

Supplementary Movie Captions:

Supplementary Movie 1: Isolated short rods localize to the membrane-polymer interface.

Time-lapse movie of an edge-on colloidal membrane consisting of unlabeled *fd-wt* and a low volume fraction of fluorescently labeled Litmus rods. Short rods are localized to the membrane-polymer interface to reduce the number of defects in the smectic ordering of the background membrane. The arrow indicates the isolated short rods that hop between the top and bottom interface of the membrane. The number ratio of *litmus* to *fd-wt* is 1:3000 at dextran concentration of 50mg/mL. Scale bar, 2 μm .

Supplementary Movie 2: Dynamics of short rods in a binary colloidal membrane.

Time-lapse movie of a 2D colloidal membrane consisting of weakly fluorescently labeled *fd-wt* (blue) and unlabeled *litmus* rods. The bilayer droplets are enriched in *litmus* rods. A low volume fraction of *litmus* rods (1:10,000) are highly labeled (bright blue) and are observed as bright points in the membrane. They continuously exchange between the dense *litmus* enriched droplets and the background membranes demonstrating the structure fluidity. $n_{\text{mem}}=1.5$ and dextran concentration is 50 mg/mL; Scale bar, 5 μm .

Supplementary Movie 3, Lateral phase separation in binary colloidal membranes.

Image series of a 2D membrane comprised of fluorescently labeled *fd-wt* (blue) and unlabeled *litmus* rods. *Litmus* droplets are stable and coalescence of droplets is never observed. Dextran concentration is 55 mg/mL, $n_{\text{mem}}=1$; scale bar is 5 μm .

Supplementary Movie 4, Absence of bilayer droplet coalescence.

A laterally phase-separated 2D membrane comprised of fluorescently labeled *fd-wt* (blue) and unlabeled *litmus* rods at $n_{\text{mem}}=2$. Increasing fraction of short rods (n_{mem}) increases the average droplet size. Even at these high droplet area fractions the bilayer droplets do not coalesce, indicating the existence of repulsive interactions between the droplets. Dextran concentration is 53 mg/mL; $n_{\text{mem}}=2$; scale bar is 5 μm .

Supplementary Movie 5: Crystallization of background monolayer membrane.

The monolayer phase in a phase-separated 2D membrane crystallizes on long time scales. Droplets are comprised of dimers of *litmus* rods. Once a critical nucleus forms, the crystal grows throughout the monolayer membrane over the course of tens of minutes. The membrane consists of

fluorescently labeled *fd-wt* monomers (blue) and unlabeled *Litmus* rods. The video was taken five days after sample preparation. Dextran concentration is 55 mg/mL; $n_{\text{mem}}=1$; scale bar is 5 μm .

Supplementary Movie 6, Liquid-solid coexistence of binary colloidal membranes. A time-lapse movie of a 2D membrane in which the background monolayer membrane has formed a 2D crystal. A low fraction of *litmus* rods (1:10,000) are labeled (bright blue) and are observed as bright points in the membrane. The dynamics of *litmus* rods reveals that the bilayer droplets coexisting with the solid background membranes remain liquid-like. The membrane consists of fluorescently labeled *fd-wt* monomers (blue) and unlabeled *Litmus* rods. Dextran concentration is 55 mg/mL; $n_{\text{mem}}=1$; scale bar is 5 μm .

Supplementary Movie 7, Colloidal membranes with spatially heterogeneous thickness. A colloidal membrane with spatially varying thicknesses consisting of fluorescently labeled *litmus* rods (yellow) and unlabeled *fd-wt*. The dark region corresponds to the thick membrane enriched in unlabeled *fd-wt* monomers, while light yellow region is formed of monomer *litmus* rods. The bright yellow represents the transition region where the membrane thickness changes from 400 to 880 nm and is populated by a litmus bilayer that twists as it crosses the interface. Dextran concentration is 55 mg/mL; $n_{\text{mem}}=4$; scale bar is 4 μm .

Supplementary Movie 8, Fluctuations of the interface between the bilayer droplets and the background membrane (*fd-wt*). A bulk phase-separated membrane that was used to analyze the fluctuations of the bilayer droplet interface. Fluorescently labeled (yellow) *Litmus* rods form a dimeric droplet in the background of monomeric *fd-wt* rods. The interface of *litmus* bilayer and *fd-wt* monolayer is not twisted and undergoes a thermal fluctuations. Dextran concentration is 50 mg/mL, $n_{\text{mem}}=2.5$, scale bar is 5 μm .

Supplementary Movie 9, Fluctuations of the interface between the bilayer droplets and the background membrane (*fd-Y21m*). A bulk phase separated-membrane with the ratio of *litmus* to *fdY21M* $n_{\text{mem}} = 2.5:1$. Fluorescently labeled (yellow) *litmus* rods assemble into dimeric structures and wet the membranes edge. The dark region of the membrane is enriched in *fd-Y21M* monomers. The interface between the *litmus* bilayer and *fd-Y21M* monolayer is not twisted and undergoes significant thermal fluctuations. Dextran concentration is 53 mg/mL, scale bar is 5 μm .