Electronic Supplementary Information (ESI)

Exploration of interactions between membrane proteins embedded in supported lipid bilayers and their antibodies by reflectometric interference spectroscopy-based sensing

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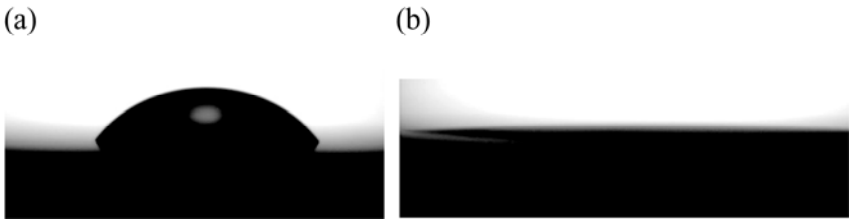
Contact Angle Measurements

The ZrO_2 surface was exposed to oxygen plasma under air at ca. 5 Pa, applying a current of 1-2 mA for 30 s (SEDE-GE, Meiwafosis Co., Ltd., Tokyo, Japan). Contact angles with water were subsequently determined using the FTA32 software package (First Ten Angstroms, Inc., VA, USA). During these measurements, the volume of a water droplet on the surface was adjusted to 1 μL and the static contact angle was measured at 0 (i.e., immediately), 1, 2, 3, 6 and 12 hours after plasma treatment. Each measurement was performed at least three times and the mean value was reported as the contact angle. The contact angles of the droplets shown in Figures S1a and S1b were 56.8° ± 0.8° and 4.4° ± 0.3°, respectively.



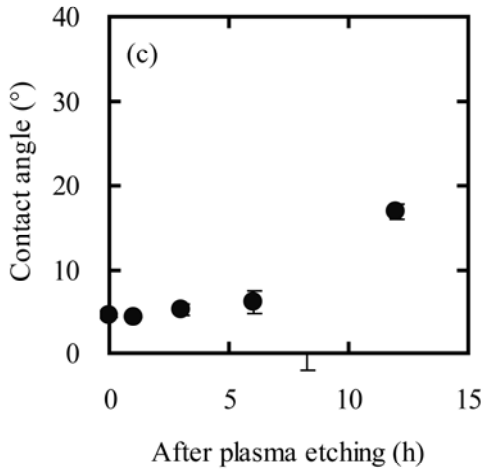


Fig. S1 Static contact angle water measurements on the ZrO_2 surface: (a) before plasma treatment, (b) immediately after plasma treatment and (c) a plot of contact angle as a function of time after plasma treatment.

Ellipsometry

The optical constants of silicon and ZrO_2 samples were measured using a V-VASE ellipsometer (J.A. Woollam Co., Inc., NE, USA). Ellipsometric measurements were carried out over 400-1000 nm at incident angles of 70°, 75° and 80° for silicon and 65°, 70° and 75° for ZrO_2. Si.mat and Cauchy models were applied to determine the experimental ellipsometric Ψ and Δ for silicon and ZrO_2, respectively.

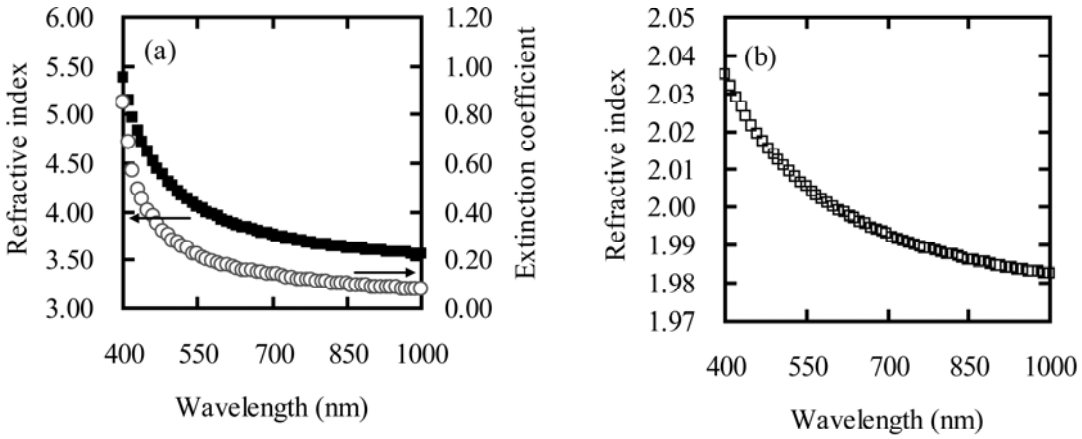


Fig. S2 Optical constants of (a) silicon (solid square: refractive index, open circle: extinction coefficient) and (b) ZrO_2, as measured by ellipsometry.

Dynamic Light Scattering (DLS)

Vesicle size distribution was determined using a Zetasizer Nano-ZS (Malvern Instruments Ltd., Worcestershire, UK) over the size range of 0.4 nm to 10 μm. Vesicle solutions were prepared by 1:100 dilution in 10 mM Tris-HCl buffer (pH 7.4, 2.7 mM KCl, 140 mM NaCl). Each sample (200 μL) was placed in a disposable plastic cuvette (SARSTEDT AG & Co., Nümbrecht, Germany) and measured at 25 ºC. The particle size distribution of each sample was determined from the average of 13 to 21 cycles.

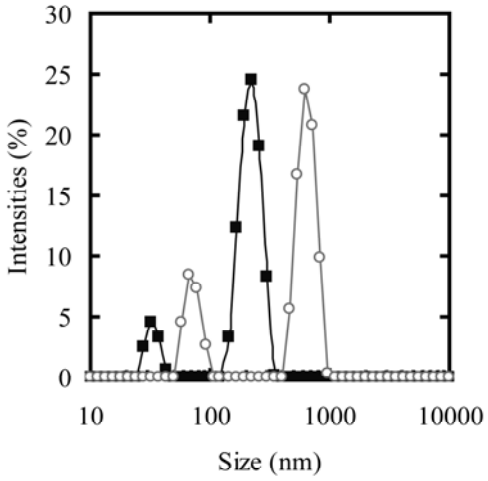


Fig. S3 Vesicle size distribution by DLS measurements. Solid squares: MRP1, open circles: control.

Transmission Electron Microscopy (TEM)

TEM to analyze the vesicle structures was carried out using a JEM-2000FX (JEOL Ltd., Tokyo, Japan) operating at 200 kV. The samples were incubated at 37 ºC for 5 min before observations (IN602, Yamato Scientific Co., Ltd., Tokyo, Japan). Samples were prepared by depositing an aliquot of the vesicle solution (5 mg/mL, 3 μL) onto a carbon-coated copper grid which had been hydrophilized by plasma etching (JP-170, JEOL Ltd.) for 60 s. Visualization of the vesicles was achieved by negative staining with a 2% phosphotungstic acid aqueous solution (pH 7.0).

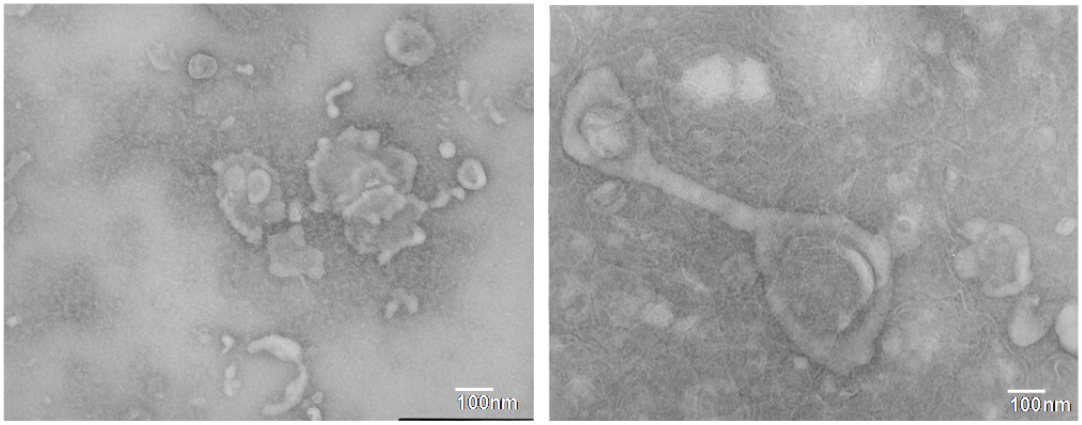


Fig. S4 TEM images of (a) MRP1 and (b) control vesicles.

Topology of MRP1

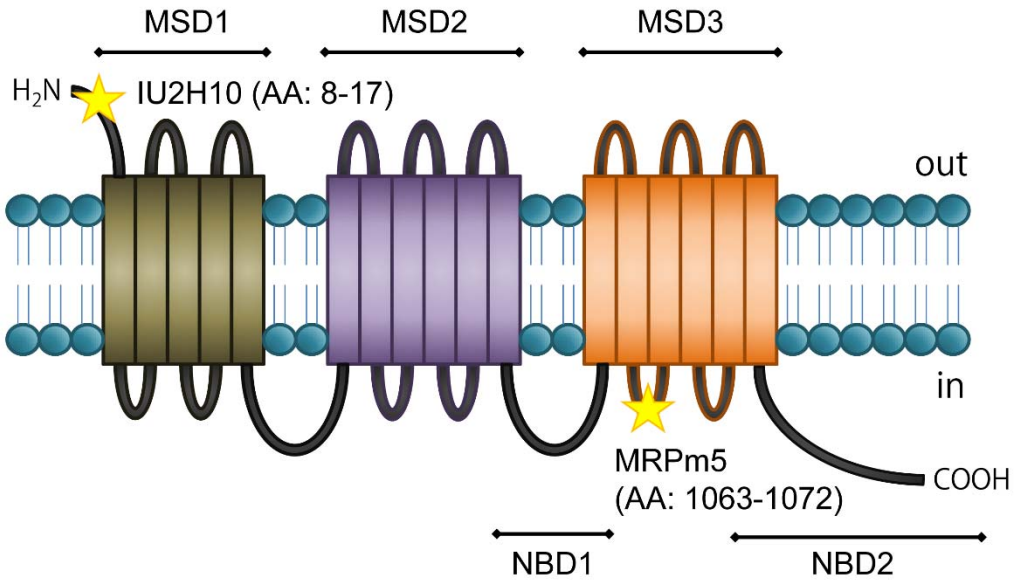


Fig. S5 Schematic of the MRP1 topological structure showing the sequence of the putative extracellular amino termini (AA: amino acids, MSD: membrane spanning domain, NBD: nucleotide binding domain).