



SUPPLEMENTARY FIGURE 1. Asymmetric two-phase lipid bilayers with the inner leaflet containing 1% (A) or 5% (B) bPIP<sub>2</sub>. The outer leaflet monolayers (A1 and B1) are composed of bPC/bSM (1:1) + 20% cholesterol, supported on a polymer cushion and visualized with 0.25% NBD-DPPE. The inner leaflet monolayers are composed of bPC + 15% POPS + 20% cholesterol + 1% PIP<sub>2</sub> (A2) or 5% bPIP<sub>2</sub> (B2) and are labeled with 0.02% Rh-DPPE. The same regions of double labeled bilayers are shown in A1 and A2, B1 and B2. Scale bar: 40 μm.