

Note

Ethanol Sensor Using Two Chloride Ion-Selective Polymeric Membrane Electrodes

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An ethanol sensor using two chloride ion-selective polymeric membrane electrodes (Cl-ISEs) was developed. Two different polymeric membranes were used, one with *o*-nitro-phenyloctyl ether (NPOE) and the other with *n*-decylalcohol (DA) as the plasticizer. The electrical potential of the Cl-ISE fell linearly with increasing ethanol concentration at a given concentration of Cl⁻. The electrical potential equation is viewed as a function of the concentration of ethanol and Cl⁻. The responses to ethanol and Cl⁻ differed between plasticizers. The two Cl-ISEs showed nearly Nernstean slopes for Cl⁻ in ethanol solution (0–20%). In addition, each electrode had good linearity for ethanol with a slope of -2.51 mV/ethanol % (NPOE) and -1.01 mV/ethanol% (DA), respectively. The concentration of ethanol and Cl⁻ can be estimated by solving the simultaneous equations derived from the electrical potentials of the two different Cl-ISEs. To decrease the interference from co-existing components, KCl solution was added to the sample until the total concentration was 30 mM. The ethanol sensor using the two Cl-ISEs showed good accuracy in the measurement of ethanol in sake and shochu.

Keywords: ethanol, chloride ion-selective polymeric membrane electrode, plasticizer, quaternary ammonium salts

A wide variety of measuring methods for ethanol have been developed because of the importance of determining ethanol in food processing and control. The measuring methods include the gravity, acid-base reactions, enzymatic reactions, and gas chromatography. As sensing systems, biosensors using an immobilized enzyme (Matsumoto *et al.*, 1990) and gas sensors using a semi-conductor (Samuta *et al.*, 1992) are well known. However, biosensors are accompanied by an inevitable fault in regard to the long-term stability of the enzymes, and gas sensors require expensive monitoring devices and complicated maintenance. An alternative method is required to overcome these problems and to measure ethanol concentration simply and correctly.

Anion-selective polymeric membrane electrodes are based on classical ion exchangers such as quaternary ammonium salts, phosphonium salts, neutral carriers, etc. A large number of polymeric membrane electrodes have been proposed for the analysis of Cl⁻, NO₃⁻, I⁻ and other inorganic anions as well as organic anions (Nakamura, 1994; Wakida, 1997). However, the application of these electrodes for the determination of non-ionic substrates has never been examined.

We previously reported that the electrical potential of a chloride ion-selective polymeric membrane electrode (Cl-ISE) using quaternary ammonium salts decreases linearly with an increase in the concentration of ethanol at a given concentration of Cl⁻. The electrical potentials of the Cl-ISE were found to depend on the concentrations of ethanol and Cl⁻ (Tsukatani & Toko, 1998; Arikawa *et al.*, 1996). Furthermore, investigation of the effect of plasticizers on the response to ethanol and the selectivity for Cl⁻ showed that the response to ethanol and Cl⁻ differed from one plasticizer to

another (Tsukatani & Toko, 1999). This means that the simultaneous equations which include a function of the concentration of ethanol and Cl⁻ can be derived from the electrical potentials of two different Cl-ISEs. These results suggested that the Cl-ISEs could be utilized to measure the concentration of ethanol and Cl⁻ simultaneously. The purpose of this research was to develop a sensor that is capable of measuring ethanol concentration in sake and shochu.

Materials and Methods

Reagents *o*-Nitro-phenyloctyl ether (NPOE) was obtained from Dojindo Laboratories (Kumamoto). Tridodecyl-methylammonium chloride (TDMA) was bought from Tokyo Kasei (Tokyo). *n*-Decylalcohol (DA) and polyvinyl chloride (PVC) were purchased from Wako Pure Chemical Industries (Osaka). All other chemicals were of analytical reagent grade and were used without further purification.

Preparation of polymeric membrane electrode Two different polymeric membranes were examined, one consisted of 5.3 wt% TDMA, 52.6 wt% NPOE, and 42.1 wt% PVC and the other of 6.7 wt% TDMA, 40.0 wt% DA, and 53.3 wt% PVC. The polymeric membrane was prepared by dissolving the three membrane components in about 18 ml of tetrahydrofuran and pouring the solution into a glass plate (90 mm i.d.). After solvent evaporation, the polymeric membrane was cut into small disks and mounted onto a hollow-cylinder electrode.

Cell assembly and measuring apparatus Cells of the type Ag/AgCl|3 M KCl||salt bridge (agar)||sample solution|polymeric membrane|3 M KCl|Ag/AgCl were used for the

potentiometric measurement. The Cl-ISE was made of an Ag wire whose surface was plated with Ag/AgCl and had an internal cavity filled with 3 M KCl solution. Each Cl-ISE was pre-conditioned with 100 mM KCl solution overnight before the measurements.

The measuring apparatus used in this work is shown in Fig. 1. The electrical signal from each electrode was converted to a digital code by a digital voltmeter (DMM2000, Keithley Instruments Inc., Cleveland, OH) through a high-input-impedance amplifier and a multi-channel scanner, and taken into a computer. The electrical potential difference between a Cl-ISE and a reference electrode was measured at a temperature of $25 \pm 1^\circ\text{C}$.

Measurement of ethanol First of all, the simultaneous equations were derived from the electrical potentials measured by two different Cl-ISEs for known ethanol and KCl solutions. Then, KCl solution was added to samples to decrease the interference from co-existent components such as organic acids and amino acids. The concentration of ethanol and Cl^- was estimated by solving the simultaneous equations when the electrical potential for samples was measured. The stabilizing times of the electrical potential were 60 s until its change became within ± 0.1 mV.

Analytical method The ethanol concentration was measured by a gas chromatograph (Model GC-6A, Shimadzu Corp., Kyoto). The titrable acidity and the amino acidity were determined by the official method of analysis of the National Tax Administration Agency of Japan.

Results and Discussion

Linearities for ethanol and Cl^- We previously reported that the response of a Cl-ISE to ethanol was based on the change in transfer activity coefficient of Cl^- from water to ethanol (Tsukatani & Toko, 1998). The electrical potential relative to the concentration of ethanol and Cl^- would be given by

$$\Delta E = S_{\text{Transfer}} [\text{Ethanol}] + S_{\text{Nernst}} \log [\text{Cl}^-]$$

where the slope for [Ethanol] is defined as

$$S_{\text{Transfer}} \equiv S_{\text{Nernst}} \log \gamma_i (\text{Cl}^-, \text{water} \rightarrow \text{ethanol}) \times 1/100$$

$\log \gamma_i (\text{Cl}^-, \text{water} \rightarrow \text{ethanol})$, which is called the transfer activity coefficient of Cl^- from water to ethanol, is known to increase linearly with increasing ethanol concentration (Marcus, 1985). S_{Nernst} represents the Nernstean slope (mV/decade). [Ethanol] and $[\text{Cl}^-]$ are the concentration of ethanol (%) and Cl^- (mM). The electrical potential equation of the Cl-ISE is considered to be a function of the concentration of ethanol and Cl^- .

Figure 2 shows the electrical potentials of the Cl-ISEs relative to the concentration of ethanol and Cl^- . TDMA was used as the sensing material and NPOE or DA was used as the plasticizer. The electrical potentials of two Cl-ISEs fell linearly with increasing ethanol concentration, and all slopes of response lines were equal in different KCl solutions from 1.0 to 100 mM. From the multiple regression analysis, the simultaneous equations for the electrical potential of two different Cl-ISEs could be derived in the following expressions:

$$\text{NPOE: } E = -2.51 [\text{Ethanol}] - 56.1 \log [\text{Cl}^-] + 3.214$$

$$\text{DA: } E = -1.01 [\text{Ethanol}] - 53.8 \log [\text{Cl}^-] + 1.398$$

The responses of both electrodes had nearly Nernstean slopes for Cl^- in ethanol solution (0–20%). In addition, each electrode had good linearity for ethanol with a slope of -2.51 mV/ethanol % (NPOE) and -1.01 mV/ethanol % (DA), respectively. These results mean that the response to ethanol and Cl^- differs from one plasticizer to another. Especially, there was a considerable difference between NPOE and DA from the point of view of the slope for ethanol. One explanation for this may be that hydrogen bond by a protic solvent such as DA to anions affect the ion-exchange reaction of the Cl-ISE because the protic solvent can solvate the anions more strongly than an aprotic solvent such as NPOE. An

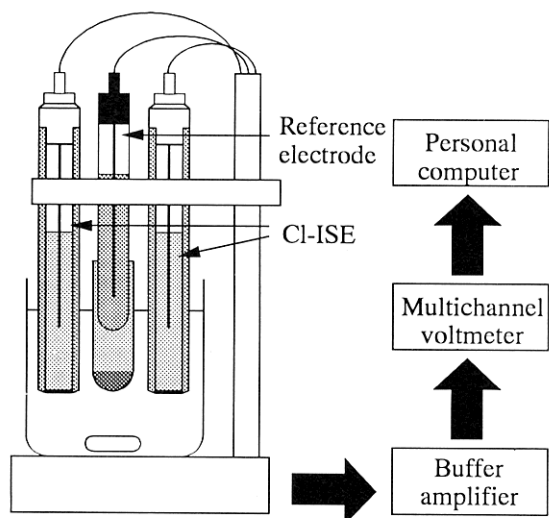


Fig. 1. Experimental apparatus for measuring the electrical potential.

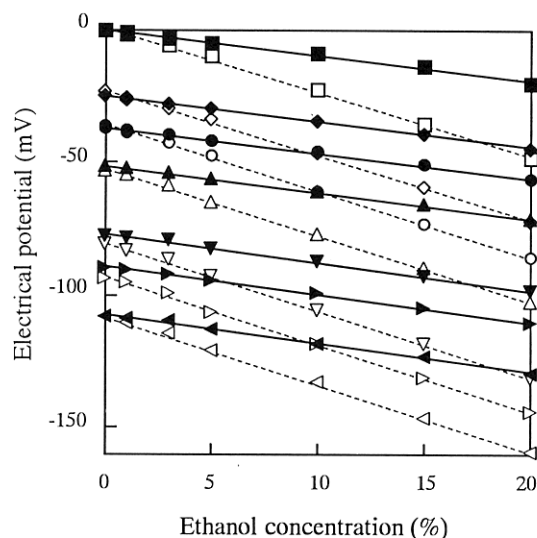


Fig. 2. Response of the Cl-ISEs to ethanol and Cl^- . Plasticizer: (.....) NPOE, (—) DA. KCl: (\square , \blacksquare) 1, (\diamond , \blacklozenge) 3, (\circ , \bullet) 5, (\triangle , \blacktriangle) 10, (∇ , \blacktriangledown) 30, (\triangleright , \blacktriangleright) 50, (\triangleleft , \blacktriangleleft) 100 mM.

aprotic solvent having little ability of hydrogen bond to the anions, in contrast, hardly affects the ion-exchange reaction.

Accuracy and reproducibility for the ethanol measurement Standard mixtures containing ethanol and Cl^- at various concentrations were measured to elucidate the accuracy of the present method, and the results are shown in Fig. 3. The measurement was performed according to the method mentioned in the previous section. The results obtained by the present method (y) were compared with those obtained by gas chromatography (x). A straight line, defined by the equation $y=1.014x-0.084$, with a correlation coefficient $r=0.999$ ($n=36$), was obtained, so that the present technique was found to show good accuracy in the measurement of ethanol.

The reproducibility of the ethanol measurement was determined in various concentration ranges of ethanol and Cl^- . The variation coefficients for ten measurements were less than 1.31%.

Interference study Organic acids and amino acids in sake and shochu affected the responses of the Cl-ISEs based on the ion exchange reaction by quaternary ammonium salts (Matsui & Freiser, 1970; Shibata *et al.*, 1994). Selectivity coefficients of the Cl-ISEs for 3 kinds of organic acids (succinic acid, lactic acid and malic acid) and 14 kinds of amino acids (glycine, alanine, valine, leucine, isoleucine, phenylalanine, proline, serine, threonine, cysteine, aspartic

acid, glutamic acid, lysine and arginine) were determined by the mixed solutions method (Umezawa *et al.*, 1995). The obtained $\log K^{\text{POT}}_{\text{Cl}^-,j}$ values were -1.64 to -2.10 for organic acids and -1.38 to -2.89 for amino acids. The interference could not be ignored in some cases. Thus, the effect of the addition of KCl solution to commercial sake, shochu and synthetic sake on the ethanol measurement was investigated to reduce the interference from co-existing components. The synthetic sakes contain 3 kinds of organic acids and 14 kinds of amino acids at different concentrations. As shown in Table 1, the difference between the values measured by the Cl-ISEs and a gas chromatograph decreased with increase in the total concentration of KCl, and a good agreement was then obtained above 30 mM. This result shows that the interference from co-existing components can be ignored in ethanol solution containing over 30 mM KCl. Thus, KCl solution was added to the sample until the total concentration was 30 mM in subsequent work.

Measurement of ethanol in sake and shochu The ethanol sensor using two different Cl-ISEs was applied to determine ethanol concentration in sake and shochu. Sake was mixed with 3 M KCl solution at a ratio of 99 : 1. While, shochu was diluted 2 times with 60 mM KCl solution. The quality of the present method as regards accuracy was

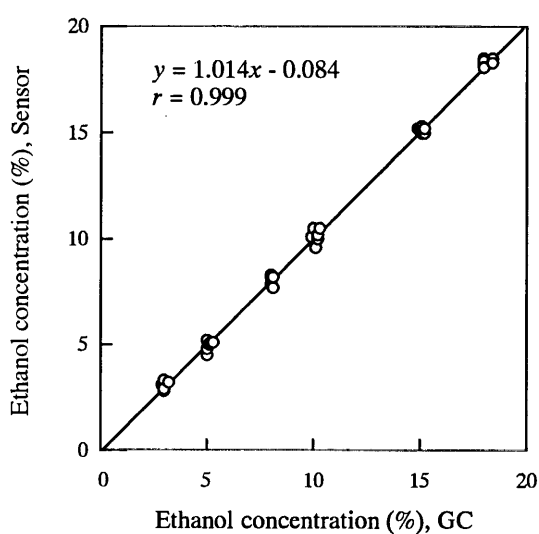


Fig. 3. Measurement of ethanol concentration in standard mixtures.

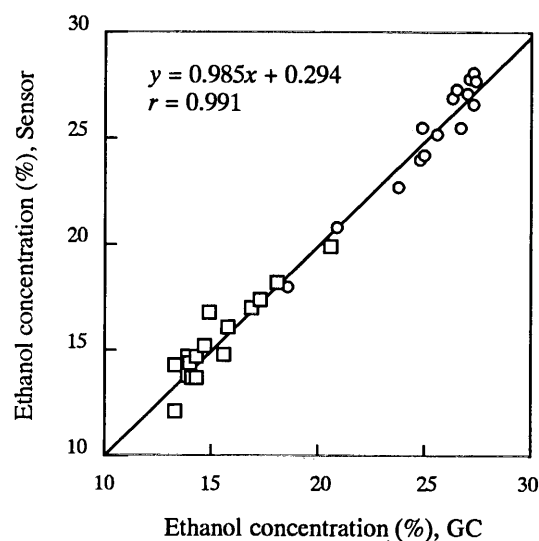


Fig. 4. Measurement of ethanol concentration in sake and shochu. Sake (\square), Shochu (\circ).

Table 1. Effect of KCl concentration on the ethanol measurement.

Total KCl (mM)	EtOH (%) ^{a)}						EtOH (%) ^{b)}	Titrable acidity	Amino acidity
	0	5	10	30	50	100			
Sake 1	35.43	21.32	18.80	17.40	17.32	17.38	17.34	1.36	0.90
Sake 2	35.01	23.11	20.17	18.20	17.99	17.98	18.13	1.20	1.02
Sake 3	35.57	18.11	17.09	16.10	15.95	17.02	15.82	1.22	1.21
Shochu 1	20.11	30.09	30.17	27.70	27.75	29.72	27.38	0.01	0.00
Shochu 2	17.55	37.65	30.65	27.11	27.13	28.74	26.98	0.04	0.00
Synthetic sake 1	34.50	18.78	17.42	15.57	15.80	16.22	15.16	2.72	0.86
Synthetic sake 2	30.31	18.08	16.54	15.32	15.40	16.01	15.10	1.68	0.55

^{a)}Ethanol concentration measured by the sensor, ^{b)}Ethanol concentration measured by GC.

investigated by a comparison of methods. Gas chromatography was chosen for the comparison. Figure 4 shows the results of the determination of ethanol in sake and shochu. A straight line, defined by the equation $y=0.985x+0.294$, with a correlation coefficient $r=0.991$ ($n=32$), was obtained. The ethanol sensor using two Cl-ISEs was found to show good accuracy in the measurement of ethanol in sake and shochu.

However, the developed sensor contains several problems in actual usage, such as adsorption of non-ionic substances to the polymeric membrane and intricacy in sensing module. Membrane components must be improved to reduce the adsorption of non-ionic substances. An ion sensitive field effect transistor is a widely used device in various ion-selective sensors and biosensors (Moriizumi & Nakamoto, 1997), and enables one to reduce the size of the sensing module. For environmental protection, other materials must be substituted for PVC as the polymer. We are confident that the developed sensor will be put into practice when these problems are overcome.

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