Determination of Glutaraldehyde by Photometric Microtitration

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Addition reaction with sodium sulfite was used for determination of low concentrations of glutaraldehyde. The amount of released hydroxide ions was proportional to the content of glutaraldehyde and was objectively determined by means of spectrophotometrically observed neutralization microtitration in a special apparatus.

The deteriorating quality of the environment and consequently stricter emission limits force the producers of pollutants to modernize their current technologies, to reduce technologies with toxic materials or to minimalize emissions.

The industrial use of formaldehyde in various fields such as textile, wood, leather, and food processing industry causes a major environmental problem. Formaldehyde is a highly toxic matter which, due to its volatility, not only contaminates working environment but also remains in residual concentration in final products, so that it endangers consumers. One of the possibilities to solve this problem is to replace formaldehyde with less volatile aldehydes, *e.g.* glutaraldehyde. For this case it will be necessary to have a fast, accurate and reliable method for its determination even in very low concentrations.

In papers published so far on glutaraldehyde determination various methods are mentioned. For example, the absorption of ultraviolet radiation is observed. Glutaraldehyde sample shows two maximums at the wavelengths of 235 nm and 280 nm. Absorption band at 235 nm is ascribed to polymer products which originate in glutaraldehyde solutions. Their absorption coefficient is about 160 times higher than that of glutaraldehyde and that is why they dominate in the spectrum [1]. Glutaraldehyde can also be determined by spectrophotometry after reaction with hydroxylamine, hydrazine or methylamine [2]. The polarographic method of determination consists in the conversion of glutaraldehyde to the imino form by reaction with ammonia. This form is more easily reducible than the original compound and in the DPP regime it shows a peak with maximum in the potential range from -1.2 V to -1.3 V against SCE. The position of the maximum is influenced by the value of pH [3]. Another method is gas chromatography. It can be realized in column filling with stationary phase Carbowax 20 M. Its usage is suitable namely if there are interfering materials in the analyzed solution. Furthermore, gas chromatography with mass detector is capable to solve completely the composition of glutaraldehyde solution [1]. For the determination of higher concentrations of glutaraldehyde its reaction with sodium hydrogen sulfite is used where appropriate bisulfite compound is formed. The unreacted amount of sodium hydrogen sulfite is determined by iodometric titration [4]. This determination is loaded with a systematic error. The reaction is reversible and during the titration of the excess of sodium hydrogen sulfite with iodine the addition product partially decomposed into aldehyde and sodium hydrogen sulfite. The magnitude of the error depends on the equilibrium constant of the reaction of aldehyde used and on the reaction period, on the amount of addition compound, and on the amount of iodine. Besides, for the determination an everyday check-up of the concentration of the unstable sodium hydrogen sulfite solution is necessary.

In this work, for the glutaraldehyde determination its reaction with sodium sulfite was applied, based on the following scheme

$$OHC(CH_2)_3CHO + 2\ SO_3^{2-} + 2\ H_2O \rightleftharpoons$$
$$\rightleftharpoons O_3^-S(HO)HC(CH_2)_3CH(OH)SO_3^- + 2\ OH^-$$
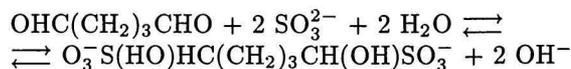
The amount of glutaraldehyde is measured on the basis of objectively determined amount of hydroxide ions.

EXPERIMENTAL

Model standard solutions of glutaraldehyde were prepared by the dilution of technical product the accurate concentration of which was determined by the Ponndorf method. This method is based on the oxidation of aldehyde groups by the excess of silver oxide. The unconsumed silver oxide after dissolution in di-
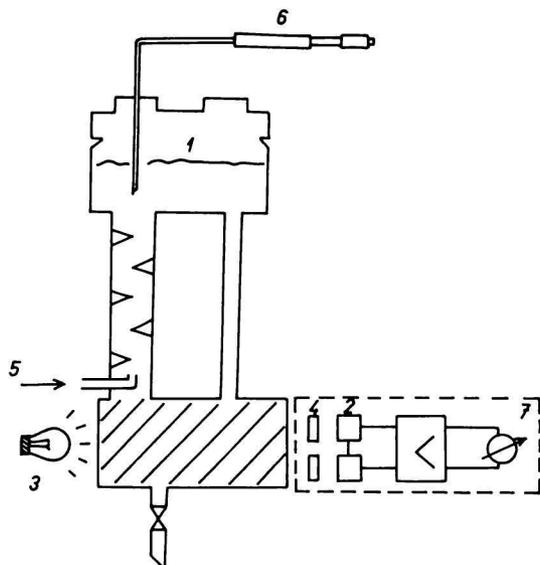
Fig. 1. The scheme of the device for determination of glutaraldehyde by the sulfite method. *1.* Reaction chamber; *2.* photoelectric sensors; *3.* source of light; *4.* optical filters; *5.* inert gas; *6.* manual or automatic titration burette; *7.* detector.

luted sulfuric acid was titrated with sodium chloride using potentiometric indication by a cell made of silver and reference silver chloride electrodes. The quantitative course of the reaction was supported firstly by the enhancement of the active area of Ag_2O with simultaneous precipitation of magnesium hydroxide, secondly by gradual increase of basicity of solution, by intensive stirring, and by completing the reaction in ultrasonic bath followed by heating to the boiling point.

All chemicals, supplied in anal. grade by Lachema, Brno and highly pure water obtained by equipment MILI Q (Millipore, Bedford, USA) were used for preparation of standard solutions.

The determination of glutaraldehyde by the sulfite method was performed on the apparatus the schematic diagram of which is in Fig. 1. The reaction itself took place in a small all-glass device [5] of the total volume of 80 cm^3 the bottom part of which was modified as a cuvette. The length of optical line was 25 mm. The inert gas argon was introduced into the vessel which not only ensured the stirring of the reaction mixture but also eliminated the influence of atmospheric CO_2 on the acid-base changes of the reaction solution. The volume of reaction mixture (or sample) containing 0.3 mass % of sodium sulfite, 0.001 M thymol blue and determined glutaraldehyde had to be selected between 30 cm^3 and 50 cm^3. If the volume was smaller then bubbles penetrated the solution and interfered with the spectrophotometric observation. The upper limit of the recommended volume is related to the total volume of the device and also to the quality of homogenization of reacting components.

Released hydroxide ions equivalent to the determined glutaraldehyde increased the basicity of the solution indicated by the change of colour of the acid-base indicator, thymol blue. This change of colour was scanned spectrophotometrically on an apparatus which utilized two light wavelengths corresponding to the absorption maximums of both colour forms of indicator and two sensors. During the process of determination, a change of sensors signal in relation to the change of colour of the reacting solution was observed. The original colour and thus also the original signal was achieved by manual or automatic dosing of the solution of 0.01 M-HCl. The amount of glutaraldehyde can be calculated from the total consumption of acid. The determination period did not exceed 5 min. Measurements of pH were performed on an MV 870 pH-meter (Präcitronic).

RESULTS AND DISCUSSION

The addition reaction of sulfite with aldehyde groups of glutaraldehyde is simple and does not need any special conditions. For its quantitative and sufficiently fast completion it is necessary to ensure a suitable sulfite ions concentration in the reaction solution.

The suitable concentration was found experimentally on the basis of analysis of known amount of glutaraldehyde with the usage of increasing concentration of sulfite ions. The consumption of acid which reflects the results achieved with different amount of SO_3^{2-} is shown in Fig. 2. With smaller concentrations of sulfite ions the reaction was probably not quantitative, which is shown by smaller consumptions of acid. The consumptions increased with rising concentration of SO_3^{2-} ions up to the value of approximately 0.2 mass %. The rise of actual concentration of sulfite up to the level of 1 mass % did not affect the consump-
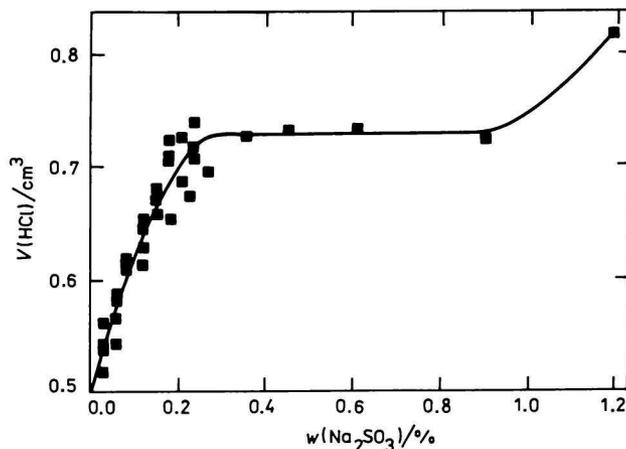

Fig. 2. Determination of glutaraldehyde at different concentrations of sodium sulfite using the sulfite method. (Given: 365.2 μg glutaraldehyde; c(thymol blue) = 0.001 mol dm^{-3}; c(HCl) = 0.01 mol dm^{-3}.)

Table 1. Reproducibility of the Determination of Glutaraldehyde Using the Sulfite Method
$w(Na_2SO_3) = 0.32$ %; $c(Thymol\ Blue) = 0.001$ mol dm^{-3};
$c(HCl) = 0.01$ mol dm^{-3}; given: 365.2 μg glutaraldehyde

Experiment	Consumption of HCl	m(Glutaraldehyde)	Δ	δ
	mm^3	μg	μg	%
1	744.5	374.2	9.0	2.46
2	737.5	370.6	5.4	1.48
3	726.0	364.9	−0.3	−0.08
4	741.0	372.4	7.2	1.97
5	739.5	371.6	6.4	1.75
6	743.5	373.7	8.5	2.33
7	738.5	371.1	5.9	1.62
8	720.1	361.9	−3.3	−0.90
9	746.0	374.9	9.7	2.66
10	737.0	370.4	5.2	1.42

Arithmetic mean: $x = 370.6$ μg
Estimation of standard deviation: $s_r = 4.136$ μg
Relative standard deviation: RSD = 0.011
Absolute error: $\Delta = 5.4$ μg
Relative error: $\delta = 1.48$ %
Interval of reliability for the significance level $1 - \alpha = 0.95$: (367.6—373.5) μg

Table 2. Determination of Different Glutaraldehyde Concentrations Using the Sulfite Method
$w(Na_2SO_3) = 0.32$ %; $c(Thymol\ Blue) = 0.001$ mol dm^{-3};
$c(HCl) = 0.01$ mol dm^{-3}

m(Glutaraldehyde) given	m(Glutaraldehyde) found*	Relative error	Standard deviation	Relative standard deviation
μg	μg	%	μg	
8.5	8.2	−3.5	0.473	0.058
17	17.2	1.2	0.391	0.023
34	34.6	1.8	0.713	0.021
68	66.9	−1.6	0.365	0.005
102	100.8	−1.2	0.718	0.007

*Mean calculated from five determinations.

tion of acid. Further increase of sulfite concentration in reaction solution caused higher values, which can be explained so that due to a high ionic strength of the solution and also due to its higher buffering ability the indicator system became less sensitive. On the basis of the acquired knowledge we recommend to determine milligram amounts of glutaraldehyde in a 0.25 up to 1 mass % solution of sodium sulfite. For all determinations the initial value of pH must be set up within the range of 8.5—8.6.

Reproducibility of the method under the recommended conditions was studied by repeated analyses of known amount of glutaraldehyde (365.2 μg). Achieved results are given in Table 1. For the calculation of the amount of glutaraldehyde from the consumption of the acid, the stoichiometric coefficient 2:1 (two moles of OH$^-$ ions released by one mole of glutaraldehyde) was taken into account. From the collected values and the following statistical calculations it can be seen that the method yields satisfactory results. The acquired data file was run by the program

Adstat [6]. It was verified to yield normal distribution. That is why conventional estimates of parameters were used for further evaluation in predictive analysis of the data. The average value of found amount of glutaraldehyde was 370.6 μg. The determination error did not exceed 3 %.

Further analyses proved that the consumption of acid reflects linearly the increasing amount of the determined glutaraldehyde. The amount was varied from 0.01 mg up to 0.5 mg. By the evaluation of the experimental data direct proportion equation $y = 1.99\ x$ was found which proves the passage through point 0. The accuracy of the calibration model was also confirmed by the high value of the correlation coefficient 0.99946.

The reliability and accuracy of the determination of low concentrations of glutaraldehyde is documented by Table 2. From the given values, also the limit of determination of the studied method can be set which is about 10 μg of glutaraldehyde in 30 cm^3 of the analyzed solution, i.e. 300 μg dm^{-3}. In case of analyses of smaller amounts the change of colour accompany-

Table 3. Determination of Glutaraldehyde by the Ponndorf Method
$c(AgNO_3) = 0.029$ mol dm^{-3};
$c(NaCl) = 0.5$ mol dm^{-3};
$c(KOH) = 0.22$ mol dm^{-3};
$c(MgSO_4) = 0.015$ mol dm^{-3}

Experiment	c(Glutaraldehyde) found
	mol dm^{-3}
1	0.2555
2	0.2511
3	0.2549
4	0.2505
5	0.2475

Arithmetic mean: $x = 0.2519$ mol dm^{-3}
Estimation of standard deviation: $s_r = 3.314 \times 10^{-3}$ mol dm^{-3}
Relative standard deviation: RSD $= 0.013$
Interval of reliability for the significance level $1 - \alpha = 0.95$:
(0.2478—0.2560) mol dm^{-3}

Table 4. Comparative Determinations of Glutaraldehyde by the Ponndorf and Sulfite Methods

Method	c(Glutaraldehyde) found
	mol dm^{-3}
Ponndorf	0.2519
Sulfite	0.2556

ing the addition reaction was less distinctive, which led to an unreliable determination of the end point and consequently to a higher determination error.

The results of the standardization of the basic glutaraldehyde solution by the Ponndorf method are presented in Table 3. The found value was considered correct.

Comparing the results of the determination of glutaraldehyde by this independent method with the sulfite method (Table 4), we can say they agree quite well. The sulfite method yields the result which is on average for about 1.6 % higher. This difference may be caused by the fact that the sulfite method was realized with a small amount of glutaraldehyde (376 μg), whereas the conventional titrimetric Ponndorf method could be applied only with much higher concentrations (about 30 times). Relative standard error was in both cases comparable.

For the realization of the designed method, the fact that the method is not selective as it is based on the reaction of aldehyde group, must be taken into account. Therefore it is suitable for the analysis of samples the qualitative composition of which is known. Interfering factors are materials of acid-base character. Unless they function in the given working pH area as buffers, they can be easily eliminated by the modification of pH of the sample.

The proposed method was applied for the determination of glutaraldehyde in hardening solutions used in the production of artificial guts. These solutions are either acidic (pH 2—3) or alkaline (5 % NaHCO$_3$). That is why it was necessary in the presence of thymol blue indicator first to alter pH to its working value between 8.5 and 8.6. Only then addition of sulfite having the same value of pH as that of working value used in the method could follow in order that it might react with aldehyde groups to release OH$^-$ ions. It was shown that this determination in case of alkaline samples containing a high concentration of sodium hydrogen carbonate was more correct when these ions were mostly eliminated by acidification of the solution and argon bubbling.

The results for the selected samples applied varied within 0.04 and 0.05 mass % of glutaraldehyde.

In conclusion, the designed alternative of sulfite method enables easy, fast, and correct determination of glutaraldehyde even in small concentrations. The method is suitable especially for not completely equipped service laboratories where the common iodometric sulfite method used is subject to aforementioned systematic error.

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Translated by J. Pšenička