# *Article*

# Turbidimetric Determination of Sulfate in Rainwater Employing a LED Based Photometer and Multicommuted Flow Analysis System with In-Line Preconcentration

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No presente trabalho é proposto um procedimento automático para determinação turbidimétrica de sulfato em águas de chuvas, baseado no processo de multicomutação em análise em fluxo. O equipamento fotométrico foi desenvolvido no laboratório, e inclui um LED ($\lambda$ = 420 nm), uma cela de fluxo com caminho óptico de 100 mm e um fotodiodo. Uma etapa de preconcentração foi incluída para melhorar a sensibilidade. Após estabelecer as condições operacionais adequadas, o procedimento apresentou as seguintes características: resposta linear na faixa de concentração entre 0,1 a 2,0 mg L$^{-1}$ de sulfato, limite de detecção (critério 3$\sigma$) de 0,04 mg L$^{-1}$ de sulfato, desvio padrão relativo de 1,5% (n = 7) para uma solução com 0,5 mg L$^{-1}$ de sulfato, consumo de 17,0 mg de cloreto de bário por determinação, volume de efluente de 7.3 mL por determinação e frquência de amostragem de 13 determinações por hora.

In the present work, an automatic procedure for turbidimetric determination of sulfate in rainwater, based on the multicommuted flow analysis approach, is proposed. The photometer set-up was developed at the laboratory, and includes: a LED ($\lambda$ = 420 nm), a long pathlength flow cell (100 mm), and a photodiode. A preconcentration step is included in order to improve sensitivity. After establishing the adequate operational conditions, the proposed procedure presented the following useful features: a linear response ranging from 0.1 mg L$^{-1}$ up to 2.0 mg L$^{-1}$ sulfate, a detection limit (3$\sigma$ criterion) of 0.04 mg L$^{-1}$ sulfate, a relative standard deviation of 1.5% for 0.5 mg L$^{-1}$ sulfate solution (n = 7), barium chloride consumptions of 17.0 mg per determination, and waste generation of 7.3 mL *per* determination. Accuracy assessment was achieved by applying the paired t-test between results obtained using the proposed procedure and ICP-OES and showed that there is no significant difference at 95% confidence level.

**Keywords**: sulfate, turbidimetry, multicommuted flow analysis, LED-based photometer, ion exchange resin, rainwater

## Introduction

During the last three decades, rainwater acidity and composition have been widely studied in order to take measures against acid rain. This type of water produces deterioration of historic buildings and acidification of lakes.[1] It may also adversely affect ecosystems and, indirectly, human health.[2] For these reasons, it is very important to know the rainwater composition in order to prevent atmospheric pollution. Acid rain has as its main constituent sulfate ion and its principal contributor to pollution is sulfur dioxide, which is oxidized in the atmosphere to sulfate and converted into an environmentally harmful strong acid ($H_2SO_4$).[3-6]

Traditionally, the most commonly used procedures for sulfate determination have been based on measurements of barium sulfate turbidity,[7] methylthymol blue colorimetry,[8,9] or ion chromatography.[10,11] Sulfate has been also determined by inductive coupled plasma optical emission spectrometry (ICP-OES).[12] Turbidimetric methods based on flow analysis approaches have also been implemented for the determination of sulfate in river water,[13-16] and these have shown good characteristics, such as high sample throughput, low reagents consumption, high precision, and accuracy.

The detection limit of about 2 mg L$^{-1}$ is not enough for some water samples. Therefore, improvement of the flow system has been proposed, in order to overcome this drawback. For example, Santos Filha *et al.*[17] proposed the utilization of an anion exchange resin column, thereby allowing sulfate determination in river water at low concentration levels. Nevertheless, the flow system presented high reagents consumption and waste generation. These disadvantages can be overcome by employing an analytical procedure based on multicommutation flow analysis process (MCFA).[18-20]

In the present work, we intend to develop a multicommuted flow analysis procedure for sulfate determination in rain waters. The method is based on the reaction between sulfate anions and barium cations, resulting in the formation of a precipitate (barium sulfate), and its turbidimetry is monitored at 420 nm using a LED-based photometer developed at the laboratory. The system set-up is designed to afford the following facilities: automation, low cost instrumentation, high sensitivity, low reagent consumption, and robustness. The flow system includes a set of solenoid valves, which are assembled to work as independent commutation devices in order to allow an efficient handling of sample and reagent solutions.

## Experimental

### Apparatus

The equipment set-up consisted of an IPC-8 Ismatec (619 Oak Street, Oak Harbor, WA, USA) peristaltic pump equipped with Tygon pumping tubes; five three-way solenoid valves HP161T031 and a dual three-way solenoid valve 360T041 (NResearch, 267 Fairfield Ave, West Caldwell, NJ, USA); a solenoid micro-pump 090 SP (BioChem. 85 Fulton Street, Boonton, NJ, USA), nominal volume of 8 μL *per* pulse; a homemade LED-based photometer,[21] equipped with a boron-silicate glass flow cell with an 100 mm optical path-length and a 2.0 mm inner diameter, a blue LED (λ = 420 nm, high-bright and narrow radiation beam) and a photodiode; a homemade resin column machined in an acrylic block as depicted below; a microcomputer furnished with an electronic interface card PCL-711S (Advantech Inc, 38 Tesla Street, 100 Irvine, CA, USA), a software written in QuickBasic 4.5 and a homemade electronic interface to drive the solenoid valves and the solenoid pump,[19] joint devices of acrylic, and reaction coil and flow lines made of polyethylene tubing (0.8 mm i.d.). To allow accuracy assessment, a model Optima 3000DV sequential inductively coupled plasma optical was used.emission spectrometer (Perkin-Elmer - Norwalk, Connecticut, USA) was used for the determination of sulfate in rainwater, which was operated following its operation manual recommendation.

### Reagents and samples

All reagents were of analytical grade and all solutions were prepared with purified water (conductivity < 0.1 μS cm$^{-1}$). A 1000 mg L$^{-1}$ sulfate stock solution was prepared by dissolving 1.376 g of ammonium sulfate in 1000 mL of water. A 300 mg L$^{-1}$ sulfate solution was obtained by rigorous dilution of the previous one. This was used to prepare the working standard solutions with concentrations of 0.10, 0.25, 0.50, 0.75, 1.00, 1.50 and 2.00 mg L$^{-1}$ SO$_4^{2-}$ and was done by appropriate dilution with water. A 0.3% (m/v) EDTA (disodium salt) in 0.2 mol L$^{-1}$ sodium hydroxide was used as cleaning solution. A 20% (m/v) barium chloride solution was employed as a precipitating agent, containing 0.1% Tween 80 as a stabilising agent. Eluent solution was composed of 1.0 mol L$^{-1}$ sodium nitrate in a 0.01 mol L$^{-1}$ nitric acid solution. A 0.5 mol L$^{-1}$ sodium chloride solution in a 0.01 mol L$^{-1}$ hydrochloric acid medium was used as a carrier fluid. Anion exchange resin AG1-X8 (Bio-Rad, 200-400 mesh) (chloride form) was utilized for sulfate in-line concentration. Before use, the resin was converted to nitrate form has been described elsewhere.[17]

A set of rainwater samples was collected at different points at the Piracicaba city using polyethylene trays, which were previously treated with a 10% nitric acid solution. Afterwards, the trays were rinsed two times with water and maintained sealed up to the used. After collection, samples were filtered and transferred to polyethylene flasks and maintained in a freezer at 4 °C. Before analysis, samples were equilibrated the laboratory temperature.

### Flow system and procedure

The column to pack the ion exchange resin was machined using an acrylic block and its exploded view is shown in Figure 1. To prevent fluid leakage, the screws (Sw)
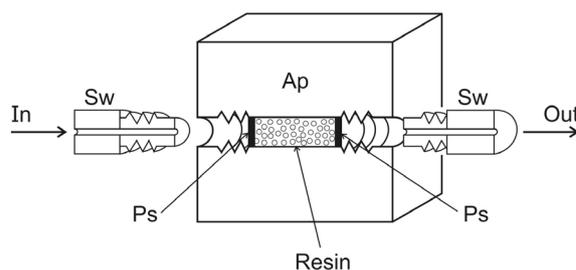


**Figure 1.** Exploded view of the resin column. Sw = screw of acrylic; Ap = acrylic plat; Ps = disk of polyester cloth; In and Out = input and output of fluid, respectively. The hole to pack the ion exchange resin is 20 mm long and 3 mm inner diameter.

**Table 1.** Sequence of the analytical run followed by the microcomputer

| Step | $V_1$ | $V_2$ | $V_3$ | $V_4$ | $V_5$ | $V_6$ | $P_1$ | time / s |
|---|---|---|---|---|---|---|---|---|
| $SO_4^{2-}$ pulses (4) | 0 | 1 | 1 | 0 | 0 | 0 | 1/0 | 0.1 |
| Preconcentration | 1 | 1 | 1 | 1 | 0 | 0 | 0 | 90 |
| Elution pulses (40) | 0 | 0 | 1 | 0 | 0 | 1/0 | 0 | 0.125 |
| Analytical signal | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 60 |
| EDTA | 0 | 0 | 1 | 0 | 1 | 0 | 0 | 60 |
| Washing | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 60 |

0 indicates that valve was OFF while 1 means that the valve was switched ON.

were capped with a Teflon sheet. The disks of polyester cloth prevented the resin beads from being carried away by the stream of solutions.

The flow-diagram of the flow system is shown in Figure 2. In this configuration, all valves were switched OFF, so that all solutions were pumped towards their storing recipient, expect for the carrier solution (Cs) that flowed through valves $V_3$, $V_1$ and $V_2$ towards the detector (Det).
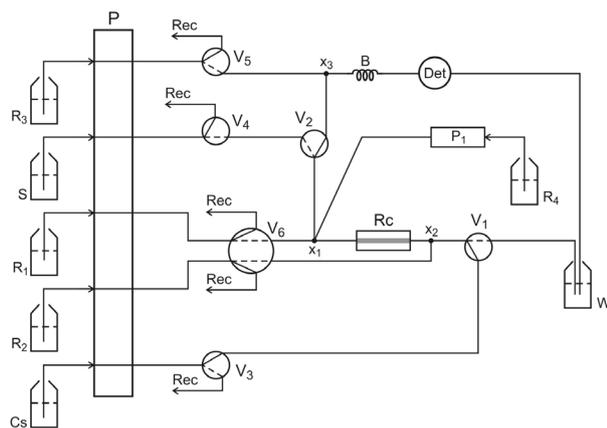


**Figure 2.** Diagram of the flow system. $V_1$-$V_5$ = three-way solenoid valves; $V_6$ = dual three-way solenoid valve; Cs = carrier solution, flow rate of 1 mL min$^{-1}$; $R_1$ = 20% barium chloride solution, of 1 mL min$^{-1}$; $R_2$ = eluent solution, of 1 mL min$^{-1}$; $R_3$ = EDTA solution, of 1 mL min$^{-1}$; $R_4$ = 300 mg L$^{-1}$ sulfate standard solution; S = sample, of 3.4 mL min$^{-1}$; P = peristaltic pump; $P_1$ = solenoid micro-pump; Rc = column resin, 20 mm long and 3.0 mm inner diameter; B = reaction coil, 100 cm long; Det = LED-based photometer. The interrupted and continuous lines in the valves symbols indicated the solutions pathway when the valves were switched ON or OFF, respectively.

When the control software was run, a preset step not shown in Table 1 was carried out in order to fill all flow lines with the appropriate solution. The microcomputer read the signal generated by the LED-based photometer through the analog/digital interface. This operational condition was maintained until a stable signal was achieved. Afterwards, the microcomputer switched ON/OFF the solenoid valves following the pattern depicted in Table 1.

In the first step, the solenoid valve $V_3$ was switched ON to direct the carrier solution towards its storing vessel, while mini-pump $P_1$ was switched ON/OFF four times to insert into the resin column (Rc) an aliquot of 32 μL of sulfate

solution ($R_4$). In the second step, $V_1$, $V_2$, $V_3$ and $V_4$ were switched ON at the same time to direct the sample stream through the resin column (Rc) towards the waste (W), thereby allowing the sulfate ions to be retained by the resin. After a pre-set loading time, these valves were switched OFF, and $V_6$ was switched ON/OFF to perform the eluting step. The dual three-way solenoid valve ($V_6$) permitted the insertion of both the eluent solution ($R_2$) and the barium chloride solution ($R_1$) to be done using a single signal control. The eluted sulfate ions merged with barium chloride solution in the confluence $x_1$, so that the reaction to form barium sulfate proceeded while the mix was being displaced by the carrier solution through the reaction coil (B) towards the waste (W). The signal generated by the photometer (Det), related with turbidity caused by the barium sulfate suspension, was read by the microcomputer through the analog input of the PCL-711 interface card, which converted the input to digital format. The digital data were stored as an ASCII file to permit further treatment. While measurements were performed, a plot of the signal was displayed as a time function on the microcomputer screen to allow its visualization in real time. The fifth step consisted in the introduction of the EDTA solution with the aim of cleaning the flow system. Finally, all the solenoid valves and the micro-pump were switched OFF, and the carrier solution was propelled through the analytical path for 60 s, followed by another analytical run.

The variables assayed to find the best operational conditions included: sample loading time, reagent concentration, and eluting and washing times, which are summarised in Table 1. With the aim of proving the effectiveness of the proposed procedure, samples of rainwater were processed using the best operational conditions. To allow accuracy assessment, samples were also analysed by ICP OES.

## Results and Discussion

### Effect of the sulfate addition

The turbidimetric procedure for sulfate determination based on the reaction with barium chloride and the detection

by photometry presents a limiting factor to improve sensitivity: the initial sulfate concentration required to start the nucleation process. Santos Filha *et al.*[17] have shown that this drawback could be overcome by inserting into the resin column a small volume of a concentrated sulfate standard solution. Since for the determination of sulfate in rainwater a high sensibility is required, this strategy was adopted for the proposed procedure. Prior to beginning each analytical replicate, a small volume of concentrated sulfate standard solution was inserted into the ion exchange resin column. In order to find the best condition for improving sensitivity, the volume of sulfate solution aliquot was varied from 8 to 40 μL, which was done by varying from one to five the number of electric pulses applied to the micro-pump $P_1$. The assays were performed using a set of sulfate standard solutions with concentrations of 100, 200, 300 and 400 mg $L^{-1}$. These experiments were performed using a resin column loading time of 180 s, yielding the results summarized in Table 2. Considering the slope and the linear coefficient as indicating parameters, we can see that best results were achieved when 32 μL of the 300 mg $L^{-1}$ sulfate solution was inserted. Thus, this value was selected.

**Table 2.** Effect of the sulfate standard solution volume inserted into the resin column

| Conc. / (mg $L^{-1}$) | Volume / μL | Intercept | Slope | Preconcentration $r^2$ |
|---|---|---|---|---|
| 100 | 24 | 0.158 ± 0.008 | 0.152 | 0.980 |
| 100 | 32 | 0.186 ± 0.012 | 0.174 | 0.982 |
| 100 | 40 | 0.240 ±0.009 | 0.183 | 0.992 |
| 200 | 24 | 0.332 ± 0.009 | 0.201 | 0.998 |
| 200 | 32 | 0.370 ± 0.015 | 0.287 | 0.998 |
| 200 | 40 | 0.486 ± 0.010 | 0.259 | 0.996 |
| 300 | 24 | 0.443 ± 0.011 | 0.224 | 0.998 |
| 300 | 32 | 0.478 ± 0.007 | 0.348 | 0.999 |
| 300 | 40 | 0.558 ± 0.009 | 0.277 | 0.997 |
| 400 | 24 | 0.453 ± 0.014 | 0.236 | 0.998 |
| 400 | 32 | 0.477 ± 0.008 | 0.325 | 0.998 |
| 400 | 40 | 0.542 ± 0.010 | 0.289 | 0.998 |

*Effect of the sample volume*

In the current manuscript, the ion exchange resin column was employed to improve sensibility. Since this parameter could be affected by the volume of sample that flowed through the resin column, a set of assays was carried out in order to select the best condition. These assays were done by varying the sample loading time from 60 up to 360 s, yielding the results shown in Table 3. As we can be observed, the signal increased up to the volume of 180 μL. The time interval of 90 s was selected as a compromise between sampling throughput and sensitivity required for sulfate determination in rainwater. These results show that samples with concentrations lower than 0.10 mg $L^{-1}$, would be analyzed increasing the sample loading time.

*Effect of the eluting time*

A 1.0 mol $L^{-1}$ sodium nitrate solution, used as an eluent, was prepared in an acidic medium (0.10 mol $L^{-1}$ $HNO_3$) to improve the kinetic of the barium sulfate formation. The eluting process and the insertion of barium chloride occurred at the same time by switching ON the dual three-way solenoid valve ($V_6$), which was assembled to direct the eluent solution stream through the resin column in the inverse direction to that of the sample solution. This eluting strategy could prevent the dispersion of the eluted sample zone through the resin column.

Furthermore, the strategy prevented the resin from compacting at the end of the column, which would have produced an excessive increase of inner pressure, causing fluid leakage, and thus hampering the preconcentration process.

In Table 1, it was shown that eluent and barium chloride solutions were inserted at the same time by switching ON the solenoid valve $V_6$, thereby permitting control over the volumes of both solutions. In order to study the effects of varying the volumes of these solutions, the time interval to maintain this valve switched ON was varied from 0.125 up to 1.0 s, and the number of ON/OFF cycles was varied from 10 up to 50. In this way, different calibration

**Table 3.** Effect of sample loading time

| Loading time / s | Volume / mL | Blank Solution (Absorbance) | Sulfate Solution (Absorbance) | Difference in Absorbance |
|---|---|---|---|---|
| 60 | 3.4 | 0.45 | 0.50 | 0.05 |
| 90 | 5.1 | 0.48 | 0.56 | 0.08 |
| 120 | 6.8 | 0.52 | 0.63 | 0.11 |
| 180 | 10.2 | 0.52 | 0.68 | 0.16 |
| 360 | 20.4 | 0.54 | 0.70 | 0.16 |

The assays were carried out using a 0.10 mg $L^{-1}$ sulfate standard solution.

curves were achieved and the best results were obtained for a time interval of 0.125 s and 40 insertion cycles. Under this condition, a linear response represented by the equation: Absorbance = (0.315 ± 0.023) x + (0.312 ± 0.053) (preconcentration $r^2$ = 0.999) was achieved. In this case, the volumes of 165 μL of both sodium nitrate solution and barium chloride solution were delivered. Taking into account the results, the time interval of 0.125 s and 40 insertion cycles were selected as the most adequate.

*The use of an EDTA cleaning solution*

Since barium sulfate suspension could precipitate into the reaction coil and flow cell, which would cause both clogging and base line drift, the EDTA solution was employed to prevent this drawback. As was indicated in Table 1, after the signal reading step, an aliquot of this cleaning solution was inserted into the reaction coil (B). In order to find the appropriate EDTA solution volume, the time interval for maintaining valve $V_5$ switched ON was varied from 30 up to 120 s. It was observed that a time interval of 60 s was enough to avoid the baseline drift. Afterwards, the carrier solution was propelled through the resin column for a time interval of 60 s to regenerate the ion exchange resin prior to beginning the next analytical run.

*Performance of the proposed system*

The assays previously described were carried out to select the working variables, which are shown in Table 1. In order to verify the performance of the proposed system, a set of sulfate standard solutions was processed using those variables. The records displayed in Figure 3 show that no base-line drift occurred, thereby indicating a good long-term stability. The calibration curves were linear within the
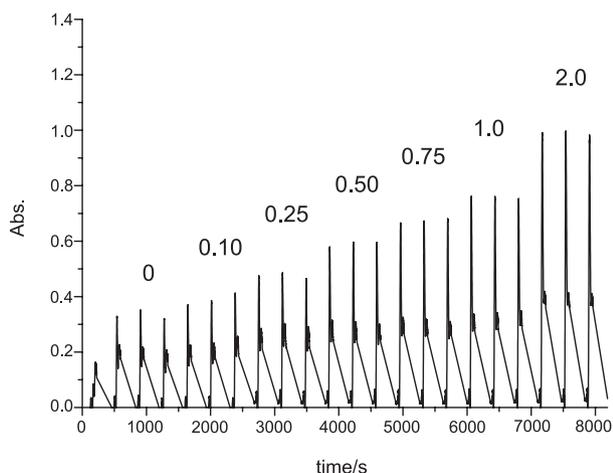
range of 0.1 to 2.0 mg $L^{-1}$ sulfate, presenting the following linear equation: y = (0.341 ± 0.023) x + (0.334 ± 0.049), and a linear correlation coefficient of preconcentration $r^2$ = 0.999. Other useful features of the procedure include a relative standard deviation of 1.5% (n = 7) obtained by processing a standard solution with a concentration of 0.5 mg $L^{-1}$ sulfate, a detection limit of 0.04 mg $L^{-1}$ sulfate, a sampling rate of 13 determinations *per* hour, barium chloride, eluent ($NO_3^-$), and EDTA consumptions of 17.0 mg, 9.3 mg, 6.0 mg, respectively, *per* determination, and waste generation of 7.3 mL *per* determination.

After establishing the hardware configuration of the instrument setup showed in Figure 2, it was used daily during six month, while the development of the analytical procedure was in course, affording similar response without requiring any adjustment. The geometry of the resin column showed in Figure 1, was used at the first time in the present manuscript. The screws attached to its ends avoided resin displacement and as well as fluid leakage. The resin loaded into the column could be used during two weeks.

In order to prove the usefulness of the proposed procedure, a set of rain water samples collected at different places in Piracicaba city (Brazil) was analyzed. In order to carry out the accuracy assessment, samples were also analyzed by ICP-OES methodology, yielding the results shown in Table 4. Applying the paired t-test between results for a 95% confidence level, the calculated value (t = −2.43) was less than the tabulated value (t = 2.31), indicating that there was no statistically significant difference between the results.

**Table 4.** Comparison of results

| Sample | Proposed system[a] | ICP-OES[a] |
|---|---|---|
| 1 | 1.405 ± 0.015 | 1.398 ± 0.018 |
| 2 | 0.829 ± 0.015 | 0.858 ± 0.010 |
| 3 | 0.613 ± 0.019 | 0.634 ± 0.038 |
| 4 | 0.670 ± 0.012 | 0.694 ± 0.039 |
| 5 | 0.943 ± 0,008 | 0.979 ± 0.041 |
| 6 | 0.761 ± 0.017 | 0.763 ± 0.054 |
| 7 | 0.897 ± 0.009 | 0.895 ± 0.073 |
| 8 | 0.602 ± 0.043 | 0.625 ± 0.019 |
| 9 | 0.674 ± 0.019 | 0.670 ± 0.086 |

[a]Mean sulfate concentration and standard deviation (mg $L^{-1}$). Results average of three consecutive measurements.

## Conclusions

Albeit, the instrument setup described in the current manuscript has been constructed using not expensive devices, which were acquired expending about US$ 1200,



**Figure 3.** Records related to sulfate standard solutions. The numbers on the records correspond to the sulfate concentrations expressed in mg $L^{-1}$.

the results discussed before showed that it presented a good performance, including long-term stability and minor operator intervention.

Low consumption of reagents (17.0 mg $BaCl_2$, 9.3 mg $NO_3^-$ and, 6.0 mg EDTA), and low volume of waste generation (7.3 mL) could be also considered as a useful feature afforded by the proposed procedures. In addition, the use of a long pathlength cell (100 mm), enhances the sensitivity of the proposed procedure.

The quality of the results obtained by using the developed MCFA turbidimetric method is similar to that obtained by applying the ICP-OES, methodology, presenting agreement between results at 95% confidence level, could be considered as an unequivocal indication of the procedure effectiveness.

Finally, the usefulness of the equipment set-up has been proved. Thus, we can affirm that the procedures should be used for routine laboratory analysis.

## Ackowledgments

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