rivatization of all formaldehyde oligomers yet the 40-μL injection resulted in a greater yield. Competing reactions, including spontaneous decomposition of BSTFA, may have marginally limited its availability for derivatization of the glycols and monomethyl ethers with the 30-μL injection.

Within the accuracy of measurement, all the formaldehyde equivalents in the vapor phase in equilibrium with formalin solution were in the form of methylal, methylene glycol, and the oligomers of poly(oxymethylene) glycol monomethyl ethers containing one, two, and three formaldehyde units.

Mass spectra for the Me_3Si derivatives of the formaldehyde condensates and methylal from the formalin headspace derivatization liquid phase appear in Figure 3 and are identical with the fragmentation patterns of the Me_3Si derivatives obtained for formaldehyde condensates in formalin solution (2).

CONCLUSIONS

Total formaldehyde content of the formalin headspace measured by a gas-phase trimethylsilylation procedure closely approximated the total formaldehyde content of the formalin solution headspace determined by chromotropic acid analysis. The vapor-phase species in equilibrium with a formalin solution were shown to be methylal, methylene glycol, and three oligomeric poly(oxymethylene) glycol monomethyl ethers.

Registry No. CH_2O, 50-00-0; H_2O, 7732-18-5; CH_3OH, 67-56-1; $CH_2(OH)_2$, 463-57-0; $HOCH_2OCH_3$, 4461-52-3; $HO(CH_2O)_2CH_3$, 19942-08-6; $HO(CH_2O)_3CH_3$, 87728-58-3; methylal, 109-87-5.

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Simple Spectrophotometric Method for Determination of Carbonyl and Sulfonyl Chlorides

Maciej Siewiński, Marianna Kuropatwa, and Apolinary Szewczuk*

Biochemical Laboratory, Institute of Immunology and Experimental Therapy, Polish Academy of Sciences, 53-114 Wrocław, Czerska 12, Poland

The method presented is based on the reaction of carbonyl and sulfonyl chlorides with sodium azide in water–acetone solution at room temperature for 10 min; the excess of azide is determined spectrophotometrically after conversion to a red complex with Fe^{3+}. Reactive anhydride can also be determined by this method. The application of the new method was demonstrated for monitoring conversion of sulfonyl chlorides to sulfonyl fluorides and for controlling the acylation of amino compounds with phthaloyl-L-glutamic anhydride.

A variety of methods for determination of carbonyl and sulfonyl chlorides have been described. Among them differential acid titrations, differential chloride titrations, and reductive titrations are the most precise (*1*). Spectrophotometric methods for determination of carbonyl chlorides are based on reaction with hydroxylamine, giving hydroxamic acid converted to red complexes with Fe^{3+} ions (*2*), or on reaction with 2-(nitrophenyl)hydrazine giving hydrazides, which in alkaline solution yield intensive colors (*3*). Carbonyl chlorides were also determined by high-performance liquid chromatography after their conversion to methyl esters (*4*).

During our studies on covalent inhibitors of penicillin amidase from *Escherichia coli* (*5*) we developed a new, precise, and fast method for determination of sulfonyl chlorides. This method is based on the reaction described by Curtius and Haas (*6*). Excess sodium azide can be easily determined spectrophotometrically, after its conversion to a red ferriazide complex. The last one was used for determination of inorganic azide (*7*) and for determination of organic acid azides after their alkaline hydrolysis (*8*, *9*). So the new procedure for determination of acid chlorides proceeds according to the following two reactions:

$$R\text{-}SO_2Cl\ (R\text{-}COCl) + NaN_3 \rightarrow R\text{-}SO_2N_3\ (R\text{-}CON_3) + NaCl \quad (I)$$

$$N_3^- + Fe^{3+} \rightleftharpoons (FeN_3)^{2+}\ \text{(red ferrazide complex)} \quad (II)$$

EXPERIMENTAL SECTION

Materials. Reagents synthesized in our laboratory are hexanesulfonyl chloride (*10*) redistillated at 114 °C/10 mmHg, phenylacetyl chloride (*11*), phenylmethanesulfonyl chloride (*12*) recrystallized from benzene–cyclohexane, para-substituted phenylmethanesulfonyl chlorides (*10*, *12*) and phthaloyl-L-glutamic anhydride (*13*). Commercial reagents acetyl chloride, butyryl chloride, butyric anhydride, 5-(dimethylamino)-1-naphthalenesulfonyl chloride, methynesulfonyl chloride, propionic anhydride, *p*-toluenesulfonyl chloride were purchased from Fluka AG. Chloroacetamide was obtained from Schering VEB Adlershof, phenylmethanesulfonyl fluoride was from Sigma Chemical Co., and sodium azide (over 99%) was from Serva. Other reagents were from POCh, Gliwice.

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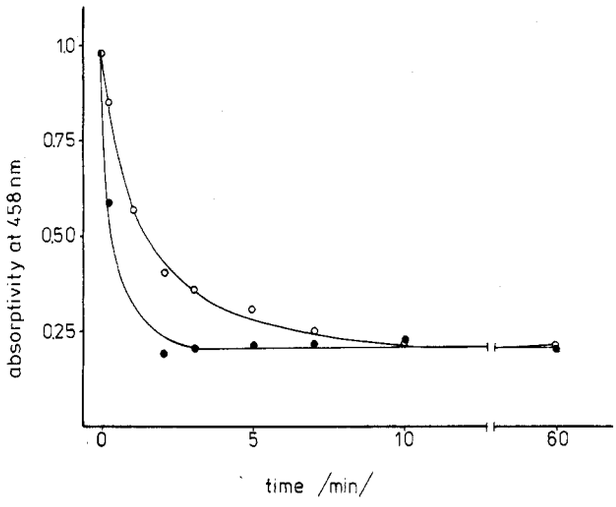

Figure 1. Effect of incubation time on the consumption of sodium azide (measured as ferroazide complex) by *p*-toluenesulfonyl chlorides (black circles) and hexanesulfonyl chloride (open circles). Details of determination are given in the Analytical Procedure section.

In all reagents the content of sulfonyl and carbonyl chlorides was determined with the morpholine method (*14*) and acid anhydrides were determined by the acid titrimetric method (*15*). Reagent solutions were (A) 0.1 M sodium azide (656.6 mg in 100 mL of water), (B) 0.05 M ferric sulfate (20 of g anhydrous $Fe_2(SO_4)_3$ dissolved in 1000 mL of water and filtered after cooling), and (C) 0.02 M organic acid chlorides in dried acetone. Solutions A and B are stable in the dark at room temperature. Solution C was prepared before using.

Analytical Procedure. Sodium azide (0.1 M, 0.025 mL) was mixed with 0.1 mL of 0.02 M organic acid chloride in a small test tube (1 × 10 cm) and left for 10 min at room temperature. Then 2.5 mL of 0.05 M ferric sulfate was added, the tube was well shaken, and after 2–10 min the absorption was measured at 458 nm against the blank. The amount of remaining sodium azide was read from the calibration curve. Molar extinction for NaN_3 at 458 nm was 10^6 cm^2 M^{-1}. The content of determined chloride was calculated by using

$$[(2.5\ \mu\text{mol} - \text{remaining } NaN_3, \mu\text{mol}) \times \text{mol wt of chloride}]/(\text{concn of sample, mg/mL})$$

RESULTS AND DISCUSSION

The reaction of *p*-toluenesulfonyl chloride with sodium azide in water–acetone solution at room temperature was completed in 2 min. Other sulfonyl and carbonyl chlorides also react fast with sodium azide. The slowest reaction was noted for hexanesulfonyl chlorides, but it was also completed within 10 min. Prolonged incubation of the reaction mixture up to 60 min did not affect the results (Figure 1). At the conditions given in the analytical procedure a linear relationship between the amount of sulfonyl chlorides and consumption of inorganic azide was observed. This is evidence for utility for this reaction in analysis. The reaction of *p*-toluenesulfonyl chloride with sodium azide was performed in 80% acetone in water solution, if the water content up to 50% did not affect the determination of *p*-toluenesulfonyl chloride. Higher concentration of water decreased consumption of sodium azide. When the new spectrophotometric method was used, several sulfonyl and carbonyl chlorides were determined in commercial and synthesized reagents, and results are presented in Table I. Some reactive anhydrides reacted also very fast with sodium azide and could be determined by the new method. A good agreement was noted between the results obtained by the new method and those obtained by morpholine and acid titrimetric methods (Table I). It suggests that only acetic anhydride is determined by the spectrophotometric method, contrary to the titrimetric method where the acetic anhydride and acetic acid present in the commercial reagent were measured as well. Standard errors for determination of these acid chlorides and anhydrides by the new method were calculated to be from ±0.5% to 1.6%. Some substituted phenylmethanesulfonyl chlorides (*p*-chloromethyl-, *o*-chloromethyl-) were also measurable by the spectrophotometric method.

Table I. Determination of Organic Acid Chlorides and Acid Anhydrides

reagent	mean content (%) determined: spectrophotometrically	mean content (%) determined: titrametrically[a]
carbonyl chlorides		
acetyl chloride	96.0	92.9
benzoyl chloride	98.3	99.1
phenylacetyl chloride	98.0	96.8
sulfonyl chlorides		
benzenesulfonyl chloride	99.1	99.4
5-dimethyl-1-naphthalenesulfonyl chloride	94.0	92.7
hexanesulfonyl chloride	99.5	97.5
methanesulfonyl chloride	95.5	95.8
phenylmethanesulfonyl chloride	97.6	96.7
p-toluenesulfonyl chloride	100.0	100.0
anhydrides		
acetic anhydride	85.5	95.4
butyric anhydride	97.4	96.6
phthaloyl-L-glutamic anhydride	92.8	94.9
propionic anhydride	99.1	97.8

[a] Methods are given in the Experimental Section.

Phenylmethanesulfonyl fluoride, phthalic anhydride, and succinic anhydride did not react with sodium azide, and, therefore, they could not be determined by the spectrophotometric method. No consumption of inorganic azide was observed for some carbonyl acids (acetic, benzoic, and phenylacetic), alkyl chlorides (benzyl chloride and phenylethyl bromide), esters (ethyl acetate, phenylacetyl methyl ester, and *p*-toluenesulfonyl ethyl ester), and chloroacetamide. Carbonyl acids did not affect the assay of corresponding carbonyl chlorides and anhydrides. The presence of strong acids like hydrochloric acid and *p*-toluenesulfonic acid as well as phosphorus trichloride diminished the quantity of determined acid chlorides, but addition of sodium acetate reduced this negative effect.

The new spectrophotometric method for determination of carbonyl and sulfonyl chlorides is precise, sensitive, fast, and very simple, in comparison with methods described in literature. Those methods utilized about 100 times greater samples, their accuracy sometimes is much lower, and they use potentiometric or titrimetric equipment. This suggests that the new method can be applied for routine measurements.

The observation that phenylmethanesulfonyl fluoride cannot be measured by the spectrophotometric method suggested application of this method for monitoring conversion of phenylmethanesulfonyl chloride to fluoride in conditions described by Gold and Fahrney (*12*). During the heating of the suspended sodium fluoride in the solution of phenylmethanesulfonyl chloride in dry acetone at 90–95 °C, a slow disappearance of the chloride was observed. After 6 h 50% of the chloride and after 13 h only 3% remained in solution. It suggested that a 6-h heating time recommended in the literature (*12*) is not sufficient for complete conversion of the chloride to fluoride. During similar treatment of *p*-toluenesulfonyl chloride with sodium fluoride in acetonitrile, disappearance of the chloride was very slow. After 20 h, concentrations of the chloride was decreased only by 4%. This result

was confirmed by analysis of the solid product isolated from the reaction mixture.

The new method was also applied for controlling the condensation of phthaloyl-L-glutamic anhydride with several amino compounds used in synthesis of γ-glutamyl amino acids (*16*) and γ-glutamyl arylamides (*17*). It was observed that heating of 0.2 M phthaloyl-L-glutamic anhydride with an equimolar amount of *p*-aminobenzoic acid in dry dioxane for 60 and 120 min at 40 °C caused the decrease of the anhydride concentration by 60% and 85%, respectively. The reaction with β-naphthylamine was much faster—after 10, 60, and 120 min 50%, 10%, and 2% of the remaining anhydride was observed, respectively. During these two condensation procedures the anhydride concentrations were almost equal to the remaining free arylamines. Also condensation of phthaloyl-L-glutamic anhydride with L-alanine was monitored by the new spectrophotometric method. When 0.2 M anhydride in glacial acetic acid was heated at 60 °C with the amino acid, after 60 min a 50% decrease of initial concentration of the anhydride was observed and after 120 min 35% of the initial anhydride was found in the reaction mixture.

Registry No. NaN_3, 26628-22-8; Fe, 7439-89-6; acetyl chloride, 75-36-5; benzoyl chloride, 98-88-4; phenylacetyl chloride, 103-80-0; benzenesulfonyl chloride, 98-09-9; 5-(dimethylamino)-1-naphthalenesulfonyl chloride, 605-65-2; hexanesulfonyl chloride, 14532-24-2; methanesulfonyl chloride, 124-63-0; phenylmethanesulfonyl chloride, 1939-99-7; *p*-toluenesulfonyl chloride, 98-59-9; acetic anhydride, 108-24-7; butyric anhydride, 106-31-0; phthaloyl-L-glutamic anhydride, 25830-77-7; propionic anhydride, 123-62-6; phenylmethanesulfonyl fluoride, 329-98-6; *p*-aminobenzoic acid, 150-13-0; β-naphthylamine, 91-59-8; L-alanine, 56-41-7.

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Microdroplet Titration Apparatus for Analyzing Small Sample Volumes

A. W. Steele and G. M. Hieftje*

Department of Chemistry, Indiana University, Bloomington, Indiana 47405

A computer-controlled, automatic titrator has been developed from a microdroplet dispensing system originally designed for sample application in electrothermal atomic absorption spectrometry. The system is capable of performing high-precision titrations using total volumes of sample solution and titrant in the microliter range. With the device, titrant droplets are formed by the repetitive insertion and withdrawal of a 150-μm glass needle into and from a small reservoir of titrant solution. Unlike other microtitration instruments, this apparatus permits operation with as little as 100 μL of bulk titrant solution. The control system employed for dispensing the droplets is inherently digital, thereby simplifying computer-supervised operation of the apparatus. Several chemical systems were used to evaluate the device in terms of precision and ease of operation. Potential application to other analytical problems will also be discussed.

Automated titration systems range from simple push-button titrant delivery units to microprocessor-controlled instruments that handle titrant delivery, titration monitoring, end-point detection, sample changing, and data calculations. However, such systems generally must use large sample volumes, on the order of milliliters to tens-of-milliliters. There is considerable interest today in titrating smaller sample volumes, on the order of 5–100 μL.

One approach to titrating small sample volumes is simply to dilute the sample with a sufficiently large volume of solvent to allow the use of standard macroscale titration equipment. This method has two drawbacks. First, the chemistry of the titration reaction is often changed at high dilutions, and the titration results might be difficult to analyze. Second, most end-point detection devices are concentration dependent, so precision of the end-point determination decreases as the titrate becomes more dilute.

As a result, the best approach to microtitration is usually to miniaturize the solution-dispensing apparatus and maintain the reagents at their original concentrations (*1*). In order to achieve this goal, the analyst needs a device capable of delivering microliter to submicroliter aliquots of titrant with high accuracy and precision. Many instruments have been developed to perform microliter range titrations (*2–7*), but most are not easily automated and seldom offer precision better than 2–5% RSD.

The device described in the present paper overcomes most of these limitations. It operates by producing precise droplets of titrant that are approximately 2 nL in volume. This type of operation is ideal for microtitration applications, since the titrant resolution is high and titrant delivery rate is adjustable over a broad range. In addition, operation of the device is inherently digital and is therefore easily automated. In the present paper, the operation of the device will be discussed and its performance on simple acid–base titration systems will

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