X-ray Reflectivity Studies on the DNA Adsorption by DPPG Monolayer with Divalent Cations

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Recently, there are many studies on the lipid-DNA complex for developing the delivery systems for nonviral gene therapy. Most of these studies focused on the structure and interaction of cationic lipid-DNA complex in solution. In this research, we have studied the anionic lipid monolayer and its interaction with DNA by measuring the surface pressure- area isotherm and X-ray reflectivity. The LB films were prepared from the DPPG [1,2-dipalmitoyl-*sn*-glycero-3-phospho-(1'-*rac*-glycerol)] monolayer with 1 μ M DNA in the subphase with different amounts of calcium ions added. The effect of adding divalent ions on the DNA adsorption by anionic lipid monolayer at the air-water interface was investigated by X-ray reflectivity for Langmuir-Blodgett (LB) films supported on silicon wafers. It is found in this study that adding divalent ions, such as calcium ions, can enhance the DNA adsorption to interfaces. The adsorbed DNA layer thickness was found to increase with the increase of divalent ion concentrations.

Figure 1 shows the isotherms of DPPG and DPPG/DNA monolayers and the X-ray reflectivity curves of the corresponding LB films supported on silicon wafer. It is interesting to note that a plateau appears in the isotherm of DPPG/DNA system, which indicates the DPPG monolayer can interact with the DNA molecules in the solution. A plateau region existed in the DPPG/DNA isotherm. It is not expected for such a negative charge lipid to have condensation interaction with negative charge polyions (DNA) without the help of multivalent cations. However, the reflectivity curve of the DPPG/DNA LB film is almost identical to the pure DPPG monolayer case. There is only slight difference between these two reflectivity curves which indicates the DNA molecules have some effects on the DPPG monolayer at the air-water interface but DNA molecules do not form a stable adsorbed layer underneath the DPPG monolayer. The interaction between the DPPG monolayer and DNA must be weak.

Recent studies indicated that negative charged lipid bilayer membrane could also interact with DNA through the addition of multivalent ions. The presence of calcium ions in the solution seems to have little effect on the isotherm of the DPPG and the reflectivity curves of the DPPG LB films. Figure 2 shows the isotherms of DPPG monolayers in the presence of $CaCl_2$ in the subphase and the X-ray reflectivity curves of the corresponding LB films supported on silicon wafer. The thickness of the DPPG monolayers seems to have little change with the addition of calcium ions in the solution during the preparation of the DPPG LB monolayer supported on silicon wafer. Figure 3 shows the isotherms of DPPG/DNA monolayers in the presence of $CaCl_2$ in the subphase and the X-ray reflectivity curves of the corresponding LB films supported on silicon wafer. In the presence of DNA in the sub-phase, the isotherms of the DPPG/DNA/$CaCl_2$ are significantly different from the DPPG/$CaCl_2$ system. This indicates that DPPG monolayer can interact strongly with the DNA in the solution assisted by the calcium ions. Upon the addition of calcium ions, DNA molecules are adsorbed by the DPPG monolayers and the thickness of the adsorbed DNA layer seems to increase with the increase of the calcium ion concentration.

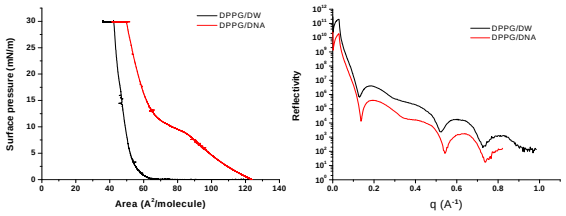

Fig. 1: Isotherms of DPPG and DPPG/DNA monolayers and the X-ray reflectivity curves of the corresponding LB films supported on silicon wafer.

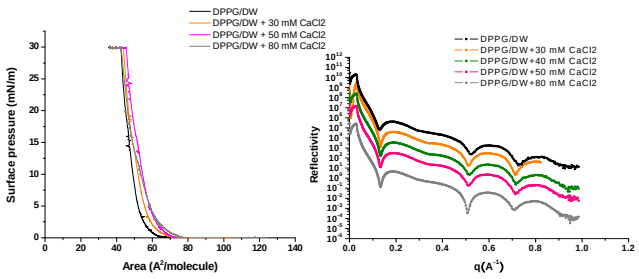

Fig. 2: Isotherms of DPPG monolayers in the presence of $CaCl_2$ in the subphase and the X-ray reflectivity curves of the corresponding LB films supported on silicon wafer.

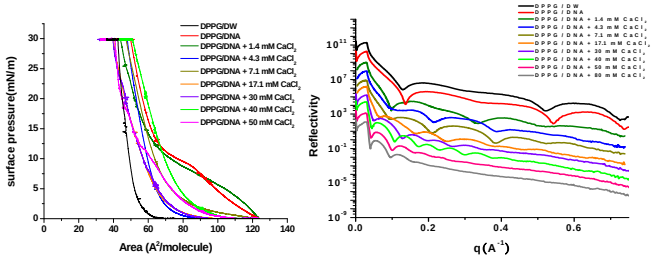

Fig. 3: Isotherms of DPPG/DNA monolayers in the presence of $CaCl_2$ in the subphase and the X-ray reflectivity curves of the corresponding LB films supported on silicon wafer.