml ethanol.

Potassium bromate standard stock solution (0.0100 mol/l) was prepared by dissolving 0.4175 g of potassium bromate in distilled water and then diluted to 250 ml. Working standard solutions were prepared by appropriate dilution before use.

Potassium thiocyanate solution (0.10 mol/l) was prepared in distilled water.

H_2SO_4 solution (9.0 mol/l) was prepared in distilled water.

2.3. Procedure

The FIA conditions were given in Table 1. Carrier stream (C) and reagent stream (R1 and R2) were pumped into the flow system as shown in Fig. 1, and the sample can not be injected into the flow system until a steady baseline has been obtained. Sample concentration was evaluated directly from a calibration curve obtained under the same conditions.

3. Results and discussion

3.1. The stability of the product

As mentioned above, this method is based on the unstable violet product produced by the reaction of bromate, thiocyanate and 3,5-dibromo-PADAP in a strongly acidic solution. The plot of the absorption of the violet product vs. time was shown in Fig. 2. As can be seen from Fig. 2, the violet product is not stable. Therefore, the reaction can not be used for achieving reproducible quantitative analyses with a batch approach. However, the precise timing characteristic of FIA makes it ideally suited for the determination of bromate by the proposed method.

3.2. Effects of different kinds of acids and acidity on sensitivity

Preliminary tests showed that H_2SO_4 is the most effective acids tested: H_2SO_4, HCl and HNO_3. Hence H_2SO_4 was selected as the medium in this work.

Fig. 3 showed that the peak height was found to increase rapidly with increasing concentration of H_2SO_4 up to 4.5 mol/l and then slightly increased. So, a H_2SO_4 concentration of 4.5 mol/l was used in the subsequent experiments.

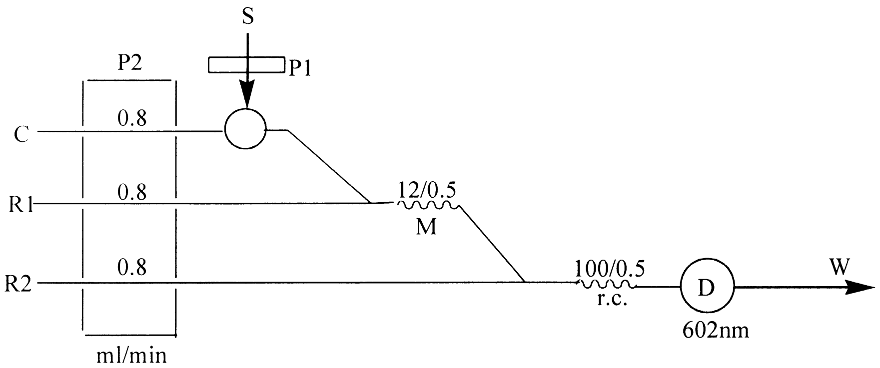

Fig. 1. Schematic diagram of the flow-injection system for the determination of bromate. P: peristaltic pump; C: distilled water; R1: color reagent; R2: KCNS; M: mixed coil; r.c.: reaction coil; D: detector; S: sample; W: waste.

Table 1
Conditions for FIA

Composition and flow rate of C	Distilled water, 0.80 ml/min
Composition and flow rate of R1	9.34×10^{-5} mol/l 3,5-dibromo-PADAP and 4.5 mol/l H_2SO_4 mixture solution, 0.80 ml/min
Composition and flow rate of R2	0.040 mol/l thiocyanate, 0.80 ml/min
λ	602 nm
Gain factor	1.00
Sample volume	100 µl
Mixing coil length	12 cm
Reaction coil length	100 cm

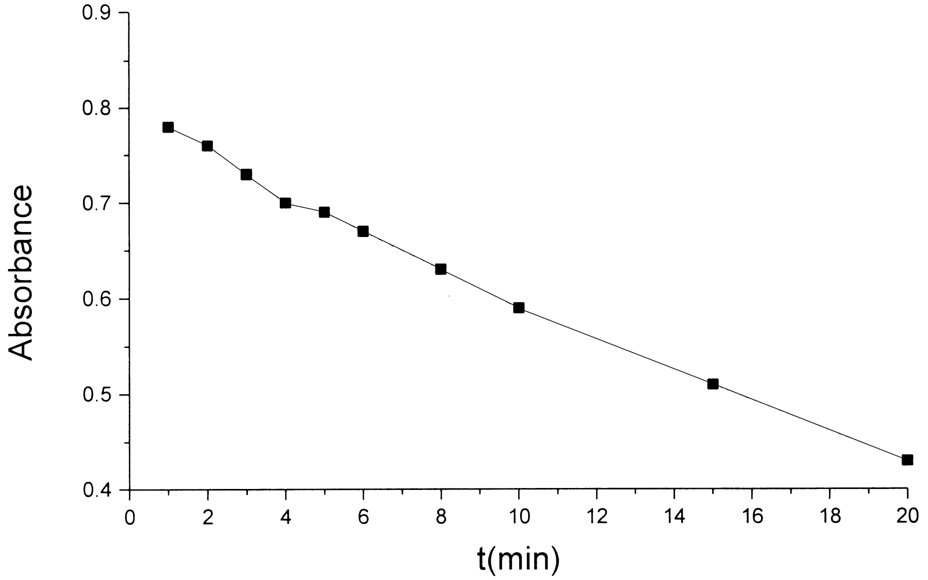

Fig. 2. The plot of the absorption of violet vs. time.

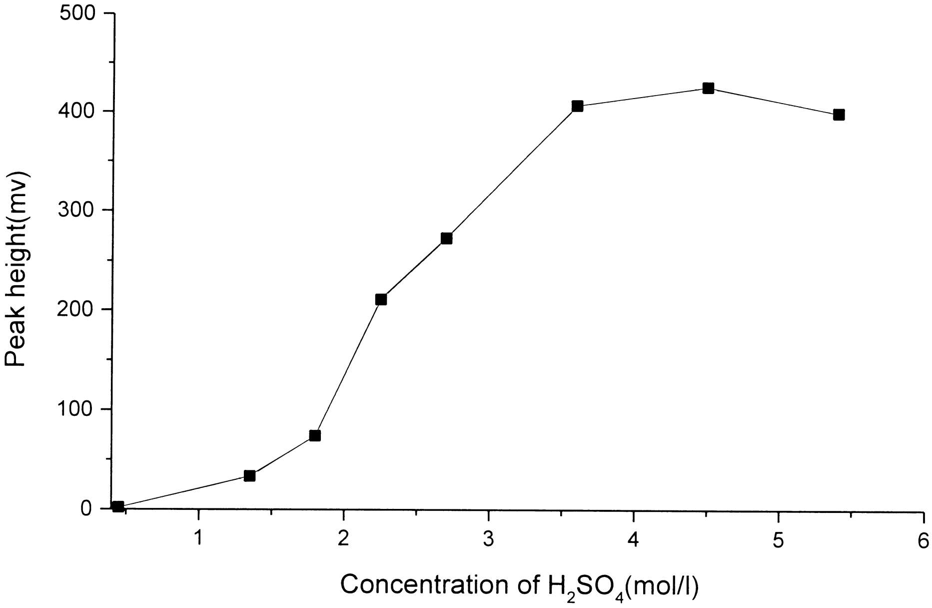

Fig. 3. Influence of H_2SO_4 concentration.

3.3. Effects of the concentration of 3,5-dibromo-PADAP and thiocyanate

The effect of concentration of 3,5-dibromo-PADAP on sensitivity was evaluated in the range 2.33×10^{-5}–1.87×10^{-4} mol/l. The results obtained were shown in Fig. 4. Fig. 4 showed that the peak height was independent of the 3,5-dibromo-PADAP concentration in the range 9.34×10^{-5}–1.87×10^{-4} mol/l. Hence, a working concentration of 9.34×10^{-5} mol/l was chosen for further experiments.

It was shown from Fig. 5, the peak height increased rapidly with increasing the concentration of thiocyanate in the range 2.50×10^{-3}–2.00×10^{-2} mol/l. When the concentration of thiocyanate exceeded 5.00×10^{-2} mol/l, the signal decreased slightly and a steady baseline can not easily be obtained. So, a thiocyanate concentration of 4.00×10^{-2} mol/l was selected.

3.4. Operative conditions for FIA

It was shown from the experimental results that the longer reaction coil, the higher is the sensitivity. However, using too long a reaction coil the sensitivity will decrease because of higher dispersion. In addition, the

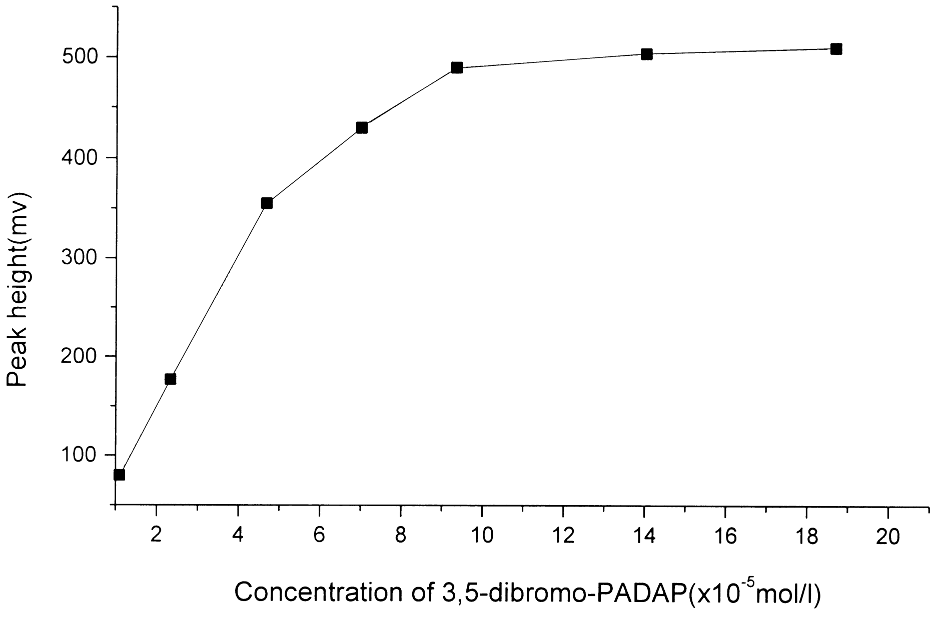

Fig. 4. Influence of 3,5-dibromo-PADAP concentration.

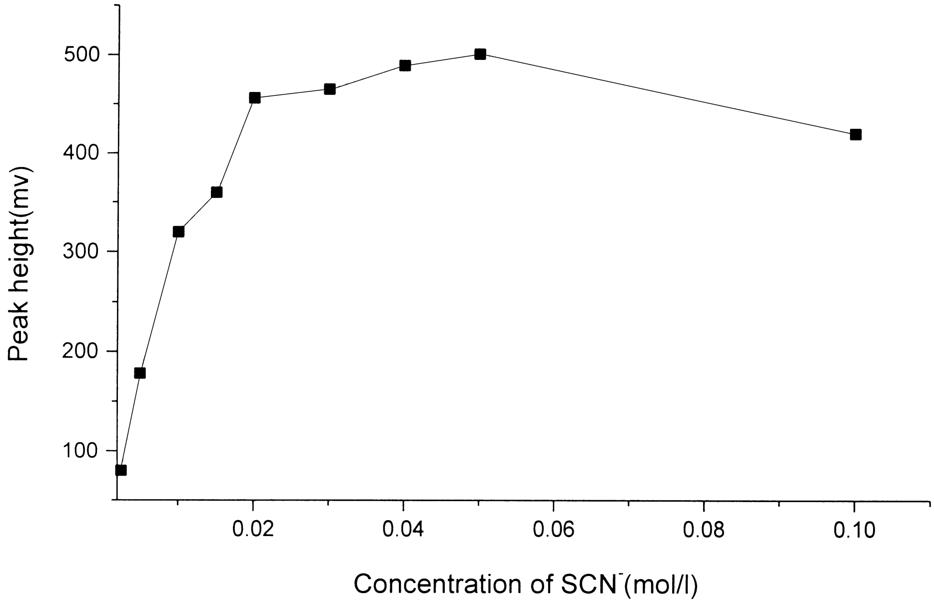

Fig. 5. Influence of thiocyanate concentration.

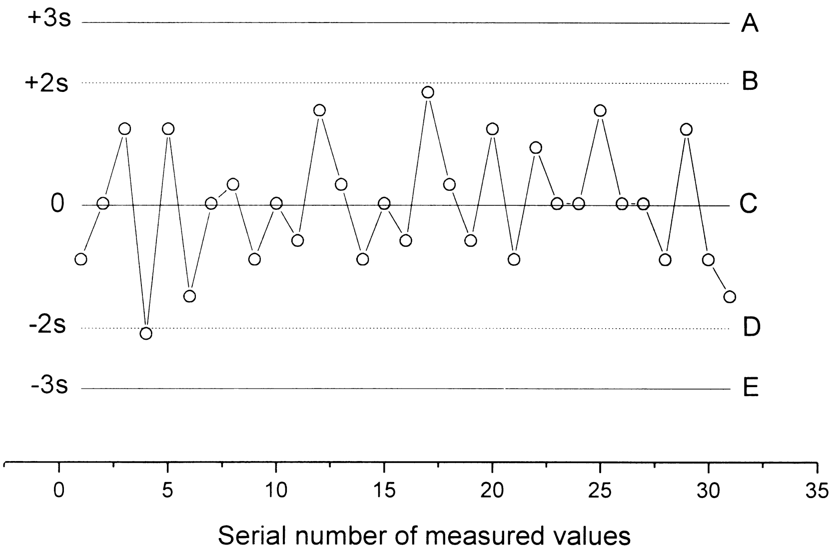

Fig. 6. Control chart. A: upper alarm limit ($\bar{x}+3s$), B: upper control limit ($\bar{x}+2s$), C: center line ($\bar{x}$), D: lower control limit ($\bar{x}-2s$), E: lower alarm limit ($\bar{x}-3s$).

sensitivity is not sensitive to variations in the length of mixing coil. For the above reasons, 100 and 12 cm were used as the length of reaction coil and mixing coil, respectively. In a certain range, the sensitivity increases with increasing sampling volume. 100 µl was chosen as sampling volume because it provided the best compromise between sensitivity and sampling frequency. The operative conditions for FIA were as follows: total flow rate of 2.4 ml/min, injection volume of 100 µl, reaction coil length of 100 cm and mixing coil length of 12 cm. The sampling frequency was 90 h^{-1}. The optimization of the operative conditions ensures the maximum signal to noise ratio.

In this work, the calibration graph was linear over the range of 2.00×10^{-6}–2.10×10^{-5} mol/l and the detection limit was 8.00×10^{-7} mol/l which was calculated as three times the standard deviation of noise. The regression calibration equation obtained under optimum conditions was $H=44.6507+2.3631\times10^{-7}C$ ($n=11$, $P<0.0001$), where H and C represent the peak height (mv) and concentration (mol/l), respectively. The correlation coefficient of the calibration curve was 0.9998.

3.5. Precision

The precision was evaluated by measuring the peak height of bromate for 1.80×10^{-5} mol/l. The relative standard deviation was 0.69% in the same day (7 times under the experimental conditions) and 1.74% for day to day. In addition, variability of signal generated in different days was drawn in a control chart (Fig. 6).

3.6. Interference

The effect of interfering ions on the determination of 1.00×10^{-5} mol/l bromate was shown in Table 2. The criterion used for interfering was a ±5% deviation of expected signal.

Table 2
Effect of some foreign ions on the determination of 1.00×10^{-5} mol/l bromate[a]

Interfering ions	Tolerated concentration (µg/ml)
F^-, Cl^-, NO_3^-, $H_2PO_4^-$, Zn^{2+}, tartrate	5000
Citrate	4000
Mg^{2+}	3000
Br^-	2000
Al^{3+}, NH^{4+}, Mn^{2+}	1000
Cu^{2+}, Ni^{2+}	300
Co^{2+}, $C_2O_4^{2-}$	200
Hg^{2+}	100
NO_2^-	5
I^-, Fe^{3+}	2
Cr^{3+}	1.5
IO_3^-, $Cr_2O_7^{2-}$	0.5

[a] Deviation of ±5% was allowed in the peak height.

Table 3
Results of the recovery

No.	Added (µg/g)		Found[a] (µg/g)		Recovery (%)	
1	5.76	9.60	5.56	9.64	96.0	100.4
	12.16	14.72	12.16	14.68	100.0	99.7
2	3.84	7.68	3.84	7.88	100.0	102.7
	10.24	12.80	10.48	12.72	102.4	99.4
3	5.12	8.96	4.76	8.68	93.0	96.8
	11.52	14.08	11.22	13.64	97.4	96.9

[a] Average of 7 determinations.

Table 4
Analysis of bromate in bread additives and flours

Samples	Amount of bromate[a] (mg/g)	Added (mg/g)	Found[a] (mg/g)	Recovery (%)
Bread additive	84.00	56.00	53.20	95
		12.0	12.12	101
Bread additive	16.00	2.56	2.41	94
		5.12	5.22	102
Bread additive	33.00	15.00	14.40	96
		30.00	27.90	93
Flour[b]	31.20	30.00	28.80	96
		60.00	63.60	106
Flour[b]	35.30	30.00	28.20	94
		60.00	57.60	96
Flour[b]	40.10	40.00	38.40	96
		80.00	80.80	101

[a] Average of 7 determinations.

[b] Expressed as µg/g.

3.7. Applications

The proposed method has been applied to check the percentage recovery of bromate from untreated unblended flour that was produced by different factories. Different amounts of potassium bromate were added to a 5.0 g sample of flour then 25.00 ml water added. The mixture was subjected to ultrasonic extraction at room temperature for 10 min followed by centrifugal separation. The supernatant was filtered and percentage recovery was determined directly by the proposed method. The results were given in Table 3.

The method described in this work was successfully applied to the determination of bromate in commercial bread additives and flour samples. Bread additives extract was prepared by vigorous shaking of 0.5000 g of the sample for 5 min with 100 ml of distilled water, then diluted to 250 ml and followed by centrifugation. The supernatant was filtered and analyzed. Five gram flour samples were weighed and the analytical procedure was the same as untreated unblended flour described above. The results were summarized in Table 4.

4. Conclusions

The reaction exploited in this work is unstable and difficult to use for achieving reproducible analytes with a batch approach. However, the adaptation of the reaction of a FIA system proved to be fairly simple. An excellent precision of 0.67% (R.S.D.) was achieved at a sampling frequency of 90 h^{-1}, giving a detection limit of 8.00×10^{-7} mol/l. We wish to emphasize that this newly devised method is simple, sensitive and reliable and applicable for the measurement of the bromate content in commercial bread additives and flours.

Acknowledgements

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