

Supporting Information for

Atomic force microscopy study on stiffness of nanosized

liposomes containing charged lipids

Yuki Takechi-Haraya,¹ Yukihiro Goda,² and Kumiko Sakai-Kato^{1,*}

¹Division of Drugs, National Institute of Health Sciences, 3-25-26 Tonomachi, Kawasaki-ku, Kawasaki City, Kanagawa 210-9501, Japan, ²National Institute of Health Sciences, 3-25-26 Tonomachi, Kawasaki-ku, Kawasaki City, Kanagawa 210-9501, Japan

*To whom correspondence should be addressed:
kumikato@nihs.go.jp

Number of pages: 5

Number of Table: 1

Number of Figures: 3

Table S1. Heights and area-equivalent diameters of liposomes analyzed by AFM

Lipid composition	Height (nm) ^a	AD (nm) ^a
DOPC (100)	46 ± 3.6	84 ± 2.8
DOPC/DOTAP (50/50)	55 ± 1.0	90 ± 18
DOPC/DOPG (50/50)	46 ± 1.8	117 ± 7.9
DSPC/DSTAP (90/10)	66 ± 2.2	97 ± 4.6
DSPC/DSTAP (50/50)	45 ± 8.9	88 ± 17
DSPC/DSPG (50/50)	44 ± 14	84 ± 11

All values are means ± S.D of three independent samples. The values in brackets are %mol. DOPC, 1,2-dioleoyl-*sn*-glycero-3-phosphocholine; DOTAP, 1,2-dioleoyl-3-trimethylammoniumpropane; DOPG, 1,2-dioleoyl-*sn*-glycero-3-phospho-(1'-*rac*-glycerol); DSPC, 1,2-distearoyl-*sn*-glycero-3-phosphocholine; DSTAP, 1,2-distearoyl-3-trimethylammoniumpropane; DSPG, 1,2-distearoyl-*sn*-glycero-3-phospho-(1'-*rac*-glycerol). ^aRegarding each parameter, there were no significant differences between the liposome samples by Bonferroni's multiple comparison test.

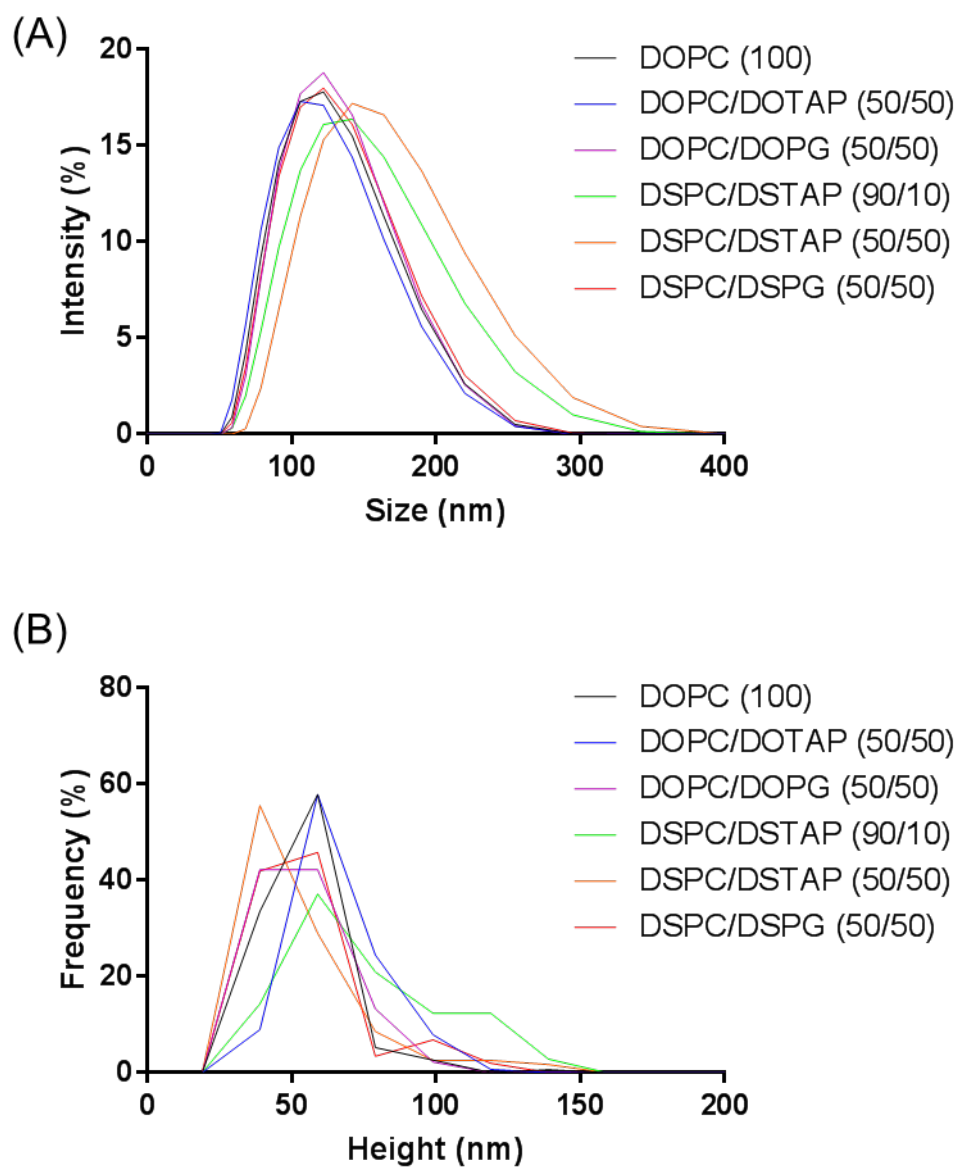


Figure S1. Size distribution of liposomes. (A) The light scattering intensity-weighted hydrodynamic diameter distribution of liposomes obtained by DLS. (B) The number-weighted height distribution of liposomes analyzed by AFM.

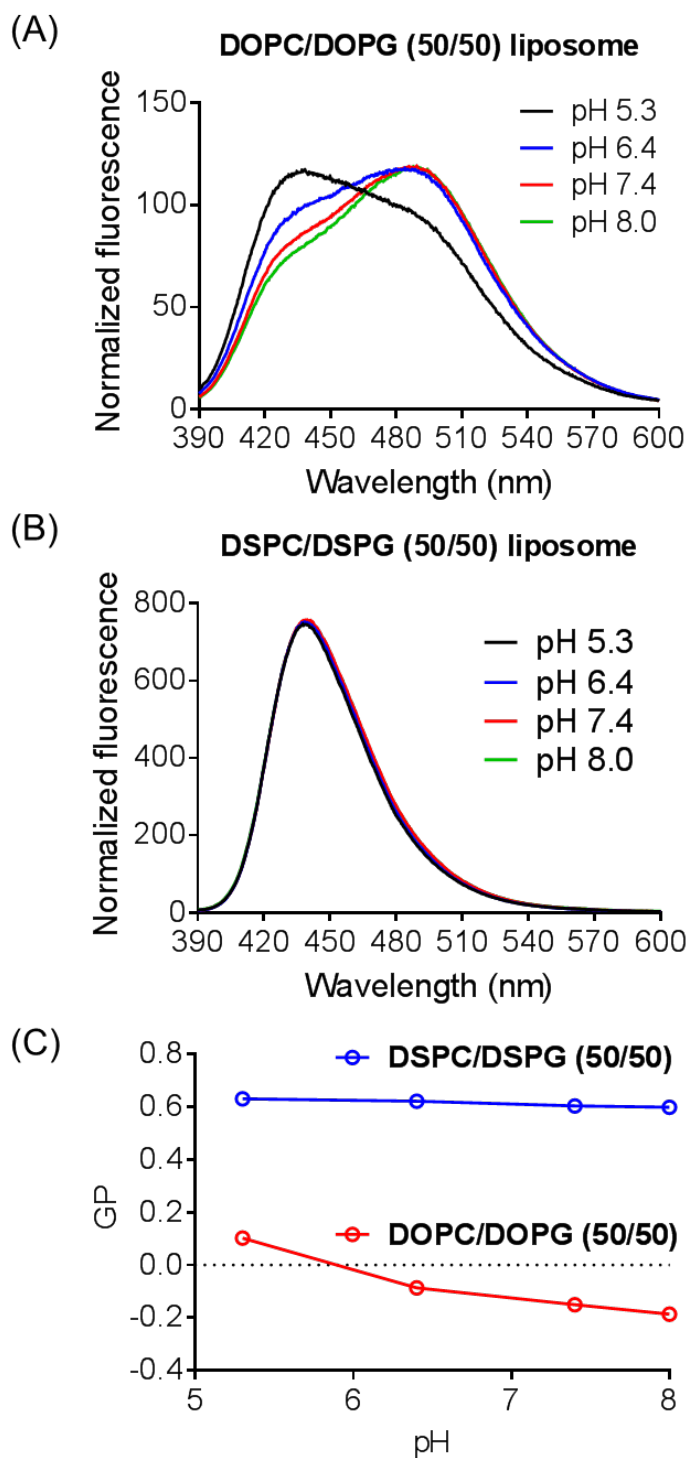


Figure S2. Fluorescence measurement of liposomes in 5% w/w glucose aqueous solution at various pH. (A) Fluorescence spectra of Laurdan in DOPC/DOPG (50/50) liposomes. (B) Fluorescence spectra of Laurdan in DSPC/DSPG (50/50) liposomes. (C) Plot of Laurdan GP values against pH values in DOPC/DOPG (50/50) and DSPC/DSTAP (50/50) liposomes.

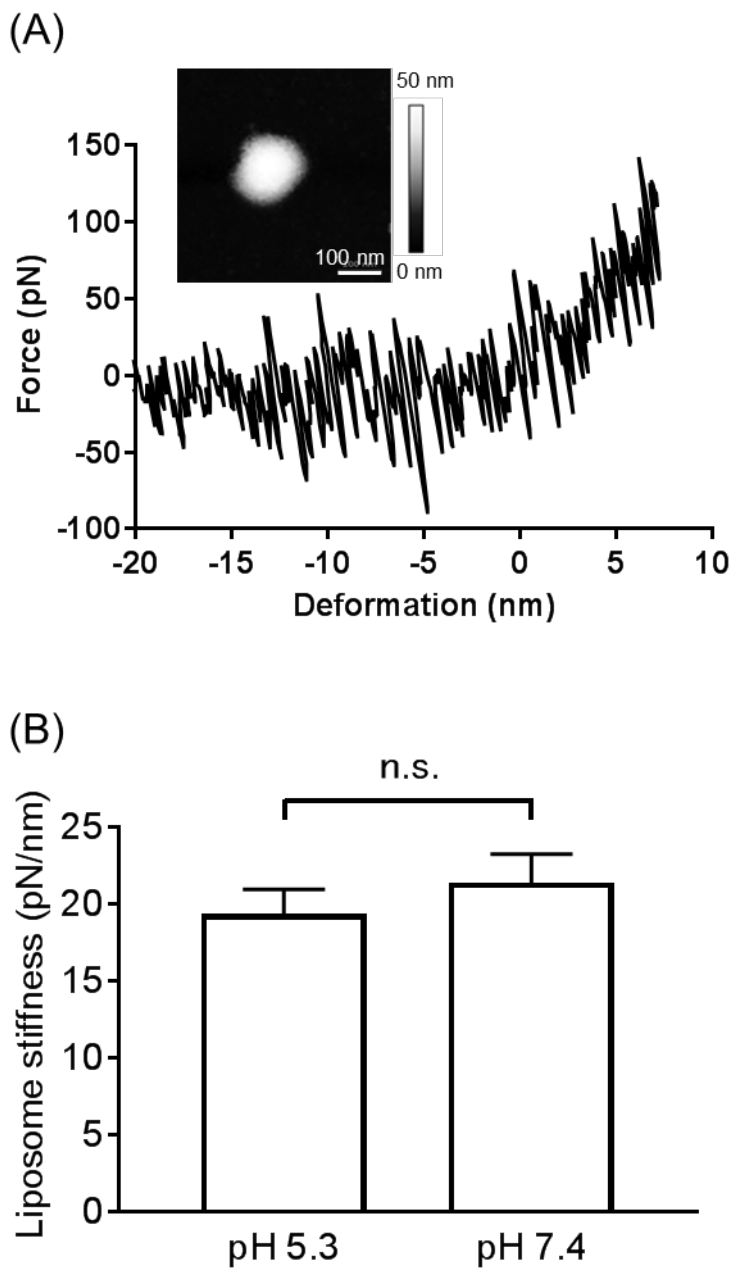


Figure S3. (A) AFM measurement of DOPC/DOPG (50/50) liposomes in 5% w/w glucose aqueous solution at pH 7.4. (B) Comparison of stiffness of DOPC/DOPG (50/50) liposomes in 5% w/w glucose aqueous solution at pH 5.3 and 7.4. n.s., not significant by *t*-test. The total number (*N*) of analyzed liposomes: *N* = 52 at pH 5.3, *N* = 36 at pH 7.4.