

Permeability of Synthetic Poly(α -amino Acid) Membranes to Oxygen Dissolved in Water

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Synopsis

Membranes of synthetic poly(α -amino acids), namely, poly(γ -methyl L-glutamate) (PMLG), poly(γ -benzyl L-glutamate) (PBLG), poly(L-glutamic acid) (PLGA), poly(L-methionine) (PLM), and poly(N ϵ -carbobenzoxy-L-lysine) (PCLL), were prepared and their permeabilities of oxygen dissolved in water were measured in the 8–50°C temperature range using an oxygen electrode. Permeation curves for the poly(α -amino acid) membranes did not approach steady-state currents because of membrane degradation. To eliminate this, the membranes were laminated between polystyrene membranes; thus, the poly(α -amino acid) membranes came in direct contact with neither cathode surface nor electrolyte solution. No effect of membrane thickness on the permeability was observed. The Arrhenius plots of permeability coefficients for PCLL appear to change slope at about 22°C. This is consistent with the diffusion of oxygen in PCLL through the side-chain regions between helices. Comparisons between the permeability of oxygen dissolved in water and permeability of gaseous oxygen obtained by the high-vacuum method and between the activation energy of permeation of dissolved oxygen and that of gaseous oxygen were made in order to elucidate the effect of water on the oxygen permeation of each polymer. The permeability of the poly(α -amino acid) membranes to dissolved oxygen appears to depend on the properties of the side chains of the polymers.

INTRODUCTION

The permeation of dissolved oxygen and carbon dioxide through membranes has been reported.^{1–5} We developed an apparatus for measuring the permeation of dissolved oxygen through a membrane and studied the effect of water on oxygen permeation in ethylene–vinyl acetate copolymer membranes and their hydrolyzed membranes.⁶ The permeation of dissolved oxygen through synthetic poly(α -amino acid) membranes is of special interest because of its importance as a biomaterial for artificial lungs, skin, and corneas.

This investigation developed an experimental method for measuring permeation of dissolved oxygen through synthetic poly(α -amino acid) membranes and to elucidate the effect of water on oxygen permeation through them.

EXPERIMENTAL

Materials

Poly(γ -benzyl L-glutamate) (PBLG), poly(N ϵ -carbobenzoxy-L-lysine) (PCLL), and poly(L-methionine) (PLM) were prepared by the polymerization of the corresponding N-carboxyamino acid anhydride in dichloromethane, dioxane, and dichloromethane, respectively, at room temperature. Triethylamine was used as initiator.

Poly(γ -methyl L-glutamate) (PMLG), supplied by Kyowa Hakkō K. K. (Japan), was prepared by the polymerization of N-carboxyamino acid anhydride.

Membranes of PMLG, PBLG, PLM, and PCLL were prepared by casting dilute solutions of polymer onto glass plates, allowing the solvents to evaporate slowly at room temperature, and drying under vacuum at 80°C for one day. The casting solvents used are summarized in Table I.

Poly(L-glutamic acid) (PLGA) membrane, supplied by Ajinomoto K. K. (Japan), was prepared by the following method: Poly(sodium L-glutamate) (PSLG) was prepared by the saponification reaction of poly(γ -methyl L-glutamate). The PLGA membrane was then prepared by casting the 20% PSLG aqueous solution, immersing in citric acid solution, washing with water and methanol, and drying in air.

The degrees of polymerization of the polymers are listed in Table I. All of the membranes had α -helical structures on the basis of infrared absorption spectra.⁷ The thicknesses of the water-swollen membranes were measured and are listed in Table I.

The degree of hydration of the membrane in Table I is the volume fraction of water of the water-swollen membrane and was determined as follows: The membrane was swollen, blotted, and weighed repeatedly until constant weight of the swollen membrane was obtained within experimental error at 20°C. The membrane was then dried to constant weight under vacuum at 80°C.

Polystyrene membranes were obtained from Mitsubishi Jushi K. K. (Japan). This membrane was strained biaxially and transparent. It was confirmed to be uniform in thickness and free of pinholes before use. The thickness of the wet membrane was 0.019 mm.

Poly(vinyl alcohol) membranes were prepared by casting the aqueous solution of the polymer [Wako Pure Chemical Industries, Ltd. (Japan)] on a poly(methyl methacrylate) plate at room temperature, drying in air, and then irradiating at a dose rate of 1 Mrads/hr from a ⁶⁰Co source at room temperature in air to a total dose of 5 Mrad. The thickness of the wet membrane was 0.300 mm.

Permeation Experiment of Dissolved Oxygen

A cross-sectional view of the oxygen electrode is given in Figure 1. The electrode has an anode (A) of silver tubing (about 150 mm in length and 10 mm in diameter) and a platinum disk (about 5 mm in diameter) as cathode (F). The electrodes are sheathed by an outer tube of stainless steel (B). An electrolyte

TABLE I
Poly(α -amino Acid) Samples

	PMLG	PBLG	PLGA	PLM	PCLL
Degree of polymerization	1120	1820	800	160	1700
Casting solvent	DCE ^a	DCM ^b	Water	DCM	THF ^c
Thickness of wet membrane, mm	0.035	0.035	0.072	0.025	0.027
Degree of hydration	0.109	0.021	0.959	0.020	0.063

^a Dichloroethane.

^b Dichloromethane.

^c Tetrahydrofuran.

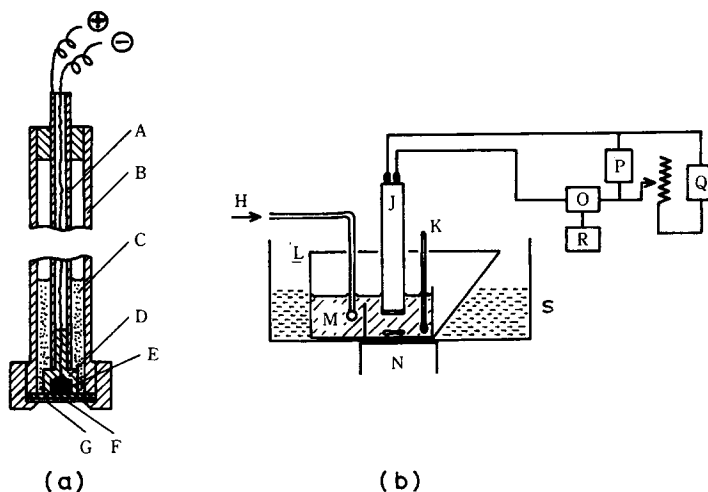


Fig. 1. (a) Cross-sectional view of electrode and (b) schematic diagram of experiment: A, Ag anode; B, outer tube (stainless steel); C, 0.5N KCl solution; D, insulator; E, Pt cathode; F, membrane; G, support; H, N₂ or O₂ inlet; J, electrode; K, thermometer; L, glass equipment; M, distilled water; N, magnetic stirrer; O, microammeter; P, voltmeter; Q, dry cell; R, recorder; S, water bath.

solution (0.5N KCl aqueous solution) partially fills the annular space (C). The electrode was designed for easy change of membranes, to ensure favorable contact between the cathode and the membrane (F) which is at the end of the outer tube, and to permit measurements on water-swollen membranes.

Prior to each run, the cathode was polished to minimize contamination. A new membrane was used for each run. The electrode was inserted vertically into distilled water (m) (Fig. 1). Distilled water was saturated with nitrogen gas to displace oxygen and other dissolved gases and then saturated with oxygen gas. The electrode was operated at -0.7 V, and the reduction current was measured by use of a Hokuto Denko HM-101 ammeter.

Figure 2 shows a schematic permeation curve. From the steady-state current i_{∞} of the permeation curve, the permeability coefficient P [cm^3 (S.T.P.)- $\text{cm}/\text{cm}^2\text{-sec-cm Hg}$] can be calculated by the following equation⁸:

$$P = i_{\infty} L / NFAP_s \quad (1)$$

where L is the thickness of the membrane (cm), N is the number of electrons per molar unit of reaction (four in this case), F is Faraday's constant, A is the area of the cathode (0.190 cm^2 in this work), and P_s is the oxygen partial pressure in the distilled water (1 atm in this work).

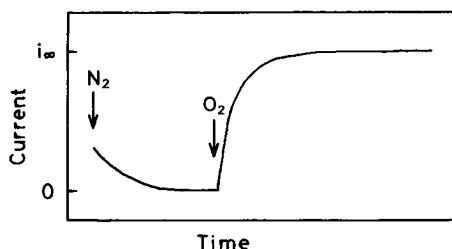


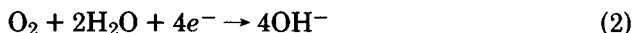
Fig. 2. Schematic permeation curve.

Prior to measurement, the membranes were immersed in distilled water for about one month. Sorption equilibrium with respect to water is assumed.

RESULTS AND DISCUSSION

Establishment of Experimental Method

Permeation curves for the PMLG membrane at 20 and 31°C are given in Figure 3. The currents continued to increase with time and did not approach steady-state values. The higher the temperature of the experiment, the larger was the rate of the current increase. After the measurements, parts of membranes in contact with the cathode of the electrode became opaque. In the case of the PLGA membranes, the permeation curves did not approach steady-state currents, and parts of the membranes dissolved in the electrolyte solutions. Such phenomena were not observed for vinyl polymers. The half-cell reaction for the reduction of oxygen at the cathode surface of platinum may be written as⁹



Accordingly, the hydroxyl ion may hydrolyze the peptide bonds of the poly(α -amino acid) so that the membrane becomes thin with time.

To prevent this, the poly(α -amino acid) membrane (L_1) was laminated to polystyrene membranes as shown in Figure 4. A water layer (L_2) between the membranes must be considered. The poly(α -amino acid) membranes contact neither cathode surface nor electrolyte solution. Figure 5 shows the permeation curves for the composite PMLG membrane. All the permeation curves reached steady-state currents.

Generally, in the three-layer composite membrane the permeability coefficients of the composite membrane, P_{123} , is related to that of the single-layer membrane, P_i , as follows¹⁰:

$$\frac{L_{123}}{P_{123}} = \frac{L_1}{P_1} + \frac{L_2}{P_2} + \frac{L_3}{P_3} \quad (3)$$

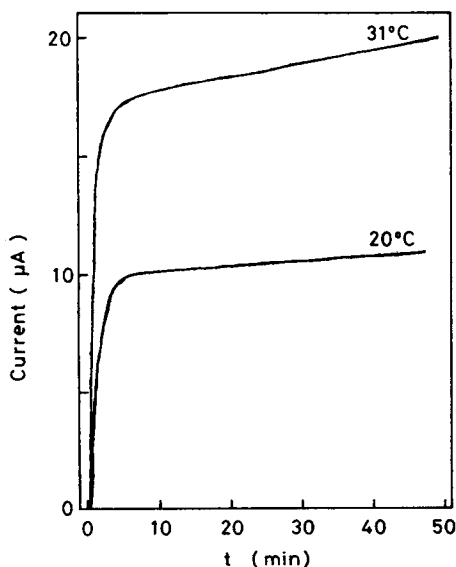


Fig. 3. Permeation curves for PMLG membranes.

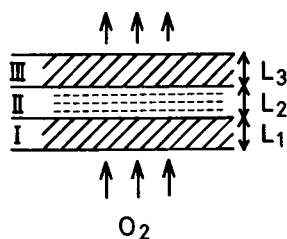


Fig. 4. Profile of laminated membrane: I, poly(α -amino acid) membrane; II, water layer; III, polystyrene membrane.

where L_{123} is the thickness of the composite membrane and L_i is that of the single-layer membrane ($L_1 + L_2 + L_3 = L_{123}$).

In this study (Fig. 4), the thickness of the water layer, L_2 , is much thinner than those of the membranes, L_1 and L_3 ; and the permeability coefficient of the water layer, P_2 , is much larger than those of the membrane, P_1 and P_3 . So, L_2/P_2 in eq. (3) is negligibly small, and eq. (3) approximately gives

$$\frac{1}{P_1} = \frac{1}{L_1} \left(\frac{L_{123}}{P_{123}} - \frac{L_3}{P_3} \right) \quad (4)$$

From eq. (4) we can obtain the permeability coefficient of the poly(α -amino acid) membrane, P_1 , by measuring both the permeability coefficient of the composite membrane, P_{123} , and that of the polystyrene membrane, P_3 .

The validity of eq. (4) was confirmed by measuring the permeabilities of single polystyrene and poly(vinyl alcohol) membranes and laminated polystyrene and poly(vinyl alcohol) membranes. The permeabilities of the polystyrene and poly(vinyl alcohol) membranes which were obtained for single and laminated membranes are in good agreement (Table II). Accordingly, the laminated membrane method should be applicable for determining the permeability coefficient of the poly(α -amino acid) membranes.

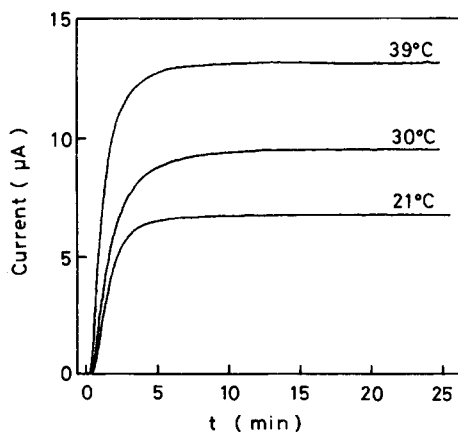


Fig. 5. Permeation curves for composite PMLG membrane.

TABLE II
Comparison of Experimental Method

Membrane	Permeability coefficient, $\text{cm}^3 \text{ (S.T.P.)} \cdot \text{cm} / \text{cm}^2 \cdot \text{sec} \cdot \text{cm Hg} \times 10^{10}$
Polystyrene	1.87
Polystyrene (I) ^a	1.84
Poly(vinyl alcohol)	27.8
Poly(vinyl alcohol) (I) ^b	29.0

^a Laminated method (polystyrene (I) + polystyrene (II)), as shown in Fig. 4.

^b Laminated method (poly(vinyl alcohol) (I) + polystyrene (II)), as shown in Fig. 4.

Permeability of Polystyrene Membranes

Figure 6 shows the temperature dependence of the permeability coefficient of the polystyrene membranes. A single straight line could be drawn through all the points in the temperature range of 8 to 54°C. The values of permeability coefficient of the polystyrene membrane to dissolved oxygen agreed well with those to gaseous oxygen obtained by the high-vacuum method.¹¹ The resistance of any boundary layer to the transport of oxygen dissolved in water through a polystyrene membrane is negligible. This is reasonable for a system with low permeability [$P = 1.87 \times 10^{-10} \text{ cm}^3 \text{ (S.T.P.)} \cdot \text{cm} / \text{cm}^2 \cdot \text{sec} \cdot \text{cm Hg}$]. This contrasts with silicone rubbers for which oxygen permeabilities are very great [$P = 4.5 \times 10^{-8} \text{ cm}^3 \text{ (S.T.P.)} \cdot \text{cm} / \text{cm}^2 \cdot \text{sec} \cdot \text{cm Hg}$] and resistance of the aqueous boundary layer to transport of oxygen is appreciable.⁴⁻⁶

Effect of Membrane Thickness

The reciprocal of the permeability of the PMLG membrane is plotted against the reciprocal of thickness of the PMLG membrane at the constant thickness of the polystyrene in Figure 7. Permeability coefficients are independent of the membrane thickness. This indicates that PMLG membranes prepared are homogeneous and further that the resistance of the boundary layer is also negligibly small in the composite membrane system.

Permeability of Poly(α -amino Acid) Membrane

Logarithms of permeability coefficients of the poly(α -amino acid) membranes obtained by the laminated method are plotted against the inverse temperature in Figure 8. These Arrhenius plots are linear for all polymers measured, except

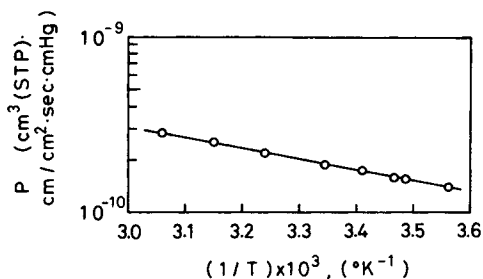


Fig. 6. Temperature dependence of permeability coefficients for polystyrene membrane.

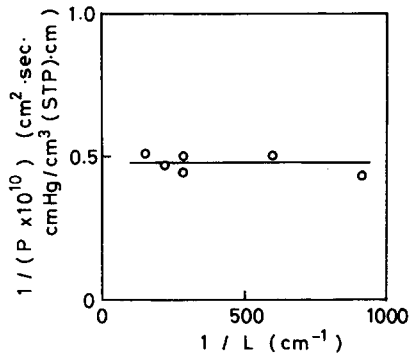


Fig. 7. Effect of membrane thickness on permeability coefficients of PMLG membrane.

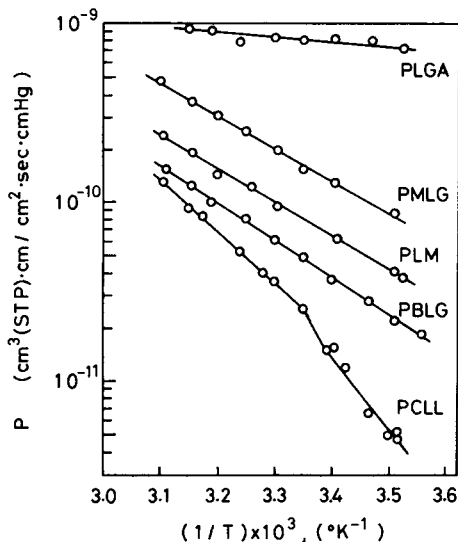


Fig. 8. Temperature dependence of permeability coefficients for poly(α -amino acid) membrane.

for PCLL, which appears to have break in the plot at about 22°C. The glass transition temperature T_g of the PCLL determined by dilatometry¹² is about 20°C; this is associated with molecular motions of the side chains.¹³⁻¹⁵ Accordingly, the break suggests that the oxygen permeation through the water-swollen PCLL membrane is affected by the motions of side chains and, consequently, that the diffusion of oxygen in PCLL occurs through the side chain regions between helices.

Since temperature dependences of permeabilities are linear, the activation energies of permeation of dissolved oxygen, E_{P_w} , were calculated from Figure 8. These are given in Table III with the activation energies of permeation of gaseous oxygen, E_{P_g} , obtained by the high-vacuum method.¹⁶

A comparison between the permeabilities of dissolved oxygen, P_w (water-membrane-water system), and that of gaseous oxygen, P_g (gas-membrane-gas system), obtained by the high-vacuum method¹⁶ is given in Table III. From the ratio of P_w and P_g , the influence of water on oxygen permeation can be observed.

TABLE III
Comparison Between Permeability of Dissolved Oxygen (P_w) and That of Gaseous Oxygen (P_g) at 20°C and Activation Energy of Permeation of Dissolved Oxygen (E_{P_w}) and That of Gaseous Oxygen (E_{P_g})

	Permeability coefficient, cm ³ (S.T.P.)-cm/cm ² -sec-cm Hg		P_w/P_g	Activation energy, kcal/mole	
	$P_w \times 10^{10}$	$P_g \times 10^{10}$		E_{P_w}	E_{P_g}
PMLG	1.26	0.867	1.5	8.3	8.5
PBLG	0.36	0.275	1.3	9.5	12.5
PLGA	7.80	0.0006	13000	1.4	14.8
PLM	0.62	0.403	1.5	8.7	7.9
PCLL	0.13	0.0234	5.6	13.4	15.3 ($T > T_g'$)
				18.1	16.8 ($T < T_g'$)

For PLGA the value of the ratio P_w/P_g is very large because the permeability coefficient P_w of the PLGA membrane is larger than that of the other membranes and the permeability coefficient P_g of the PLGA membrane is very much smaller than that of the other membranes. The activation energy of permeation of dissolved oxygen for PLGA is very small compared with that of gaseous oxygen. The degree of hydration of PLGA is observed to be very high (Table I). We have suggested that sorption and diffusion of small molecules in poly(α -amino acid) membranes with α -helical structure takes place in the side chain regions between helices.^{17,18} From these results the behavior of PLGA can be interpreted as follows: In the *dry* state the side chains of PLGA are linked by hydrogen bonds, so that the motions of the side chains are limited. When wetted, however, the hydrogen bonds are broken by water molecules and the PLGA membrane swells, so that oxygen molecules can diffuse more easily.

For PCLL, the value of the ratio P_w/P_g is larger than that for PMLG, PBLG, and PLM, though the degree of hydration is comparable. This indicates that the water in PCLL acts as a plasticizer and increases the motion of the long side chains of PCLL.

As can be seen in Table III, the P_w/P_g ratio in PMLG, PBLG, and PLM is 1.3–1.5. Compared with PCLL and PLGA, the effect of water on oxygen permeation through PMLG, PBLG, and PLM is small.

We conclude that the permeability of synthetic poly(α -amino acid) membranes to dissolved oxygen depends on the motion of the side chains of the polymers.

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