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pH-RESPONSIVE PERMEATION OF BILAYER-COATED CAPSULE MEMBRANES BY AMBIENT pH CHANGES[1)]

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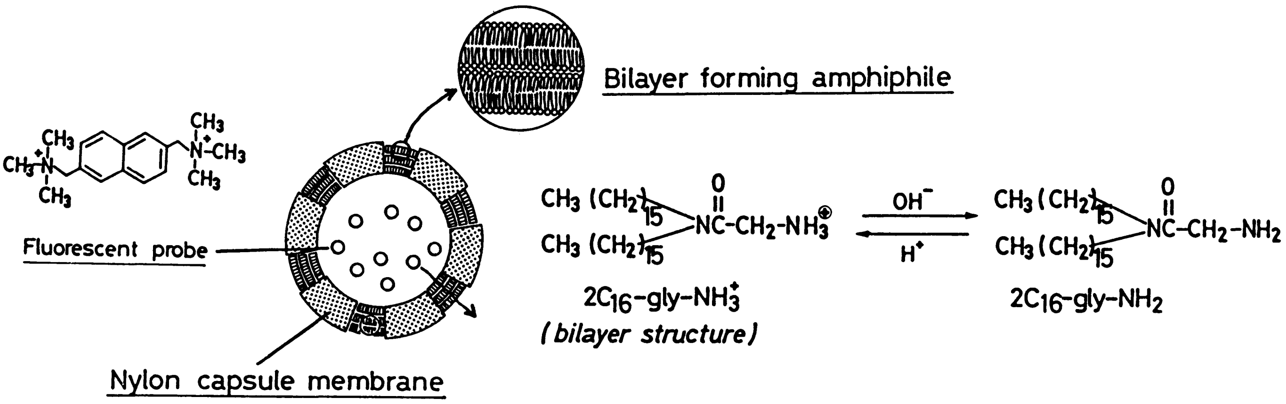
Nylon capsule membranes coated with synthetic bilayers containing an dissociative, cationic head group ($2C_{16}$-gly-NH_3^+) were prepared. Permeation through the membrane of a fluorescent probe trapped in the inner aqeous phase was reversibly controlled by pH changes of the outer medium. This pH-sensitive permeation occured only at the temperature above T_c of coating bilayers, but not below their T_c.

Recently we prepared novel functional nylon capsules whose porous membranes were coated with synthetic bilayers.[2-10)] The capsule is formed by physically strong, ultrathin nylon membranes and the coating shows characteristics of bilayer vesicles. Permeation through the membrane of NaCl trapped in the inner aqueous phase was reversibly controlled by outside effects such as temperature,[2-10)] photoirradiation,[4)] ultrasonic power,[6)] and interaction with divalent cations.[5,9)] Their signal-receptive permeability control is explained by changes in the physical state of the coating bilayers which act as an 'ion gate'. Aqueous synthetic bilayer vesicles as well as liposomes could not achieve such a reversible permeability control because of their easily breakable bilayer walls.

In this communication, we describe that an ambient pH can also act as a signal which causes the reversible permeation control from nylon capsules coated with synthetic bilayers containing a dissociative head group. A schematic illustration of the capsule is shown below.



Large nylon-2,12 capsules (diameter: 2 mm, membrane thickness: 5 μm) were obtained by an interfacial polymerization and dialyzed against aqueous solution of 0.01 M NaCl containing 2,6-bis(trimethylammoniomethyl)naphthalene dibromide as an fluorescent probe. The capsules were coated with N,N-dihexadecyl-α-ammonioacetoamide chloride[11)] ($2C_{16}$-gly-NH_3^+) in dodecane solution at 60 °C according to the previous method.[2-10)] The porous, ultrathin membrane of a capsule was observed by scanning electron microscopy to be entirely covered with the amphiphiles. It was also confirmed by X-ray diffraction analysis and transmission electron microscopy (TEM) that amphiphiles on the capsule membrane exist as well-oriented, multiple-bilayers.[9,10)] The bilayer-coat was also proved to have a phase transition temperature (T_c=51 °C) between crystal and liquid crystal, as in the case of aqueous bilayer vesicles. Amphiphile contents on the capsule were obtained to be 5.0 ± 0.2 μg per capsule.

Permeability of capsules toward the fluorescent probe trapped in the inner aqouous phase was measured by detecting increases in the relative fluorescence intensity at 340 nm (exitation at 280 nm) of the outer water phase after dropping one capsule into 3 ml aqueous solution (pH 7) in a qualtz cell. pH Values of the outer medium was changed by adding aliquots of aqueous 0.1 M HCl or NaOH solution. Fig. 1 shows typical time courses of permeations of the fluourescent probe at different pH's of the outer medium. In the case of the uncoated, semipermeable capsule, the permeation was very fast and not affected with the ambient pH 7 or 11. When the capsule coated with $2C_{16}$-gly-NH_3^+ bilayers was employed, the probe permeation was remarkably reduced at an ambient pH 7 at both 60 °C and 25 °C, which means the cationic $2C_{16}$-gly-NH_3^+ bilayer-coats provide a high barrier to the permeation of a cationic probe. Upon changing the pH of the outer medium from 7 to 11 at 60 °C, the permeability was immediately enhanced by a factor of 7-8 and reduced again nearly to the original slow rate by returning the ambient pH to 7. This permeability regulation by ambient pH changes could be repeated many times without damaging both coating bilayers and capsule membranes, until most of probes had permeated. It is obvious from the inserted pH-rate profile (curve b) in Fig. 1 that the permeability enhancement in the basic medium at 60 °C is due to the neutralization of cationic head groups of coating $2C_{16}$-gly-NH_3^+ bilayers (pK_a= 10). Thus, the $2C_{16}$-gly-NH_3^+ coatings provide a high barrier to the probe permeation in the cationic bilayer form below pH 9, but not in the neutral form above pH 11.

Only a minimum effect of the ambient pH on the permeation was noted at 25 °C (curves c of Fig. 1 and the inserted figure), against at 60 °C. It is well known that the bilayer property varies largely between below and above T_c of coating bilayers.[2-10)] Permeability coefficients were obtained in a range of 10 - 70 °C at both ambient pH's 7 and 11, and Arrhenius plots were shown in Fig. 2. In the case of the uncoated capsule, the plot of log P vs. T^{-1} gave a simple straight line. On the contrary, Arrhenius plots of the cationic $2C_{16}$-gly-NH_3^+ bilayer-coated capsule gave inflections, at both pH's 7 and 11, near 51 °C, which consist with T_c of coating bilayers obtained from DSC measurements (shown as an arrow in the figure). The effect of the ambient pH on the permeability was not observed below T_c of coating bilayers, but clearly above their T_c.

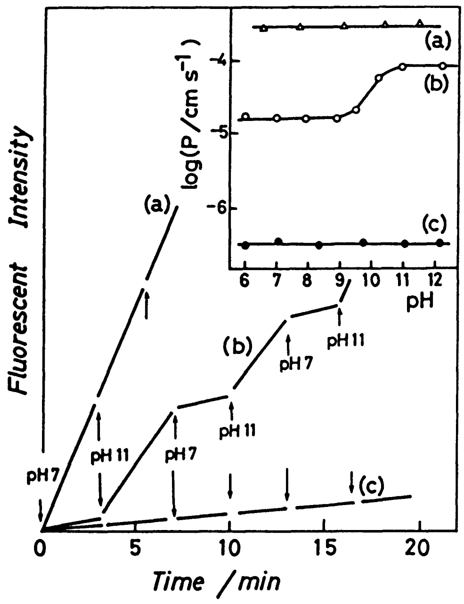


Fig. 1. pH-Sensitive permeation from nylon capsule membranes of a fluorescent probe by pH changes of the outer medium. The inserted figure shows pH-rate profiles of permeations.

(a): The uncoated capsule at 60 °C,

(b): The $2\mathrm{C}_{16}$-gly-NH_3^+-coated capsule at 60 °C (above T_c),

(c): The $2\mathrm{C}_{16}$-gly-NH_3^+-coated capsule at 25 °C (below T_c).

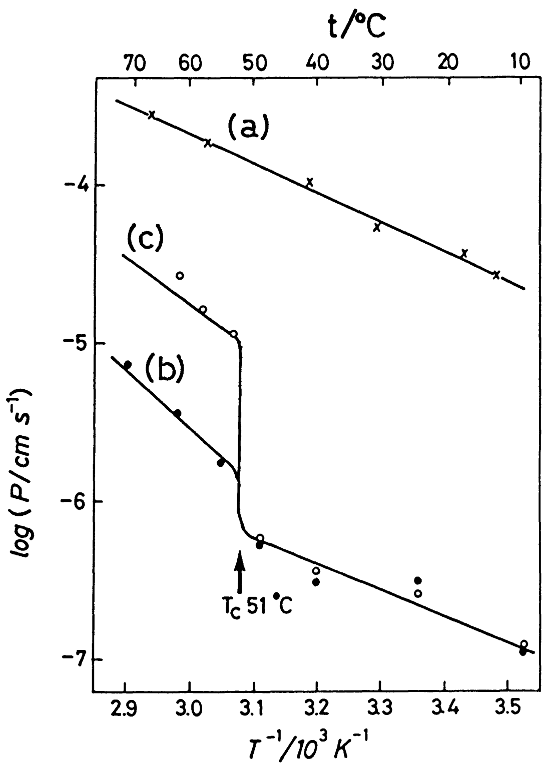


Fig. 2. Arrhenius plots of the probe permeation from capsules.

(a): The uncoated capsule in pH 7,

(b): The $2\mathrm{C}_{16}$-gly-NH_3^+-coated capsule in pH 7,

(c): The $2\mathrm{C}_{16}$-gly-NH_3^+-coated capsule in pH 11.

This different effect of the ambient pH below and above T_c may be explained as follows. In the neutral (or acidic) medium, $2\mathrm{C}_{16}$-gly-NH_3^+ amphiphiles exist as the cationic bilayers (providing a high barrier to the permeation), and the inflection near T_c is account for the phase transition from the rigid gel to the fluid liquid crystal of coating bilayers (Fig. 2, curve b). The permeation enhancement above T_c was frequently observed in other bilayer-coated capsules.$^{2-10)}$ In the basic medium (pH 11), the permeability was drastically enhanced above T_c, but not below T_c. At the temperature above T_c, the fluid cationic bilayers are immediately neutralized in the basic medium, and the permeability is drastically enhanced probably because of the non-, or disturbed-bilayer structure of the neutral $2\mathrm{C}_{16}$-gly-NH_2 coatings. In the rigid gel state below T_c, only the surface of multiple-bilayer-coats seems to be neutralized and the inner, most of bilayer-coats still exist as an cationic form (providing a high barrier to the permeation) even at the ambient pH 11.

The capsule coated with $2\mathrm{C}_{16}$-gly-NH_3^+ bilayers (T_c= 51 °C), which was immersed in the basic aqueous solution (pH 11) above their T_c, showed an endothermic peak at 33 °C (melting point of $2\mathrm{C}_{16}$-gly-NH_2 amphiphiles), instead at 51 °C (T_c of cationic bilayers) by DSC measurements. X-ray diffraction patterns of bilayer spacings were not clearly observed in this neutralized capsule. When the capsule was immersed in the basic solution below T_c, coating amphiphiles still showed the original bilayer characteristics. These results strongly suggest the above explanations.

pH-Sensitive, $2C_{16}$-gly-NH_3^+ bilayer-coated capsule membrane can be also applied to the permeation of other water-soluble substances, such as NaCl and glucose. Yatvin et al.12) reported that pH-sensitive liposomes in which dissociative amphiphiles are incorporated would be useful for clinical implications; they would release encapsulated drug when passing around tumor cells that have the considerably lower pH than that of normal tissues. It is difficult, however, to realize the reversible permeation control in liposomal membranes, because they are easily damaged and fused each other by continuous changes of ambient pH's. On the contrary, the bilayer-coated capsule membranes are not damaged by continuous pH changes, because lipid bilayers are supported by the physically strong capsule wall.

In conclusion, although nylon capsule membranes are simply semipermeable, the capsule coated with the dissociative, synthetic bilayers can reversibly control the permeation by pH changes of the outer medium, depending on the phase transition temperature (T_c) of coating bilayers. This is the first example of reversibly pH-sensitive permeation control across capsule membranes. These pH-sensitive, bilayer-coated capsule membranes could be useful for biological and industrial uses.

References

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