

Chemiluminescence System with Air Pump as a Sensor for Determination of Metal Levels in Rain

Hyuckgi Hong and H. B. Lim*

Department of Chemistry, Dankook University, Seoul 140-714, Korea. *E-mail: plasma@dankook.ac.kr

Received June 30, 2005

A lab-made chemiluminescence system with air pump was developed for monitoring of some metal levels in rain. The air pump enabled injection of 17.7 μg samples into a glass cell filled with luminol- H_2O_2 reagent of typically 300 μL for chemiluminescence measurement. The monitored trend of total metal ions in the rain collected in our campus was compared with analytical results of each metal ion from GFAAS. The system was also demonstrated to determine Cr^{6+} by reduction to Cr^{3+} using SnCl_2 . The limit of detection for Cr^{6+} obtained by 4 measurements was 85.0 pg mL^{-1} with a relative standard deviation of 3.4%. Although this system doesn't have selectivity due to the characteristics of chemiluminescence, application of it to environmental monitoring as a sensor for some transition metal ions was demonstrated.

Key Words : Chemiluminescence, Determination of metal ions, Rain analysis, Air pump

Introduction

The importance of ultra-sensitive determination of metal ions in the environmental field has been established.¹⁻⁴ Analytical results from rain drops can indicate metal pollution levels in air since most airborne particles emitted by human activity contain water-soluble metal salts. Although common analytical techniques that have been used for ultra-trace metal determination in the environmental samples, especially in rain are ICP-MS^{5,6} and GFAAS,⁷⁻⁹ chemiluminescence can be used as a sensor for the air pollution levels of any metal ions that react with the luminescent chemicals. Among the metal ions that can have a chemiluminescence reaction, the determination of Cr is of importance in environmental analysis.¹⁰⁻¹² Various methods have been developed for chemiluminescent detection of Cr^{6+} .^{13,14}

In this work, a chemiluminescence system for environmental analysis is manufactured. For micro liter sample injection, a lab-made air pump is developed for the first time to obtain a fast sampling time with reproducible sample injection. This is commonly required in environmental analysis due to the high number of samples in routine analysis. For an application, the metal ions that respond to H_2O_2 -luminol are monitored in rain. In aqueous alkaline solution, luminol is oxidized to 3-aminophthalate in the presence of H_2O_2 and metal ion as a catalyst, with the emission of light. The emission is blue, centered about 425 nm, and is from excited state of 3-aminophthalate. The emission intensity is directly proportional to the concentration of metal ion. The results are compared with those of GFAAS. Furthermore, application of the system to determine Cr^{6+} is discussed in this work.

Experimental Section

Instruments. The schematic diagram of a batch-type

chemiluminescence system with air pump is shown in Figure 1. The system is equipped with a peristaltic pump (Miniplus 3, Gilson, France) for delivering luminol and H_2O_2 to the reaction cell. A Y-shaped element is used for mixing luminol and H_2O_2 , which is set at the inlet of the reaction cell. The cell has a cylindrical body (10 mm i.d. and 8 mm in height) and a flat quartz window bottom. The top of the reaction cell has a wide opening for sample injection. The inlet was positioned at 6 mm from the bottom of the cell and the outlet was near to the bottom.

No magnetic stirring system or other convective method was employed due to the limited cell volume. Emission from the cell was collected by a PMT (R4632, Hamamatsu, Japan) powered by a HV power supply (-900V, C3830, Hamamatsu, Japan). The lab-made PMT housing was attached to the bottom of the cell. The reaction cell and PMT housing were sealed in a dark box (50 \times 50 \times 120 mm). The output of the PMT was fed into a current pre-amplifier ($\times 10^7$, Oriel, model 70710) and transferred to a data acquisition board. The data acquisition system consisted of

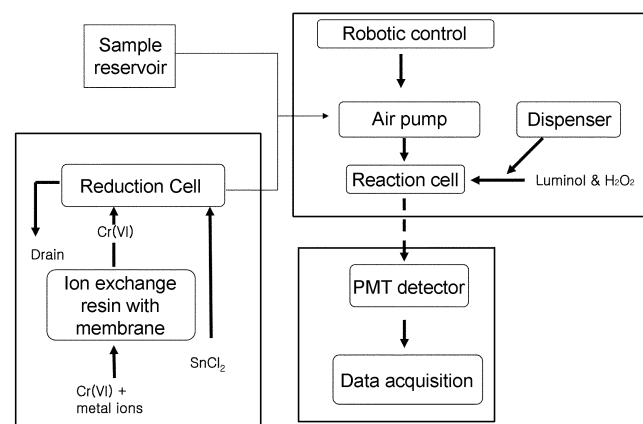


Figure 1. Schematic diagram of a batch-type chemiluminescence system with air pump.

an I/O interface (± 15 V input) with A/D converter and a computer with lab-made graphics software.

Air pump. The air pump was developed for microliter sample injection and consisted of a capillary and a glass tube with a tungsten filament inside. Sample was taken up into the capillary by capillary action from a sample reservoir of about 1.0 mL. The sampled volume, therefore, depended on various factors such as capillary material, solution viscosity, and air temperature of the pump. The injection driving force was air expansion in the glass tube. The cylindrical tube was 38 mm in length with a 4.7 mm i.d. The tip of the capillary (1.5 mm i.d., about 100 mm long, bent) attached to the tube was coated with a Teflon (J-308, O&V Lub Chem. Co., Korea) in order to prevent droplet formation on the outer surface during suction and injection. For injection, the air expansion of the pump tube must be at least 177 mm^3 assuming the capillary was fully filled with sample solution, meaning that a temperature increase of about 20.7°C is required for complete discharge. About 3 A of current with 2V (Power supply: HS-1505D, HwaSung electric. Co., Korea) was applied to the filament (Tungsten, 0.2 mm diam. and 110 mm in length) for few seconds, which ensured complete discharge from the capillary. After injection, the tube was cooled down quickly to room temperature by an air fan for the next measurement. A cycle of the process, *i.e.*, suction for sampling, movement to the reaction cell, and injection was automatically controlled and repeated by a lab-made robotic system manually programmed by Labview 6.0 (National Instrument, USA).

Rain analysis. For an application, the system was used to determine the concentration of metal ions in rain, and the analytical results were compared with those obtained by GFAAS (Model: SOLAAR GF-AAS, Thermo elemental, England, UK). The GFAAS manufacturer's recommended temperature programming for water analysis was used. The rain sample was collected using a funnel (about 300 mm in radius) positioned at ~ 5 m above the ground outside the lab and was delivered by gravity to about 1.0 mL glass reservoir placed inside the lab through a tygon tube. The sampled volume for a measurement in GFAAS was $50 \mu\text{L}$. The overflowed rain water was then delivered to the other reservoir inside the chemiluminescence system using a peristaltic pump for chemiluminescence measurement. Four measurements were performed for each data point throughout this experiment.

Separation column and reduction coil for Cr^{6+} measurement. Two steps such as cation exchange with resin and reduction of Cr^{6+} to Cr^{3+} were required to ensure the elimination of interfering cations and the complete reaction. For interference elimination, the Teflon tube (20 mm diam. and 30 mm in length) with porous PE membrane filter was packed with cation exchange resin ($0.4\text{--}0.55$ mm in effective size, 1.9 meq mL^{-1} for capacity, Samyang Co., Korea). Below the Teflon tube, a reaction coil (30 mm in length and 1.5 mm i.d.) with injection port for SnCl_2 and a filter (Model: 6779 1304, PVDF syringe filter, Whatman, England) were used for the complete reduction reaction of Cr^{6+} . The

reduced Cr solution was then pumped into the reservoir for measurement using the chemiluminescence system described above.

Reagents. Luminol (5-amino-2,3-dihydrophthalazine-1,4-dione, Aldrich Chem. Co., USA) of 0.05 M was prepared in borate buffer. Hydrogen peroxide (Dongwoo Fine Chemical, LTD, Korea) of 0.1 M was mixed with luminol in a 1 : 1 volume ratio and then placed into the reaction cell. All solutions and standard working solutions were prepared using doubly distilled deionized water of $18.3 \text{ M}\Omega$. Stock solutions of $1,000 \mu\text{g mL}^{-1}$ for Fe and Cu were prepared from chloride salts in 1% HCl solution. Cr^{3+} and Cr^{6+} were prepared by dissolving CrCl_3 and Potassium dichromate (99.99%, TEDIA Company, Inc, USA). The working solutions for each different matrix were prepared daily. Tin(II) chloride (General grade, Samchun pure chemical industries LTD, Korea) of 0.1M was used as a reducing agent for Cr^{6+} .

Results and Discussion

Air pump. Several control factors must be considered in order to achieve a reproducible sample volume from the air pump, such as heating current and the physical properties of the capillary surface, which would be affected by temperature and the expansion coefficient. In order to verify the reproducibility of injection volume, the cumulative mass added to a weighing bottle was measured with a balance and plotted, as shown in Figure 2. The average mass of each injection (droplet) was found to be $17.6 \mu\text{g}$ with a reproducibility of $< \pm 0.2\%$ relative standard deviation. It indicated that, unlike with a peristaltic pump, no fluctuation of the amount of sample uptake or discharge, *i.e.*, injection volume, was observed. In addition, between ambient temperature and 30°C , no change in signal intensity was observed as a function of temperature variation of the reactants. Above 30°C the intensity decreased with increasing the temperature probably because of some hydrogen peroxide decomposition.¹⁵ This system also exhibited high throughput (about $100 \text{ samples h}^{-1}$) and low reagent consumption.

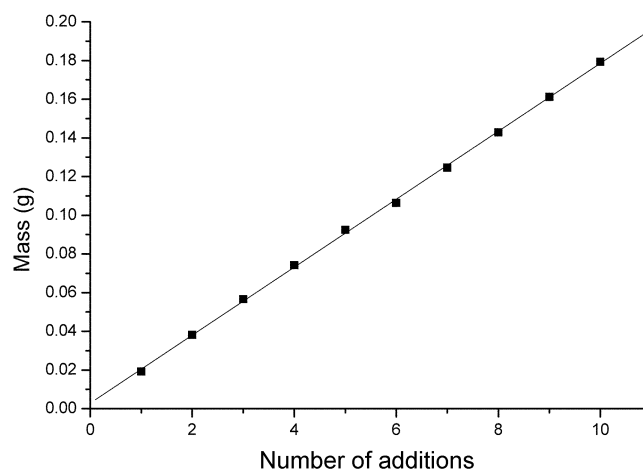


Figure 2. Cumulative mass of sampled volume added to a weighing bottle using air pump.

About 300 μL of the reagent mixture was placed in the cell for a measurement, so the ratio of sample volume to the volume of luminol and hydrogen peroxide was about 0.057. It meant that all analytes of each sample droplet react instantly with the luminol-hydrogen peroxide reagent. The sampled volume would be a trade-off between signal intensity and lifetime of the reagent in the cell. Up to a certain point, a larger sample volume would provide the higher signal. However, if too much sample was used, repeated measurements wouldn't be feasible due to high background with a long tailing of the peak and short lifetime of the reagent. In this experiment the reagent was drained out and refilled after single droplet measurement. The integrated area of each time-resolved profile was used for quantitative analysis throughout this experiment.

Optimization and chemiluminescence reaction. Assuming the mixing process in chemiluminescence is not rate-limited and the luminescence lifetime is long, the following equations are valid.¹⁶

$$I_{\text{CL}} = \Phi_{\text{CL}} k [\text{L}]$$

where, I_{CL} is a measured intensity, $[\text{L}]$ is the analyte concentration as a function of time, k is the rate constant, and Φ_{CL} is the efficiency of the chemiluminescent reaction.

For this type chemiluminescence system, the assumptions can be true only if the diffusion rate of a droplet injected onto the surface of the luminol- H_2O_2 using the air pump is much faster than the reaction rate. Since the diffusion coefficient of water is $\sim 2.2 \times 10^{-5} \text{ cm}^2 \text{ s}^{-1}$, the chemiluminescent reaction can be limited at very small area. Therefore, in this batch mode without any convection, a low concentration of analyte with small amount of sample is required for the study of reaction kinetics. Otherwise the diffusion-limited reaction will prevail and the profiles for all analytes will be similar to each other under the same experimental condition, which is unlikely in this case. Typical kinetic parameters, such as the chemiluminescence formation and decay rates, can be easily estimated from the straight segments of the rising and falling portions of the intensity profile.¹⁷ Both parameters are directly related to analyte concentration.

To test the analytical performance, some 10 ng mL^{-1} solutions of reactive analytes, such as Co^{2+} , and Cr^{3+} , were tested using the lab-made system. A peak profile for each metallic ion was obtained, and the signal intensity of the analyte was recorded in time. The peak profile of Cr^{3+} was quite different from that of Co^{2+} . For example, Figure 3 shows the plots for Co^{2+} and Cr^{3+} . The kinetic response curves obtained as intensity profiles correspond to a first order sequence of two consecutive steps, generation of the light-emitting product by mixing and formation of the end product. As shown in the figure, the generation rate of the light-emitting product for Cr^{3+} by mixing with luminol- H_2O_2 was slower than that of Co^{2+} . However, the formation rate of the end product for Cr^{3+} was faster than that of Co^{2+} because Cr^{3+} decayed faster than Co^{2+} .

Determination of Cr (III) and Cr (VI). Peak areas were integrated for quantitative analysis. The calibration curve for

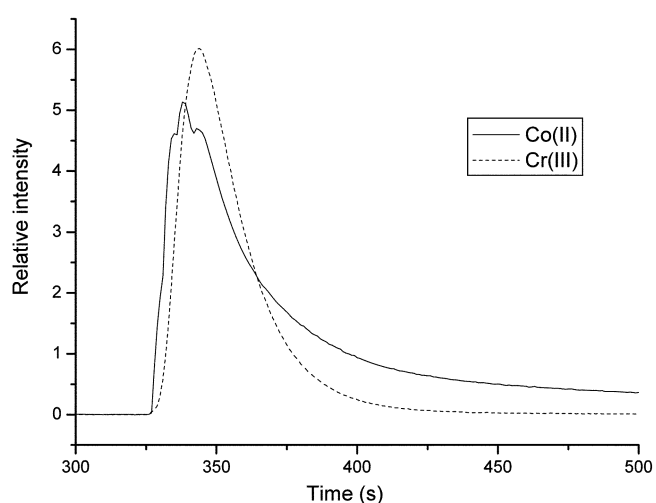


Figure 3. Modified signal profiles of Cr (10 ng mL^{-1}) and Co (2 ng mL^{-1}).

Cr^{3+} using manually modified standard solutions in the range of 0.5 ng mL^{-1} to 20 ng mL^{-1} was obtained with a linear calibration coefficient of 0.9910. From the curve, the limit of detection (3s of blank) was found to be 11.1 pg mL^{-1} with a 4.4% RSD. For the concentrations greater than 20 ng mL^{-1} , the calibration curve became non-linear with positive deviation. This limit of detection for Cr^{3+} was lower than that of Fe^{2+} and Cu^{2+} , which were usually in the range of 20–50 pg mL^{-1} .

For the determination of Cr^{6+} , it was reduced to Cr^{3+} by SnCl_2 . The completeness of the reaction and spectral interference were checked by measuring the absorbance of Cr^{6+} at 350 nm and 440 nm and that of Cr^{3+} at 573 nm and 402 nm using a spectrophotometer. The 0.1 M SnCl_2 solution didn't absorb at wavelength over 310 nm. The absorbance of Cr^{3+} increased after the reduction reaction. If the concentration of SnCl_2 became high, a precipitate stuck to the filter and blocked the flow, resulting in poor reproducibility. The calibration curve for Cr^{6+} was obtained using this online measurement technique in the concentration range of 5 to 50 ng mL^{-1} . The limit of detection was 85.0 pg mL^{-1} with a 3.4% RSD.

Application to rain samples. The system was used for monitoring of trace metal ions in rain drops for environmental application. The sampling position was about 5 m above the ground in the Natural Science Building of Dankook University in the center of Seoul, Yongsan-ku, Hannam-dong. Since the signal was generated by the reaction of active metal ions dissolved in the rain drops with luminol- H_2O_2 , no elemental selectivity can be gained from the result. As shown in Figure 4a, the output of the system was recorded as a voltage that was saturated at 10 V. The monitored signal obtained using this system decreased significantly for the first 30 min and then increased to the saturation point of the detector 80 min after the rain began.

In order to compare the data with the separate information about metal ions, the concentrations of Fe, Cr, Pb, and Cu were determined by GFAAS (Fig. 4b). The same trend seen

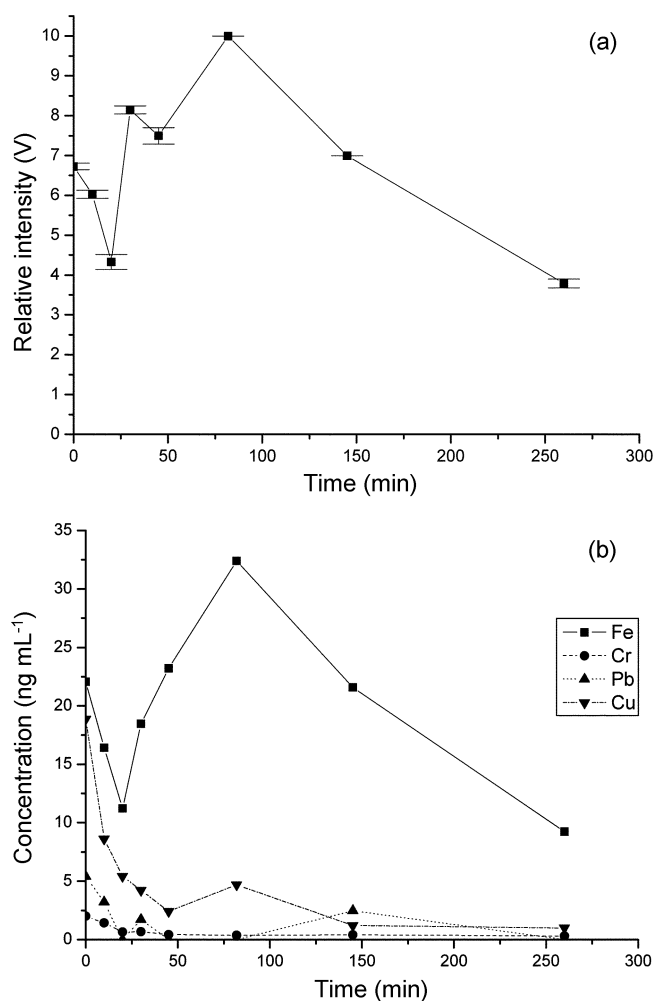


Figure 4. Concentration change of trace metal ions in rain (rainfall 78.5 mm, humidity 86%, temperature 24.1 °C): obtained with (a) chemiluminescence and (b) GFAAS.

in the chemiluminescence measurement was observed for the Fe concentration measured by GFAAS. The concentration of Cu increased slightly near 80 min but not as significantly as Fe. The signals for other metals decreased with time. This disparate behavior of each metal in the rain can be explained by the change of soluble particles in the air as a function of time during a rain storm. When rain begins, soluble metal ions in the air are dissolved into rain drops. As those metals dissolve, their concentrations decrease with time. This is why all the signals decrease for the first 30 min. Meanwhile, rain drops hitting the ground can cause a turbulent cloud of fine particles to rise gradually into the air. Since the major components of the cloud were from the soil, which contained mostly silicon and iron oxide, the signal from the dissolved Fe ions in rain increased until the ground was completely wet. Si ions weren't reactive in this chemi-

luminescence reaction. The particle cloud would be washed out gradually, which caused the Fe signal decrease seen in Figure 4b. Presumably, the chemiluminescence signal from the rain drops in this experiment was mostly caused by the reaction of luminol-H₂O₂ reagent with Fe ions although interference due to other metal ions might be significant.

In conclusions, the air pump for micro sample injection was manufactured for chemiluminescent detection of ultra-trace metals in environmental samples. The pump was operated by the capillary action for suction and heating for injection of small and reproducible sample volumes without any pulsation that is common in peristaltic pumps. This system exhibited high throughput (about 100 samples h⁻¹) and low reagent consumption. For an application, Cr⁶⁺ was determined after the reduction to Cr³⁺ using SnCl₂. This system can also be used as a sensor for online monitoring of trace metal ions in rain drops for environmental application due to its high sensitivity. Although it doesn't have selectivity for each metal ion, it will give us information of total contaminated metal level for the control of environmental guideline.

Acknowledgement. This work was supported by Internal Research Fund of Dankook University, 2004.

References

- Chon, H. T.; Ahn, J. S.; Jung, M. C. *Environ. Geochem. Health* **1998**, *20*, 77.
- Hirata, S.; Yoshihara, H.; Aihara, M. *Talanta* **1999**, *49*, 1059.
- Croot, P. L.; Laan, P. *Anal. Chim. Acta* **2002**, *466*, 261.
- Sekaly, A. L. R.; Chakrabarti, C. L.; Back, M. H.; Gregoire, D. C.; Lu, J. Y.; Schroeder, W. H. *Anal. Chim. Acta* **1999**, *402*, 223.
- Santosa, S. J.; Tanaka, S. *Anal. Lett.* **1995**, *28*, 509.
- Hoffmann, P.; Karandasev, V. K.; Sinner, T.; Ortner, H. M. *Fresenius' J. of Anal. Chem.* **1997**, *357*, 1142.
- Baechmann, K.; Haag, I.; Steigerwald, K. *Atmosph. Environ.* **1995**, *29*, 175.
- Hiraide, M.; Chen, Z. S.; Kawaguchi, H. *Anal. Sci.* **1995**, *11*, 333.
- Kawakubo, S.; Iwatsuki, M. *Anal. Sci.* **2000**, *16*, 945.
- Escobar, R.; Lin, Q.; Guiraum, A. *Analyst* **1993**, *118*, 643.
- Steitz, W. R.; Suydam, W. W.; Hercules, D. M. *Anal. Chem.* **1972**, *44*, 957.
- Paleologos, E. K.; Vlessidis, A. G.; Karayannis, M. I.; Evmiridis, N. P. *Anal. Chim. Acta* **2003**, *477*, 223.
- Zhang, Z.; Qin, W.; Liu, S. *Anal. Chim. Acta* **1995**, *318*, 71.
- Yang, W.-P.; Zhang, Z.-J.; Deng, W. *J. of Chromato. A* **2003**, *1014*, 203.
- Yang, W.-P.; Zhang, Z.-J.; Deng, W. *Anal. Chim. Acta* **2003**, *485*, 169.
- Schulman, S. G.; Schulman, J. M.; Rakicioglu, Y. *Chemiluminescence in Analytical Chemistry*; Garcia-Campana, A. M.; Baeyens, W. R. G., Eds.; Marcel Dekker, Inc.: New York, 2001; Chap. 3, p 77.
- Murillo Pulgarin, J. A.; Garcia Bermejo, L. F.; Rubio Aranda, J. A. *Anal. Chim. Acta* **2004**, *517*, 111.