Electronic Supplementary Information (ESI)

Simple Sugars Shape Giant Vesicles into Multispheres with Many Membrane Necks

Tripta Bhatia, Simon Christ, Jan Steinkühler, Rumiana Dimova and Reinhard Lipowsky*

Theory and Bio-Systems, Max Planck Institute of Colloids and Interfaces, Science Park Golm, 14424 Potsdam, Germany

Supplementary Tables corresponding to Figs. 7 and 9

Table S1: Parameter values for sugar asymmetry I. The first column displays the GUV index number used in Fig. 7, the second column the number of large and small spheres, $N_l + N_s$. The subsequent columns provide the radii R_l and R_s of the large and small spheres, the neck curvatures M_{ls} and M_{ss} of the ls- and ss-necks as given by Eq 7, the parameter $B_{\rm up}$ defined in Eq 9, and the volume-to-area ratio v in Eq 3

#	shape	R_l [μm]	R_s [μm]	M_{ls} [μm^{-1}]	M_{ss} [μm^{-1}]	$B_{\rm up}$ [μm^{-1}]	v
1	$(1+1)$	11.40 ± 0.25	3.12 ± 0.25	0.20 ± 0.01	–	1.32 ± 0.11	0.92
2	$(1+1)$	3.39 ± 0.25	1.5 ± 0.25	0.47 ± 0.06	–	3.56 ± 0.78	0.82
3	$(1+1)$	2.66 ± 0.25	1.64 ± 0.25	0.49 ± 0.06	–	4.76 ± 1.55	0.76
4	$(1+7)$	10.17 ± 0.25	2.24 ± 0.25	0.27 ± 0.03	0.44 ± 0.05	1.72 ± 0.19	0.69
5	$(1+2)$	8.90 ± 0.25	1.92 ± 0.25	0.31 ± 0.03	0.52 ± 0.06	1.98 ± 0.27	0.89
6	$(2+2)$	2.35 ± 0.25	0.94 ± 0.25	0.74 ± 0.16	1.06 ± 0.28	5.31 ± 1.73	0.60
7	$(1+3)$	2.65 ± 0.25	0.72 ± 0.25	0.88 ± 0.26	1.38 ± 0.48	5.70 ± 2.12	0.78
8	$(1+3)$	3.00 ± 0.25	0.71 ± 0.25	0.86 ± 0.26	1.4 ± 0.5	5.51 ± 2.02	0.82

Table S2: Parameter values for sugar asymmetry II. The first column displays the GUV index number used in Fig. 9, the second column the number of large and small spheres, $N_l + N_s$, and the subsequent columns the same quantities as in Table S1.

#	shape	R_l [μm]	R_s [μm]	M_{ls} [μm^{-1}]	M_{ss} [μm^{-1}]	B_{up} [μm^{-1}]	v
1	$(1+1)$	3.48 ± 0.25	1.11 ± 0.25	0.59 ± 0.11	–	3.96 ± 0.99	0.89
2	$(1+1)$	6.14 ± 0.25	1.62 ± 0.25	0.39 ± 0.05	–	2.51 ± 0.41	0.92
3	$(1+1)$	25.0 ± 0.25	2.75 ± 0.25	0.2 ± 0.01	–	1.22 ± 0.11	0.98
4	$(1+1)$	13.57 ± 0.25	2.51 ± 0.25	0.23 ± 0.02	–	1.46 ± 0.15	0.96
5	$(1+2)$	3.01 ± 0.25	0.71 ± 0.25	0.86 ± 0.26	1.4 ± 0.49	5.50 ± 2.02	0.87
6	$(1+2)$	6.87 ± 0.25	1.17 ± 0.25	0.5 ± 0.09	0.85 ± 0.18	3.08 ± 0.67	0.93
7	$(1+2)$	6.95 ± 0.25	0.98 ± 0.25	0.58 ± 0.13	1.01 ± 0.26	3.55 ± 0.91	0.95
8	$(1+2)$	6.85 ± 0.25	0.97 ± 0.25	0.59 ± 0.13	1.1 ± 0.2	3.60 ± 0.94	0.95
9	$(1+2)$	6.45 ± 0.25	0.96 ± 0.25	0.59 ± 0.14	1.1 ± 0.2	3.67 ± 0.97	0.94
10	$(1+3)$	5.46 ± 0.25	2.57 ± 0.25	0.28 ± 0.02	1.1 ± 0.2	2.20 ± 0.29	0.61
11	$(1+3)$	3.18 ± 0.25	0.92 ± 0.25	0.69 ± 0.16	1.08 ± 0.29	4.56 ± 1.33	0.76
12	$(1+3)$	5.92 ± 0.25	1.40 ± 0.25	0.44 ± 0.07	0.71 ± 0.13	2.80 ± 0.52	0.82
13	$(1+4)$	4.43 ± 0.25	1.02 ± 0.25	0.6 ± 0.1	0.97 ± 0.24	3.81 ± 0.97	0.78
14	$(2+2)$	2.98 ± 0.25	2.19 ± 0.25	0.39 ± 0.04	–	5.18 ± 2.12	0.51
15	$(2+2)$	1.66 ± 0.25	0.80 ± 0.25	0.92 ± 0.24	1.24 ± 0.38	7.21 ± 3.23	0.57

GUV morphologies for reversed sugar asymmetries

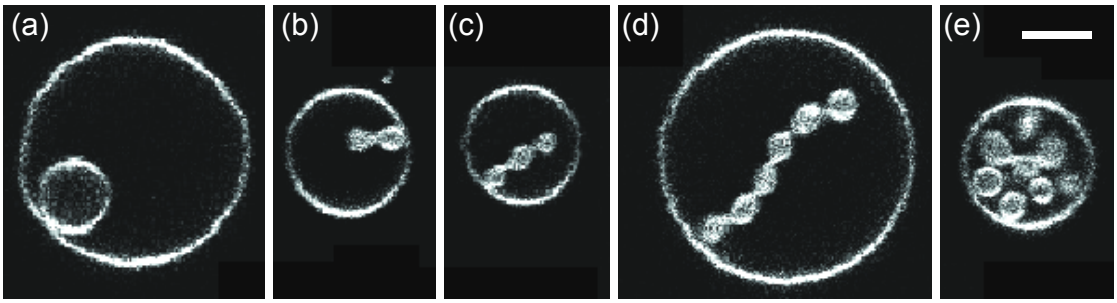

Figure S1: In the main text, we studied GUVs for which the inner leaflets were exposed to sucrose only and the outer leaflets were in contact with a glucose-sucrose solution. When we reverse the sugar asymmetry by preparing the GUVs in glucose and exposing them to an exterior glucose-sucrose solution in the observation chamber, we observe a variety of multispherical shapes with inward-pointing buds and chains of small spheres, as displayed here: (a) GUV with single in-bud; (b-e) GUVs with in-tubes consisting of chains with (b) two, (c) three, and (d) six small spheres; and (e) GUV filled with many small spheres. All of these shapes imply that a reverse sugar asymmetry across the GUV membranes does indeed generate a negative spontaneous curvature as predicted by the theory of curvature elasticity. The scale bar in (e) is 5 μm and applies to all panels.

Movie Captions

Movie1. Time-dependent shape evolution of the GUV displayed in Fig. 2. The GUV was prepared in 172 mM sucrose and then transferred into an observation chamber filled with 200 mM glucose. The movie is composed of two frame sets. The first set consists of frames 1 to 10 and displays about 23 s, see time stamps. The second set consists of frames 11 to 21 and covers about 26 s. The first image in Fig. 2, labeled by time $t = 0$ in this figure, corresponds to frame 11 in the movie, with a time stamp of about 60 s, the last image in Fig. 2 to frame 20 in the movie. At frame 11, the resolution is changed and the image is zoomed out. The scale bar during the first 10 frames is $5\,\mu$m.

Movie 2. Shape evolution of a GUV shape that consists of one large and five small spheres, with four small spheres forming a chain. The GUV was prepared in 172 mM sucrose and then transferred into an observation chamber filled with 200 mM glucose. The first frame of the movie corresponds to 30 mins after adding the aliquot of sucrose-GUV solution to the observation chamber. During the first 12 s of the movie, the chain is linear but then transforms into a branched chain, which is clearly visible after 69 s, thereby illustrating the mobility of the small spheres even for number of large and small spheres. Scale bar is 5 μm.

Movie 3. Additional optical images of the GUV in Fig. 5(a). The movie displays a time series of 30 such images. Close inspection of these images confirms that the multisphere in Figure 5a consists of $N_* = 14$ equally sized spheres that form a branched chain. The images also illustrate the practical difficulty to determine the precise value of N_* because the chain undergoes thermal fluctuations that move the individual spheres in and out of the optical focus. A chain of 14 equally sized spheres has the volume-to-area ratio $v = 1/\sqrt{14} = 0.267$, see Eq 5 in the main text, irrespective of the sphere radius R_*.

Movie 4. Additional optical images of the GUV in Fig. 5(c). The movie displays a time series of 130 such images. Close inspection of these images confirms that the multisphere in Figure 5c consists of $N_* = 15$ equally sizes spheres that form a branched chain. The images illustrate the practical difficulty to determine the precise value of N_* because the chain undergoes thermal fluctuations that move the individual spheres in and out of the optical focus. A chain of 15 equally sized spheres has the volume-to-area ratio $v = 1/\sqrt{15} = 0.258$, see Eq 5 in the main text, irrespective of the sphere radius R_*.

Movie 5. During the first 58 s, this movie displays a GUV shape consisting of one large sphere and a necklace-like tube that resembles a capped unduloid with four bellies and three open necks. After about 79 s, the unduloid starts to undergo peristaltic fluctuations that lead to a linear chain of $N_s = 5$ small spheres. The GUV was prepared

in 172 mM sucrose and then transferred into an observation chamber filled with 200 mM glucose. The initial time $t = 0$ s (frame number 1) in the movie corresponds to 30 min after adding the aliquot to the observation chamber. The image is zoomed in and out for frame numbers 94 (after 75.2 s) and 138 (after 107.6 s), respectively. Initial scale bar is 5 μm.

Movie 6. The first frame of Movie6 displays a shape that resembles a thick letter 'C' which then develops two constrictions and three bellies as well as a spherical bud which is connected to the belly in the middle. After about 30 s, the bud starts to move towards the upper constriction between the belly in the middle and the upper belly. After about 50 s, these two large bellies are clearly separated which implies that the small sphere has moved in between the two bellies. After about 58 s, the large belly in the middle starts to develop yet another constriction which transforms this belly into a two-sphere shape with a large and a small sphere. After about 78 s, the GUV has relaxed into a multispherical shape consisting of three large and two small spheres, arranged as an linear $(lslsl)$-necklace connected by four ls-necks. The GUV was prepared in 186 mM sucrose and then transferred into the observation chamber filled with 427 mM glucose. The first frame at time $t = 0$ corresponds to 30 min after adding the aliquot to the observation chamber. Scale bar is 20 μm.

Movie 7. The initial frame of Movie7 displays a shape that resembles a three-armed starfish. which transforms into a (3+1)-sphere, for which the three large spheres are all connected to the small sphere in the middle, resembling a three-leaved trefoil. The latter process leads to the formation of three ls-necks and takes about 60 s. The GUV was prepared in 186 mM sucrose and then transferred into the observation chamber filled with 427 mM glucose. The first frame at time $t = 0$ corresponds to 30 min after adding the aliquot to the observation chamber. The scale bar is 20 μm.