## Characterization and regulation of 2D-3D convertible lipid membrane transformation

Wancheng Zhang,<sup>ab</sup> Yuta Uei,<sup>b</sup> Tomoaki Matsuura<sup>\*a</sup> and Atsushi Maruyama<sup>\*b</sup>

<sup>a</sup> Earth-Life Science Institute, Tokyo Institute of Technology, Ookayama 2-12-1, Meguro-Ku, Tokyo 152-8550, Japan

<sup>b</sup> School of Life Science and Technology, Tokyo Institute of Technology, B-57 4259 Nagatsuta-cho, Midori-ku, Yokohama, Kanagawa 226-8501, Japan

\*Correspondence:

Tomoaki Matsuura

Email: matsuura\_tomoaki@elsi.jp

Atsushi Maruyama

Email: amaruyama@bio.titech.ac.jp

	Molecular weight	PEG / wt%	SA / mol% (to PAA monomer)
	PAA / g mol <sup>-1</sup>		
PAA-g-PEG	5,000	89	-
PAA-g-PEG-SA	5,000	89	8

 Table S1. Composition of PAA-g-PEG and PAA-g-PEG-SA.

## E5 H - GLFEAIAEFIEGGWEGLIEG - OH E5-pal H – E5– OOC-( $CH_2$ )<sub>14</sub> $CH_3$



Figure S1. Amino acid sequences of E5 and E5-pal.

Figure S2. Chemical structures and 1H-NMR profiles of a) PAA-g-PEG and b) PAA-g-PEG-SA.



Figure S3. Confocal images of GUVs with different fluorescent compositions.



**Figure S4.** a) FSC and SSC histograms of standard beads measured by FCM. b) Relationship between FSC peak intensity of standard beads in the histograms and their diameters.



**Figure S5**. The compressed dots only constituted less than 0.5% of the population of the GUVs with FITC-dextran inside and rhodamine-labeled membranes.



**Figure S6.** FCM data of GUVs treated with E5-pal and different concentrations of PAA-*g*-PEG-SA. Plots of a) log FSC intensity versus log SSC intensity, b) log FSC intensity versus log rhodamine fluorescence intensity, and c) Q2 region of b) which were used for analysis. The Q2 region was considered containing the lipid membranes.



**Figure S7.** The compressed dots only constituted less than 10 % of the population of lipid membranes treated with different concentration of PAA-g-PEG-SA.



**Figure S8.** Confocal microscopic images and FCM profiles of GUVs treated with or without BSA or AF488-α-BSA. Scale bar: 10 μm.



**Figure S9.** The compressed dots only constituted less than 5% of the population of the BSA-modified GUVs treated with AF488-α-BSA.



**Figure S10.** FCM data of log FSC intensity versus rhodamine intensity for GUVs prepared with indicated mol% of DSPE-PEG-NHS functionalized with BSA. Data with or without E5/PAA-*g*-PEG treatment followed by PVS treatment after incubation with AF488-α-BSA are shown.



**Figure S11.** Confocal images of BSA-decorated GUVs with indicated mol% DSPE-PEG-NHS functionalized with BSA that were untreated (left), then treated with E5/PAA-*g*-PEG (middle), and then treated with PVS (right). Approximately 100% sheets were observed after treating with E5/PAA-*g*-PEG, and 100% reconversion of sheets to vesicles were found after subsequent treatment with PVS, indicating BSA decoration has little influence on sheet formation and conversion. Scale bar: 5 µm.