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A Highly Sensitive and Selective Spectrofluorimetric Method for the Determination of Nitrite in Food Products

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Abstract This paper describes a simple, sensitive, and selective spectrofluorimetric method to determine trace amount of nitrite in food products using 4-amino-5-methylamino-2′,7′-difluorofluorescein diacetate (DAF-FM DA) as a fluorescent probe. The reaction of DAF-FM DA with nitrite in acidic medium resulted in triazolofluorescein (DAF-FM T), a highly fluorescent reagent in neutral medium. The fluorescence enhancement was proportional to nitrite concentration in the range of 5.0×10^{-8} to 1.5×10^{-6} mol L^{-1} with a detection limit of 3.3×10^{-8} mol L^{-1} (S/*N*=3). The proposed method has been applied to the determination of nitrite in real food samples, with relative standard deviation (RSD) (*n*=6) less than 5.1 % and recoveries in the range of 86.4~102.9 %.

Keywords Nitrite · Spectrofluorimetric · Determination · 4-Amino-5-methylamino-2′,7′-difluorofluorescein diacetate (DAF-FM DA)

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Introduction

Nitrite, a chemically active substance, exists widely in the environment and food products. In order to suppress the propagation of *Clostridium botulinum* and to improve the flavor of meat, it has been widely used in meat preservation and processing (Li et al. 2003). Excessive amount of nitrite in food can be fatal, especially for pregnant women and infants. It can interfere with the body's oxygen delivery system, decreasing the ability of hemoglobin to carry oxygen (Seike et al. 2004; Cotton and Wilkinson 1988). It can also react with the secondary amines and amides in the stomach to form carcinogenic *N*-nitrosamines (Burden 1961). These hazards of nitrite make its determination and quantification of great interest.

A large number of analytical methods for the detection of nitrite have been developed, including spectrophotometric (Burakham et al. 2004; Nagaraja et al. 2010; Senra-Ferreiro et al. 2010; Tsikas 2007), electrochemical (Menart et al. 2015; Azad et al. 2014; Ramdane-Terbouche et al. 2014; Wang et al. 2013), capillary electrophoresis (Della Betta et al. 2014; Troška et al. 2013; Merusi et al. 2010; Wang et al. 2012b; Tanaka et al. 2004), chromatographic (Wang et al. 2012a; He et al. 2011; Akyüz and Ata 2009; Pagliano et al. 2014), and spectrofluorimetric methods (Lee and Field 1984; Guo et al. 2013; Liu et al. 2009; Huang et al. 2006; Wang et al. 2000). Each of them has its own merits, but each method also offers some drawbacks. Spectrophotometry is the most widely used method for the determination of nitrite. The most common approach to the spectrophotometric detection of nitrite is the Griess method which involves a diazotization-coupling procedure. However, this method suffers from poor sensitivity and interference from other chromophores and anions. Chromatography and capillary electrophoresis are always expensive and time-consuming; thus, the wide utilization of these methods is largely limited.

In comparison to these methods, spectrofluorimetry possesses more merits due to its convenience, simplicity, high sensitivity and selectivity, low limits of detection, and low-cost. Spectrofluorimetric methods are mainly based on the reaction of nitrite with various fluorescent probes such as 5-aminofluorescein (Axelrod and Engel 1975), 4-hydroxycoumarin (Ohta et al. 1986), unsymmetrical rhodamine (Liu et al. 2009), and murexide (Biswas et al. 2004). Probably the most successful fluorescent probes for detection of nitric oxide (NO) and nitrite have been aromatic vicinal diamines such as 5,6-diamino-1,3-naphthalene disulphonic acid (DANDS) (Wang et al. 2000), 2,3-diaminonaphthalene (DAN) (Damiani and Burini 1986; Tarigh and Shemirani 2014; Wiersma 1970), and diaminofluoresceins (DAFs) (Kojima et al. 1998; Kojima et al. 1999). It is believed that these probes can effectively react with NO in the presence of oxygen or with nitrite ion in acidic conditions to yield highly fluorescent triazole compounds (Itoh et al. 2000; Jourd'heuil 2002; Nagano and Yoshimura 2002; Zhang et al. 2004; Zhang et al. 2003).

4-Amino-5-methylamino-2′,7′-difluorofluorescein diacetate (DAF-FM DA), firstly synthesized by Nagano and co-workers in 1999 (Kojima et al. 1999), is the latest generation of fluorescent probes for quantitative detection of nitric oxide. DAF-FM DA is even more useful than other diaminofluoresceins since it is more sensitive for NO with a detection limit of 3 nM. The fluorescence intensity of the triazole form of DAF-FM DA is essentially independent of pH above pH 5.8 (Kojima et al. 1999). Furthermore, DAF-FM DA has been commercially available and frequently used for the detection of nitric oxide in living cells (Nagano and Yoshimura 2002). To the best of our knowledge, there is no report about its application in detection of nitrite. Considering the fact that NO could be generated from nitrite under acidic conditions, we speculated that the determination of nitrite with DAF-FM DA should be applicable and valuable. In this paper, the feasibility of this speculation was confirmed. In acidic medium, DAF-FM DA reacted with nitrite ion to yield highly fluorescent triazolofluorescein (DAF-FM T) and its fluorescence enhancement was directly proportional to the concentration of nitrite ion in the solution. The detection limit was estimated to be 3.3×10^{-8} mol L^{-1} (S/*N*=3). It was demonstrated that DAF-FM DA could be applied to the determination of nitrite in real food samples such as meat products and pickled vegetables, with good precision, accuracy, and reproducibility.

Experimental

Apparatus

All fluorescence measurements were performed with a Cary Eclipse Fluorescence Spectrophotometer (Agilent Technologies, USA) equipped with a 1 cm×1 cm quartz cell. The slit widths in terms of wavelength were 5 nm for excitation and emission, respectively. A FEP20-FiveEasy Plus pH meter (Mettler-Toledo Instruments (Shanghai) Co., LTD.) was used for the measurements of the pH. A thermostat bath model WB-2000 (Changchengkegongmao Co., Zhengzhou, China) maintained at the desired temperature was used for the experiments. All solutions were prepared in ultrapure water (R=18.2 MΩ) purified by a Milli-Q Gradient system (Millipore, Molsheim, France). A Cary 50 UV–visible Spectrophotometer (Agilent Technologies, USA) with 1 cm matched quartz cell was used for measuring the absorbance. A LC-20A liquid chromatography (Shimadzu, Kyoto, Japan) with a Shimadzu LC-20AT pump, a Shimadzu SPD-20A UV–VIS detector (operated at 220 nm) and a SIL-20A autosampler was employed. A Waters Symmetry C18 column (150 mm×4.6 mm i.d., 5 μm particle size) and mobile phase consisted of 12 mM tetrabutylammonium hydroxide, 0.125 vol.% methanol, and 10 mM KH_2PO_4 (pH 7.00) at a flow rate of 1.2 mL min^{-1} were used for separation of nitrite. Data acquisition and processing were accomplished with Shimadzu LC solution software.

Reagents

Chemicals of analytical reagent grade and Milli-Q water were used throughout.

A 1.0×10^{-3} mol L^{-1} stock solution of nitrite was prepared by dissolving 69.0 mg of sodium nitrite (pre-dried at 110 °C for 4 h) in Milli-Q water. A small amount of sodium hydroxide (about 5.0 mg) was added to prevent its decomposition. Twenty drops of chloroform (0.3 mL) were also added to inhibit the bacterial growth and thus make the nitrite solution stable (Lew 1977). The resulting solution was made up to the mark in a 1000-mL volumetric flask. This standard stock solution was prepared weekly and kept in a refrigerator at 4 °C. Working solutions were prepared freshly by an appropriate dilution of the stock solution.

Hydrochloric acid (0.4 mol L^{-1}) was prepared from concentrated hydrochloric acid.

A 1.0 mol L^{-1} Tris–HCl buffer solution (pH 8.0) was purchased from Beijing Solarbio Company (Beijing, China).

The 5.0×10^{-5} mol L^{-1} DAF-FM DA solution was prepared by diluting 40 μL of 5 mM DAF-FM DA solution (Beyotime Institute of Biotechnology, Haimen, Jiangsu, China) to 4 mL with the corresponding diluents (Beyotime Institute of Biotechnology, Haimen, Jiangsu, China).

The salts including NaH_2PO_4, Na_2HPO_4, Na_3PO_4, $NaNO_3$, $NaHCO_3$, Na_2CO_3, Na_2SO_4, NaBr, CH_3COONa, NH_4Cl, KCl, LiCl, $CaCl_2\cdot2H_2O$, $Zn(CH_3COO)_2$, $FeCl_3\cdot6H_2O$, $FeCl_2\cdot4H_2O$, $CuCl_2\cdot2H_2O$, and $MgCl_2\cdot6H_2O$ were purchased from Beijing Chemical Works.

Four kinds of meat products (ham, sausage, beef, and salted fish) and three kinds of pickled vegetables (Korean Kimchi,

Chinese sauerkraut, and preserved szechuan pickle) were randomly collected from local supermarket in Beijing, China.

p-Aminobenzenesulfonic acid solution (4 g L^{-1}) and *N*-(1-naphthyl)-ethylenediamine solution (2 g L^{-1}) were prepared by dissolving 0.4 g *p*-aminobenzenesulfonic acid or 0.2 g *N*-(1-naphthyl)-ethylenediamine in 100 mL water.

Tetrabutylammonium hydroxide was used as the ion-pairing agent for the HPLC separation of nitrite, and it was obtained as a 0.4-mol L^{-1} water solution from Acros (NJ, USA).

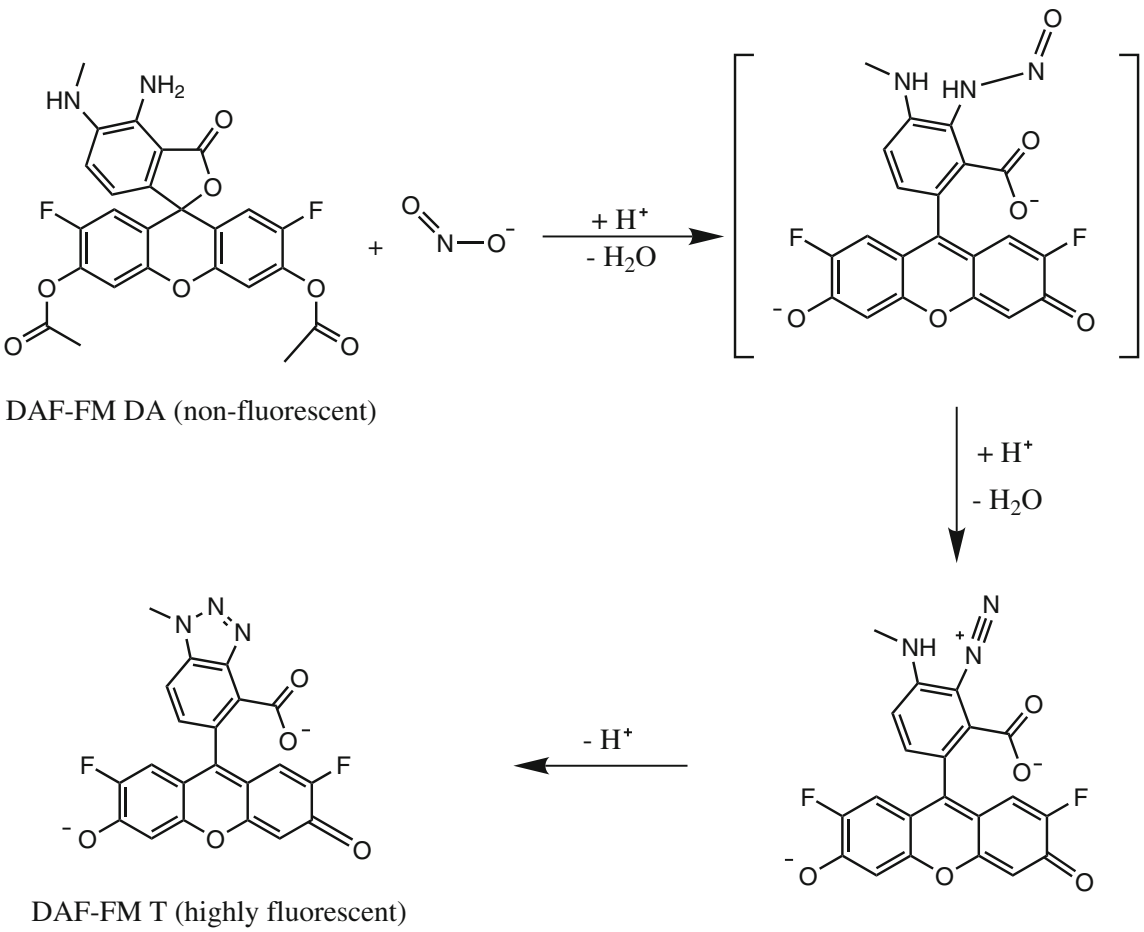
Procedure

Fifty microliters of 5.0×10^{-5} mol L^{-1} DAF-FM DA solution and 0.5 mL of 0.4 mol L^{-1} hydrochloric acid were added to a 10-mL colorimetric tube. An appropriate quantity of working solution of 1.0×10^{-5} mol L^{-1} sodium nitrite or sample solution was added. The mixture was diluted to 1.0 mL with Milli-Q water and mixed well. Then the tube was placed in a thermostat water bath at 60 °C for 30 min, cooled to room temperature, and made up to 3.0 mL with 1.0 mol L^{-1} Tris–HCl buffer solution (pH 8.0). The final pH of the solution was 7.8. The fluorescence intensity of the solution was measured at 517 nm, with excitation at 495 nm. All fluorescence intensity measurements were corrected with a blank.

Sample Preparation

Meat products were purchased from local supermarket in Beijing and triturated. Ten grams of each sample was weighed and placed in a beaker, then 25 mL of $Na_2B_4O_7\cdot10H_2O$ solution (5 %) and 350 mL of hot water were added. The mixture was placed in a hot water bath (80~90 °C) for 15 min, then cooled to room temperature, and transferred to a volumetric flask. Ten milliliters of potassium ferrocyanide aqueous solution and 10 mL of zinc acetate aqueous solution were added to precipitate the protein, and the volume was made up to 500 mL, followed by filtration through filter paper. The filtrate was stored at about 4 °C. Each time, an 80-μL portion of the filtrate was determined with the method described above.

Pickled vegetables were cleaned and dried with a blower in room temperature and crushed before use. Then 10.0 g of pickled vegetables was weighed and placed in a beaker, then 25 mL of $Na_2B_4O_7\cdot10H_2O$ solution (5 %) and 350 mL of hot water were added. The mixture was placed in a hot water bath (80~90 °C) for 15 min, then cooled to room temperature, and



Scheme 1 Interaction of DAF-FM DA with nitrite in HCl solution to form DAF-FM T

transferred to a volumetric flask. Ten milliliters of potassium ferrocyanide aqueous solution and 10 mL of zinc acetate aqueous solution were added, and the volume was made up to 500 mL, followed by filtration through filter paper. The filtrate was stored at about 4 °C. Each time, a 20-μL portion of the filtrate was determined with the method described above.

Results and Discussion

Spectral Characteristics

The proposed method is based on the diazotization reaction of DAF-FM DA with nitrite in HCl solution as shown in Scheme 1.

The excitation and emission spectra of DAF-FM DA in the presence and in the absence of nitrite were shown in Fig. 1. The excitation and emission maxima were found to be at 495 and 517 nm, respectively. Enhancement of fluorescence intensity was found when nitrite was added. Similar enhancement of fluorescence intensity was also found when DAF-FM DA serves as a fluorescent probe for nitric oxide (Kojima et al. 1999). This result suggested that DAF-FM DA was a potential probe for determining trace amount of nitrite in acidic conditions.

Optimization of Reaction Conditions of DAF-FM DA with Nitrite

In order to find the optimum conditions for the method, the effects of variables such as solution acidity, interaction time, and temperature on the fluorescence enhancement of DAF-FM DA in the presence of nitrite were then systematically investigated.

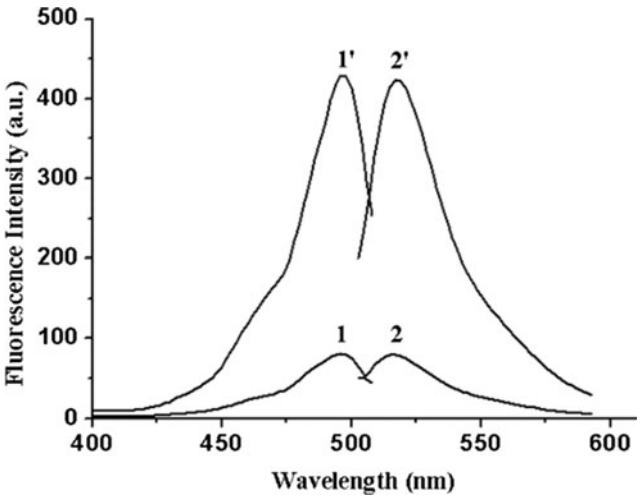


Fig. 1 Fluorescence excitation and emission spectra of DAF-FM DA in the absence (*1* and *2*) and presence (*1′* and *2′*) of nitrite. $C_{\text{DAF-FM DA}}$= 5.0×10^{-6} mol L^{-1}; C_{HCl}=0.2 mol L^{-1}; C_{nitrite}=1.0×10^{-6} mol L^{-1}; reaction time, 30 min; reaction temperature, 60 °C

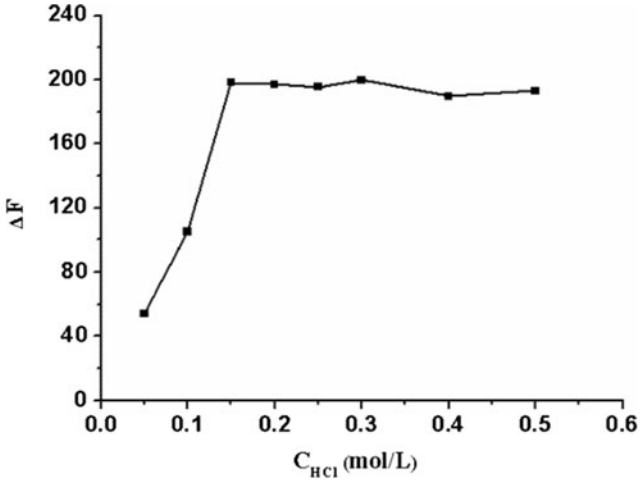


Fig. 2 Effect of HCl concentration on the fluorescence enhancement. $C_{\text{DAF-FM DA}}$=2.5×10^{-6} mol L^{-1}; C_{nitrite}=8×10^{-7} mol L^{-1}; reaction time, 30 min; reaction temperature, 60 °C

Effect of Reaction Acidity

The reaction between DAF-FM DA and nitrite takes place in acidic medium. In the present work, HCl solution was chosen to obtain an acidic medium. The effect of acidity on the fluorescence enhancement was investigated by varying the concentration of HCl from 0.05 to 0.50 mol L^{-1}. As depicted in Fig. 2, the fluorescence enhancement increased with the increase of HCl concentration from 0.05 to 0.15 mol L^{-1} and then kept constant above 0.15 mol L^{-1}. Based on this observation and consideration of reasonable time necessary for a speedy analysis, 0.20 mol L^{-1} HCl solution was chosen in the subsequent experiments.

The pKa value of the phenolic OH group of DAF-FM was 4.38±0.05, and the fluorescence intensity was stable above

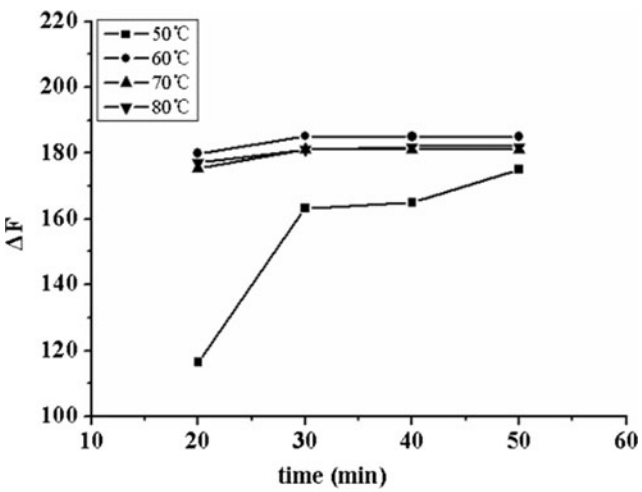


Fig. 3 Effect of time and temperature on the fluorescence enhancement. $C_{\text{DAF-FM DA}}$=2.5×10^{-6} mol L^{-1}; C_{HCl}=0.20 mol L^{-1}; C_{nitrite}=8×10^{-7} mol L^{-1}

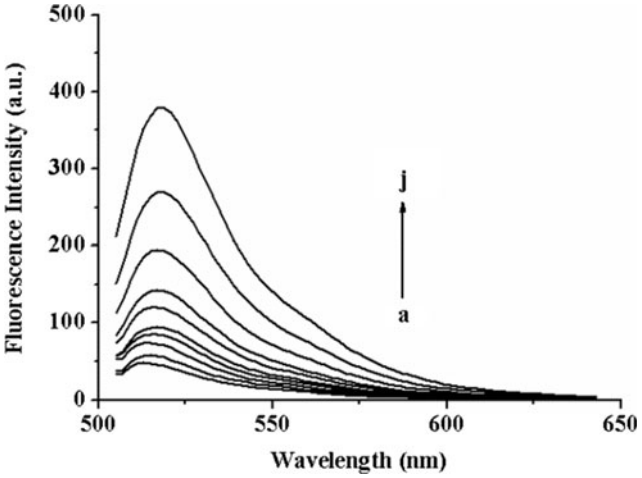


Fig. 4 Emission spectra for DAF-FM DA in the presence of nitrite ($\times 10^{-6}$ mol L^{-1}). $C_{\text{DAF-FM DA}}=2.5\times10^{-6}$ mol L^{-1}; $C_{\text{HCl}}=0.20$ mol L^{-1}; reaction time, 30 min; reaction temperature, 60 °C; (*a*) 0; (*b*) 0.05; (*c*) 0.10; (*d*) 0.15; (*e*) 0.20; (*f*) 0.30; (*g*) 0.40; (*h*) 0.60; (*i*) 1.00; (*j*) 1.50

pH 5.8 (Kojima et al. 1999). Finally, the pH of the system was adjusted to 7.8 with 1.0 mol L^{-1} Tris–HCl buffer solution (pH 8.0).

Effect of Reaction Times and Temperatures

At the very low concentrations where the measurements were carried out, the reaction between DAF-FM DA and nitrite was found to be time- and temperature-dependent. Hence, the completion of the reaction was obtained by placing the colorimetric tube in a hot water bath. The effects of reaction time and temperature on the reaction of DAF-FM DA with nitrite were shown in Fig. 3. It was found that a higher temperature was necessary to get obvious fluorescence enhancement. But when the temperature was raised from 60 to 80 °C, the fluorescence enhancement was almost constant. At 60~80 °C, the fluorescence enhancement was almost unchanged after 30 min of reaction. Therefore, the optimized conditions for the analyzing process were found to be at 60 °C for 30 min.

Linearity and Detection Limit

Under the optimized conditions, fluorescence spectra of DAF-FM DA in the absence and in the presence of nitrite were shown in Fig. 4 in the concentration range of 0 to 1.5×10^{-6} mol L^{-1}. The fluorescence enhancement was linear over nitrite concentration. Based on it, a linear calibration curve could be constructed in the range of 5.0×10^{-8} to 1.5×10^{-6} mol L^{-1}. The concentration of nitrite could be calculated from the linear regression equation: $\Delta F=220.8096\,c+5.1622$ ($R^2=0.9975$), where ΔF is the fluorescence enhancement and c is the concentration of nitrite. The relative standard deviation (RSD) for six replicate determinations is 1.3 % for 5.0×10^{-7} mol L^{-1} nitrite. The limit of detection (LOD) is 3.3×10^{-8} mol L^{-1}, which was evaluated using $3\sigma/s$, where σ is the standard deviation of the blank signals and s is the slope of the linear calibration plot.

In comparison with previously reported spectrofluorimetric methods in determination of nitrite, such as neutral red (Li et al. 2003), 5-aminofluorescein (Axelrod and Engel 1975), 4-hydroxycoumarin (Ohta et al. 1986), ADMND (Chen et al. 2007), and RB-PDA (Xue et al. 2012), the current method is more sensitive and simple.

Interference of Foreign Ions

In order to evaluate the selectivity of this new method, the effects of various common coexisting inorganic ions on the determination of nitrite were also studied by adding various foreign species into the solutions containing 0.5 μmol L^{-1} nitrite. The tolerance limits for the ions studied were summarized in Table 1. Most of anions can be allowed at concentrations of 2500 μmol L^{-1}. The 1000 μmol L^{-1} of NH_4^+, K^+, Li^+, Ca^{2+}, and Zn^{2+} and 10 μmol L^{-1} of Fe^{3+}, Fe^{2+}, Cu^{2+}, and

Table 1 Effect of foreign ions

Foreign ion	Concentration (μmol L^{-1})	Relative error (%)	Foreign ion	Concentration (μmol L^{-1})	Relative error (%)
Na, $H_2PO_4^-$	2500	+4.2	NH_4^+, Cl^-	1000	+1.7
Na, HPO_4^{2-}	2500	+3.7	K^+, Cl^-	1000	−0.7
Na, PO_4^{3-}	2500	+3.5	Li^+, Cl^-	1000	+0.3
Na, NO_3^-	2500	−2.6	Ca^{2+}, Cl^-	1000	−0.7
Na, HCO_3^-	2500	+2.9	Zn^{2+}, CH_3COO^-	1000	−2.9
Na, CO_3^{2-}	2500	−4.2	Fe^{3+}, Cl^-	10	+6.0
Na, SO_4^{2-}	2500	−0.5	Fe^{2+}, Cl^-	10	+4.9
Na, Br^-	2500	+2.1	Cu^{2+}, Cl^-	10	−4.6
Na, CH_3COO^-	2500	+1.2	Mg^{2+}, Cl^-	10	+5.0

Table 2 Determination of nitrite in food samples by the proposed method

Sample	Added (μg/g)	Found by the proposed method[a] (μg/g)	SD	RSD (%, n=6)	Recovery (%)	Found by Griess method[a] (μg/g)	Found by HPLC method[a] (μg/g)
Ham 1	0.00	3.57	0.18	5.1	–	3.80	3.45
	3.45	6.75	0.20	3.0	92.2	7.13	7.01
Ham 2	0.00	3.11	0.15	4.9	–	3.38	3.23
	3.45	6.09	0.22	3.7	86.4	6.06	6.32
Sausage 1	0.00	4.83	0.19	4.0	–	4.58	4.73
	3.45	8.05	0.20	2.5	93.3	8.17	8.06
Sausage 2	0.00	5.23	0.17	3.3	–	5.18	5.24
	3.45	8.55	0.16	1.9	96.2	8.53	8.69
Beef 1	0.00	4.91	0.16	3.3	–	4.98	5.14
	3.45	8.07	0.18	2.3	91.6	8.40	8.61
Beef 2	0.00	5.67	0.15	2.7	–	5.23	5.49
	3.45	8.97	0.20	2.3	95.6	5.68	8.92
Salted fish 1	0.00	5.38	0.14	2.7	–	5.30	5.42
	3.45	8.65	0.19	2.2	94.8	8.61	8.65
Salted fish 2	0.00	5.52	0.19	3.5	–	5.60	5.49
	3.45	9.07	0.16	1.8	102.9	9.23	8.89
Korean kimchi 1	0.00	13.78	0.13	1.0	–	14.61	14.04
	10.35	24.01	0.16	0.7	98.8	24.93	24.17
Korean kimchi 2	0.00	8.97	0.20	2.3	–	9.08	9.02
	10.35	19.27	0.18	1.0	99.5	19.21	19.37
Chinese sauerkraut 1	0.00	7.45	0.18	2.5	–	7.65	7.28
	6.90	14.29	0.12	0.9	99.1	14.35	14.19
Chinese sauerkraut 2	0.00	8.83	0.19	2.2	–	9.08	8.92
	6.90	15.73	0.12	0.8	100.0	15.85	15.72
Canned cucumber 1	0.00	6.48	0.14	2.2	–	6.21	6.52
	3.45	9.80	0.14	1.5	96.2	9.58	9.98
Canned cucumber 2	0.00	5.89	0.15	2.6	–	5.83	5.69
	3.45	9.27	0.13	1.5	98.0	9.28	9.03

SD standard deviation, *RSD* relative standard deviation

[a] Each result is the average of six replicate determinations

Mg^{2+} had negligible interference. The results indicate that the selectivity of the present method is good.

Determination of Nitrite in Real Food Samples

To evaluate the viability of the proposed method for routine analysis, the proposed method was applied to determine nitrite in four kinds of meat products and three kinds of pickled vegetables. And for each kind of food, two different samples randomly collected in markets were analyzed. The recovery test was carried out by sparking the samples at one level, and the recoveries of nitrite ranged from 86.4 to 102.9 %. All samples were analyzed in sextuplicate, and the obtained RSD was below 5.1 %. In order to further validate the accuracy of the proposed method, all the samples were also analyzed simultaneously by Griess method and ion-pairing HPLC method. The results were given in Table 2. It can be seen that there were no significant differences between the results obtained from the proposed method and the two reference methods, indicating that the proposed method was reliable.

Conclusions

A novel and sensitive spectrofluorimetric method for the determination of trace amount of nitrite with DAF-FM DA was developed. The mechanism for the fluorescence enhancement involves the formation of triazolofluorescein. The present method exhibited good selectivity, high sensitivity, and avoidance of coexisting substances interferences. The detection limit of this new method was lower or comparable to most of reported spectrofluorimetric methods. It

was also proved useful in the detection and quantification of nitrite in real food samples.

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Conflict of Interest Qiuhua Wang declares that she has no conflict of interest. Haiwei Huang declares that he has no conflict of interest. Baoming Ning declares that he has no conflict of interest. Minfeng Li declares that he has no conflict of interest. Lan He declares that she has no conflict of interest. This article does not contain any studies with human or animal subjects.

References

Akyüz M, Ata Ş (2009) Determination of low level nitrite and nitrate in biological, food and environmental samples by gas chromatography–mass spectrometry and liquid chromatography with fluorescence detection. Talanta 79:900–904

Axelrod HD, Engel NA (1975) Fluorometric determination of subnanogram levels of nitrite using 5-aminofluorescein. Anal Chem 47:922–924

Azad UP, Turllapati S, Rastogi PK, Ganesan V (2014) Tris (1,10-phenanthroline) iron (II)-bentonite film as efficient electrochemical sensing platform for nitrite determination. Electrochim Acta 127: 193–199

Biswas S, Chowdhury B, Ray BC (2004) A novel spectrofluorimetric method for the ultra trace analysis of nitrite and nitrate in aqueous medium and its application to air, water, soil and forensic samples. Talanta 64:308–312

Burakham R, Oshima M, Grudpan K, Motomizu S (2004) Simple flow-injection system for the simultaneous determination of nitrite and nitrate in water samples. Talanta 64:1259–1265

Burden E (1961) The toxicology of nitrates and nitrites with particular reference to the potability of water supplies. A review. Analyst 86: 429–433

Chen T, Tong AJ, Zhou YM (2007) 2-Amino-5,7-dimethyl-1,8-naphthyridine as a fluorescent reagent for the determination of nitrite. Spectrochim Acta A 66:586–589

Cotton FA, Wilkinson G (1988) Advanced Inorganic Chemistry, 5th Edit, Wiley-Interscience, John Wiley & Sons, New York:588

Damiani P, Burini G (1986) Fluorometric determination of nitrite. Talanta 33:649–652

Della Betta F, Vitali L, Fett R, Costa ACO (2014) Development and validation of a sub-minute capillary zone electrophoresis method for determination of nitrate and nitrite in baby foods. Talanta 122:23–29

Guo YX, Zhang QF, Shangguang X, Zhen G (2013) Spectrofluorimetric determination of trace nitrite with o-phenylenediamine enhanced by hydroxypropyl-β-cyclodextrin. Spectrochim Acta A 101:107–111

He LJ, Zhang KG, Wang CJ, Luo XL, Zhang SS (2011) Effective indirect enrichment and determination of nitrite ion in water and biological samples using ionic liquid-dispersive liquid–liquid microextraction combined with high-performance liquid chromatography. J Chromatogr A 1218:3595–3600

Huang KJ, Wang H, Guo YH, Fan RL, Zhang HS (2006) Spectrofluorimetric determination of trace nitrite in food products with a new fluorescent probe 1,3,5,7-tetramethyl-2,6-dicarbethoxy-8-(3′,4′-diaminophenyl)-difluoroboradiaza-s-indacene. Talanta 69: 73–78

Itoh Y, Ma FH, Hoshi H, Oka M, Noda K, Ukai Y et al (2000) Determination and bioimaging method for nitric oxide in biological specimens by diaminofluorescein fluorometry. Anal Biochem 287: 203–209

Jourd'heuil D (2002) Increased nitric oxide-dependent nitrosylation of 4, 5-diaminofluorescein by oxidants: implications for the measurement of intracellular nitric oxide. Free Radical Bio Med 33:676–684

Kojima H, Nakatsubo N, Kikuchi K, Kawahara S, Kirino Y, Nagoshi H et al (1998) Detection and imaging of nitric oxide with novel fluorescent indicators: diaminofluoresceins. Anal Chem 70:2446–2453

Kojima H, Urano Y, Kikuchi K, Higuchi T, Hirata Y, Nagano T (1999) Fluorescent indicators for imaging nitric oxide production. Angew Chem Int Ed 38:3209–3212

Lee SH, Field LR (1984) Postcolumn fluorescence detection of nitrite, nitrate, thiosulfate, and iodide anions in high-performance liquid chromatography. Anal Chem 56:2647–2653

Lew R (1977) Elimination of sulphite interference in the spectrophotometric determination of nitrite. Analyst 102:476–479

Li R, Yu JC, Jiang ZT, Zhou RH, Liu HY (2003) A solid-phase fluorescent quenching method for the determination of trace amounts of nitrite in foods with neutral red. J Food Drug Anal 11:251–257

Liu QH, Yan XL, Guo JC, Wang DH, Li L, Yan FY, Chen LG (2009) Spectrofluorimetric determination of trace nitrite with a novel fluorescent probe. Spectrochim Acta A 73:789–793

Menart E, Jovanovski V, Hočevar Samo B (2015) Silver particle-decorated carbon paste electrode based on ionic liquid for improved determination of nitrite. Electromchim Commun 52:45–48

Merusi C, Corradini C, Cavazza A, Borromei C, Salvadeo P (2010) Determination of nitrates, nitrites and oxalates in food products by capillary electrophoresis with pH-dependent electroosmotic flow reversal. Food Chem 120:615–620

Nagano T, Yoshimura T (2002) Bioimaging of nitric oxide. Chem Rev 102:1235–1270

Nagaraja P, Al-Tayar NGS, Shivakumar A, Shrestha AK, Gowda AK (2010) A simple and sensitive spectrophotometric method for the determination of trace amounts of nitrite in environmental and biological samples using 4-amino-5-hydroxynaphthalene-2,7-disulphonic acid monosodium salt. Spectrochim Acta A 75:1411–1416

Ohta T, Arai Y, Takitani S (1986) Fluorometric determination of nitrite with 4-hydroxycoumarin. Anal Chem 58:3132–3135

Pagliano E, Meija J, Mester Z (2014) High-precision quadruple isotope dilution method for simultaneous determination of nitrite and nitrate in seawater by GCMS after derivatization with triethyloxonium tetrafluoroborate. Anal Chim Acta 824:36–41

Ramdane-Terbouche CA, Terbouche A, Djebbar S, Hauchard D (2014) Electrochemical sensors using modified electrodes based on copper complexes formed with Algerian humic acid modified with ethylenediamine or triethylenetetramine for determination of nitrite in water. Talanta 119:214–225

Seike Y, Fukumori R, Senga Y, Oka H, Fujinaga K, Okumura M (2004) A simple and sensitive method for the determination of hydroxylamine in fresh-water samples using hypochlorite followed by gas chromatography. Anal Sci 20:139–142

Senra-Ferreiro S, Pena-Pereira F, Lavilla I, Bendicho C (2010) Griess micro-assay for the determination of nitrite by combining fibre optics-based cuvetteless UV–Vis micro-spectrophotometry with liquid-phase microextraction. Anal Chim Acta 668:195–200

Tanaka Y, Naruishi N, Fukuya H, Sakata J, Saito K, S-i W (2004) Simultaneous determination of nitrite, nitrate, thiocyanate and uric acid in human saliva by capillary zone electrophoresis and its application to the study of daily variations. J Chromatogr A 1051:193–197

Tarigh GD, Shemirani F (2014) Development of a selective and pH-independent method for the analysis of ultra trace amounts of nitrite

in environmental water samples after dispersive magnetic solid phase extraction by spectrofluorimetry. Talanta 128:354–359

Troška P, Chudoba R, Danč L, Bodor R, Horčičiak M, Tesařová E, Masár M (2013) Determination of nitrite and nitrate in cerebrospinal fluid by microchip electrophoresis with microsolid phase extraction pre-treatment. J Chromatogr B 930:41–47

Tsikas D (2007) Analysis of nitrite and nitrate in biological fluids by assays based on the Griess reaction: appraisal of the Griess reaction in the L-arginine/nitric oxide area of research. J Chromatogr B 851:51–70

Wang H, Yang W, Liang SC, Zhang ZM, Zhang HS (2000) Spectrofluorimetric determination of nitrite with 5,6-diamino-1,3-naphthalene disulfonic acid. Anal Chim Acta 419:169–173

Wang N, Wang RQ, Zhu Y (2012a) A novel ion chromatography cycling-column-switching system for the determination of low-level chlorate and nitrite in high salt matrices. J Hazard Mater 235:123–127

Wang X, Adams E, Van Schepdael A (2012b) A fast and sensitive method for the determination of nitrite in human plasma by capillary electrophoresis with fluorescence detection. Talanta 97:142–144

Wang X, Li H, Wu M, Ge SL, Zhu Y, Wang QJ et al (2013) Simultaneous electrochemical determination of sulphite and nitrite by a gold nanoparticle/graphene-chitosan modified electrode. Chin J Anal Chem 41:1232–1237

Wiersma JH (1970) 2,3-Diaminonaphthalene as a spectrophotometric and fluorometric reagent for the determination of nitrite ion. Anal Lett 3: 123–132

Xue ZW, Wu ZS, Han SF (2012) A selective fluorogenic sensor for visual detection of nitrite. Anal Methods 4:2021–2026

Zhang X, Wang H, Li JS, Zhang HS (2003) Development of a fluorescent probe for nitric oxide detection based on difluoroboradiaza-*s*-indacene fluorophore. Anal Chim Acta 481:101–108

Zhang X, Chi RA, Zou J, Zhang HS (2004) Development of a novel fluorescent probe for nitric oxide detection: 8-(3′,4′-diaminophenyl)-difluoroboradiaza-S-indacence. Spectrochim Acta A 60:3129–3134