

Supporting Information for Role of polyplex charge density in lipopolyplex

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Table S1 | The bond and angle parameters involving CO and COO beads

	Length (nm)	Force constant (kJ/(mol nm ²))
Bond (COO-CO)	0.425	1250.0
Bond (CO-SP1)	0.425	1250.0
	Angle (degree)	Force constant (kJ/(mol rad ²))
Angle (COO-CO-SP1)	180.0	25.0

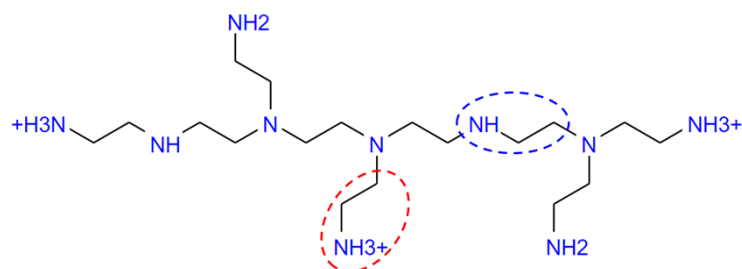


Figure S1 | Martini coarse-grained mapping for PEI. The red and blue ellipses contain the positively charged and neutral repeat unit of PEI (i.e., C-C-N group), which were assigned to Martini bead type Qd and P2, respectively¹.

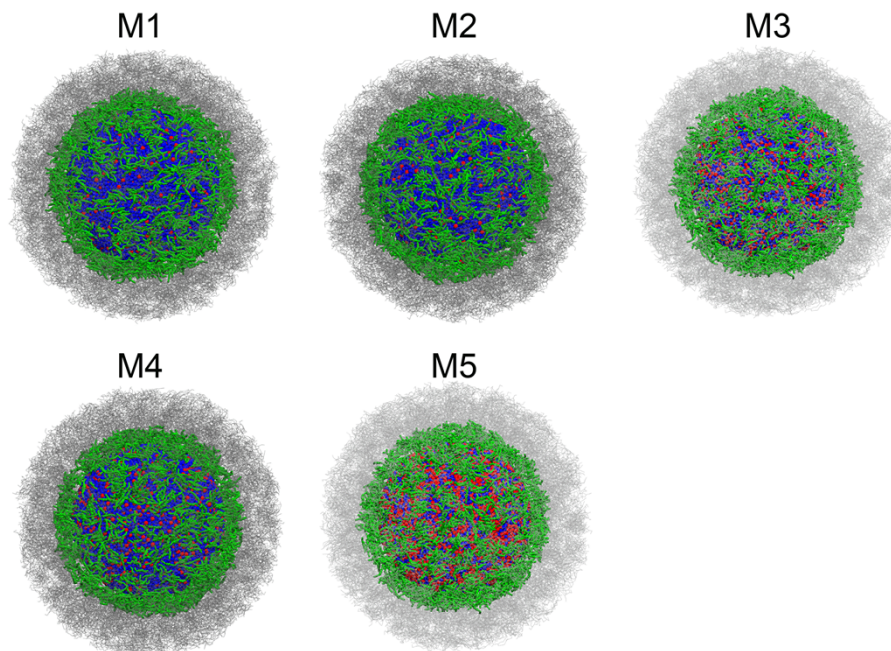


Figure S2 | Snapshots of initial configurations of the five (M1 to M5) systems. The DOPE and CHEM are shown in gray lines and green Licorice representations, respectively. Packmol² was employed to insert the CHEMS molecules within 3 nm from the polyplex surface. Subsequently, DOPE molecules were inserted within 5.5 nm from the polyplex surface.

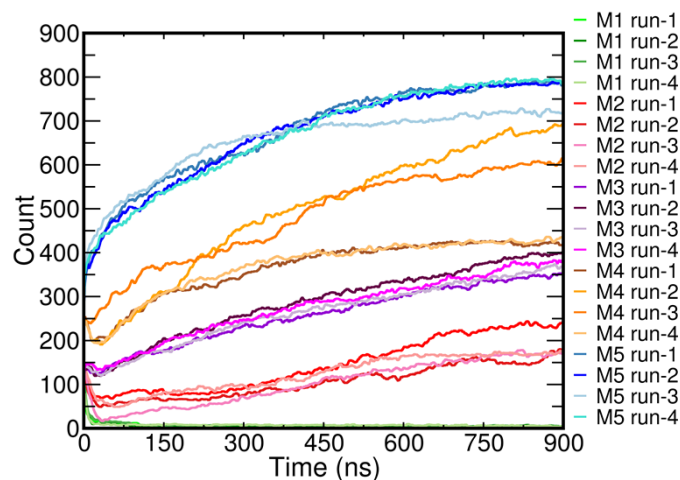


Figure S3 | Time evolution of CHEMS's COO beads number within 0.7 nm. The cutoff distance of 0.7 nm corresponds to the first hydration layer.

Discussion of Figure S3: In Fig. S3, one can observe that, initially, the number of contacts declines in all five systems, corresponding to the initial relaxing of liposome lipids. In the M1 polyplex, the contact numbers remain stable from ~ 25 ns to the end of the simulation, whereas the contacts increase after the initial relaxing for M2 to M5 systems, respectively. For example, the number of contacts increases dramatically for the M4 polyplex as compared to the M3 polyplex. Notably, two repeated simulations of M4 system (run-3 and run-4) show quite slow self-assembly kinetics. The slow self-assembly kinetics in the repeated simulations of M4 system would not change the main results and conclusions of this study. For consistency, results presented in the manuscript were calculated based on the representative run-1 of each system.

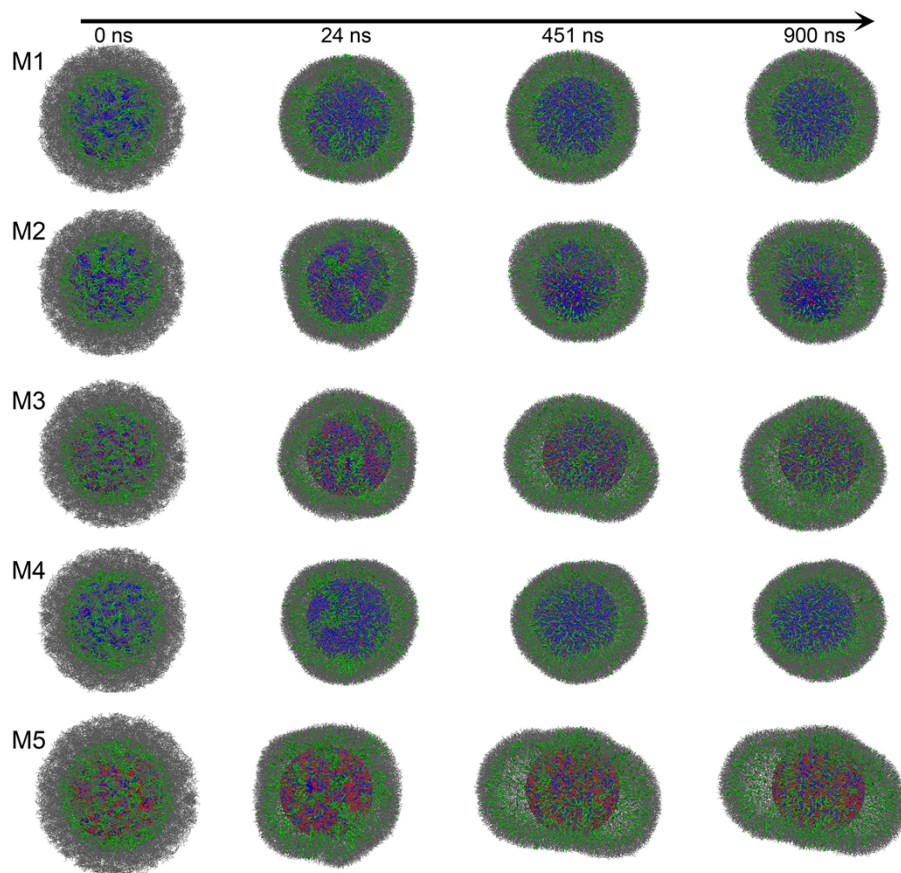


Figure S4 | Representative structures during the self-assembly of liposomes (consisting of DOPE and CHEMS lipids) on the polyplex with different charge densities. The DOPE and CHEM are shown in green lines and green Licorice representations, respectively.

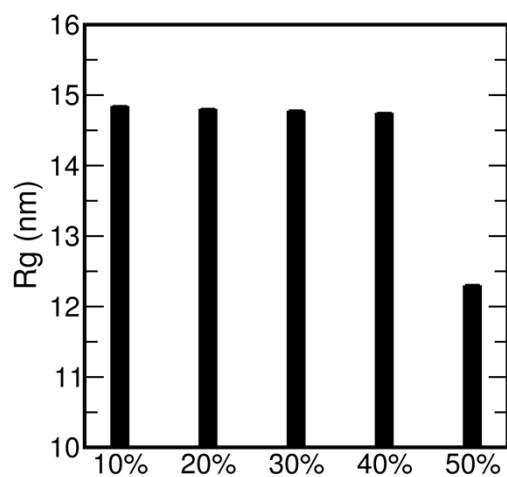


Figure S5 | Radius of gyrations of the five self-assembled liposomes, with higher charge density ones showing smaller radius of gyration. Error bar denotes the standard deviation.

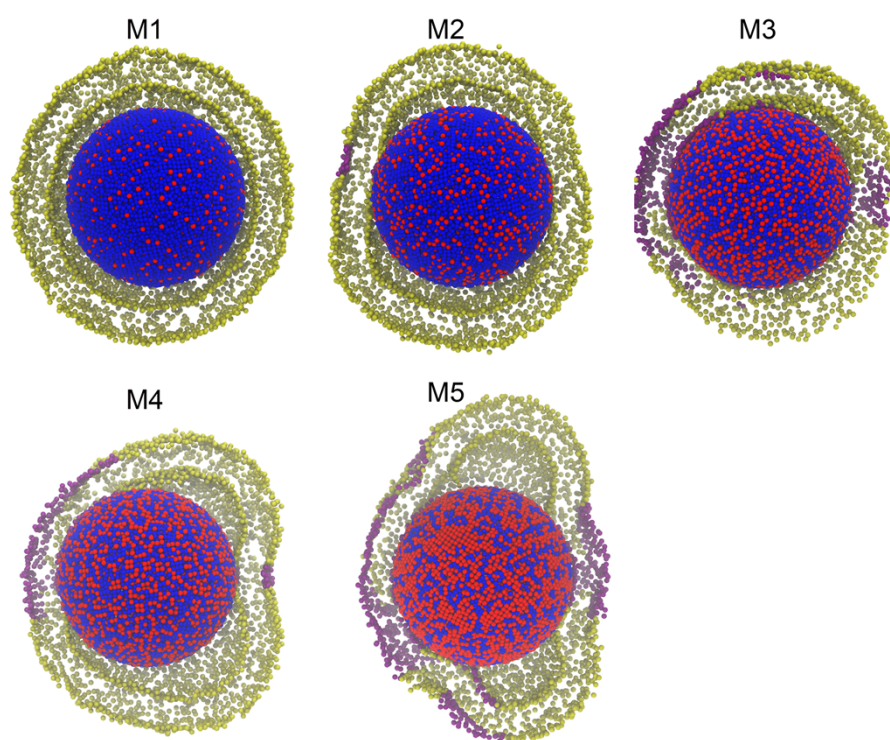


Figure S6 | Local membrane thickness for the five simulated systems. The local membrane thickness is defined as the radial distance between opposing head groups of the two leaflets (i.e., the head bead (NH₃) of DOPE). In order to estimate the local membrane thickness, the liposome membrane was divided into patches by using spherical coordinates of polar angle θ and azimuthal angle φ . The θ and φ ranges were chosen to be [0, 30, 60, 90, 120, 150, 180, 210, 240, 270, 300, 330, 360] and [0, 20, 45, 65, 80, 90, 100, 115, 135, 160, 180], respectively. The head bead is shown either in yellow where the local membrane thickness is greater or equal to 3.7 nm or in purple where the local membrane thickness is less than 3.7 nm.

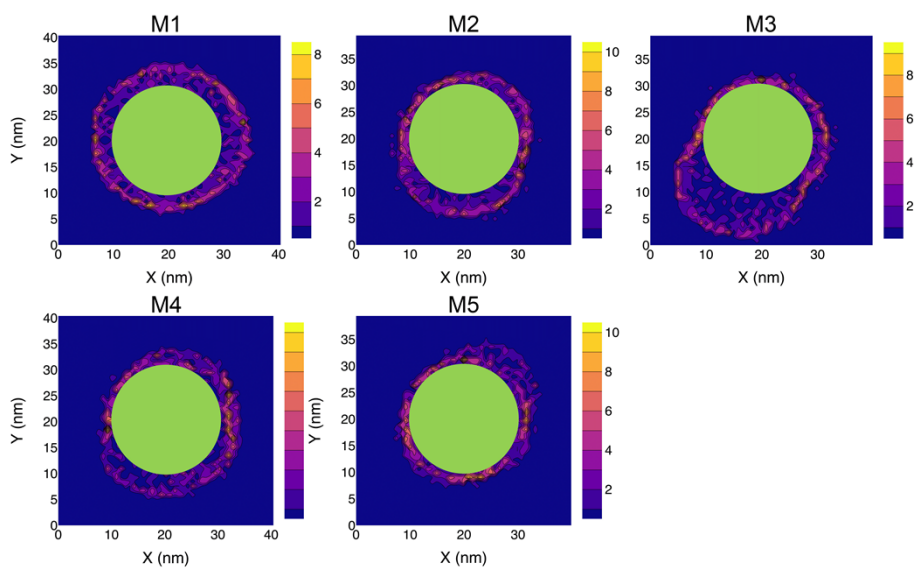


Figure S7 | Two dimensional heatmaps of CHEMS lipid distributions. The regions of polyplexes are shown in green.

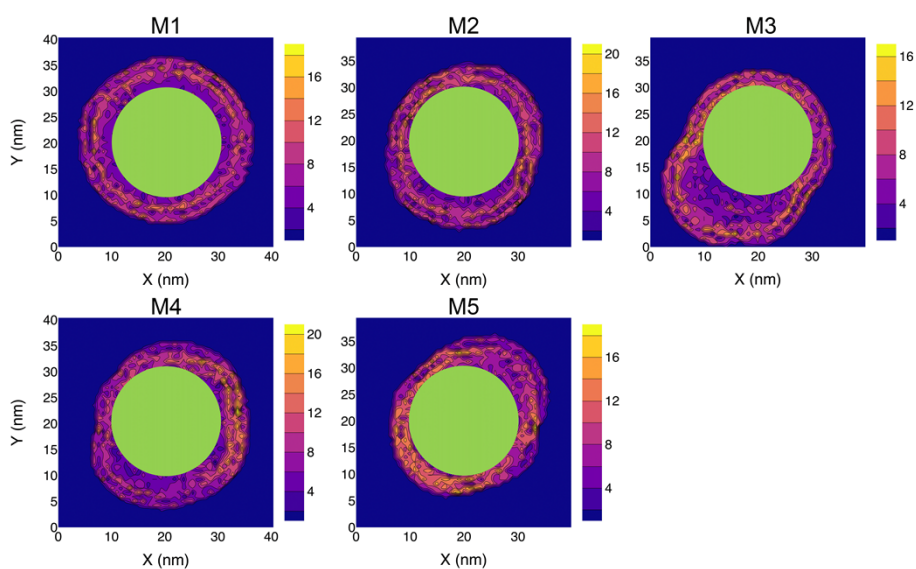


Figure S8 | Two dimensional heatmaps of DOPE lipid distributions. The regions of polyplexes are shown in green.

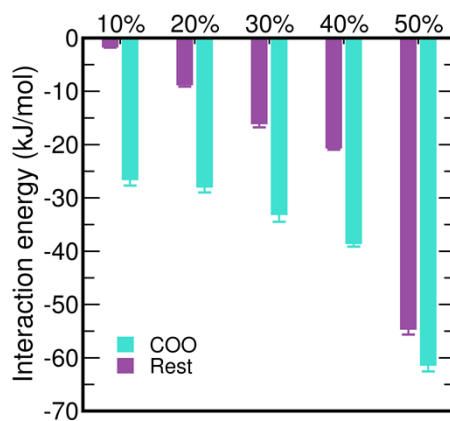


Figure S9 | Interaction energy with polyplex experienced by COO bead of CHEMS and the rest of the CHEMS. Error bar denotes the standard deviation.

Polyplex Model Construction: the polyplex models were constructed with such a method that it maps the Fibonacci lattice onto the surface of a sphere via equal-area projection (see main text). A Python script with a README file to build the polyplex models are provided as a supplementary material.

1. S. Mahajan and T. Tang, *J. Comput. Chem.*, 2019, **40**, 607-618.
2. L. Martínez, R. Andrade, E. G. Birgin and J. M. Martínez, *J. Comput. Chem.*, 2009, **30**, 2157-2164.