

Synthesis of 5-Antipyriylazosalicylfluorone and its Colour Reaction with Molybdenum

YING-HONG LIU*, WEI-XING MA, YAN-HUI LI and JUAN LIU

College of Chemical Engineering, Huaihai Institute of Technology, Lianyungang-222005, Jiangsu Province, P.R. China

*Corresponding author: Tel: +86 518 85857922; E-mail: liuyh9506@163.com; yinghong1978@yahoo.com.cn

(Received: 14 July 2010;

Accepted: 17 December 2010)

AJC-9401

The synthesis of a new reagent, 5-antipyriylazosalicylfluorone was reported. The colour reaction between 5-antipyriylazosalicylfluorone and molybdenum(VI) was studied. In weak acid medium and in the presence of surfactant cetyltrimethylammonium bromide, Mo(VI) reacted with 5-antipyriylazosalicylfluorone to form a complex with the maximum absorption wavelength at 530 nm. Beer's law was obeyed in the range of 0-1.4 $\mu\text{g mL}^{-1}$ for Mo(VI). The value of Mo(VI) content in food samples determined by this method was accordant with that measured by atomic absorption spectrophotometry. The relative standard deviation was 1.3-2.1 % for 7 replicate determinations. The recovery of the standard addition was 99.8-103.0 %.

Key Words: 5-Antipyriylazosalicylfluorone, Molybdenum, Spectrophotometry.

INTRODUCTION

Molybdenum is one of the necessary microelements for life. Molybdenum, as an important component of several enzymes, plays a role of prevention dental caries and anemia. Studies showed that esophageal cancer, liver cancer, colorectal cancer, cervical cancer, breast cancer *etc.*, were all caused due to the lack of molybdenum to a certain extent. So it is significant to study on the determination method of molybdenum. The methods for determining trace molybdenum at present mainly include spectrophotometry¹⁻³, atomic absorption spectrophotometry⁴⁻⁶, atomic emission spectrophotometry⁷, flow injection⁸, voltammetry⁹ and so on. Spectrophotometry was widely used for its simple operation and low cost.

1,2,4-Trihydroxyfluorone reagents are very sensitive to some metals, such as germanium^{10,11}, molybdenum^{12,13}, tungsten¹⁴, indium¹⁵, tin¹⁶ and manganese¹⁷ *etc.* A new reagent of 5-antipyriylazosalicylfluorone (APASF) was synthesized in this study. Its properties and colour reaction with Mo(VI) was also investigated. The method had been applied to the determination of trace Mo(VI) in soybean and redbean with satisfactory results.

EXPERIMENTAL

A 722-N spectrophotometer (Shanghai precision and scientific instrument Co., Ltd., China) was used for the determination of absorbance. A PHS-3D pH-meter (Shanghai precision and scientific instrument Co., Ltd., China) was used for the pH measurement.

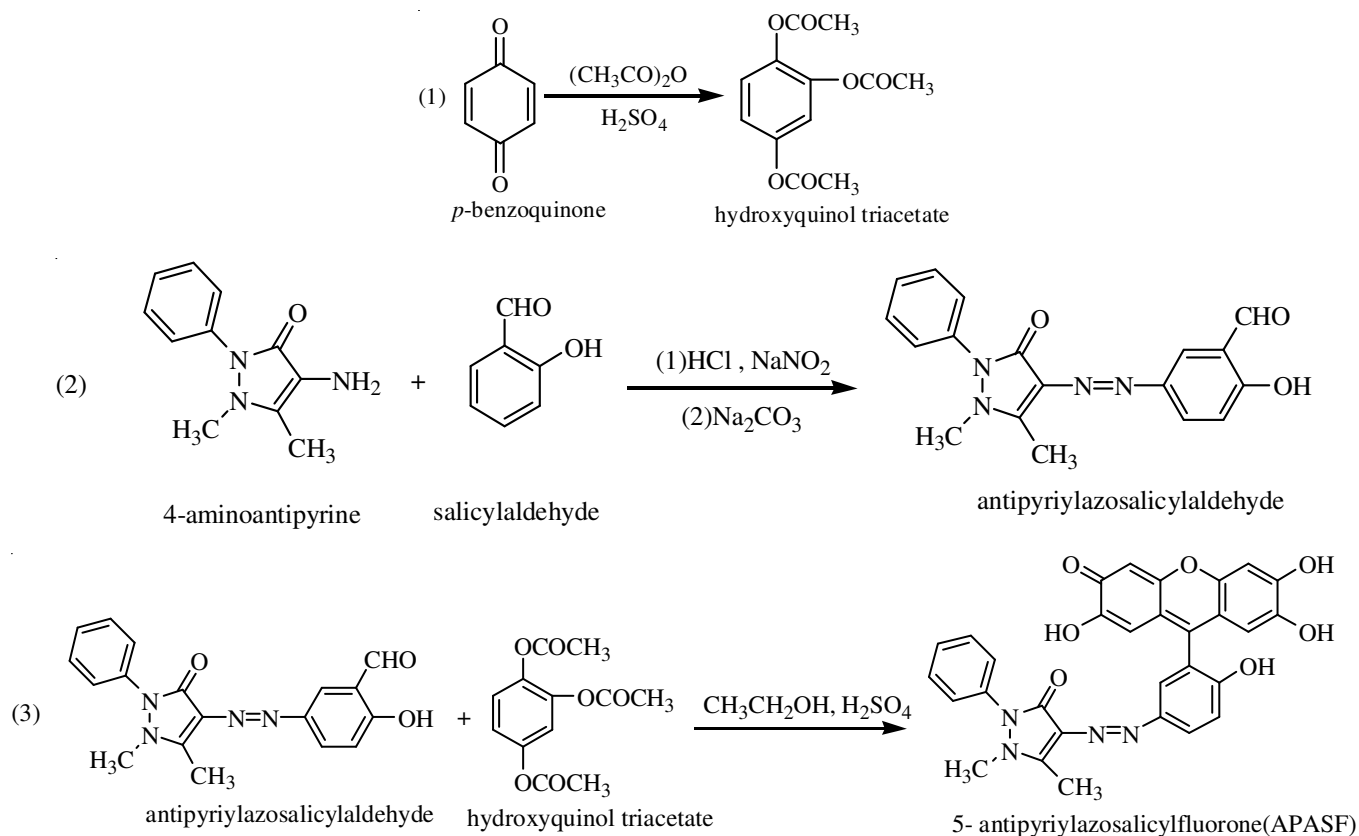
0.2058 g of ammonium molybdate was dissolved in 100 mL of water to get 0.1 mg mL⁻¹ Mo(VI) stock solution. The required concentration standard working solution (10 $\mu\text{g mL}^{-1}$) was obtained by diluting the above stock solution. 5-Antipyriylazosalicylfluorone (APASF) solution was prepared by dissolving 0.5021 g of APASF in 100 mL of ethanol after 1 mL of sulfuric acid (1:1 v/v) being added. HOAc-NaOAc buffer solution of pH 6.0 and 0.1 mol L⁻¹ cetyltrimethylammonium bromide (CTMAB) solution were used. All the reagents used were of analytical grade and the water was distilled water.

Synthesis and identification of 5-antipyriylazosalicylfluorone

The synthesis route of 5-antipyriylazosalicylfluorone (APASF) was showed as follow.

Synthesis of hydroxyquinol triacetate: Hydroxyquinol triacetate was synthesized according to the reported method¹⁸. 1.0 g of *p*-benzoquinone was dissolved in 7 mL of acetic anhydride and 1 mL of concentrated sulfuric acid when stirring at 45 °C in water bath. After being cooled to room temperature, the solution was diluted by 100 mL of water and stirred intensely until white precipitate appeared. Hydroxyquinol triacetate could be obtained by filtration.

Synthesis of antipyriylazosalicylaldehyde: 5.0 g of 4-aminoantipyrine was dissolved in 30 mL of hydrochloride solution (1:1 v/v) and cooled to about 0 °C. Kept for 15 min after 20 mL of 0.4 g L⁻¹ sodium nitrite solution being added by dropping to obtain diazonium salt solution. 3.0 mL of salicylaldehyde was dissolved in 100 mL of 50 g L⁻¹ sodium



carbonate solution and cooled to 0 °C or so. The above diazonium salt solution was added and the value of pH was adjusted to about 2.0 with hydrochloride solution after keeping for 1 h. After filtration, the precipitation was completely washed with water and then dried at 60–70 °C.

Synthesis of 5-antipyrilazosalicylfluorone: 13.0 g of hydroxyquinol triacetate, 6.0 g of antipyrilazosalicylaldehyde, 100 mL of ethanol (1:1 v/v) and 5 mL of concentrated sulfuric acid was added into a triple-mouth flask. The solution was heated to dissolve completely at 85 °C in water bath and then placed statically for 2 weeks. After filtration, water washing and drying, crude product could be obtained. Pure produce could be obtained by recrystallizing the crude product in ethanol.

Identification of 5-antipyrilazosalicylfluorone: The characteristic absorption peaks in IR spectrum (cm^{-1}) include: aromatic ring ~3060, 1580; carbonyl group 1639; ether group 1289, 1040; phenonic group 3248, ~1200, 693; azo group 1492; multi-substituted benzene: 823, 616. These results indicated that the synthesized product was 5-antipyrilazosalicylfluorone.

Procedure: Two 10 mL colorimetric tubes were taken. A suitable amount of sample solution or standard Mo(VI) working solution was added into one tube (complex solution), while the other was not added (reagent blank solution). Then 1.0 mL of HOAc-NaOAc buffer solution, 1.5 mL of CTMAB solution and 1.5 mL of APASF solution were added into each tube in order, which were diluted up to the mark with water. After placing for 15 min, the absorbance A of complex solution was measured at 530 nm with 1.0 cm cells by using reagent blank solution as reference.

RESULTS AND DISCUSSION

Absorption spectra: The absorption spectra of APASF and the complex of APASF and Mo(VI) were measured according to the experimental procedure, which were shown in Fig. 1. It could be seen that the sensitivity enhanced obviously in the presence of CTMAB. The complex and APASF had peaks at 530 and 480 nm, respectively in the presence of CTMAB. The maximal absorption wavelength red shifted 50 nm. The measuring wavelength was 530 nm in this study.

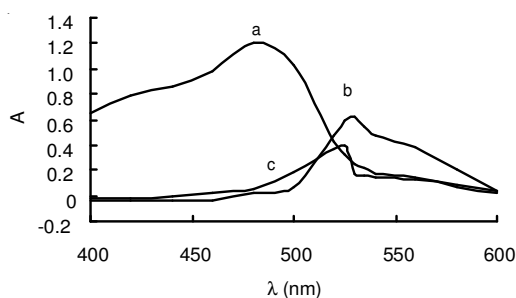


Fig. 1. Absorption spectra: (a) APASF (against water); (b) Mo(VI) + APASF + CTMAB (against reagent blank solution); (c) Mo(VI) + APASF (against reagent blank solution); [Mo(VI)] = 1 $\mu\text{g mL}^{-1}$; [APASF] = 50 $\mu\text{g mL}^{-1}$; [CTMAB] = 0.01 mol L⁻¹; pH = 6.0; reaction time t = 15 min

Effect of acidity: The colour reaction of APASF and Mo(VI) was investigated in different medium with different acidity, such as HCl, H₂SO₄, HAC-NaAc, Na₂B₄O₇-HCl, NH₃·H₂O-NH₄Cl, NaOH and *etc.* The results showed that HOAc-NaOAc was the best. When its pH was in the range of 5.0–8.0, the system had maximal and steady absorbance (Fig. 2).

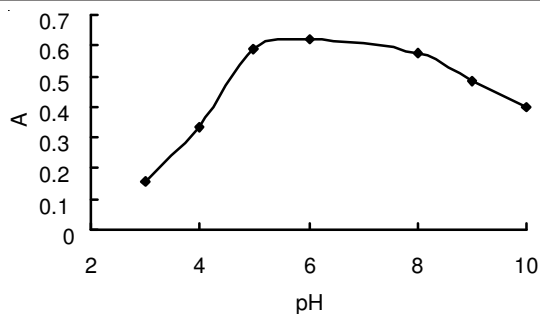


Fig. 2. Effect of acidity: $[\text{Mo(VI)}] = 1 \mu\text{g mL}^{-1}$; $[\text{APASF}] = 50 \mu\text{g mL}^{-1}$; $[\text{CTMAB}] = 0.01 \text{ mol L}^{-1}$; reaction time $t = 15 \text{ min}$

The best scope of buffer solution dose was in the range of 1.0-2.0 mL. Therefore, 1.0 mL of HOAc-NaOAc buffer solution at pH 6.0 was used in this study.

Effect of colour reagent dose: The results shown as Fig. 3 suggested that the system had maximal and steady absorbance when the dose of colour reagent was in the range of 1.0-3.0 mL. Therefore, 1.5 mL of APASF solution was chosen in present study.

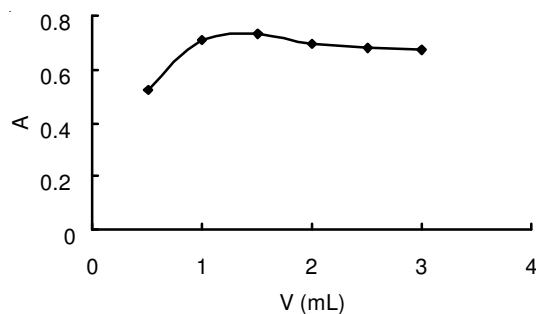


Fig. 3. Effect of colour reagent amount: $[\text{Mo(VI)}] = 1 \mu\text{g mL}^{-1}$; $[\text{CTMAB}] = 0.01 \text{ mol L}^{-1}$; $\text{pH} = 6.0$; reaction time $t = 15 \text{ min}$

Effect of surfactants: The effect of various surfactants, such as *p*-octyl polyethylene glycol phenyl ether, Tween 80, CTMAB, cetylpyridinium bromide, sodium dodecylsulfonate and *etc.*, on the system were investigated. The results showed that CTMAB had the best solubilization and sensitivity-enhancing functions on colour reaction. When the dose of CTMAB solution was in the range of 1.5-2.0 mL, the system had maximal and steady absorbance (Fig. 4). Therefore, 1.5 mL of CTMAB solution was used in present study.

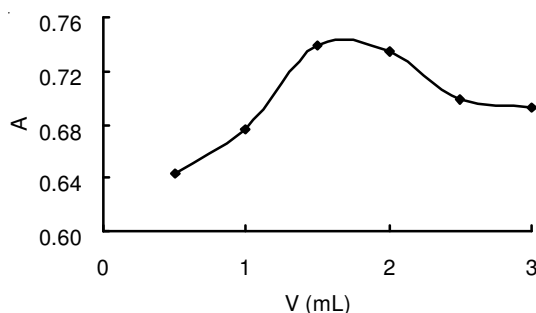


Fig. 4. Effect of surfactant amount: $[\text{Mo(VI)}] = 1 \mu\text{g mL}^{-1}$; $[\text{APASF}] = 50 \mu\text{g mL}^{-1}$; $\text{pH} = 6.0$; reaction time $t = 15 \text{ min}$

Reaction time and stability of complex: As shown in Fig. 5, Mo(VI) reacts with APASF quantitatively at room temperature in 15 min. In this study, the colour system was

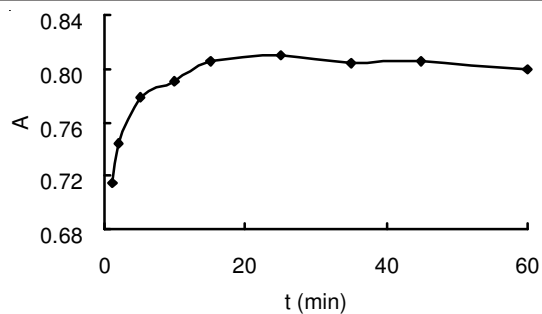


Fig. 5. Effect of reaction time: $[\text{Mo(VI)}] = 1 \mu\text{g mL}^{-1}$; $[\text{APASF}] = 50 \mu\text{g mL}^{-1}$; $[\text{CTMAB}] = 0.01 \text{ mol L}^{-1}$; $\text{pH} = 6.0$

placed for 15 min at room temperature. The formed complex remained steady in at least 1 h.

Effect of adding reagent order: The absorbance of solution was measured when changing the addition order of reagents. The results were shown in Table-1. It was shown that the addition order of reagents had a certain influence on absorbance. The addition order in the present experimental procedure of Mo(VI) + buffer + CTMAB + APASF had the best result, so it was applied in this study.

TABLE-1 EFFECT OF ADDING REAGENT ORDER	
Order of adding reagent	A
Mo(VI) + buffer + CTMAB + APASF	0.712
Mo(VI) + buffer + APASF + CTMAB	0.688
Mo(VI) + APASF + buffer + CTMAB	0.678
Mo(VI) + APASF + CTMAB + buffer	0.685
Mo(VI) + CTMAB + APASF + buffer	0.696
Mo(VI) + CTMAB + buffer + APASF	0.672

Working curve: A series of standard Mo(VI) solutions with different concentration were prepared. Under the chosen experimental conditions, the absorbance of these solutions was measured. The working curve was drawn and shown in Fig. 6. The results showed that Beer's law was obeyed in the range of $0-1.4 \mu\text{g mL}^{-1}$ for Mo(VI). The linear regression equation was $A = 0.5346c + 0.1614$ with the regression coefficient $\gamma = 0.9990$. The reagent blank was determined 11 times and the detection limit determined was $0.09 \mu\text{g mL}^{-1}$ by $3S/K$ method (S is the standard deviation of the reagent blank for 11 times determination, K is the slope of the working curve).

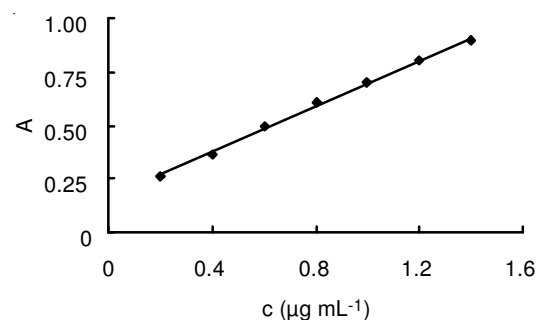


Fig. 6. Working curve: $[\text{APASF}] = 50 \mu\text{g mL}^{-1}$; $[\text{CTMAB}] = 0.01 \text{ mol L}^{-1}$; $\text{pH} = 6.0$; reaction time $t = 15 \text{ min}$

Effect of concurrent ions: $10 \mu\text{g}$ of Mo(VI) was determined according to experimental procedure. When the relative error was within $\pm 5 \%$, the allowable amounts of concurrent

ions were (μg): Na^+ , K^+ , Ca^{2+} , Mg^{2+} (> 1000); Cl^- , SO_4^{2-} , NO_3^- (500); Ni^{2+} , Zn^{2+} , Hg^{2+} (350); Fe^{2+} , Cu^{2+} (150); Co^{2+} (80); Al^{3+} , Fe^{3+} (150, F^-); Bi^{3+} , Cr^{3+} (30); F^- (> 1000).

Analysis of sample: Some soybean and redbean samples were grinded after being dried at 110 °C in oven. 5.0 g of soybean and redbean samples which had been grinded was weighed into a crucible, respectively and carbonized on electric furnace until there was no heavy smoke volatilizing. And then the samples were incinerated in muffle furnace at 700 °C for 3 h. After cooling, added 4 mL of hydrochloride solution (1:1 v/v) into the solid and adjusted pH to 7.0 with sodium hydroxide after being dissolved completely. The solutions were decanted into 50 mL volumetric flask and diluted to scale as the sample solutions. 1 mL of the above soybean or redbean sample solution was transferred into a 10 mL colorimetric tube. The content of Mo(VI) was determined by the present method and atomic absorption spectrophotometry for comparison. The standard addition recovery experiments were made also. The analysis results were shown in Table-2.

TABLE-2
ANALYTICAL RESULTS OF SAMPLES (n = 7)

Sample	Found ($\mu\text{g g}^{-1}$)		RSD (%)	Added (μg)	Recovered (μg)	Recovery (%)
	AAS	This method				
Redybean	3.25	3.28	1.3	3.00	3.09	103.0
Soybean	5.68	5.66	2.1	5.00	4.99	99.8

Conclusion

A new reagent of 5-antipyrylazosalicylfluorone (APASF) was synthesized and the spectrophotometric determination

method of Mo(VI) using APASF was investigated. From the results of analysis, it is suggested that the method is accurate, selective and stable. Hence, the assay is worthy of use for the determination of food samples.

ACKNOWLEDGEMENTS

This work is supported by Nature Science Research Project of Huaihai Institute of Technology, China (No. 2010150014).

REFERENCES

- L. Zaijun, P. Jiaomai and T. Jian, *J. Anal. Bioanal. Chem.*, **374**, 1125 (2002).
- M.J. Ahmed and M.E. Haque, *Anal. Sci.*, **18**, 433 (2002).
- B. Keshavan and P. Nagaraja, *Microchim. Acta*, **86**, 379 (2005).
- P.-G. Su and S.-D. Huang, *J. Anal. At. Spectrom.*, **13**, 641 (1998).
- P. Liang, Q. Li and R. Liu, *Microchim. Acta*, **119**, 164 (2009).
- A.M. Pashadzhinov, *Zh. Anal. Khim.*, **61**, 767 (2006).
- P. Liang, Y. Liu and L. Guo, *J. Anal. At. Spectrom.*, **19**, 1006 (2004).
- S.A. Barakat and T.Y. Mahmoud, *Acta Chim. Slov.*, **50**, 799 (2003).
- J.C. de Andrade, A.M. de Almeida, A.R. Coscione and L.M. Aleixo, *Analyst*, **126**, 892 (2001).
- K. Kania, *Chem. Anal. (Warsaw)*, **36**, 911 (1990).
- Z.J. Li, J.M. Pan and J. Tang, *Anal. Chim. Acta*, **445**, 153 (2001).
- S. Liu and J. Pan, *Anal. Lett.*, **32**, 1225 (1999).
- Z.J. Li, J.M. Pan and J. Tang, *Analyst*, **126**, 1154 (2001).
- V.A. Nazarenko, V.P. Antonovich and N.A. Veshchikova, *Talanta*, **34**, 215 (1987).
- Q. Wei, B. Du, H. Zhang, Y. Li and Z. Li, *Anal. Chem.*, **60**, 868 (2005).
- H. Kruihara and H. Kuwabara, *Anal. Chem.*, **33**, E199 (1984).
- W. Wanda, B. Franciszek and A. Agata, *J. Anal. Bioanal. Chem.*, **36**, 934 (2003).
- Y.-P. Huang and X.-Y. Li, *J. Metallur. Anal.*, **17**, 5 (1997) (in Chinese).