

Full Paper

Design of a Gramine-Selective Membrane Sensor

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Abstract- The interaction of organic cation gramine with methyl orange was investigated with the help of thermal studies. These slightly soluble associates were used as electrode active substances (EAS) in plasticized polyvinyl chloride (PVC) membranes of ion-selective electrodes (ISE), sensitive to organic cation gramine. The best response characteristics were obtained using the composition: ion pair 7%, 59% TCP 34% PVC. The electrochemical sensor shows a linear dynamic range 2.0×10^{-5} – 1.0×10^{-1} mol/L and a Nernstian slope of 45.0 mV/decade with a detection limit of 1.3×10^{-5} mol/L. It has a fast response time < 15 s and can be used for at least 7 weeks without any considerable divergences in its potential response. Based on the experimental data, a new method of gramine determination in model solution by direct potentiometry with the help of membrane sensor was developed.

Keywords- Gramine, Membrane sensor, Potentiometry, Ion pair, Methyl orange

1. INTRODUCTION

A rapid increase in the development and production of sensors during the last three decades signifies a fundamental change in chemical analysis tools. The most numerous group of these chemical sensors is composed of potentiometric sensors that include ion-selective electrodes (ISEs) [1-3]. Potentiometric detection based on ion-selective membrane electrodes as a simple method, offers several advantages, such as fast response, easy preparation, simple

instrumentation, wide linear dynamic range, relatively low detection limit, reasonable selectivity, application in colored and turbid solutions and low costs [4-9]. Active development of ionometry began with the studies of new chemical sensors and their promising use in pharmaceutical analysis [10-21]. Currently a wide assortment of commercially available ISEs is being enriched with new developments and improvements in the known types of sensors. The task of analytical chemistry in this challenge is the development and the implementation of efficient methods of the quality control of the substances.

Gramine is a naturally occurring indole alkaloid. Despite being found in several plant species, gramine is far easier to synthesize directly from indole via a Mannich reaction with dimethylamine and formaldehyde [22].

In recent years, effective methods to prevent algal blooms were developed and explored. That is why natural plant toxins such as gramine are so popular which seriously influence algal photosynthetic activity by destroying photosynthetic pigments [23].

There is a large number of approaches to qualitative and quantitative determination of gramine. They include colorimetric method [24], gas-liquid chromatography [25], fluorimetry [26] and others methods [27-29]. There are no reports on determination of gramine using ion selective electrode based on ion pair.

Therefore, the development of new potentiometric sensors using ion pair is of current interest. The simultaneous study of chemical-analytical characteristics of isolated solid ion pair, the effect of the electrode active substances (EAS) used, pH of the analyzed solution, properties of the membrane plasticizer, etc. are also of importance. The combination of those factors which define the electrochemical properties of ISEs, and the determination and consideration of the relations between certain parameters would permit, in our opinion, the improvement of the electrochemical characteristics of electrodes and would predict the properties of other ISEs in their development for new classes of substances.

2. EXPERIMENTAL

All EMF measurements were carried out with the following cell assembly. An AI-123 (Ukraine) model pH/mV meter with a Ag-AgCl saturated reference electrode was used for the measurements of potential difference at 25.0 ± 1.0 °C. All chemicals were of analytical-reagent grade. Distilled water was used to prepare all solution and in all experiments.

The modeling of the membrane composition of the ion-selective sensors utilized high molecular weight polyvinylchloride (PVC) (Sigma-Aldrich), dibutyl phthalate (DBP) (South African Republic), dibutyl sebacate (DBS) (Merck), dioctyl phthalate (DOP) (Merck), dinonyl phthalate (DNP) (Merck), tricresyl phosphate (TCP) (Acros organics, USA), diethyl phthalate (DEP) (Merck), cyclohexanone (CHN) (Shostka, Ukraine) and tetrahydrofuran (THF)

(Labsan Ltd, Ireland).

The ionic strength of the solutions was adjusted with 0.2 mol/l LiCl solution. The pH value of solutions was maintained with the use of a buffer mixture (0.04 mol/l CH₃COOH, H₃BO₃, H₃PO₄, and a 0.2 mol/l NaOH solution) and monitored by potentiometry with a glass electrode. Stock solution (0.01 mol/l) of gramine was prepared as follows: accurately weighed portion of the substance was added to 5 ml of buffer solution with pH 3.7 and diluted to 50 ml with 0.2 mol/l solution of LiCl. It was further diluted to produce working standard solutions with the concentration range 1×10^{-1} – 1×10^{-7} mol/l.

An ion pair of gramine methyl orange was prepared by mixing equimolar quantities of 1×10^{-2} mol/l gramine and anionic dye methyl orange (MO) (Fig. 1). The solution was settled for 2 h and the IP sediment was filtered (quantitative rapid filter paper).

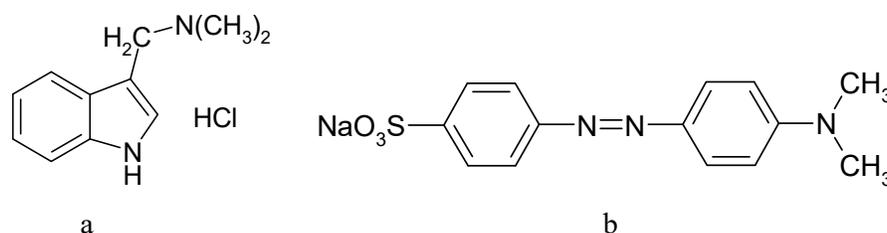


Fig. 1. Chemical structure of gramine (a) and methyl orange (b)

This residue was treated with 50 ml of cold distilled water. The precipitate was dried and used as an electrode-active substance for preparing the gramine-sensitive sensor.

The generally accepted technique of preparing a plasticized membrane consists of mixing of the electrode-active substance with PVC dissolved in cyclohexanone or tetrahydrofuran followed by the evaporation of the solvent in a glass ring. PVC requires plasticization and places a constraint on the choice of mediator. Plasticized PVC membranes were prepared according to the recommendations [30]. The sensing membrane was prepared by mixing 70 mg of PVC powder and 15 mg of IP with 0.12 ml of a plasticizer. The mixture was stirred until the PVC was well moistened, and then the mixture was dispersed in 0.5 ml THF or CHN. The resulting mixture was transferred into a glass dish of 25 mm diameter. The solvent was evaporated slowly at room temperature until a solid membrane of about 0.3 mm thickness was formed. A desired piece of the membrane was cut and then was attached to an end of polyethylene tube using viscous solution of PVC as an adhesive. The resulting sensor was then filled with an internal solution of 1.0×10^{-2} mol/l gramine and conditioned for 15 min. Then a copper wire was immersed into the tube.

The thermal studies (TG, DTG and DTA) were carried out on an apparatus for complex dynamic thermal analysis under the following conditions: temperature range 20–700 °C, heating rate 5 K/min, sample weight 27.3 mg, static nitrogen medium, channel sensitivities:

DTA–150 mV, TG–50 mV.

3. RESULTS AND DISCUSSION

Thermal behavior of obtained IP was investigated by differential thermal analysis that would show the thermal stability and the character of the decomposition of the complex. The thermolysis of MO-Gramine⁺ IP undergoes three stages that fit a theoretical interpretation. The thermogram is shown in Fig. 2 (after drying at 50 °C), Fig. 3 (before drying) and its computation are presented in Table 1.

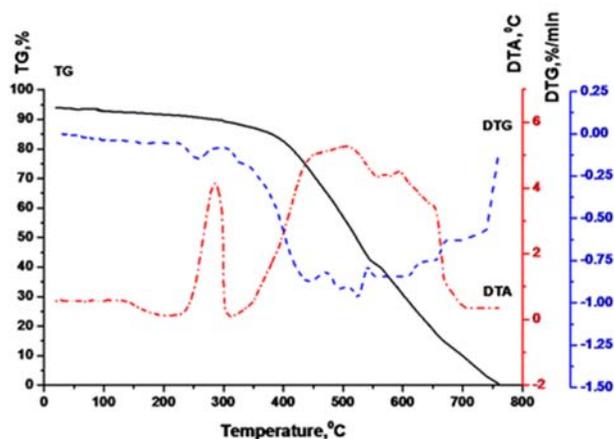


Fig. 2. Simultaneous TG, DTA curves of the MO-Gramine⁺ (after drying at 50 °C)

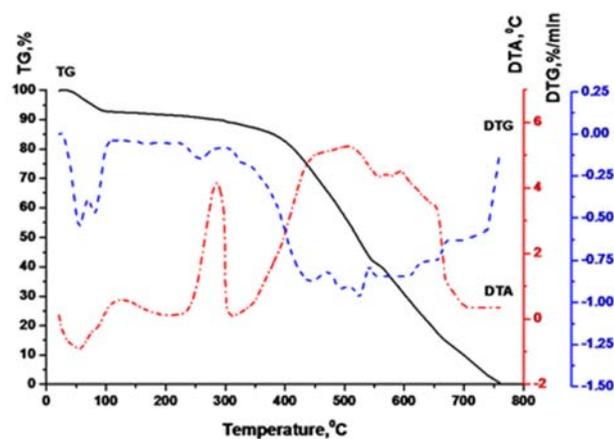


Fig. 3. Simultaneous TG, DTA curves of the MO-Gramine⁺ (before drying)

Thermal decomposition starts above 250 °C and is accompanied by the loss of mass and the discharge of the decomposition products. This is followed by carbonization and combustion of the IP, which is shown at the thermogram by big exo effects at 292 °C and a

significant loss of mass. A large amount of amorphous carbon remained as black plaque at the wall of the crucible in which the thermolysis reaction took place.

Table 1. Data of DTA curve of the ion pair of MO^- -Gramine⁺ and decomposition products

Ion pair	Temperature range, °C	Loss of mass, %		Products
		Theoretical	Experimental	
$\text{C}_{25}\text{H}_{28}\text{N}_5\text{O}_3\text{S}$ (after drying at 50 °C)	20 – 420 (+292)	20.51	20.50	SO_2
	420 – 460	10.88	11.25	$\text{CO}+2\text{N}_2$
	460 – 490	8.74	9.00	$\text{NH}(\text{CH}_3)_2$
	490 – 760 (-560)	59.87	59.25	(22C+15H)
Total loss of mass		100.00	100.00	

In this paper new plasticized PVC membrane sensors on the base of IP gramine – methyl orange are investigated. The developed sensor exhibit a good selectivity with respect to related substances, additives in dosage forms and heavy metals. The conventional design was prepared, characterized and compared according to IUPAC recommendations.

Table 2. Effect of EAS content and plasticizer nature on main electroanalytical characteristics of the developed sensors

Amount of the ion pair (%)	Plasticizer (59 %)	Slope (mV)	Linear range (mol/L)	Detection limit (mol/L)
3	TCP	32 ± 1	$1 \times 10^{-1} - 3.2 \times 10^{-4}$	1.0×10^{-4}
5	TCP	33 ± 1	$1 \times 10^{-2} - 7.9 \times 10^{-5}$	6.3×10^{-5}
7	DNP	24 ± 1	$1 \times 10^{-1} - 1.0 \times 10^{-3}$	7.9×10^{-4}
7	DOP	30 ± 1	$1 \times 10^{-1} - 5.0 \times 10^{-3}$	2.0×10^{-4}
7	DBP	46 ± 1	$1 \times 10^{-1} - 7.9 \times 10^{-4}$	5.0×10^{-4}
7	DBS	40 ± 1	$1 \times 10^{-1} - 7.9 \times 10^{-4}$	6.3×10^{-4}
7	DEP	43 ± 1	$1 \times 10^{-1} - 5.0 \times 10^{-4}$	2.0×10^{-5}
7	TCP	45 ± 1	$1 \times 10^{-1} - 2.0 \times 10^{-5}$	1.3×10^{-5}
10	TCP	45 ± 1	$1 \times 10^{-1} - 4.0 \times 10^{-4}$	1.6×10^{-4}
13	TCP	44 ± 1	$1 \times 10^{-1} - 6.3 \times 10^{-3}$	3.2×10^{-4}

Generally, the sensitivity, selectivity, working range, and stability of an ion-selective electrode depend not only on nature of the IP, but are also strongly influenced by the nature and amount of the plasticizer and additives. To assess the effect of the plasticizer nature, uniform membranes were prepared with DBP, DOP, DBS, DNP, DEP and TCP. It was established that the best plasticizers for the membranes based on IP Gramine⁺ MO^- are TCP

and DEP with electrode function slope 45 and 43 mV/pC and the detection limit 1.3×10^{-5} mol/l and 2.0×10^{-5} mol/l respectively (Table 2).

The content of IP on the response of the proposed electrodes was investigated. Among the 3%, 5%, 7%, 10%, 13%, of different content of IP, which were used some significant difference on the potential response was founded. The best result was shown by electrode with tricresyl phosphate as a plasticizer and 7% of IP content used (Fig. 4).

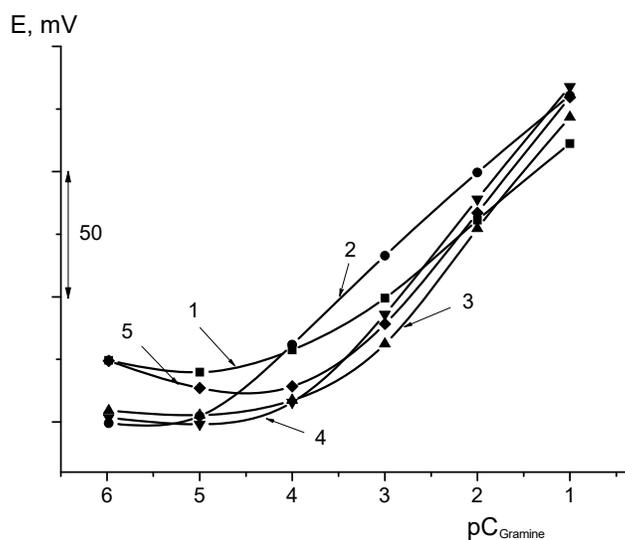


Fig. 4. Graphical dependence of the ion-selective electrode potential on the Gramine concentration $E=f(pC)$. EAS content: 1–3%; 2–7%; 3–5%; 4–10%; 5–13%

The response time was measured that is necessary for the potential of the membrane electrode to reach the value within ± 1 mV of the final equilibrium value after immersing the electrode in the solution of gramine with 10-fold difference in concentration (Fig. 5). The sensors showed rapid response within 15 seconds for drug solutions $\geq 1 \times 10^{-3}$ mol/l and 25 seconds for 1×10^{-5} mol/l.

The lifetime of the sensors were examined by re-calibration every 2 days. There was no noticeable deterioration in the sensor performance in terms of detection limit, calibration curve slope and response time over a period of 30 days.

The sensor lifetime is mainly determined by the frequency of its use and averages 10 months from the manufacture date.

According to the experimental data, the best concentration of LiCl in solution is 0.5 mol/l. Generally, the nature and concentration of the electrolyte are very important in the measurement of the electrode potential. The cation must not complete with the potential-defining cation for the place in the membrane [31].

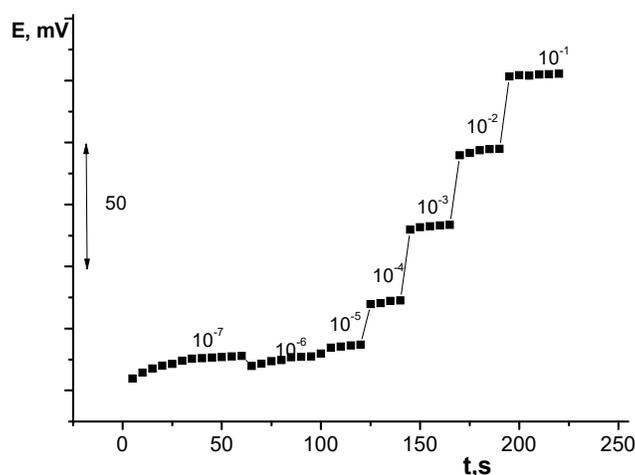


Fig. 5. Sensor response time for various gramine concentrations

The pH effect of the tested solution on the electrochemical behavior of the sensor was studied at a constant concentration of gramine and varying the content of the hydrogen ions in the pH range of 2.0–11.0 which was adjusted with HCl or NaOH solution. The results are illustrated in Fig. 6. The potential remains constant in the range of 2.0–9.0.

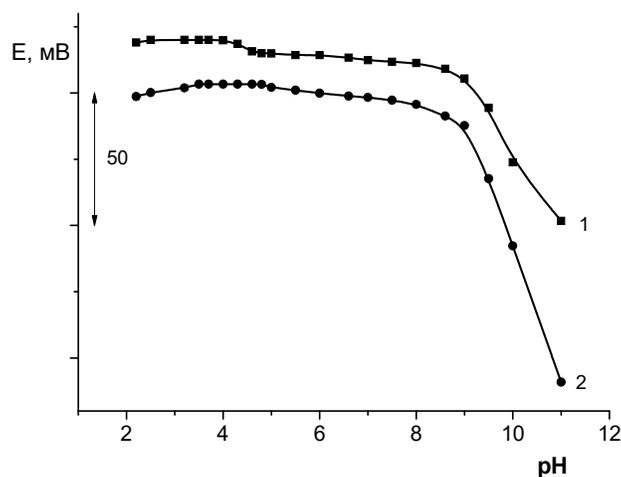


Fig. 6. the effect of the solution ph on the electrode potential at gramine pc=2 (1), pc=2.5 (2)

The potentiometric selectivity coefficients of the gramine sensors were measured. The influence of some inorganic cations was investigated by separate solution method in which the Nicolsky-Eisenman equation was used.

The potentiometric coefficients were determined using the separate solution method.

$$K_{gramine,J^{z+}}^{pot} = \frac{E_2 - E_1}{S} + \log[gramine] - \log[J^{z+}]^{1/z},$$

Where E_1 is the potential for the drug, E_2 for the interfering ion J, with charge Z and slope S of the calibration graph.

The potentiometric selectivity coefficient of the gramine-based sensors depends on the selectivity of the ion-exchange process at the membrane-sample interference, the mobility of the respective ions in the membrane. The free energy transfer of the gramine ion between the aqueous and the organic phase could also control the selectivity of the proposed sensor. No interference from ions such as NH_4^+ (>4), K^+ (>4), Na^+ (>4), Co^{2+} (4), Ba^{2+} (3,7), Ca^{2+} (4), Mg^{2+} (3.8), cetylpyridinium chloride (3.2), tetramethylammonium chloride (>4), cetyltrimethylammonium bromide (4), tetrabutylammonium iodide (2.0), benzyltriethylammonium chloride (>4) etc. was recorded. The proposed sensors exhibited high selectivity towards gramine with respect to the test ion.

Table 3. Results of the determination of gramine amounts in model solution ($F_{\text{tabl.}}=5.05$; $t_{\text{tabl.}}=2.78$)

Label amount	Found by proposed sensor			Found by potentiometric titration			<i>F</i> -test	<i>t</i> -test
	mg	<i>S</i> ²	RSD (%)	mg	<i>S</i> ²	RSD (%)		
50	49.8±0.76	1.11	2.46	50.0±0.83	1.54	1.50	0.96	1.99
100	101.1±0.78	1.32	1.47	100.2±0.97	1.39	1.24	1.89	2.20
200	200.6±0.58	1.24	1.32	200.2±1.36	1.87	1.62	1.71	2.04

Table 4. Validation of the proposed method for the determination of Gramine in pure form

Conc. (mol/l)	Recovery %	RSD % *	Error %**
Intraday precision			
1×10^{-6}	99.82	0.72	0.35
1×10^{-5}	99.45	0.21	0.25
1×10^{-4}	99.12	0.20	0.22
Interday precision			
1×10^{-6}	98.75	0.58	0.45
1×10^{-5}	99.32	0.54	0.31
1×10^{-4}	99.65	0.38	0.27

*%RSD=(S.D/Mean)100

**%Error= %RSD/ \sqrt{n}

Five portions of gramine were accurately weighted, transferred to 50-ml volumetric flask, dissolved and shaken for 15 minutes with 5 ml of 0.04 mol/l acetate buffer of pH 3.7 and diluted to the mark with 0.5 mol/l solution LiCl.

The e.m.f. of the solution was measured above and the corresponding concentration was

determined using the calibration plot. Gramine in model solution was determined by direct potentiometric measurement using these sensors. The potentials measured by those sensors were recorded and compared with the calibration graph. The proposed membrane sensor based on ion pair of gramine - methyl orange was found to work well under laboratory conditions (Table 3).

The recovery results are shown in Table 4. Three replicate determinations at different concentration levels were carried out using the three electrodes to test the precision of the method.

The standard deviations were found to be 1.0–1.5, indicating reasonable repeatability and reproducibility of the selected method. The precision of the method was calculated in terms of (intra-day and inter-day). The %RSD values of intra-day and inter-day studies for the repeated determination were less than 2% indicating good precision.

4. CONCLUSION

The results obtained in present work demonstrate that membrane potentiometric sensors based on the ion-pair of gramine and methyl orange with an internal comparison solution were developed. The proposed sensors exhibit long lifetime, good stability, sensitivity, precision, accuracy and selectivity. They are low cost and easy to prepare and use. The sensors can be used for routine analysis of gramine. All sensors have potentiometric pH-independent response in the region (2.0–9.0).

REFERENCES

- [1] M. R. Ganjali, V. K. Gupta, F. Faridbod, and P. Norouzi, *Lanthanides Series Determination by Various Analytical Methods*, 1st Edition, Elsevier (2016).
- [2] K. N. Mikhelson, *Ion-Selective Electrodes*, Springer-Verlag Berlin Heidelberg (2013).
- [3] E. Pretsch, *Trends Anal. Chem.* 26 (2007) 46.
- [4] J. Lenik, and C. Wardak, *Cent. Eur. J. Chem.* 8 (2010) 382.
- [5] M. Ardeshiri, and F. Jalali, *Mater. Sci. Eng. C* 63 (2016) 30.
- [6] J. Lenik, C. Wardak, and B. Marczewska, *Cent. Eur. J. Chem.* 6 (2008) 513.
- [7] C. Wardak, B. Marczewska, and J. Lenik, *Electrochim. Acta* 51 (2006) 2267.
- [8] J. Lenik, and R. Lyszczek, *Mater. Sci. Eng. C* 61 (2016) 149.
- [9] N. T. Abdel Ghani, R. Nashar, El Mohamed, FM Abdel-Haleem, and A. Madbouly, *Electroanalysis* 28 (2016) 1530.
- [10] V. V. Cosofret, and R. B. Buck, *Pharmaceutical Application of Membrane Sensors*, CRC Press, Boca Raton, FL, (1992).
- [11] Z. Kormosh, and O. Matviychuk, *Chin. Chem. Lett.* 24 (2013) 315.
- [12] I. Antal, Zh. Kormosh, Ya. Bazel, S. Lysenko, and N. Kormosh, *Electroanalysis* 22 (2010)

- 2714.
- [13] Zh. Kormosh, I. Hunka, and Ya. Bazel, *Chin. Chem. Lett.* 18 (2007) 103.
 - [14] Zh. Kormosh, and T. Savchuk, *Mater. Sci. Eng. C* 32 (2012) 2286.
 - [15] Zh. Kormosh, T. Savchuk, S. Korolchuk, and Ya Bazel, *Electroanalysis* 23 (2011) 2144.
 - [16] Zh. Kormosh, I. Hunka, Ya. Bazel, and O. Matviychuk, *Mater. Sci. Eng. C* 30 (2010) 997.
 - [17] Zh. Kormosh, I. Hunka, Ya. Bazel, N. Kormosh, A. Laganovsky, and I. Mazurenko, *Cent. Eur. J. Chem.* 5 (2007) 813.
 - [18] Z. Kormosh, I. Hunka, and Y. Bazel, *J. Iran. Chem. Res.* 1 (2008) 25.
 - [19] Zh. Kormosh, I. Hunka, and Ya. Bazel, *Mater. Sci. Eng. C* 29 (2009) 1018.
 - [20] Zh. A. Kormosh, I. P. Hunka, Ya. R. Bazel, *J. Anal. Chem.* 64 (2009) 853.
 - [21] Zh. Kormosh, I. Hunka, and Ya. Bazel, *Acta Chim. Slov.* 55 (2008) 261.
 - [22] L. J. Corcuera, *Phytochemistry* 33 (1993) 741.
 - [23] Y. Honga, H. Y. Hua, and X. Xiea, *Aquatic Toxicol.* 91 (2009) 262.
 - [24] A. B. Simons, and G. C. Marten, *J. Agron.* 63 (1971) 915.
 - [25] D. L. Woods, and K. W. Clark, *Canadian J. Plant Sci.* 51 (1971) 323.
 - [26] W. Majak, R. E. McDiarmid, and R. J. Bose, *Phytochemistry* 17 (1978) 301.
 - [27] H. Yoshida, H. Tsumuki, K. Kawada, and K. Kanehisa, *Bull. Res. Inst. Bioresour. Okayama Univ.* 4 (1996) 73.
 - [28] J. De Ribamar, and F. Júnior, *Talanta* 42 (1995) 1505.
 - [29] A. Q. N. Manzar, *Pakistan J. Pharm. Sci.* 1 (1988) 25.
 - [30] K. Kamman, *Work with Ion-Selective Electrodes*, Mir, Moscow (1980).
 - [31] R. P. Buck, and E. Linder, *Pure Appl. Chem.* 66 (1994) 2527.