

## Morphology of Vesicle Triplets: Shape Transformation at Weak and Strong Adhesion Limits

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Supporting Information:

### S1: Comparison between area-fixed model and two-tension model

To demonstrate the validity of the two-tension model, we show the simulation results of the area-fixed model which consists of the membrane bending energy and adhesion energy with constant membrane area and constant vesicle volume constraints. Based on the area-fixed model, the total energy of vesicle triplet is expressed by,

$$w = \sum_i w_{b,i} - \sum_i \sum_j^{ad} \gamma a_{ij}^c \quad (\text{S1})$$

where  $w_{b,i}$  is the normalized bending energy,  $\gamma$  is the normalized adhesion strength, and  $a_{ij}^c$  is the normalized contact area (see section 2.1 in the manuscript). Under the constraints of constant surface area and enclosed volume, the total energy is numerically minimized by Surface Evolver.

Figure S1a shows shape transformation of vesicle triplet with triangular topology as a function of the mean reduced volume  $\bar{v}$ , which are reproduced by numerical simulations based on the two-tension model and cavity model (as is for Fig. 1c). The two-tension model reproduced the observed triplet morphologies successfully as shown in Fig. S1a (Figs. 1a and c). On the other hand, triplet morphologies calculated by the area-fixed model failed to capture the observed shapes. In the triangle topology, area-fixed model numerically predicts the oblate-vesicle triplets at large reduced volumes and non-spherical sigmoidal-contact triplets at small reduced volumes (Fig. S1b), which are not consistent with the spherical-cap shapes observed experimentally (Fig. 1a). In the linear topology, the morphologies based on the two-tension model and cavity model again reproduced experimental observations well as shown in Fig. S2a (Figs. 2a and b). Numerical predictions based on the area-fixed model departed from the experimental observations and deformed to oblate-based triplet shapes (Fig. S2b). In the end, the area-fixed model cannot reproduce the observed morphologies of vesicle triplets.

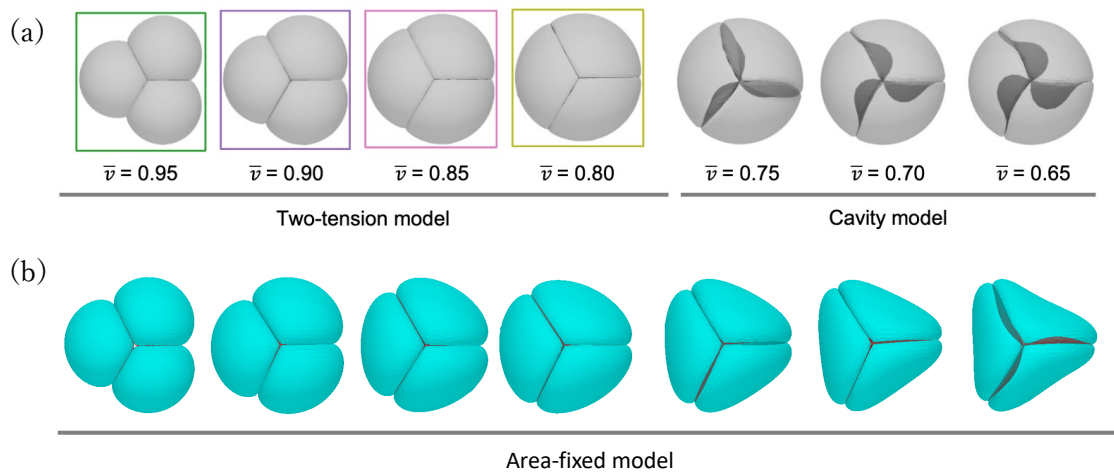


Fig. S1 Shape transformation of vesicle triplet with triangular topology as a function of the mean reduced volume  $\bar{v}$ , which are reproduced by numerical simulations using Surface Evolver. (a) Based on Two-tension and Cavity model (as is for Fig. 1c). (b) Based on area-fixed model where surface area is fixed. The adhesions strength is set as  $\gamma = 20$ .

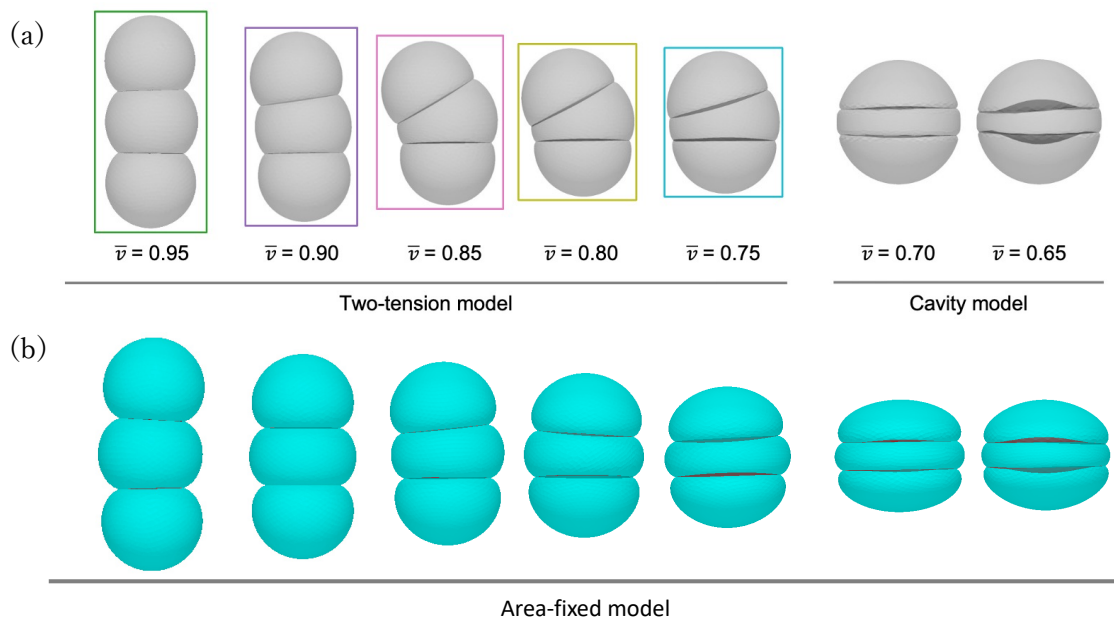


Fig. S2 Shape transformation of vesicle triplet with linear topology as a function of the mean reduced volume  $\bar{v}$ , which are reproduced by numerical simulations using Surface Evolver. (a) Based on Two-tension and Cavity model (as is for Fig. 2b). (b) Based on area-fixed model where surface area is fixed. The adhesions strength is set as  $\gamma = 20$ .

Movie S1:

Deformation of a DMPC vesicle triplet with triangular topology in KCl 200 mM solution during heating. The temperature started from 25°C and the heating rate was 1°C/min. The temperature is displayed at the upper left of the movie. Scale bar is 20  $\mu\text{m}$ .

Movies S2:

Deformation of a DMPC vesicle triplet with linear topology in KCl 200 mM solution during heating. The temperature started from 25°C and the heating rate was 1°C/min. The temperature is displayed at the upper left of the movie. Scale bar is 20  $\mu\text{m}$ .

Movie S3:

Deformation of a DMPC vesicle triplet with triangular topology in KCl 100 mM solution during heating. The temperature started from 25°C and the heating rate was 1°C/min. The temperature is displayed at the upper left of the movie. Scale bar is 20  $\mu\text{m}$ .

Movie S4:

Deformation of a DMPC vesicle triplet with linear topology in KCl 100 mM solution during heating. The temperature started from 25°C and the heating rate was 1°C/min. The temperature is displayed at the upper left of the movie. Scale bar is 20  $\mu\text{m}$ .