Chem. Anal. (Warsaw), **49**, 619 (2004)

Coulometric Titration of Thiols with Electrogenerated Chlorine

by Witold Ciesielski* and Monika Skowron

Department of Instrumental Analysis, University of Łódź, ul. Pomorska 163, 90-236 Łódź, Poland

Key words: coulometric titration, thiols, mesna, electrogenerated chlorine

Coulometric titration of thiols with chlorine in solution containing different electrolytes has been performed. Optimum determination conditions have been found. The developed method was applied to the determinaton of mesna (0.25–10 μmol), L-cysteine (0.25–2 μmol), mercaptosuccinic acid (0.5–2.5 μmol), 3-mercaptopropionic acid (0.42–8.3 μmol), cysteamine hydrochloride (0.5–3 μmol), carbimazole (0.25–2 μmol), thiopental (0.1–1 μmol), thioglycolic acid (0.87–10 μmol), D-penicillamine (0.125–5 μmol), L-glutathione (0.5–10 μmol) and mesna in pharmaceutical preparations (Mistabron and Anti–Uron). Determination error was below 1%.

Przeprowadzono miareczkowanie kulometryczne tioli z użyciem chloru jako titranta w środowisku różnych elektrolitów. Opracowano optymalne warunki oznaczenia. Za pomocą opracowanej metody oznaczono mesnę (0.25–10 μmol), L-cysteinę (0.25–2 μmol), kwas merkaptobursztynowy (0.5–2.5 μmol), kwas 3-mercaptopropionowy (0.42–8.3 μmol), chlorowodorek cysteaminy (0.5–3 μmol), karbimazol (0.25–2 μmol), tiopental (0.1–1 μmol), kwas tioglikolowy (0.87–10 μmol), D-penicyloaminę (0.125–5 μmol), L-glutation (0.5–10 μmol) oraz mesnę w preparatach farmaceutycznych Mistabron i Anti–Uron. Opracowana metoda umożliwia oznaczanie tioli z błędem poniżej 1%.

* Corresponding author. E-mail: zaiciesielski@hotmail.com; Fax: +48-42-678-39-85.

The coulometric titration method with anodically generated chlorine has been known for about 50 years [1]. It has been successively applied to the determination of many organic compounds, such as thiols (*e.g.* N-acetyl-L-cysteine [2], 6-mercaptopurine [3], 6-thioguanine [4], captopril [5]), disulfides (*e.g.* 2,2'-dithiodiglycolic acid, L-cystine, cystamine dihydrochloride [6a]) and thiophosphorous compounds (*e.g.* methamidophos, iso-malation, fenitrothion) [6b]. In this paper we present coulometric titration of cysteamine hydrochloride, thioglycolic acid, 3-mercaptopropionic acid, mercapto-succinic acid, L-cysteine, D-penicillamine, L-glutathione, mesna, carbimazole and thiopental performed with anodically generated chlorine as titrant and applying biamperometric end-point detection.

Thiols studied in this paper are commonly used in many different fields of technology, cosmetology and medicine.

Thioglycolic acid was titrated coulometrically with electrogenerated iodine using potentiometric [7,8] and biamperometric [9] end-point detection. Electrogenerated manganese(III) [10] and hypobromite ions [11] were also used as titrants. L-cysteine was titrated with anodically generated bromine and iodine, applying biampero-metric [12] and potentiometric [12,13] end-point detection. Anodically generated silver(I) ions and biamperometric end-point detection were also applied for the determination of L-cysteine [14]. D-penicillamine was titrated with mercury(II) ions generated from an amalgamated gold anode [15]. Mercaptosuccinic acid was titrated with electrogenerated manganese(III) ions, as well as with cerium(IV) ions generated from cerium(III) nitrate [16]. End-point was detected biamperometrically [10], potentiometrically [17] and bipotentiometrically [18]. L-glutathione was determined using generated silver(I) ions and biamperometric end-point detection [19]. Carbimazole was titrated coulometrically with electrogenerated iodine in alkaline medium using biamperometric end-point detection [20]. Carbimazole was also determined coulometrically involving iodine-azide reaction [21]. Thiopental was titrated coulometrically using electrogenerated bromine [22] or mercury(II) [23] as titrant. Determination was carried out in two steps. In the first one the sample was reduced at the constant potential. After that the analyte was titrated with generated mercury(II).

According to our current knowledge, there are no literature reports on the coulometric determination of 3-mercaptopropionic acid, cysteamine hydrochloride and mesna.

EXPERIMENTAL

Apparatus

Measurements were performed using a universal coulometric analyser, model OH–404, Radelkis, Hungary. An electrolysis cell with two platinum electrodes each with the area of 5 cm^2, working in the generating

circuit, and the double electroede OH–9381 working in biamperometric indicator circuit were used. Cathodic and anodic compartments of the electrolytic cell were separated with a sintered glass G–4 disc.

Reagents and solutions

Doubly distilled water was used throughout. Sulfuric acid, sodium chloride and acetonitrile were purchased from POCH, Gliwice.

The following compounds were used: cysteamine hydrochloride (98%, Aldrich) thioglycolic acid (98%, Sigma), 3-mercaptopropionic acid (99%, Aldrich), L-cysteine (98%, Roanal), D-penicillamine (99%, Sigma), mercaptosuccinic acid (97%, Aldrich), L-glutathione (98%, Sigma), mesna (98%, Aldrich), carbimazole (97%, Lancaster), thiopental (United Pharmaceutical Works, Prague), Mistabron 600 mg (PLIVA, Kraków), Anti–Uron 200 mg (Polfa, Kraków).

Analyte solutions of the concentration of 5×10^{-3} mol L^{-1} were prepared freshly before use by dissolving weighed amounts of the reagents in the solvent.

Aqueous solution containing hydrochloric acid or sulfuric acid, and sodium chloride at different concentrations (Tab. 1) served as the supporting electrolyte solution.

Table 1. Determination results of thiols in the solutions of various supporting electrolytes

Compound	Supporting electrolyte mol L^{-1}			z*	RSD %
	HCl	H_2SO_4	NaCl		
Mesna	1			5.76	0.24
		0.25	0.2	5.63	0.28
		0.5	0.1	5.53	0.23
		0.5	0.2	5.97	0.22
		0.5	0.4	5.55	0.20
Carbimazole	1			5.98	0.13
		0.25	0.2	6.10	0.16
		0.5	0.1	6.34	0.12
		0.5	0.2	6.40	0.10
		0.5	0.4	6.50	0.20
Thiopental	1			5.00	0.12
		0.25	0.2	5.05	0.20
		0.5	0.1	4.94	0.18
		0.5	0.2	4.95	0.11
		0.5	0.4	4.94	0.16

(Continuation on the next page)

Table 1. (Continuation)

	1			5.87	0.12
		0.25	0.2	6.07	0.10
3-Mercaptopropionic acid		0.5	0.1	6.00	0.09
		0.5	0.2	5.99	0.09
		0.5	0.4	5.98	0.11

* Number of electrons transferred.

Procedure

Sample solution containing the analytes was introduced into the supporting electrolyte solution (25 mL) and placed in the anodic compartment of the electrolytic cell. 150-mV potential was applied to the indicator system. After switching the mechanical stirrer on, a constant current was passed through the solution. The current in the generating circuit was adjusted between 1 and 10 mA, according to the amount of the analyte. Titration time was maintained at the level of several minutes. When the current in the indicator circuit achieved 0.04 μA, titration was completed and the charge Q was measured. The excess of chlorine that appeared in the solution after the reaction was finished allowed one to detect the end-point. In the same way the sample was titrated in the absence of the analyte (blank experiment), and the charge of 7–10 mC (Q_o) was measured.

The content [μmol] of the analyte in the sample was estimated according to the Faraday's law:

$$n = \frac{\Delta Q}{zF} \times 10^3$$

where:

$\Delta Q = Q - Q_o$ [mC]; Q_o – charge corresponding to the blank titration; z – number of electrons transferred (5 for thiopental, 6 for other compounds), F – Faraday constant (96 485 C mol^{-1}).

Determination of mesna in drugs

The contents of ten drug phials were quantitatively introduced into 100 mL which was fixed up with water. The mixture was diluted 10-fold and 100 μL of the resulting solution was introduced into 25 mL of the supporting electrolyte solution, in the anodic part of electrolytic cell. The sample was titrated similarly as pure analyte. The content of mesna in each dosage form was calculated using Faraday's laws.

RESULTS AND DISCUSSIONS

Efficiency of chlorine generation and reaction rate between chlorine and the analytes depends on the concentration of hydrogen and chloride ions in the solution. Both should be optimised experimentally [24] in order to provide accurate and reprodu-

cible results, constant number of transferred electrons, and low RSD. Thus, various concentrations of hydrochloride or sulfuric acid and sodium chloride were applied. The number of electrons transferred in the presence of various supporting electrolytes was calculated. Some of the results are given in Table 1.

The biamperometric end-point detection provided correct and reproducible results of the determination of thiols in the concentration ranges given in Table 2. However, the error increased above 1% when larger or smaller amounts of the analytes than those presented in Table 2 were determined. The literature data concerning the determination error and RSD of the coulometric titration with chlorine and visual end-point detection were less satisfactory – these factors amounted to several percent [2,4].

Table 2. Determination results of thiols; n = 6

Compound (supporting electrolyte)	Taken μmol	Found μmol	RSD %
Mesna (0.5 mol L^{-1} H_2SO_4 0.2 mol L^{-1} NaCl)	0.2500	0.2515	0.31
	2.500	2.483	0.05
	10.000	9.960	0.04
L-cysteine (0.5 mol L^{-1} H_2SO_4 0.2 mol L^{-1} NaCl)	0.1250	0.1243	0.18
	0.5000	0.5014	0.08
	2.500	2.497	0.01
Mercaptosuccinic acid (0.5 mol L^{-1} H_2SO_4 0.2 mol L^{-1} NaCl)	0.5000	0.5022	0.07
	1.000	1.005	0.04
	2.500	2.499	0.02
3-Mercaptopropionic acid (0.5 mol L^{-1} H_2SO_4 0.1 mol L^{-1} NaCl)	0.4160	0.4188	0.05
	1.108	1.108	0.03
	8.310	8.273	0.01
Cysteamine hydrochloride (0.5 mol L^{-1} H_2SO_4 0.1 mol L^{-1} NaCl)	0.5000	0.4988	0.15
	1.000	0.9988	0.06
	3.000	2.984	0.01
Carbimazole (1 mol L^{-1} HCl)	0.2500	0.2488	0.23
	1.000	1.001	0.04
	2.002	2.012	0.03

(Continuation on the next page)

Table 2. (Continuation)

Thiopental (1 mol L^{-1} HCl)	0.1001	0.0996	0.64
	0.5007	0.5001	0.12
	1.0014	1.0094	0.07
Thioglycolic acid (1 mol L^{-1} HCl)	0.8670	0.8622	0.04
	2.890	2.891	0.02
	10.11	10.13	0.01
D-penicillamine (1 mol L^{-1} HCl)	0.1250	0.1252	0.28
	1.000	1.008	0.07
	5.000	5.036	0.01
L-glutathione (0.5 mol L^{-1} HCl)	0.5000	0.5025	0.25
	2.500	2.514	0.09
	5.000	5.002	0.05

The reaction mechanism between most of the analytes and chlorine is given below:

$$RSH + 3Cl_2 + 3H_2O \longrightarrow RSO_3H + 6HCl \quad (1)$$

where:

R = $-CH_2CH_2NH_2 \cdot HCl$ for cysteamine hydrochloride

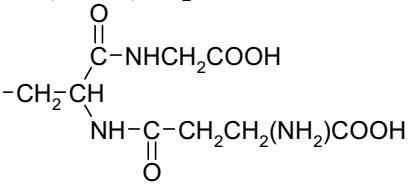
$-CH_2COOH$ for thioglycolic acid

$-CH_2CH_2COOH$ for 3-mercaptopropionic acid

$-CH_2CH(NH_2)COOH$ for L-cysteine

$-C(CH_3)_2CH(NH_2)COOH$ for D-penicillamine

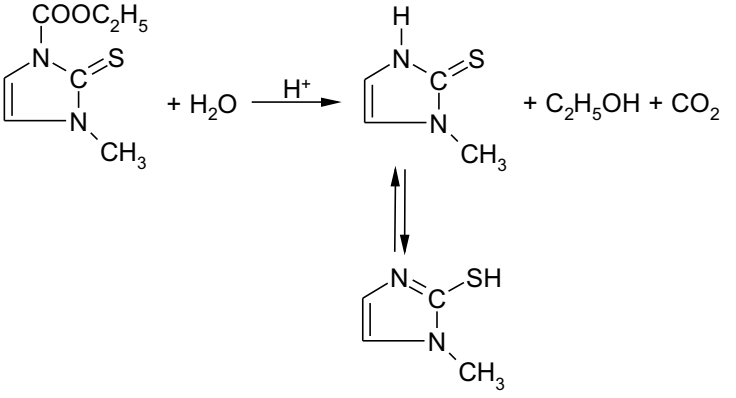
$-CH(COOH)\ CH_2COOH$ for mercaptosuccinic acid



for L-glutathione

$-C_2H_4SO_3^-Na^+$ for mesna

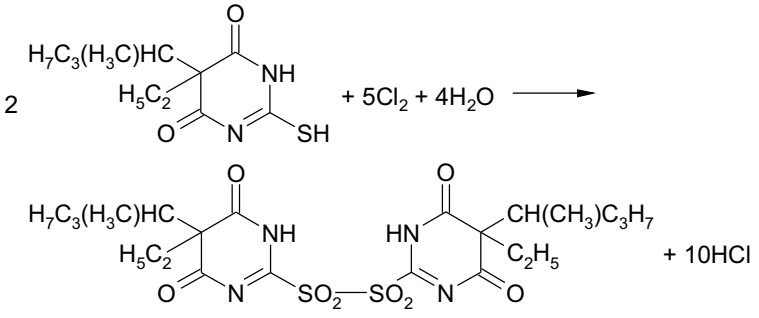
Carbimazole exists only in the thione form, but it was proved that in an acidic medium it undergoes hydrolysis and decarboxylation [25], according to the equation:



(2)

1-methylimidazole-2-thiol is formed as the product, which reacts directly with chlorine (equation 1).

Thiopental reacts with chlorine according to the following reaction scheme:



(3)

The presented method of coulometric determination of thiols is characterised by short analysis time, high precision and accuracy, and simplicity (does not involve complex chemical reactions). The method requires the use of only commonly available reagents, and can be partly automated. As an absolute method it requires no standard solutions. It was applied to the determination of mesna in Mistabron and Anti-Uron drugs. The results of these determinations are given in Table 3.

Table 3. Determination results of mesna in drugs; n = 6

Drug	Declared Content mg	Found mg	RSD %
Mistabron	600	602	0.07
Anti-Uron	200	201	0.04

Coulometric titration with chlorine can be applied to the determination of thiols, which react rapidly with chlorine. However, for those reacting slowly (*e.g.* 2-thiobarbituric acid, 3-mercapto-1,2,4-triazole, 2-mercapto-4(3*H*)quinazolinone, 2-mercaptobenzothiazole, 2-mercaptobenzimidazole, 2,5-dimercapto-1,3,4-thiadiazole) no conditions were found to determine them using coulometric titration.

REFERENCES

1. Farrington P.S. and Swift E.H., *Anal. Chem.*, **22**, 889 (1950).
2. Nikolič K.I., Vesalevič K.R. and Javanovič T., *Pharmazie*, **42**, 134 (1987).
3. Nikolič K.I. and Vesalevič K.R., *J. Pharm. Belg.*, **46**, 455 (1988).
4. Nikolič K.I., Vesalevič K.R. and Zimowa N., *Il. Farmaco*, **46**, 409 (1991).
5. Nikolič K.I., Vesalevič K.R., *Acta. Polon. Pharm.*, **48**, 5 (1991).

6a. Ciesielski W. and Skowron M., *Chem. Anal. (Warsaw)*, in press.

6b. Ciesielski W., Skowron M., Bałczewski P. and Szadowiak A., *Talanta*, **60**, 725 (2003).

7. Pastor T.J., Vajgand V.J. and Antonijevic V.V., *Mikrochim. Acta*, **3**, 203 (1983).
8. Pastor T.J. and Antonijevic V.V., *Anal. Chim. Acta*, **196**, 229 (1987).
9. Ruettinger H.H. and Spohn U., *Anal. Chim. Acta*, **202**, 75 (1987).
10. Pastor T.J., Vajgand V.J. and Antonijevic V.V., *Glas. Hem. Drus. Beograd*, **44**, 651 (1979); *Chem. Abstr.*, **92**:226164t (1980).
11. Palma R.J., Gupta H.K.L. and Boltz D.F., *Anal. Lett.*, **4**, 277 (1971).
12. Kreshkov A.P. and Oganeshyan L.B., *Zh. Anal. Khim.*, **26**, 614 (1971).
13. Kreshkov A.P. and Oganeshyan L.B., *Zh. Anal. Khim.*, **28**, 2260 (1973).
14. Ladenson J.H. and Purdy W.C., *Anal. Chim. Acta*, **64**, 259 (1973).
15. Forsman U., *Anal. Chim. Acta*, **93**, 153 (1977).
16. Pastor T.J., Vajgand V., Civic Ivan, *Magy. Kem. Foly*, **88**, 519 (1982); *Chem. Abstr.*, **98**:46241f (1983).
17. Pastor T.J. and Civic I., *Mikrochim. Acta*, **1**, 435 (1984).
18. Pastor T.J. and Antonijevic V.V., *Mikrochim. Acta*, **1,** 313 (1990).
19. Ladenson J.H. and Purdy W.C., *Anal. Chim. Acta*, **64**, 259 (1973).
20. Ciesielski W. and Krenc A.; *Anal. Lett.*, **33**, 1545 (2000)
21. Ciesielski W., Zakrzewski R. and Skowron M., *Chem. Anal.*, **46**, 873 (2001).
22. Kalinowski K. and Piotrowska A., *Acta Polon. Pharm.*, **26**, 107 (1959).
23. Mairesse-Ducarmois C.A., *J. Pharm. Belg.*, **28**, 3000 (1973).
24. Lingane J.J., *Electroanalytical Chemistry,* Interscience Publ., New York 1958, (in Polish).
25. El-Bardicy M.G., El-Saharty Y.S. and Tawakkal M.S., *Talanta*, **40**, 577 (1993).

Received October 2003
Accepted July 2004