Adhesion of Binary Vesicles Containing negative spontaneous curvature lipids

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Adhesion of cell membranes is one of the elementary processes of biological phenomena such as membrane traffic. So far, the physical aspects of the vesicle adhesion have been investigated from the view points of a key-lock interaction between ligand and receptor molecules embedded in the membranes, or the interplay of the generic interactions (repulsive hydration interaction and attractive van der Waals interaction). On the other hand, in the course of the vesicle fusion process, it is considered that the adhering vesicles form the "hemifusion" structure between upper leaflets of apposing vesicles (Fig. 1). In this study, based on the geometrical model, we tried to make two vesicles to adhere using negative spontaneous curvature lipids (DPPE and DPhPC), which prefers to form hemifusion structure.

We prepared binary giant vesicles by mixing negative spontaneous curvature lipids and zero spontaneous curvature lipids (DOPC and DPPC). In the high temperature region the two kinds of lipids mix in the vesicle homogeneously, whereas in the low temperature region they undergo a phase separation and form negative spontaneous curvature region and zero spontaneous curvature region. First, we determined the phase separation boundary using a fluorescent microscopy and then checked the adhesion behaviors using a micro–manipulation technique. In the homogeneous one phase region, two binary vesicles did not adhere each other and in the coexisting two phases region, the vesicles showed the adhesion. The adhesion-nonadhesion boundary agrees well with the immiscibility boundary as shown in Fig. 2.

The fluorescence microscope observation reveals that the adhesion takes place through the region rich in the negative spontaneous curvature lipid (Fig. 3). It should be noted that pure negative spontaneous curvature lipid vesicles didn't show adhesion. Thus the formation of negative spontaneous curvature region is responsible for the observed adhesion behavior. Based on these experimental results we propose the phase separation induced hemifusion model where the outer leaflets of the bilayers merge at the boundary of the negative spontaneous curvature domains. The fluorescence lipid transfer experiments strongly support the hemifusion model.

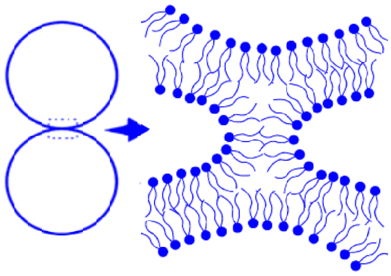

Fig. 1 Hemifusion structure

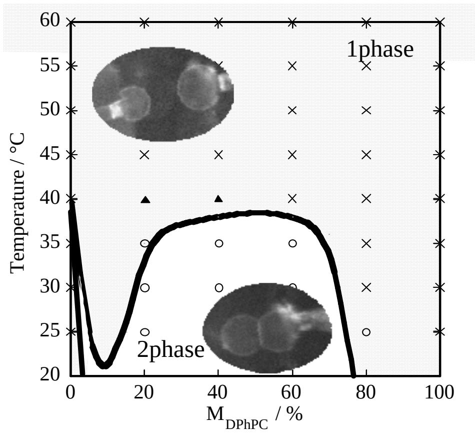

Fig. 2 Phase diagram and adhesion boundary observed in DPhPC/DPPC binary giant vesicles. Solid line indicates phase separation boundary. Open circle, cross symbol and triangle mean adhesion, non-adhesion and adhesion-nonadhesion boundary, respectively.

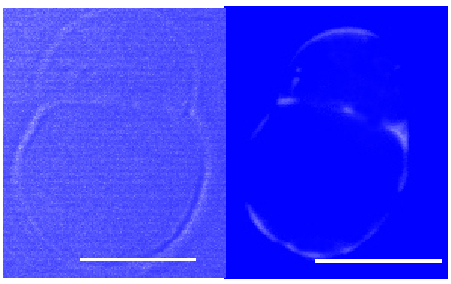

Fig. 3 Cross section images of adhering DPPE/DOPC binary vesicles. (a)phase contrast image and (b)fluorescence image (DOPC phase is dyed.)