

## Supporting information

# Physical Properties and Reactivity of Microdomains in Phosphatidylinositol-Containing Supported Lipid Bilayer

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### Diagonal illumination setup for single particle tracking

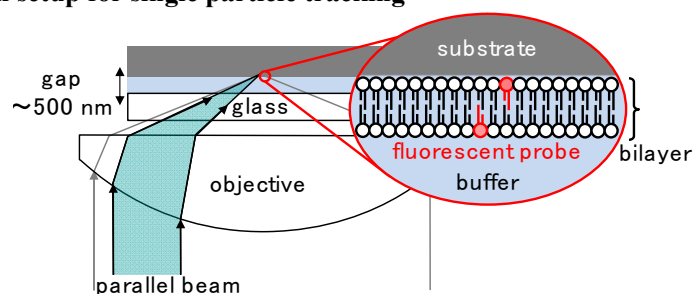


Figure S1. Schematic of the diagonal illumination setup.

### Single particle tracking results of PC-SLB without PI

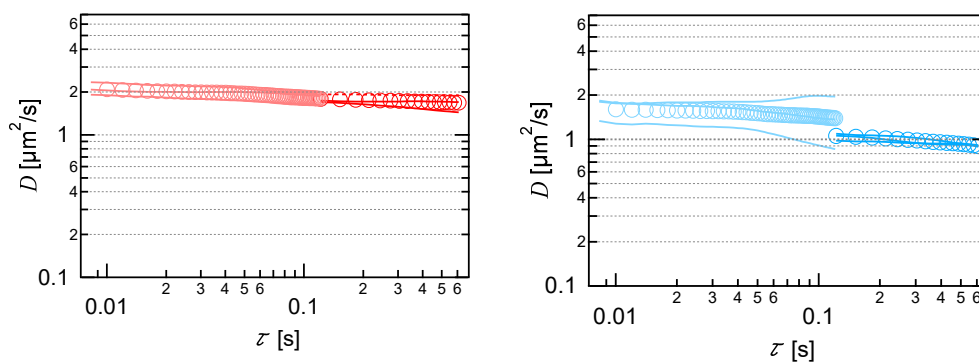


Figure S2. The dependence of  $D$  on  $\tau$  plots obtained from the analyses of the average MSD- $\tau$  plots of DiI-C18 (left) and BODIPY-HPG (right) in PC-SLB without PI.

### Analysis of AFM topography

The area of each depression domain was obtained from the AFM topographies with ImageJ, to calculate the average domain area ( $A_{\text{domain}}$ ) and the area fraction of the domain. The domain was detected according to the height threshold from the images without any filtering processes. We estimated the size of the compartment, which is the domain-free region for the microscopic lipid diffusion, as follows. We assume the circular domains with  $A_{\text{domain}}$  are allocated periodically (Figure S3). The area of the unit cell ( $A$ ) including a single domain is expressed as  $A = A_{\text{domain}} + A_{\text{surr}}$ , where  $A_{\text{surr}}$  is the area of the surrounding region around the domain.  $A_{\text{surr}}$  is calculated using the values of  $A_{\text{domain}}$  ( $1.69 \times 10^4 \text{ nm}^2$ ) and the area fraction of the domain ( $A_{\text{domain}}/A = 0.24$ ) that were obtained from the AFM topographies. We assume  $A_{\text{surr}}$  as the area of the domain-free compartment and regard its size as the approximated diameter  $2R = 2\sqrt{A_{\text{surr}}/\pi} = 261 \text{ (nm)}$ .

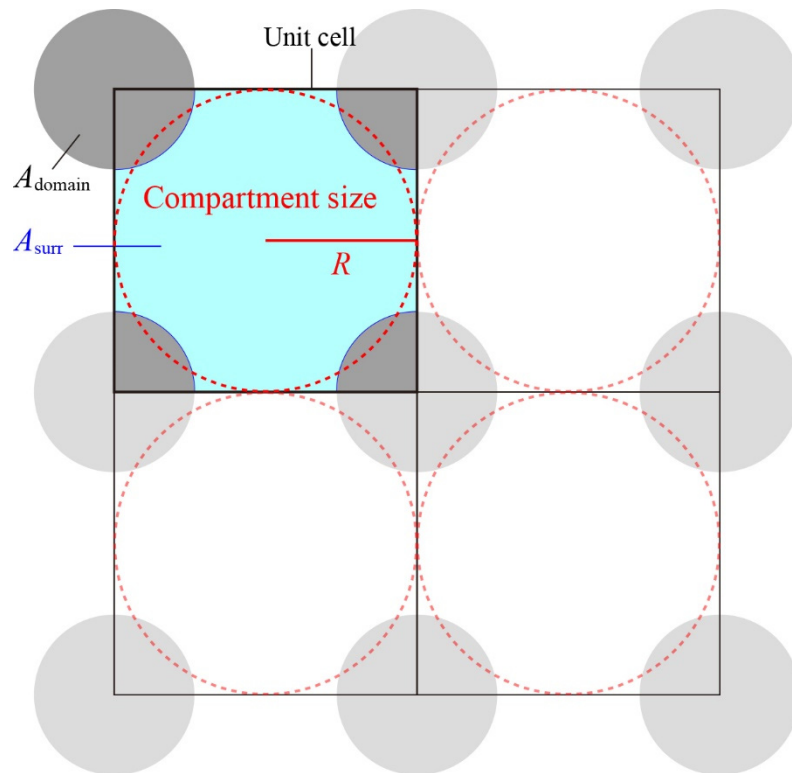


Figure S3. Schematic for estimating the domain-free compartment size.