Membrane Species Mobility under In-Lipid-Membrane Forced Convection

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Supplementary Information

Estimation of the hydrophobic portion size and the extruding hydrophilic portions of the tested membrane species.

Table 1 in the main text shows the estimated shape and size of each tested species. β-BODIPY® FL C_5-HPC (BDP), and NBD C_{12}-HPC (NBD), and Texas-Red DHPE are all lipid molecules, which shape and size can be directly estimated by the chemical structure. Their hydrophobic portion size should be all similar to the one of a regular saturated phospholipid, which structure could be viewed as a cylinder with a height of 2 nm and a diameter of 0.94 nm[1]. BDP and NBD have the same hydrophilic head group as the rest of the phospholipids in the lipid membrane and therefore their extending hydrophilic portion sizes were set to be zero. Texas-Red DHPE has an extra Texas-Red fluorophore attached to the phosphatidylcholine head group. For the calculation simplicity, we assumed the Texas-Red fluorophore is a sphere with an equivalent diameter of 1.42 nm.

As for the streptavidin (SA) complex and cholera toxin (CT) complex, we estimated the shapes and sizes of SA and CT (as shown in Figure S1) by using information previously determined from X-ray crystal structures, atomic force microscopy, electron microscopy, and neutron reflectivity.[2-6] As for the hydrophobic portion of the complex, biotin-DHPE, biotin-X-DHPE and GM1 all have saturated lipid tails and therefore each of them was viewed as a cylinder with a height of 2 nm and a diameter of 0.94 nm, same as those of the BDP, NBD, and Texas-Red DHPE. When SA or CT binds to multiple biotinylated lipid or GM1, their hydrophobic portion size increase with the number of biotinylated lipid or GM1 accordingly.

Notably, when SA binds to biotin-DHPE and CT binds to GM1, these proteins can be in proximity to the upper lipid bilayer if the biotin-DHPE and GM1 remains in the upper lipid layer plane. The contact area was estimated by using the surface area facing the lipid membrane minus the cross section area occupied by biotin-DHPE or GM1. When SA binds to biotin-X-DHPE, which contains an extra acyl chain between the phosphoethanolamine and the biotin functional group, SA is not directly in proximity to the upper layer. We assumed that the extra distance could considerably reduce the possible friction force between SA and the upper lipid bilayer and therefore set the contact area for the friction as zero.

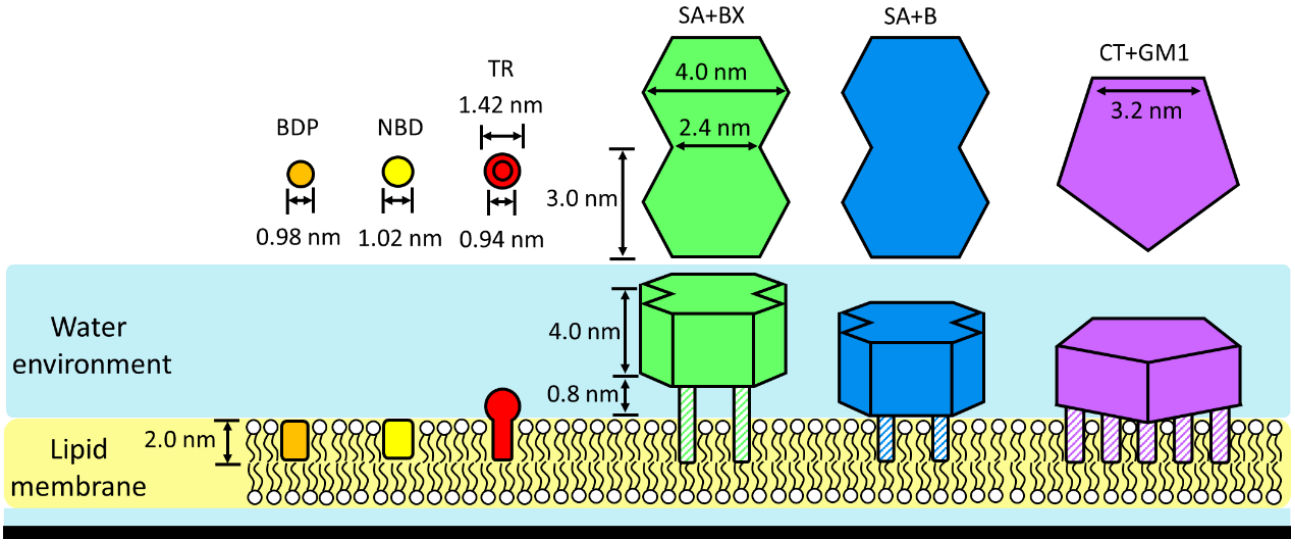

Figure S1. The detailed geometrical parameters used to calculate the extruding hydrophilic portion size and hydrophobic portion size of the tested membrane species in this study.

COMSOL simulation to obtain the hydrodynamic force.

To estimate the hydrodynamic force from bulk flow on the extruding hydrophilic portion of membrane species, we used COMSOL simulation software to calculate the flow field around the hydrophilic portion and then the overall shear force from the flow to the portion.

We viewed the extruding hydrophilic portion of membrane species as an unpenetrated solid. The velocity difference between the membrane species and the flow results in the hydrodynamic force. Since the concentration of membrane species is dilute (~1%), we assumed that the existence of each membrane species would not considerably influence the flow field around the other membrane species. As shown in the left column of Figure S2, we built box domains representing the region above the supported lipid bilayer, and the extruding portions of membrane species were set at the center of the bottom surface. We adjusted the box size to a size of 30*30*60 nm^3 so that the flow field around the species would not significantly influenced by the box boundaries. The input velocity distribution of the bulk flow inside the box was obtained by using the analytical solution of the flow velocity distribution in a rectangular microchannel[7], as shown in EQ S1.

$$v_f(y,z) \approx \frac{48Q}{h(w-0.630h)\pi^3}\sum_{n,odd}^{\infty}\frac{1}{n^3}\left[1-\frac{\cosh\left(\frac{n\pi y}{h}\right)}{\cosh\left(\frac{n\pi w}{2h}\right)}\right]\sin\left(\frac{n\pi z}{h}\right) \qquad \text{(EQ S1)}$$

where Q is the flow rate, h is the height, and w is the width of a rectangular micro-channel. The rectangular microchannel we used is 460 μm wide and 110 μm high.

The right column in Figure S2 shows the surface total stress (shear stress plus pressure) results for the extruding hydrophilic portion of TR, SA+BX, SA+B, and CT+GM1. The color towards to red color indicates smaller total stress and the color towards to blue color indicates larger total stress applied from the fluid to the hydrophilic portion. F_{hydro} for each membrane species was obtained by integrating the total stress over the entire hydrophilic portion surface area. Note that for NBD and BDP

which do not have extruding hydrophilic portion, we defined a circle area at the bottom surface, and obtained the friction from the bulk flow to that area on the bottom surface as F_{hydro}.

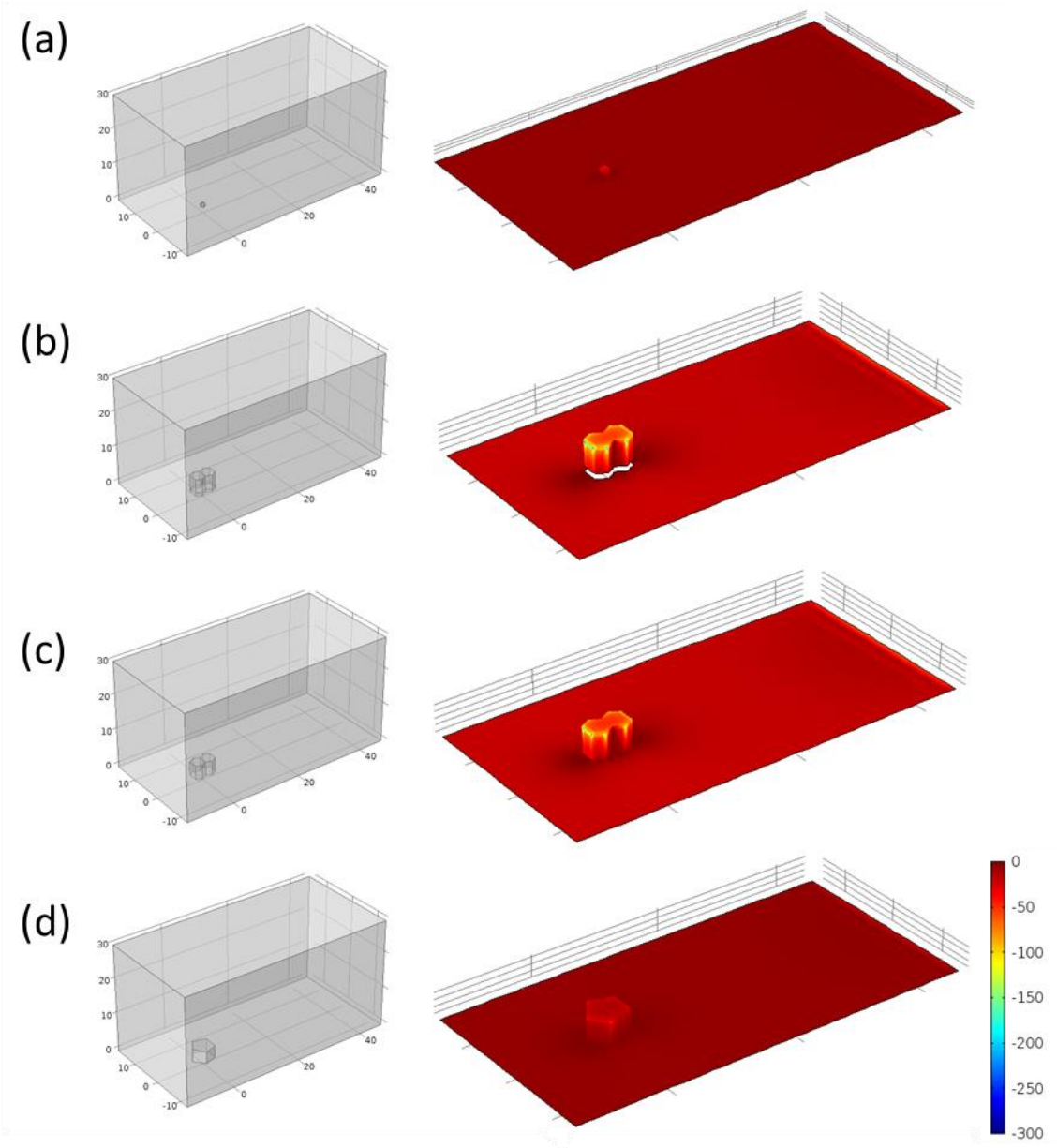

Figure S2. Left column: simulation domain of the region above the supported lipid bilayers in COMSOL software for obtaining F_{hydro}. Right column: the simulation result of the total stress distribution at the target membrane species and the SLB surface. (a) Texas red DHPE (TR), (b) SA+BX, (c) SA+B, and (f) CT+GM1.

Deriving quantitative drift velocities by using differential intensity velocity method.

We obtained the differential intensity profile for each tested membrane species from our experimental data. The differential intensity profiles were fitted to Gaussian peaks to identify the peak locations. Figure S3 shows the peak location versus the time plot for each membrane species, and the drift velocity was obtained using the slope. The differential intensity profiles of SA-biotin-DHPE

complex (SA+B) and SA-biotin-X-DHPE complex (SA+BX) samples contain two Gaussian peaks, each of which has its own velocity.

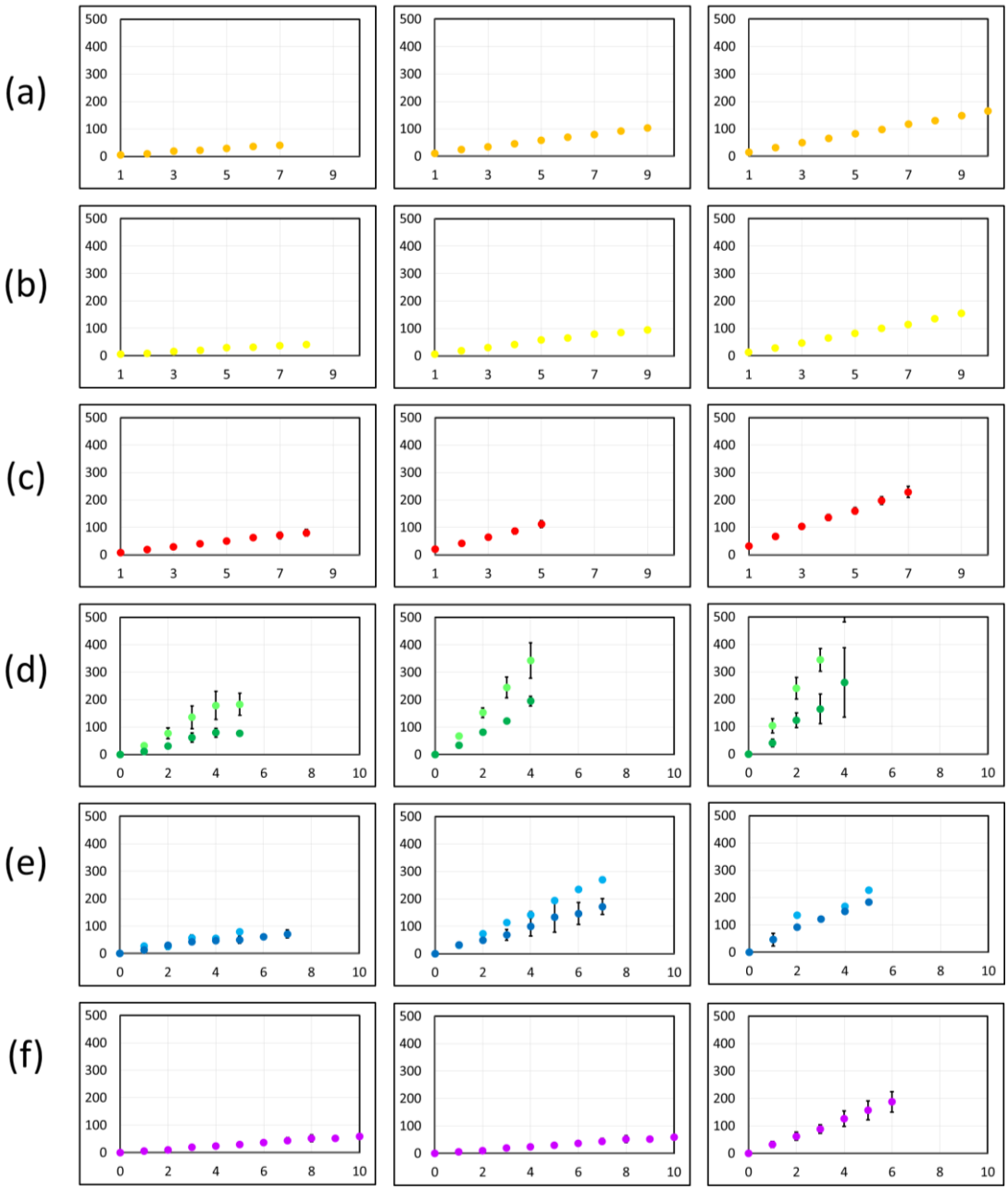

Figure S3. How the differential intensity profile peak location changes with time for each tested membrane species under the hydrodynamic flow with average flow velocity= 0.1, 0.2, and 0.3 m/s in a 460 μm wide and 110 μm high microchannel (the calculated surface shear stress τ_0= 6, 12, 18 pa). Data are represented as means with standard deviation (n = 3). The x-axis is time (min), y-axis is peak location away from the origin (μm), and the slope shows the drift velocity. (a) β-BODIPY® FL C_5-HPC (BDP), (b) NBD C_{12}-HPC (NBD), (c) Texas red DHPE (TR), (d) the complex of streptavidin and biotin-x DHPE (SA+BX), (e) the complex of streptavidin and biotin-DHPE (SA+B), and (f) the complex of cholera toxin subunit B and bovine brain ganglioside GM1(CT+GM1).

Estimation of the friction coefficient between the extruding hydrophilic portion of membrane species and top surface of the upper lipid leaflet, b_{ut}.

There is no much information about the friction coefficient between the extruding hydrophilic portion of membrane species and upper lipid leaflet when they are in contact. Therefore, we obtained the friction coefficient b_{ut} by examining which friction coefficient value can cause the minimum residual, which is the difference between the experimentally observed mobility and model-predicted mobility. In this study, we considered that SA+B and CT+GM1 complexes have large extruding hydrophilic portion in proximity to the upper lipid leaflet and calculated the sum of their residuals. Figure S4 shows how the summation of the residual changes with the guessed value of b_{ut}. The minimum residual occurred when b_{ut} was set to be $1.3 \times 10^8\ Pa\ s/m$.

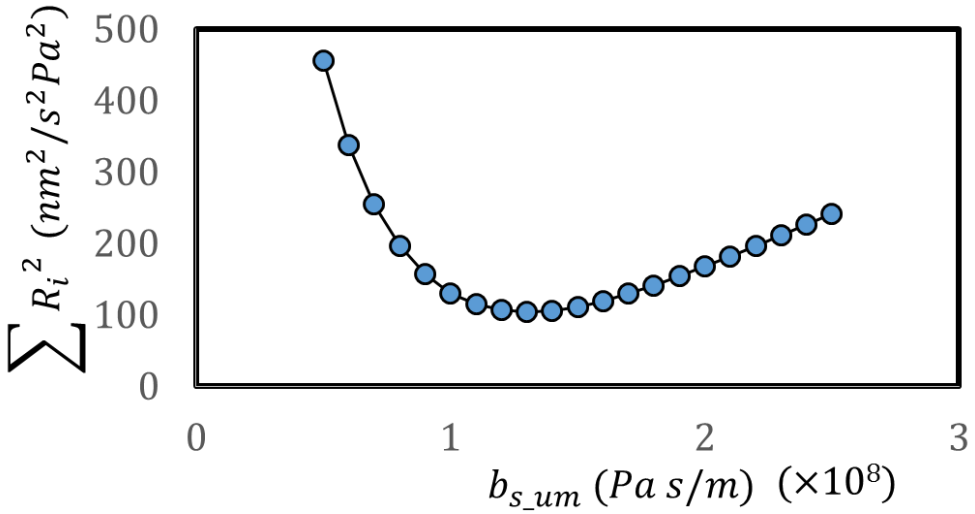

Figure S4. How the summation of the residual changes with the friction coefficient between the extruding hydrophilic portion of membrane species and top surface of the upper lipid leaflet.

Reference

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