GAS DIFFUSION-FLOW INJECTION DETERMINATION OF FREE AND TOTAL SULFUR DIOXIDE IN WINES BY CONDUCTOMETRY

Petr KUBAN[a], Pavel JANOS[b] and Vlastimil KUBAN[a1]

[a] *Department of Chemistry and Biochemistry, Mendel University of Agriculture and Forestry, Zemedelska 1, 613 00 Brno, Czech Republic; e-mail:* [1] *kuban@mendelu.cz*

[b] *Department of Analytical Chemistry, Masaryk University, Kotlarska 2, 611 37 Brno, Czech Republic*

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Sensitive flow injection analysis methods for the determination of free and total sulfur dioxide in wines are presented. The bound S(IV) was liberated by alkaline hydrolysis with 4 mol/l NaOH. All forms of S(IV) were liberated from the sample zone by sulfuric acid and subsequently transported through a microporous PVDF membrane. The penetrated gases were collected in water for preselected period and determined by conductometry with detection limit 1 mg/l and relative standard deviations 0.8 and 0.6% at 10 and 150 mg/l (n = 10) for free and total S(IV), respectively. The results are comparable with those obtained by standard titrimetric procedures with visual (Czech State Standard) and/or potentiometric indication.

Key words: SO_2 Determination; Flow injection analysis; Gas diffusion; Sulfur dioxide; Wine.

Sulfur dioxide and oxoanions of S(IV) are extensively used as food additives because of their cheapness, effectiveness and flexibility. They are easily oxidized with dissolved oxygen, which is the basis for efficient removal of oxygen from various materials. They can be further used to control numerous detrimental processes. In wines, their use is indisputable since they serve several purposes simultaneously, both during fermentation and in storage. They function as antioxidants, antimicrobial agents or enzyme inhibitors and, also, they control enzymatic and nonenzymatic browning reactions.

Due to its extensive use in foodstuff, sulfur dioxide has become a subject of legislation owing to harmful reactions that may occur in sulfite-sensitive individuals. The reactions may include asthmatic episodes, anaphylactic reactions and hypotension. The content of sulfur dioxide in wine is, of course, controlled by law in almost all countries, the levels of free and total SO_2 differing (30 and 180 mg/l) for white and red wine, respectively (40 and 200 mg/l, in the Czech Republic, see below).

The sulfite added to wines is present partly in free ionic forms (as HSO_3^-, SO_3^{2-}), H_2SO_3 and SO_2 (g) and partly complexed by the other constituents (bound to carbonyl or unsaturated compounds and/or phenol derivatives, acetaldehyde, anthocyanine pig-

ments, *etc*.). There is an enormous need for controlling the contents of different forms (speciation) of S(IV) in wines and other beverages (free, bound, total). To determine the total content of S(IV), the bound part has to be liberated before the analysis.

Numerous classical and standard methods to determine free and/or total SO_2 in wines and other beverages are available[1–6]. Determinations are performed immediately after opening a bottle to prevent any losses of the analyte. Most of the methods are based on iodometric titration with visual or potentiometric indication of the equivalence point. Free SO_2 is determined after acidification with a strong mineral acid, whereas the total amount only after hydrolysis of various sulfite complexes and sometimes distillation. The dissociation of complexes is promoted by dilution and acidification with sulfuric (water or ethanolic solution) or phosphoric acid or by alkalinization with NaOH. The dissociation can be accelerated by heating the sample in a circulating water bath. The methods are labor and time-consuming. None of them is convenient for modern routine analysis.

Several spectrophotometric flow injection analysis (FIA) methods have been proposed[7–9]. They are based either on the reactions with pararosaniline[10–15], 4-aminoazobenzene[16], Chloramine T (ref.[17]) or 5,5′-dithiodi(2-nitrobenzoic acid)[18] or on decolouration of Malachite Green[19–21]. Toxicity of the reagents, complex reaction schemes and low throughputs of the methods are the main disadvantages. Other FIA methods are based on the chemiluminiscence[22,23] and electrochemical[24–32] detection.

The simultaneous determination of CO_2 and SO_2 is based on a non-selective potentiometric detection of both analytes and subsequent selective detection of SO_2 by pararosaniline method[12]. The simultaneous determination of the free and total SO_2 usually employs soft acidification of the sample for liberation of free species and alkalization to liberate bound SO_2 with subsequent acidification to determine both forms[20]. The sample volume is divided into two independent subzones entering two separate channels of the FIA apparatus employing two detectors configured either in series or parallel arrangement. Conductometric FIA methods for determination of free and total SO_2 are described in the present paper and the results are compared with those obtained by titrimetric procedures[33,34].

EXPERIMENTAL

Chemicals and Instruments

A standard solution (1 g/l) of Na_2SO_3 was prepared from solid sodium sulfite and stabilized with 4 g of NaOH and 0.5 ml of HCHO (370 g/l) of reagent grade purity (Merck, Germany) per litre. A serious decrease in an analytical signal with time was observed when a non-stabilized solution of Na_2SO_3 was used (*ca* 10% decrease in 30 min), whereas the stabilized solution gave a stable signal for at least one day. Other chemicals (H_2SO_4, NaOH, EDTA, I_2, KI, KSCN, Na_2CO_3, $Na_2S_2O_3$, *etc*.) were commercial of analytical grade purity (Lachema, Brno, Czech Republic). Bidistilled water from

quartz apparatus Bi-18 Destamat (Heraeus Quarzschmelze, Hanau, Germany) was used for all purposes.

FIA manifolds (Fig. 1) were constructed from a peristaltic double-rotor pump (variable speed 0.05–5 ml/min, Skala, Brno, Czech Republic), a chromatographic syringe pump HPP 4001 (variable speed 0.05–5 ml/min, Laboratorni pristroje, Prague, Czech Republic), an electronically operated six-way injection valve DKR 01 (Labeco, Spisska Nova Ves, Slovakia), a reaction system mounted from Teflon capillaries (0.5 mm i.d.) and T-pieces from polypropylene (Ark-Plas Inc., Flippin, U.S.A.), a conductometric detector CD-1 with a flow-through cell (Labio, Prague, Czech Republic) and a line recorder TZ 4620 (Laboratorni pristroje, Prague, Czech Republic).

A membrane separation unit of coaxial type was mounted from an outer Teflon tube (1.7 mm i.d., 2.5 mm o.d.), polypropylene T-pieces (Ark-Plas Inc., Flippin, U.S.A.) and an inner microporous PVDF [poly(vinylidene difluoride)] capillary membrane (0.8 mm i.d., 1.0 mm o.d., 4.5 μm, 10 cm length, Enka, Wuppertal, Germany).

Potentiometric titrations[34] were performed using an electronic pH-meter pH 573 (WTW, Germany) equipped with a combined Pt–Ag/AgCl (1 mol/l KCl saturated with AgCl) electrode (Radiometer, Copenhagen, Denmark) and an electromagnetic stirrer LM2 (Laboratorni pristroje, Prague, Czech Republic).

Acidity of samples was checked with a pH-meter OP 208/1 using a combined glass–Ag/AgCl electrode OP 0808P (Redelkis, Budapest, Hungary). The pH-meter was calibrated with a set of standard buffer solutions (S 1306, S 1326, S 1336 of pH 2.18, 7.00 and 9.18 at 25 °C, Radiometer, Copenhagen, Denmark).

Working Procedures

According to the Czech State Standards (CSN 56 0216, Part 7), the content (in ppm or mg/l) of free and total SO_2 is limited for different types of wine (Table I). The content is determined by visual iodometric titration[35] with starch as an indicator. The equivalence point can be alternatively determined by potentiometry. The mean value of at least three titrations is calculated. The variance, the

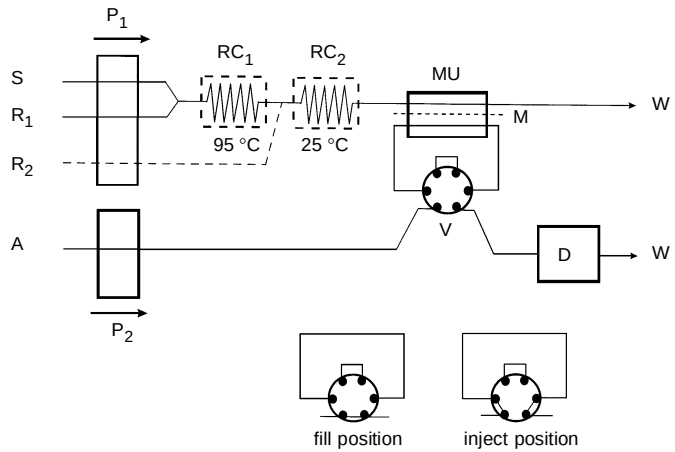

FIG. 1

Scheme of the FIA manifolds used for the determination of free and total SO_2. S sample, P_1 peristaltic pump, P_2 chromatographic pump, R_1, R_2 NaOH, H_2SO_4 (see text for details), A H_2O, RC_1, RC_2 reaction coils, MU membrane unit, M PVDF membrane, V valve, D detector, W waste

difference between the lowest and the highest values, lower than 2 and 5 mg/l for free and total SO_2, respectively, can be tolerated.

Figure 1 shows the scheme of the manifold for FIA determination of free and total SO_2. For either method, corresponding modification of the manifold was employed.

For the determination of free SO_2, R_2, RC_2 and the two termostatting baths are excluded from the manifold (Fig. 1). The FIA determination of free and ionic S(IV) is based on the evolution of SO_2 by acidification of wine (S) with sulfuric acid (R_1). The liberated SO_2 is transported through a semipermeable membrane (M) into an acceptor liquid (A). The penetrated gas forms H_2SO_3. Its zone is washed out from the membrane unit (MU) into a flow cell of a conductometric detector (D). The analytical signal is proportional to the content of S(IV) in wine.

The determination of the total (free and bound) sulfur dioxide in wine, ranging typically from 50 to 200 mg/l, is based on quantitative conversion of all forms of S(IV) to SO_2 either by acid or alkaline hydrolysis of chemically and physically bound S(IV) groups. The acid hydrolysis is performed using the manifold from Fig. 1 where R_2 is excluded. The sample (S) is mixed with 4 mol/l sulfuric acid (R_1) and heated at 95 °C in the reaction coil RC_1. The second termostatting bath is used for the temperature control of the analyte zone prior to its enter into the membrane unit. The alkaline hydrolysis (see manifold from Fig. 1, no termostatting baths are employed) is done at laboratory or slightly elevated temperature using a strong base like NaOH and KOH at a concentration of 4 mol/l (R_1). The converted sulfite is then liberated with 4 mol/l sulfuric acid (R_2) to form SO_2 (g) and non-selectively transported through a semipermeable membrane into a suitable acceptor solution.

RESULTS AND DISCUSSION

Choice of the Acceptor Media

Two different acceptor solutions are most frequently used for SO_2 collection – water or a strongly diluted hydrogen peroxide solution. In the former case, H_2SO_3 is formed, whereas in the latter sulfur dioxide is converted to H_2SO_4. The latter case seems to be more convenient for conductometric detection because of the pK_{ai} values ($pK_{a2}(H_2SO_4)$ = 1.92 while $pK_{a1}(H_2SO_3)$ = 1.77, $pK_{a2}(H_2SO_3)$ = 7.20). Despite double relative conductance of the collected S(IV) in 0.1 g/l hydrogen peroxide, water was selected as the most suitable acceptor because of the better baseline stability.

TABLE I
Limits of SO_2 mass concentration (mg/l) in wines according to the Czech State Standard

Wine	Free SO_2	Total SO_2
White	30	180
Red	40	180
Sweet	30	200
Tokay	30	200
Fruit	40	200

Optimization of the FIA Manifold Parameters

Experimental conditions were optimized using the manifolds depicted in Fig. 1. The flow rates of a carrier, Q_W, a sample solution, Q_S, and modifiers, Q_A and Q_B, the length of the dispersion capillary between an injector and a detector, L, and the time of diffusion, t_2, were optimized to obtain the highest analytical signal at the shortest time of analysis t_{ef} which is defined as the period needed to reach half of the maximum analytical signal. The parameters optimized, together with the ranges and optimum values are summarized in Table II. Maximum analytical signal (relative conductance) was chosen as the response. In the optimization study, one variable at a time was varied while the other variables were kept constant. Unless stated otherwise, 1 mol/l sulfuric acid as a modifier and 0.1 mol/l solution of Na_2SO_3 as a sample solution were used.

The relative conductance increases with decreasing flow rate of the acceptor medium (Fig. 2) and the t_{ef} also remarkably increases. The highest analytical signal was obtained at $Q_W = 0.08$ ml/min giving the optimum sensitivity but $t_{ef} = 180$ s was too long to be acceptable for the analysis. Time t_{ef} was practically independent of the flow rate at $Q_W > 0.4$ ml/min. Also the peak shape was nearly Gaussian at the flow rate $Q_W = 0.4$ ml/min. A compromise between sensitivity and speed of analysis was selected using $Q_W = 0.4$ ml/min. On the other hand, the analytical signal increases with flow rates of the sample solution and modifier (sulfuric acid) up to 0.56 ml/min and is independent of higher flow rates. The flow rates of 0.56 ml/min were selected as the best from the dependence of analytical signal on the flow rate (Fig. 2). Any excessive flow rate increases reagent consumption and thus the cost of analysis. The higher flow rates can be useful for the determination of trace concentrations of sulfur dioxide (down to 10 mg/l), when the calibration curve is nonlinear (concave) due to the low concentration of H_2SO_3 in the acceptor medium[36].

TABLE II
Parameters in the optimization study

Parameter	Range	Optimum value
Q_W, ml/min	0.08–0.80	0.40
Q_S, $Q_{A,B}$, ml/min	0.08–1.12	1.12[a], 0.56[b]
L, cm	5–40	15
t_2, s	40–100	100[a], 40[b]
c_A, mol/l	0.01–4	1[a], 4[b]
c_B, mol/l	1–4	4[b]

[a] Determination of free SO_2; [b] determination of total SO_2.

The analytical signal obtained at the optimum conditions (Q_W = 0.4 ml/min, $Q_A = Q_S$ = 0.56 ml/min) was found to be independent of the length of the dispersion capillary from the point of injection to the detector, ranging from 5 to 40 cm. A nonsignificant decrease in the analytical signal appears for longer coils due to dispersion of the sample zone. The length of 15 cm was used in further experiments because of practical aspects of manifold configuration.

Time of diffusion (preconcentration time), t_2, was varied for several concentrations of SO_2 at the constant washing time, t_1 = 60 s, which is sufficient for complete rinsing the penetrated sulfur dioxide zone from the membrane unit and flow cell of the detector. The diffusion time required for obtaining a measurable signal depends on the analyte concentration. Thus, 40- and 100-s periods were selected as the optima for concentrations above 50 mg/l and from 10 to 50 mg/l, respectively.

Interference Study

The diffusive transport of gaseous components through the microporous PVDF, PTFE, *etc*. membranes is typically non-selective. All gases have approximately the same transport efficiencies due to similar values of diffusion coefficients. All volatile and gaseous species present in a sample solution are potential interferents. Interfering effects of all ions evolving gases (carbonates, thiosulfates, cyanides, sulfides, *etc*.) have to be controlled, especially of those present in wine. Because of the high toxicity and low probability of their presence in wine, hydrogen cyanide and hydrogen sulfide were not tested. Low dissociation of these species in water solution and thus low relative conductance can be expected owing to their relatively high dissociation constants. Thiosulfate

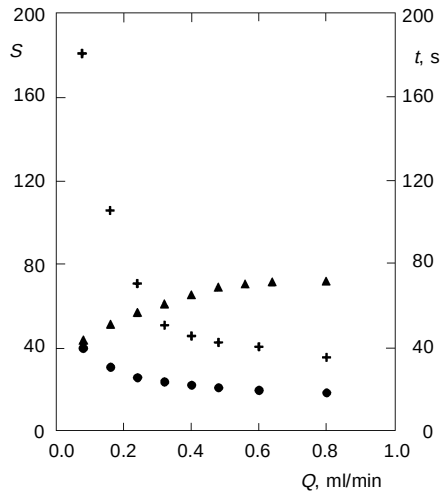

FIG. 2
Dependence of relative conductance (●, ▲) and time of analysis t_{cf} (+) on flow rate of acceptor (●, +) or donor (▲) solutions

caused serious problems at concentrations above 0.01 mol/l because of the disproportionation to elemental sulfur in acid solution according to the equation

$$S_2O_3^{2-} + 2\ H^+ \rightarrow SO_2 + H_2O + S$$

which caused membrane blocking.

Interferences of carbonates and thiocyanates were tested in detail. Analytical signal of carbonate at concentration of 10 mmol/l was independent of sulfuric acid concentration over the range 0.1 to 4 mol/l while an exponential increase of the signal for thiocyanate under the same conditions was obtained. Sulfuric acid at concentration of 1 mol/l was found to be optimum for determination of free SO_2 (see below) and thus used throughout this study. An 0.1 mmol/l solution of Na_2SO_3 corresponding to 6.3 mg/l SO_2 was used as a reference solution to check interference of carbonates and thiocyanates over the range 0.1 to 100 mmol/l (Fig. 3). No interference of the ions was observed under 10- and 50-fold concentration excess of carbonates and thiocyanates, respectively. The fact is interesting because of the dissociation constants of both species ($pK_{a1}(H_2CO_3)$ = 6.37, pK_a(HSCN) = 0.85). No interference from thiocyanates can be expected in wine since their content is zero or negligible. The carbonate interference is a more difficult problem especially when champagne or wines saturated with CO_2 are analyzed. In addition to the interfering effects, bubbles are formed in the propulsion and injection part of the manifold because of the acidity of wine and the decrease in pressure after opening the bottle. To prevent the interfering signals, carbon dioxide has to be removed by simple filtration, evacuation or sonication. Filtration was selected as the most effective procedure and no losses of sulfur dioxide were observed. The other procedures are more complicated and time-consuming.

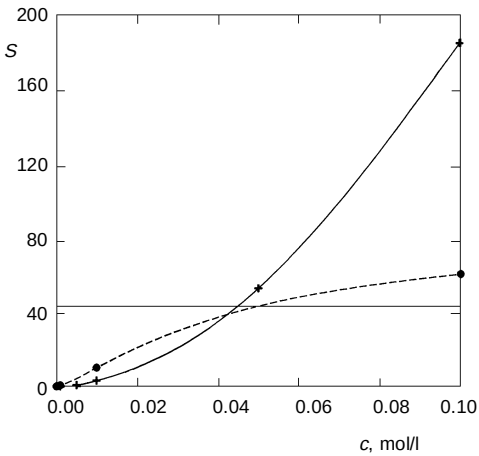

FIG. 3
Dependence of relative conductance on molar concentrations of carbonate (●) and thiocyanate (+). Horizontal line corresponds to the relative conductance signal of 0.1 mmol/l Na_2SO_3

Choice of Conditions for the Determination of Free SO_2

The concentration of sulfuric acid (c_A) as a modifier was varied over the range 0.001 to 4 mol/l at 50 mg/l SO_2 and flow rate $Q_S = Q_A = 0.56$ ml/min for sample and sulfuric acid, respectively. According to our tests, the most suitable concentration of sulfuric acid is 1 mol/l (maximum of the analytical signal).

The time of diffusion of sulfur dioxide through the membrane, t_2, is critical for its determination in wine. A non-linear calibration curve was obtained at very low concentrations and short times, its shape being nearly exponential. The longer diffusion times and/or the higher flow rates the more linear calibration curves were obtained (Fig. 4). When preconcentration time of 100 s and flow rate $Q_S = Q_A = 1.12$ ml/min were used, the calibration curve is linear from 10 to 50 mg/l. This corresponds to typical concentrations of S(IV) in wine and to the CSN (Czech State Standard) limits. Only a small curvature appears at concentrations below 10 mg/l. Using longer preconcentration time (200–300 s) widens the interval but longer times of analyses are needed.

Choice of Conditions for the Determination of Total SO_2

Two thermostats have to be used in the case of acid hydrolysis (see Fig. 1 for the manifold) since the temperature of the hydrolysis and of the final reaction mixture passing the detector are critical for the precise and accurate results. Both parameters, the yield of the hydrolytic reaction and the relative conductance of the acceptor solution strongly increase with temperature (1 and 2% per degree, respectively), and thus it must be precisely controlled. Also the heat transport from the solution to the flow cell increases its regular temperature if the length of the reaction capillary is insufficient. The necessity to use two water baths complicates the FIA manifold for the determination of the total SO_2. The results are higher than those obtained by iodometric titration

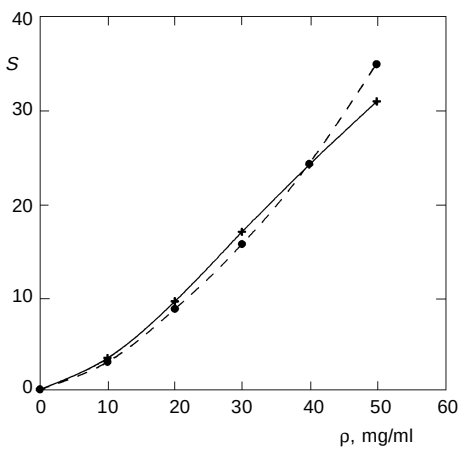

FIG. 4
Dependence of relative conductance on mass concentration of free SO_2 for different preconcentration times (s): ● 40, + 100

after alkaline hydrolysis at room temperature according to the CSN. The fact can be explained by the difference in hydrolytic destruction of sulfur bonds (probably higher efficiency of destruction) and hence a more careful study of the alkaline hydrolysis was performed.

To obtain the best sensitivity, the conditions of hydrolysis (reaction coil length, NaOH and sulfuric acid concentrations) should be optimized. The analytical signal of the conductometric detector is independent of the length of the reaction coil, RC_1, for both red and white wine (50–300 cm length was tested). Thus S(IV) is liberated quickly (immediately in a flow system) and the hydrolysis is fast enough to be highly effective in flow systems. A two-meter capillary was used for further experiments.

At least 4 mol/l NaOH (c_B) was necessary for fast and quantitative destruction of sulfur bounds but the analytical signal still increased with increasing concentration of NaOH (*ca* 2% per 1 mol/l). Consequently, 4 mol/l sulfuric acid (c_A, see Table II) was used as modifier R_2 (Fig. 1). The use of highly concentrated NaOH (*i.e.*, 8 mol/l) and, of course, sulfuric acid is inpractical due to the cost of analysis, environmental aspects and also an easier leakage of the solution caused by the low adhesion of tubes to T-pieces.

Evaluation of the Methods for Determination of Free and Total SO_2

Detection limit (x_D) is defined as the amount of an analyte producing a signal corresponding to the threefold standard deviation of the baseline signal. The x_D = 1 mg/l, was determined by successive dilution of the standard solution of Na_2SO_3.

Reproducibility of the newly developed method is one of the most important parameters which express the quality of the procedure in a single laboratory measured with a single sample. The reproducibility was better than 0.8 or 0.6% (as relative standard deviation s_r, n = 10) at 10 and 150 mg/l of SO_2 for free and total SO_2, respectively.

Calibration curves for the determination of free and total SO_2 were linear from 10 to 50 and from 50 to 200 mg/l, respectively, under optimum conditions. The corresponding correlation coefficients were r = 0.9995 and r = 0.9999, respectively.

Ruggedness of a method means the ability of a procedure to be independent of small changes in experimental conditions, *e.g.*, flow rate, concentrations of all components and temperature. The influence of the concentration of modifiers and flow rates of a sample and modifiers on the variation of an analytical signal was tested. A solution at concentration of 10 mg/l SO_2 was injected into the FIA manifold for the determination of free SO_2 and individual parameters (flow rates of water and sample, concentration of sulfuric acid) were successively changed over ±10% around the optimum. The method is robust to changes in concentration of sulfuric acid (±10% variation), no influence being observed. The changes in flow rates of the sample and acceptor streams (±10% variation) significantly affect the analytical signal (Table III). The variation exceeds the reproducibility of the method (0.8%). Similar observations were made when testing the

ruggedness of the method for determination of total SO_2. The method is very robust (at 100 mg/l SO_2) to changes in sulfuric acid concentration (±10% variation) but significant changes in the analytical signal were observed for ±10% variation in NaOH concentration and flow rates of sample and acceptor solutions (Table III). Flow rates must be strictly controlled because of small particles of Tygon tubing, any precipitate, mechanical defects, any leakage of solutions and other factors can seriously influence the flow rates and the flow rate ratios. All connections, purity of introduced solutions and quality of the propelling system must be controlled to prevent irregularities in operation of the manifold. Additionally, NaOH concentration must be controlled since the yield of the hydrolysis is strongly dependent on it.

Determination of Free SO_2 in Wine Samples.

The results (n = 7) of the FIA method for seven wine samples are comparable with those obtained by visual titrimetry (n = 3), the differences being lower than 0.9 mg/l SO_2 (Table IV). The difference is lower than the declared variance of CSN (±2 mg/l for individual determinations). It is clear that the FIA method is more than acceptable for determination of free S(IV) in wine and it can supplement or replace the tedious standard titrimetric method according to the CSN.

Determination of Total SO_2 in Wine Samples

The results of the FIA method are comparable with those obtained by standard titrimetry (Table IV) for seven different wine sorts (differences lower than 5 mg/l). Only in

TABLE III
Ruggedness of the FIA method for free and total SO_2

$c(H_2SO_4)$ mol/l	Signal mm	c(NaOH) mol/l	Signal mm	$Q(H_2O)$ ml/min	Signal mm	Q^a ml/min	Signal mm
			Free SO_2				
0.9	25.1 ± 0.1	–	–	0.36	27.5 ± 0.1	0.51	26.0 ± 0.0
1.0	25.1 ± 0.0	–	–	0.40	25.5 ± 0.1	0.56	25.0 ± 0.0
1.1	25.1 ± 0.1	–	–	0.44	23.3 ± 0.1	0.61	23.6 ± 0.0
			Total SO_2				
3.6	33.6 ± 0.1	3.6	33.6 ± 0.0	0.36	37.3 ± 0.2	0.51	33.0 ± 0.0
4.0	33.6 ± 0.1	4.0	34.5 ± 0.2	0.40	33.8 ± 0.1	0.56	35.2 ± 0.1
4.4	34.0 ± 0.0	4.4	34.0 ± 0.0	0.44	33.1 ± 0.0	0.61	36.8 ± 0.1

[a] Q(sample) = $Q(H_2SO_4)$ = Q(NaOH).

one case the difference was higher (*ca* 10 mg/l) than the CSN limit. The results of the FIA method were typically lower than those of titrimetry probably because the effectiveness of the hydrolysis is insufficient from time to time. The deviations do not exceed the value ±10 mg/l in all tested cases. The FIA method can be used for fast determination of total S(IV) in wine and only if the content of SO_2 is close to the CSN limit (180 or 200 mg/l), the titrimetric method ought to be used to verify the exact content.

CONCLUSIONS

The final test of three methods, visual titrimetric[33], potentiometric[34] and FIA, using three types of wine (white, red and white champagne) confirmed the applicability of all the methods in routine practice. All three methods give the same results (n = 3). The differences were lower than 0.8 mg/l and 5 mg/l for free and total SO_2, respectively, thus being within the limits given by CSN 56 0216. Both, FIA and potentiometric titration are suitable for routine practice and they can supplement or totally replace the classical titration with visual indication of the equivalence point.

Determination of the equivalence point is much easier and accurate in potentiometric titration, especially when red and dark white wines are analyzed and the colour transition is unclear. The time of analysis is approximately the same as for visual titration; however, a better accuracy compared with the visual method is obtained.

In addition to about seven times shorter analysis times, the FIA method allows full automation of the determination using an autosampler and a computer-operated FIA analyzer. Also, the sample volume is reduced *ca* 25 times, the reagent consumption is

TABLE IV
Comparison of the results obtained by FIA and titration methods

Sample	Free SO_2, mg/ml		Total SO_2, mg/ml	
	FIA	titration	FIA	titration
1[a]	6.7	6.7	98	100
2[a]	10.9	10.0	121	119
3[b]	25.7	26.4	136	139
4[b]	15.4	14.8	100	103
5[b]	21.3	21.6	124	128
6[a]	6.3	6.4	96	99
7[b]	11.4	11.2	100	110

[a] Red wine; [b] white wine.

significantly lower (*ca* 10–20 times) and the time and costs of analyses are drastically reduced. Last but not least, a higher throughput and simplification of laboratory operations should be mentioned.

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