Transport of Ionic Electrolytes and Proteins through Semipermeable Membrane with Effect of Surfactants

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ABSTRACT

We previously systematically reported how anionic surfactants of different hydrophilicities affected three metabolically important enzymes (namely, glutamate dehydrogenase (GDH), lactate dehydrogenase (LDH), and malate dehydrogenase (MDH)) of various molecular masses and their transport behaviors through a semipermeable membrane at a pH range (6.5-7.4) and concentrations relevant to body functions.

Reports of how electrolytes that are important to human body functions, such as Cl^-, K^+, Ca^{++}, and Na^+ , are transported through cellular membrane in a homogeneous setting are not readily available.

In this study, we systematically investigated how the surfactants of various hydrophilicities affect the interfacial transport of Cl^-, K^+, Ca^{++}, and Na^+ through a semipermeable membrane in the presence of enzymes at pH 6.5 to 7.4.

Surfactants of various hydrophilicities have definite effects on activities of enzymes and the effects are also pH dependent, but surfactants with concentrations at ppm level do have significant effect on the permeabilities of ionic electrolytes. In the presence of multiple enzymes, permeation rates of enzymes decrease when they are compared to a single enzyme solution.

Keywords: electrolytes, proteins, surfactants, permeation, membrane.

1 INTRODUCTION

Surfactants are known to influence functions of many proteins in membranes, cells and tissues. Most previous studies employed heterogeneous or complex systems to elucidate the effects of surfactants on membrane-bound proteins and cells, it is difficult to extrapolate the results of such studies to delineate the effects of surfactants on a single protein.

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We previously reported on systematic studies regarding how anionic surfactants of different hydrophilicities affected three metabolically important enzymes (namely, glutamate dehydrogenase (GDH), lactate dehydrogenase (LDH), and malate dehydrogenase (MDH)) of various molecular masses and their transport behaviors through a semipermeable membrane at a pH range (6.5-7.4) and concentrations relevant to body functions.

On the other hand, reports of how physiological relevant electrolytes that are important to human body functions, such as Cl^-, K^+, Ca^{++}, and Na^+, are transported through cellular membrane in a homogeneous setting are not readily available.

Surfactants of various hydrophilicities have definite effects on activities of enzymes and the effects are also pH dependent, but surfactants with concentrations at ppm level are not known to have much effect on the activities of ionic

electrolytes. However, the combined effect of enzymes and surfactants on transport of the electrolytes across the semipermeable membrane is not known. Understanding of such transport phenomena will help elucidate the mechanism of how ions are transported through membrane channels.

In this study, we systematically investigated how the surfactants of various hydrophilicities affect the interfacial transport of Cl^-, K^+, Ca^{++}, and Na^+ through a semipermeable membrane in the presence of enzymes at pH 6.5 to 7.4.

2 MATERIALS AND METHODS

The enzymes used in this study were GDH, LDH, and MDH. Detection of enzymes and experimental setup of the separation cell were detailed in previous reports [1,2]. The semipermeable membrane was polycarbonate with 1 micron pore size. The surfactants used were Triton X-100 (non-ionic), IB 45 (hydrophobic) and TR 70 (hydrophilic). Chloride salts of analytical grade were the source of Ca^{++}, K^+, and Na^+; all ions were detected by electrodes.

3 RESULTS AND DISCUSSIONS

3.1 Permeation rate of GDH, LDH, and MDH

Transport of MDH across the semipermeable membrane was not as fast as previous observed [2]. As shown in Figure 1, the permeation rate of the enzymes in descending order are: GDH > LDH > MDH at pH 6.95 and at room temperature. This permeation order is totally the reverse of what is predicted by conventional theory regarding the molecular sizes and thus interfacial molecular transport is controlled by multiple mechanisms, including molecular size effect [1].

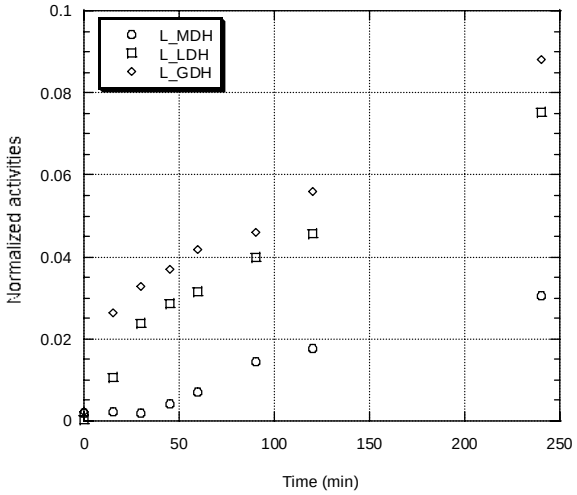

Figure1: MDH, LDH, and GDH without surfactant at pH 6.95.

3.2 GDH permeation rate in the presence of other enzymes

The permeation rate of GDH decreased in the presence of other enzyme proteins: participation of multiple enzymes appeared to slow the permeation rate of GDH more than when GDH permeated as a single enzyme (Figure 2). We observed similar phenomenon for other proteins as well. It is assumed that all the protein molecules were competing for the limited channels for transport; however, the magnitude of the decrease in permeation rate varied.

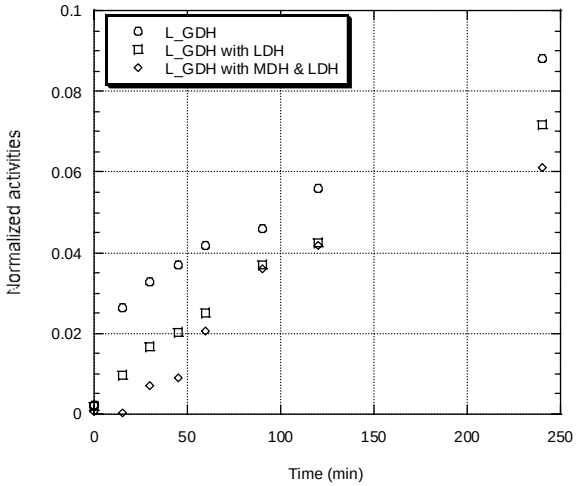

Figure 2: GDH permeation in the presence of LDH and MDH at pH 6.95

3.3 Effect of surfactants to enzyme permeation rate

We observed surfactants having definite effect on the permeation rate of enzymes and the effect differed with enzymes (size), hydrophilicity of surfactant, and pH. As shown in Figure 3, the permeation rate of GDH was more than double in the presence of 0.1 ppm of hydrophobic surfactant TR 70. This surfactant effect ~~to~~ on proteins could be important in cell signaling and biomedical applications.

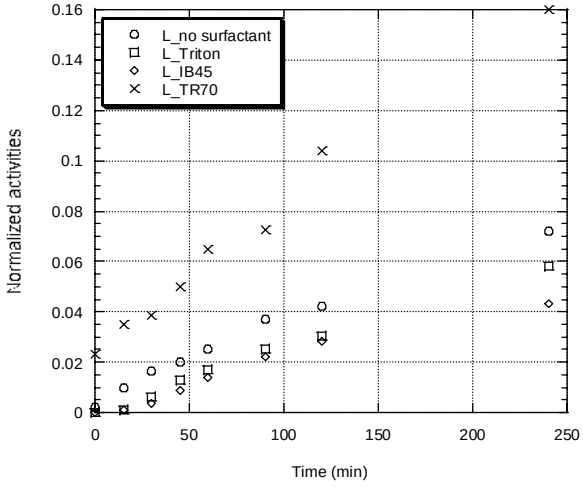

Figure 3: Permeation of GDH with surfactants of various hydrophilicity in the presence of MDH at pH 6.95

NSTI-Nanotech 2007, www.nsti.org, ISBN 1420061828 Vol. 1, 2007

3.4 Effect of surfactants to permeation rate of electrolytes

Figure 4 shows the effect of surfactants on the permeation rate of Na ion (0.095 nm) with polycarbonate membrane, which is considerably insignificant compared to hydrophilicity and surfactant concentration (0.1 ppm) that are highly influential to permeation rate of proteins. We also observed that concentration variations that are important to biological function did not alter permeation rate of Na ion significantly. Likewise, the presence of protein enzyme did not post any significant effect to the permeation rate of Na ion.

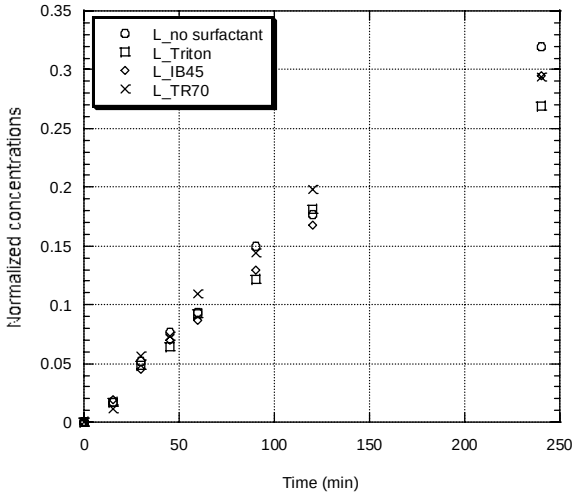

Figure 4: Permeation of 0.1 M of Na^+ ion in the presence of three surfactants with various hydrophilicity at pH 6.95.

3.5 Effect of surfactants on LDH in the presence of electrolytes

The effect of surfactants was more prominent than just the presence of NaCl, as ~~it is~~ shown in Figure 5. As mentioned previously, the influence of surfactant differs depending on the characteristics of proteins and pH. From the standpoint of cell signaling in term of interfacial protein transport, the effect of surfactant can override the effect of inorganic ions (K^+, Na^+, Ca^{++}, Cl^-).

4. CONCLUSIONS

Permeation rates of proteins with moderate molecular size (>70,000 Da) revealed reverse order than expected, in relation with molecular size. In the presence of multiple enzymes, permeation rates of enzymes decreased, compared to a single enzyme solution. Surfactants could significantly affect permeation rate of enzymes, but posted no significant effect to the permeation rate of electrolytes with the semipermeable polycarbonate membrane. To project transport behavior of DNA across semipermeable membrane, we expect DNA to exhibit different behavior from the proteins we used in this study under similar conditions.

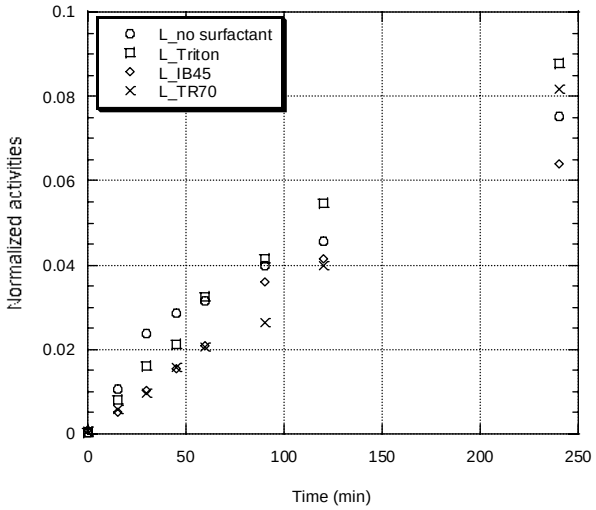

Figure 5: LDH permeation with various surfactants in the presence of 0.1 M of Na and Cl ions at pH 6.95

5 ACKNOWLEDGEMENTS

This study was supported, in part, by NIH Grant #P20 RR016454 fro the INBRE Program of the National Center for Research Resources, a Graduate Student Research Grant, and College of Engineering of Idaho State University.

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