Molecular organisation of amphotericin B in lipid monolayers with cholesterol and ergosterol

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Amphotericin B is the metabolite *Streptomyces nodosus* and is one of the oldest polyene antibiotic and is used in the treatment of invasive systemic fungal infections. Despite its over 50 year existence in clinical practice, and the recognition of amphotericin B as the gold standard in the treatment of serious systemic mycosis, it still remains one of the most toxic pharmaceuticals. The understanding of the processes at the molecular levels and the interactions between amphotericin B with lipid membranes containing sterols should elucidate the mechanisms of action and toxicity of this widely used antibiotic. In this work we use X-ray reflectivity to study the structural changes on a molecular scale accompanying the known increase in surface pressure when amphotericin B in incorporated into a ergosterol rich DPPC membrane than cholesterol rich one as shown by Langmuir isotherms. The data show that this difference is not due to higher affinity of amphotericin B towards membranes containing ergosterol but is rather due to a ~3 Angstrom corrugation of the monolayer resulting in a broadening of the X-ray SLD. Furthermore, the total quantity of amphotericin B incorporated into lipid monolayers containing cholesterol and ergosterol is the same.

Presented X-ray diffraction data shows that the amount of incorporated amphotericin B in lipid monolayers containing cholesterol and ergosterol is the same (Fig. 1). To explain this effect we introduced an extension to the generally accepted superlattice lipid-sterol model. In our model we propose that the monolayer is corrugated/buckled (Fig. 2). Such corrugation/buckling is manifested by broadening of X-ray SLD profile of lipid-cholesterol monolayer comparing to lipid-ergosterol one. This explains the smaller molecular area for a lipid-cholesterol monolayer as compared to a lipid-ergosterol one [1].

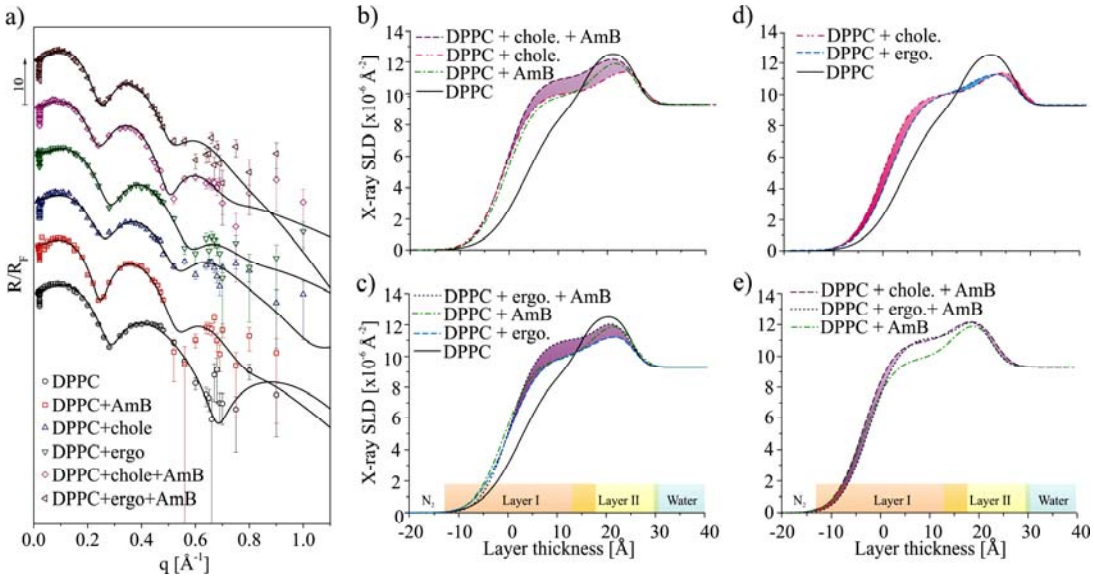

Figure 1: (a) Measured data (symbols) and model fitted curves (solid lines) normalized by Fresnel reflectivity plotted against scattering vector q_z for monolayer at surface pressure of 10 mN/m. For clarity the data have been offset vertically. (b-d) Interfacial X-ray SLD profiles calculated from the fits in a.

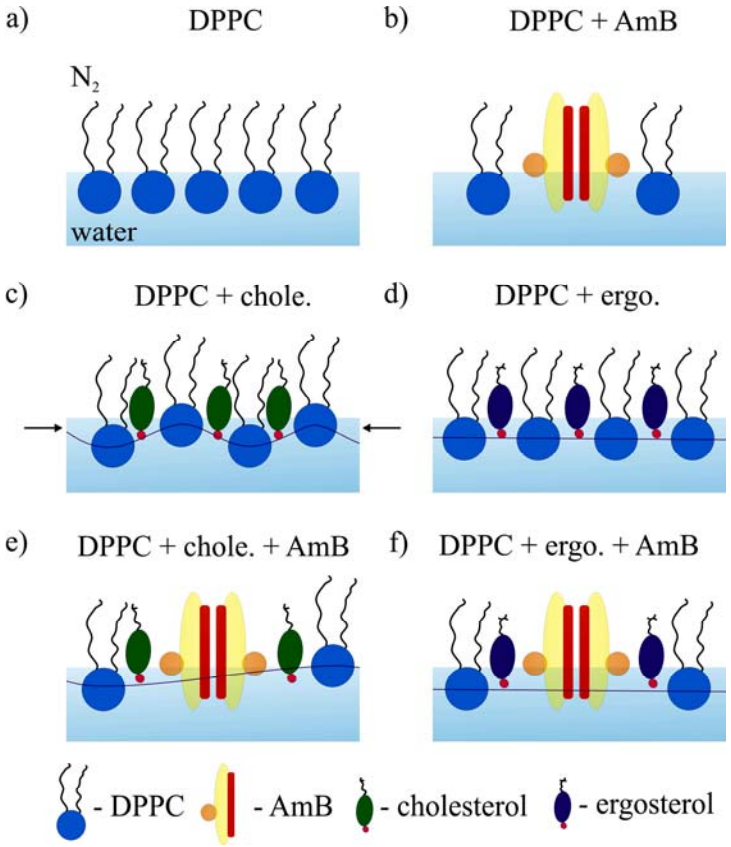

Figure 2: Simplified models of the corresponding out-of-plane electron density profiles used for explanation. Arrows indicate AmB movements comparing to the lipid layer without sterols. Red strip in case of AmB represents polyol subunit reach in –OH groups. Arrows on panel c shows surface compression caused by monolayer corrugation and thus smaller molecular surface area.

References

[1] K. Sabatini, J.P. Mattila and P.K.J. Kinnunen, Biophysical Journal, **95** (2008) 2340-2355.