Organelle-Mimicking Liposome Dissociates G-quadruplexes and Facilitates Transcription

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Supplementary Figure 12 for the melting profiles of 1crG₃.

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4cG₃.

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Supplementary Table 1. Thermodynamic parameters for formation of the G-quadruplex structures by mcG_n^a

POPC	∆ H °	TΔS°	Δ G° ₂₅	$T_{ m m}$	
Concentration (mM)	(kcal mol ⁻¹)	(kcal mol ⁻¹)	(kcal mol ⁻¹)	(°C)	
		1cG ₂			
0.0	-30.7 ± 0.4	-29.3 ± 0.4	-1.4 ± 0.1	38.5	
2.0	-26.6 ± 0.2	-25.7 ± 0.1	-0.9 ± 0.1	33.0	
	$1cG_4$				
0.0	-62.1 ± 0.4	-54.7 ± 0.3	-7.4 ± 0.1	65.5	
2.0	-38.1 ± 0.7	-33.9 ± 0.7	-4.2 ± 0.1	59.5	
		2cG ₃			
0.0	-43.1 ± 0.1	-40.2 ± 0.1	-2.9 ± 0.1	46.0	
2.0	-27.8 ± 0.4	-26.4 ± 0.4	-1.4 ± 0.1	40.5	
$3cG_3$					
0.0	-40.2 ± 0.4	-37.9 ± 0.3	-2.3 ± 0.1	42.0	
2.0	-31.7 ± 0.3	-30.3 ± 0.4	-1.4 ± 0.1	38.0	
$4cG_3$					
0.0	-43.3 ± 0.7	-41.1 ± 0.6	-2.2 ± 0.2	40.0	
2.0	-33.1 ± 0.5	-31.8 ± 0.6	-1.3 ± 0.2	36.0	

 $^{^{\}alpha}All$ experiments were carried out in a buffer containing 10 mM sodium phosphate (pH 7.0), 10 mM NaCl, and 1 mM Na₂EDTA without or with 2.0 mM POPC. Values are means \pm standard deviations from three independent measurements. Melting temperatures were measured at a total strand concentration of 20 μ M.

Supplementary Table 2. Changes in thermodynamic parameters for formation of G-quadruplex by $1cG_n$ (n = 2, 3, and 4) in the presence of 2.0 mM POPC relative to its absence

G-quadruplex	$\Delta\Delta H^{\circ}$	$\Delta(T\Delta S^{\circ})$	$\Delta\Delta G^{\circ}_{25}$
	(kcal mol ⁻¹)	(kcal mol ⁻¹)	(kcal mol ⁻¹)
$1cG_2$	+4.1	+3.6	+0.5
$1cG_3$	+17.9	+16.0	+1.9
$1cG_4$	+24.0	+20.8	+3.2

Supplementary Table 3. Thermodynamic parameters for the formation of G-quadruplex structure by $1 \text{cr} G_3^a$

POPC	ΔH°	$T\Delta S^{\circ}$	ΔG° 25	$T_{ m m}$
Concentration (mM)	(kcal mol ⁻¹)	(kcal mol ⁻¹)	(kcal mol ⁻¹)	(°C)
0.0	-45.8 ± 0.5	-40.6 ± 0.6	-5.2 ± 0.1	62.5
2.0	-23.5 ± 0.4	-22.0 ± 0.3	-1.5 ± 0.1	54.5

 $^{^{\}it a}$ All experiments were carried out in a buffer containing 10 mM sodium phosphate (pH 7.0), 10 mM NaCl, and 1 mM Na₂EDTA without or with 2.0 mM POPC. Values are means \pm standard deviations from three independent measurements. Melting temperatures were measured at a total strand concentration of 20 μ M.

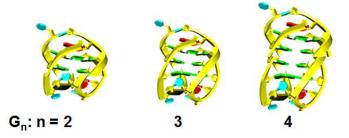
Supplementary Table 4. Thermodynamic parameters for the formation of G-quadruplex structure by $1cG_3^a$

Liposome (2.0 mM)	∆H° (kcal mol ⁻¹)	$T\Delta S^{\circ}$ (kcal mol ⁻¹)	ΔG°_{25} (kcal mol ⁻¹)	T _m (°C)
_	-40.0 ± 0.6	-36.9 ± 0.5	-3.1 ± 0.1	50.0
IM	-28.0 ± 0.5	-26.0 ± 0.4	-2.0 ± 0.1	47.0
NM	-24.9 ± 0.8	-23.4 ± 0.3	-1.5 ± 0.1	45.0
ER	-23.1 ± 0.3	-21.7 ± 0.2	-1.4 ± 0.1	42.5

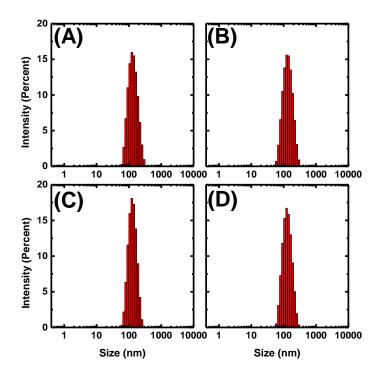
 $[^]a$ All experiments were carried out in a buffer containing 10 mM sodium phosphate (pH 7.0), 10 mM NaCl, and 1 mM Na₂EDTA without or with 2.0 mM liposome. Values are means \pm standard deviations from three independent measurements. Melting temperatures were measured at a total strand concentration of 20 μ M.

Supplementary Table 5. Changes of thermodynamic parameters of G-quadruplex formed by 1cG₃ in the presence of IM, NM, and ER mimicking liposomes and POPC liposomes

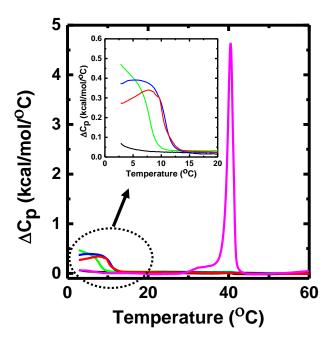
Liposome (2.0 mM)	Molar ratio of POPC (%)	ΔΔH° (kcal mol ⁻¹)	$\Delta(T\Delta S^{\circ})$ (kcal mol ⁻¹)	ΔΔG° ₂₅ (kcal mol ⁻¹)
IM	43	+12.0	+10.9	+1.1
NM	69	+15.1	+13.5	+1.6
ER	75	+16.9	+15.2	+1.7
POPC	100	+17.9	+16.0	+1.9



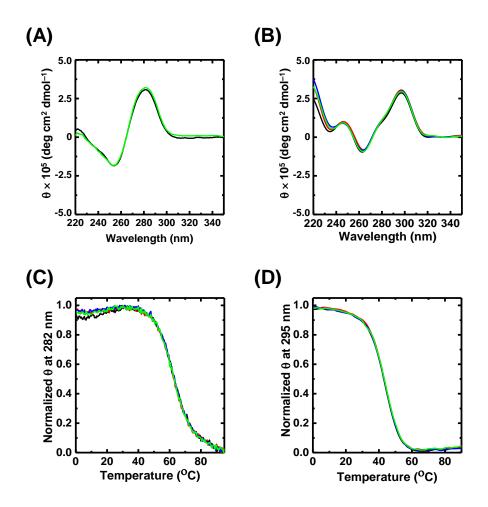
Supplementary Figure 1. Schematic representations of $G_n\ (n=2,\,3,\,\text{and}\,4)$ structures.



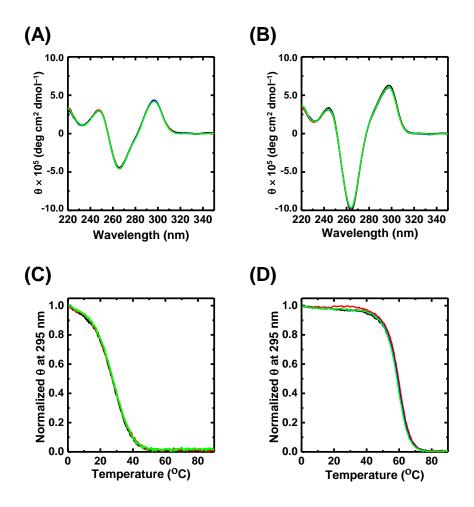
Supplementary Figure 2. Diameter distributions of **(A)** POPC, **(B)** IM, **(C)** NM, and **(D)** ER liposomes at 25 °C.



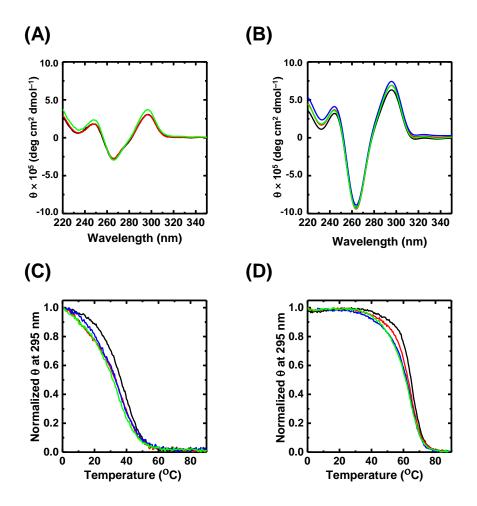
Supplementary Figure 3. DSC heating thermograms for POPC (black), IM (red), NM (blue), ER (green), and DPPC (pink) liposomes. The inset shows DSC profiles in the region from 0 to 20 °C.



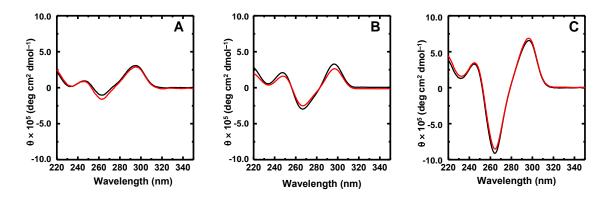
Supplementary Figure 4. CD spectra of 20 μ M (**A**) D and (**B**) G_3 at 4 °C and normalized CD melting profiles of (**C**) D at 282 nm and (**D**) G_3 at 295 nm in the absence (black) and presence of 2.0 mM IM (red), NM (blue), and ER (green) liposomes.



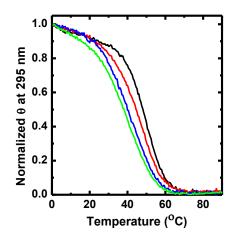
Supplementary Figure 5. CD spectra of 20 μ M (**A**) G_2 and (**B**) G_4 at 4 °C and normalized CD melting profiles of (**C**) G_2 and (**D**) G_4 at 295 nm in the absence (black) and presence of 2.0 mM IM (red), NM (blue), and ER (green) liposomes.



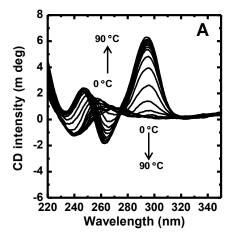
Supplementary Figure 6. CD spectra of 20 μ M (**A**) 1cG₂ and (**B**) 1cG₄ at 4 °C and normalized CD melting profiles of (**C**) 1cG₂ and (**D**) 1cG₄ at 295 nm in the absence (black) and presence of 2.0 mM IM (red), NM (blue), and ER (green) liposomes.

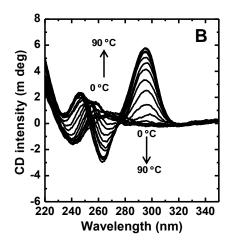


Supplementary Figure 7. CD spectra of 20 μ M (A) 1cG₃, (B) 1cG₂, and (C) 1cG₄ in the absence (black) and presence (red) of 2.0 mM POPC liposomes at 4 °C.

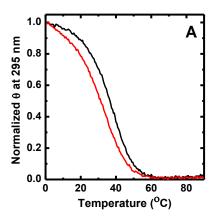


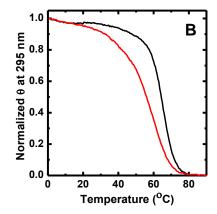
Supplementary Figure 8. Normalized CD melting profiles at 295 nm of 20 μ M total strand concentration of 1cG₃ in the absence (black) and presence of 0.5 (red), 1.0 (blue), and 2.0 mM (green) POPC liposomes.



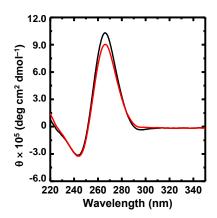


Supplementary Figure 9. CD spectra of 20 μ M 1cG₃ in (**A**) the absence and (**B**) presence of 2.0 mM POPC liposomes at temperatures from 0 to 90 °C.

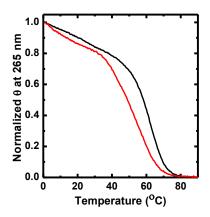




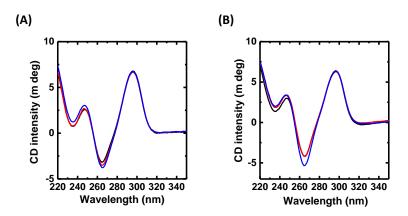
Supplementary Figure 10. Normalized CD melting profiles at 295 nm of 20 μ M total strand concentration of (**A**) 1cG₂ and (**B**) 1cG₄ in the absence (black) and presence (red) of 2.0 mM POPC liposomes.



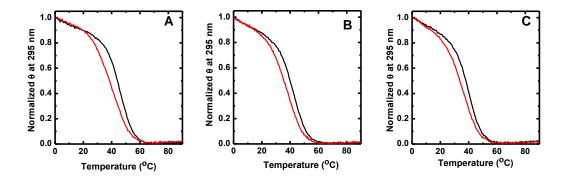
Supplementary Figure 11. CD spectra of 20 μM 1crG₃ in the absence (black) and presence (red) of 2 mM POPC liposomes at 4 °C.



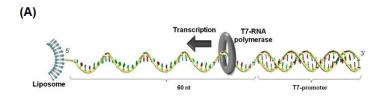
Supplementary Figure 12. Normalized CD melting profiles at 265 nm of 20 μ M total strand concentration of 1crG₃ in the absence (black) and presence (red) of 2.0 mM POPC liposomes.

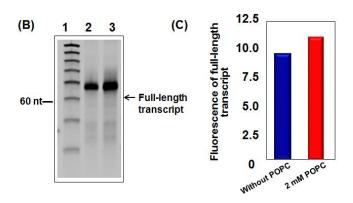


Supplementary Figure 13. CD spectra of 20 μ M 2cG₃ (black), 3cG₃ (red), and 4cG₃ (blue) in (**A**) absence and (**B**) presence of 2.0 mM POPC liposomes at 4 °C.



Supplementary Figure 14. Normalized CD melting profiles at 295 nm of 20 μ M total strand concentration of **(A)** 2cG₃, **(B)** 3cG₃, and **(C)** 4cG₃ in the absence (black) and presence (red) of 2.0 mM POPC liposomes.





Supplementary Figure 15. Effects of membrane surface on transcription of a linear template DNA sequence. (**A**) Illustration of the template DNA immobilized on a liposome. (**B**) Denaturing gel electrophoresis of products of transcription reactions performed for 90 min at 37 °C. Reaction mixtures contained 0.3 μM T7 polymerase and 1.5 μM DNA template in a buffer containing 10 mM NaCl, 40 mM Tris-HCl (pH 8.0), 8 mM MgCl₂ and 2 mM spermidine in the absence and presence of 2.0 mM POPC. Lane 1, size marker; lane 2, transcription products in the absence of POPC; lane 3, transcription products in the presence of 2.0 mM POPC. (**C**) Comparison of fluorescence intensities of full-length transcripts from gel bands in panel **B**.