Supplemental Information:

Enhanced Chemotherapeutic Toxicity of Cyclodextrin Templated Size-Tunable Rhodamine 6G nanoGUMBOS

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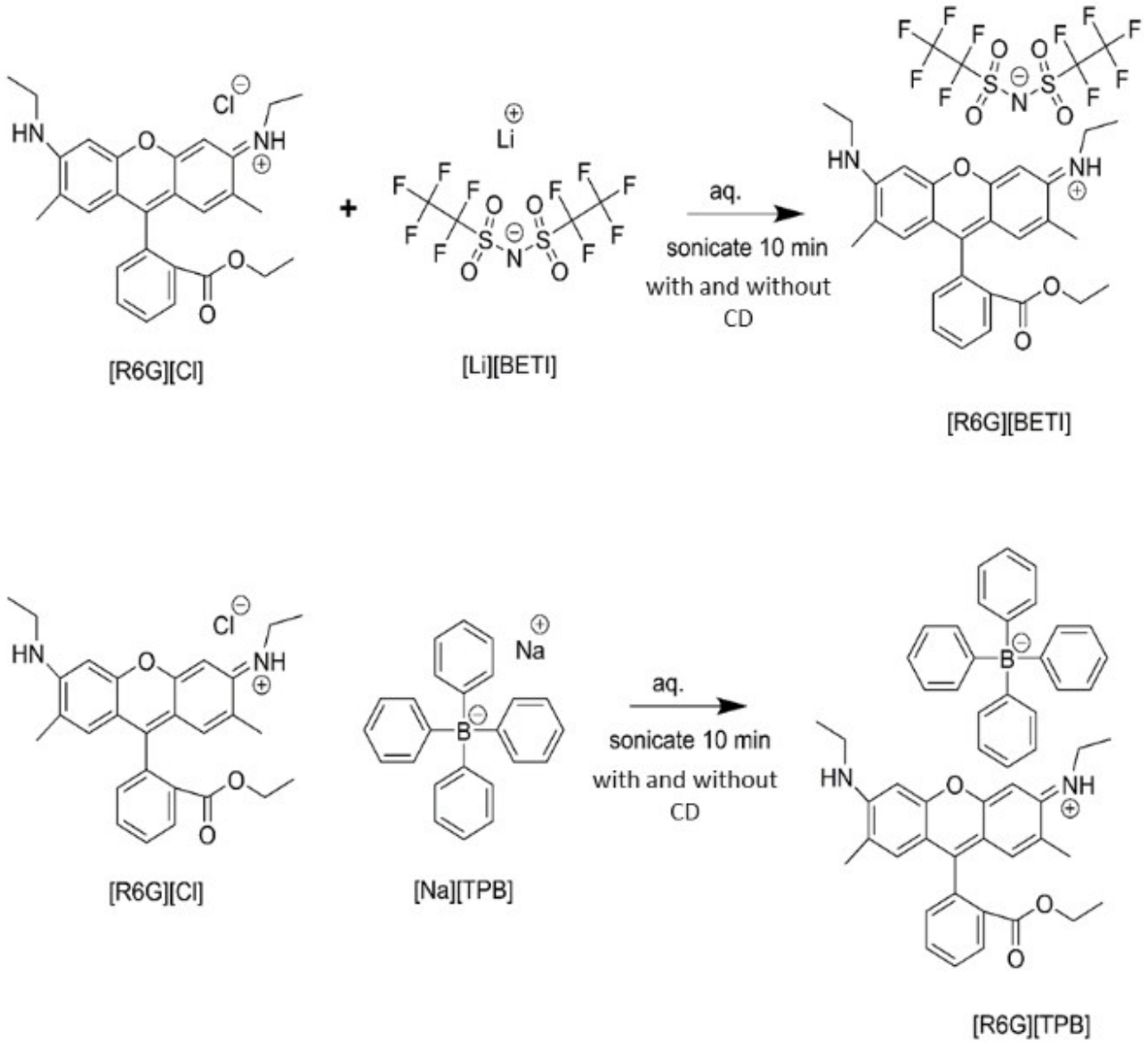

Figure S1. Synthesis of [R6G][BETI] and [R6G][TPB] nanoGUMBOS with and without CD-template using probe sonication for 5 minutes at 30 mHz and 20% amplitude.

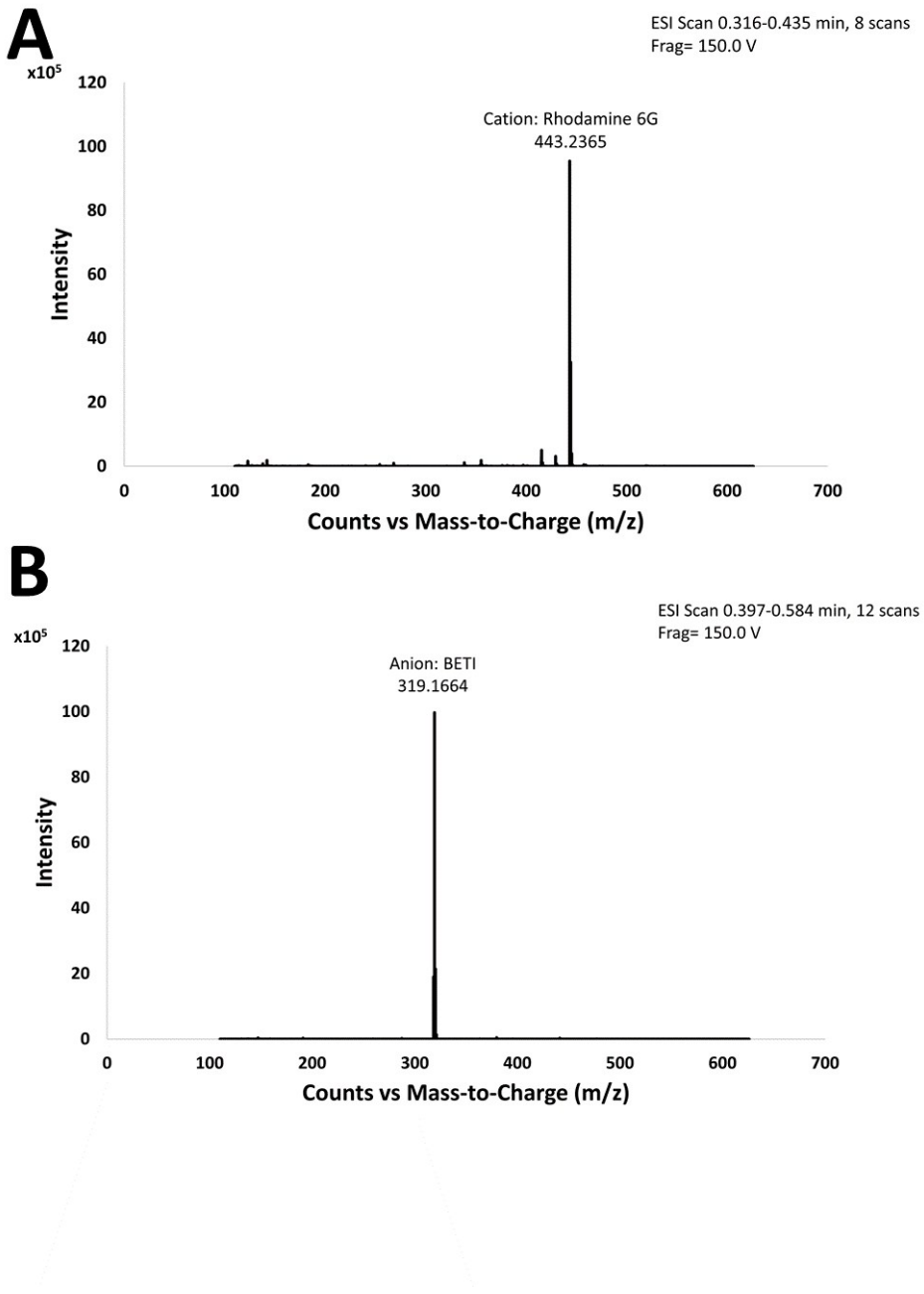

Figure S2. Electrospray Ionization Mass Spectrum in **(A)** postive and **(B)** negative mode for α-CD templated [R6G][BETI] nanoGUMBOS.

Theoretical masses of 443 and 319 obtained for cation and anion mode respectively correspond to the mass of R6G and TPB indicating successful synthesis. Similar results were observed with all other [R6G][TPB] nanoGUMBOS

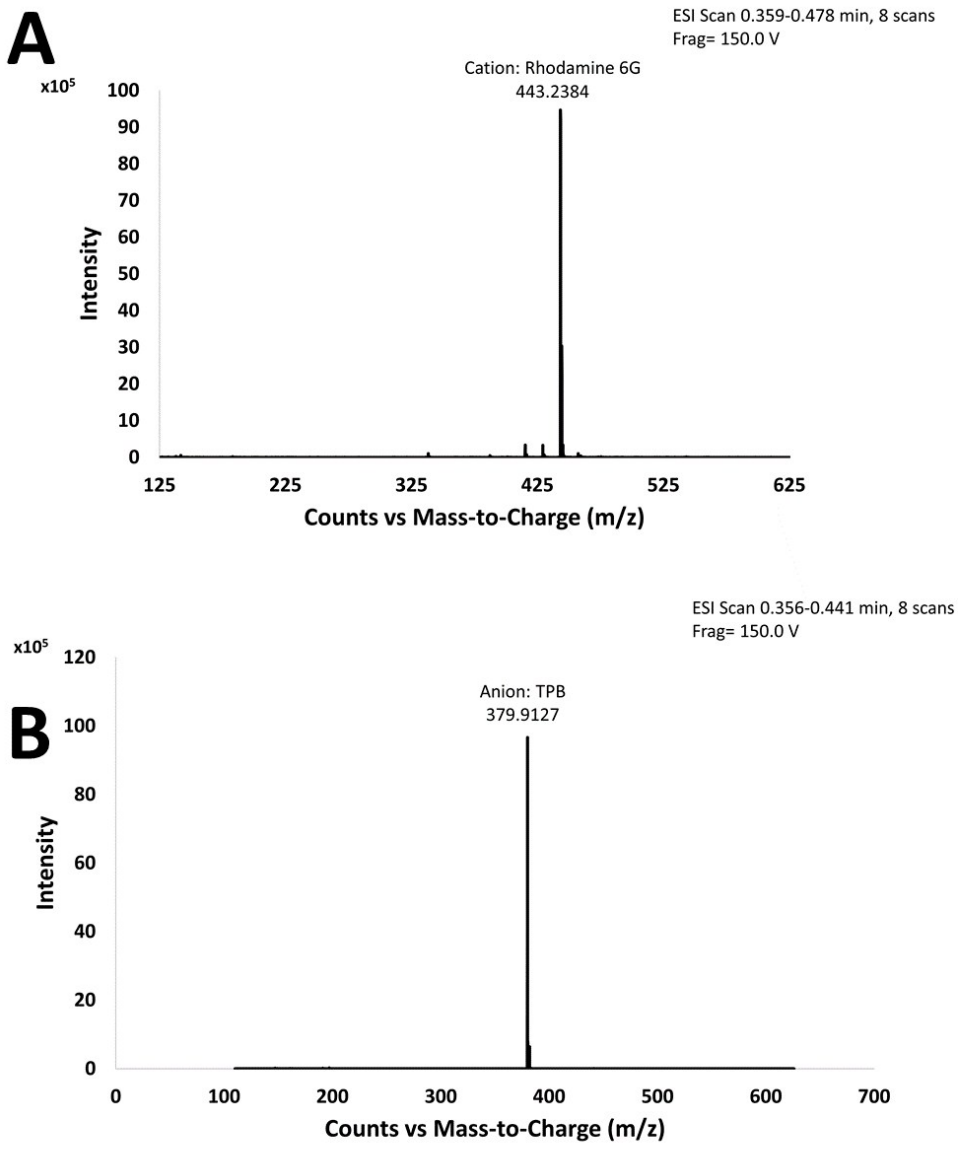

Figure S3. Electrospray Ionization Mass Spectrum in **(A)** postive and **(B)** negative mode for α-CD templated [R6G][BETI] nanoGUMBOS.

Theoretical masses of 443 and 379 obtained for cation and anion mode respectively correspond to the mass of R6G and BETI indicating successful synthesis. Similar results were observed with all other [R6G][BETI] nanoGUMBOS

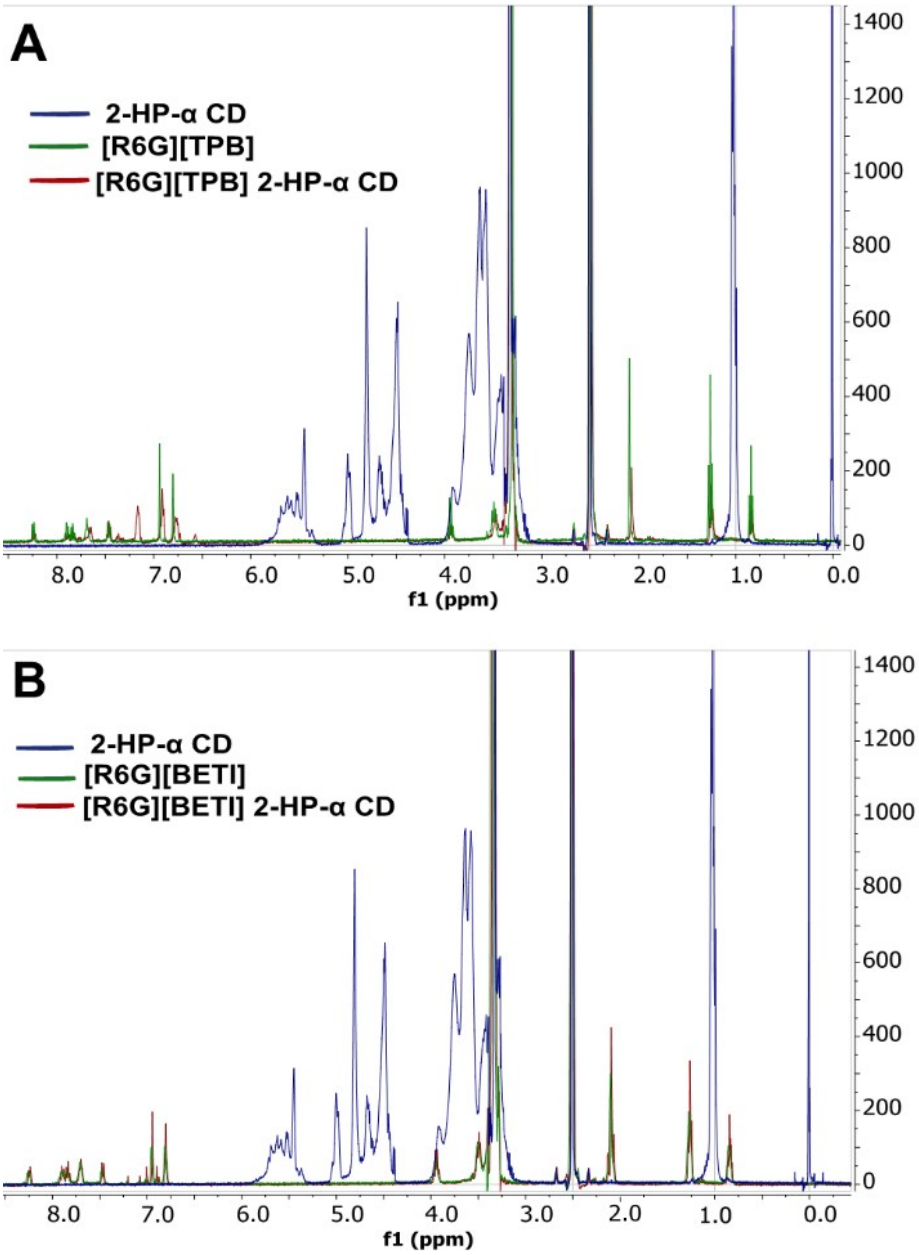

Figure S4. Overlay of NMR of [R6G][TPB] and [R6G][BETI] nanoGUMBOS with and without 2HP-α-CD-template.

The lack of the CD peaks at 5-6 ppm in the NMR spectrum of our nanoGUMBOS indicate that the CD template employed during synthesis was washed away with the byproduct. Similar results were observed for all other CD templated [R6G][TPB] and [R6G][BETI] nanoGUMBOS.

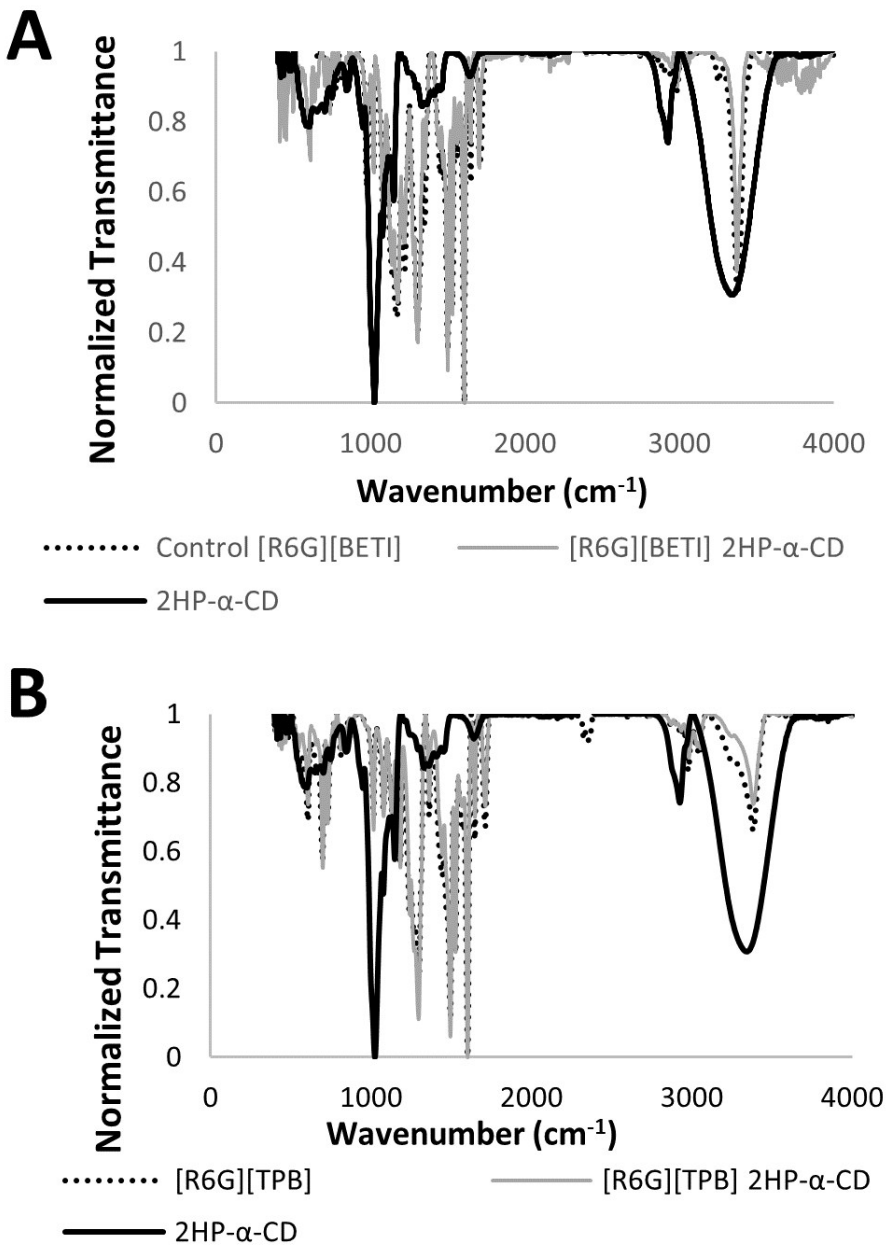

Figure S5. Overlay of FTIR of [R6G][TPB] and [R6G][BETI] nanoGUMBOS with and without CD-templating.

The lack of the CD peaks at 3600 cm^{-1} and 1000 cm^{-1} in the FTIR spectrum of our nanoGUMBOS suggests that the CD template was washed away after synthesis. Similar results were observed with all other CD-templated nanoGUMBOS.

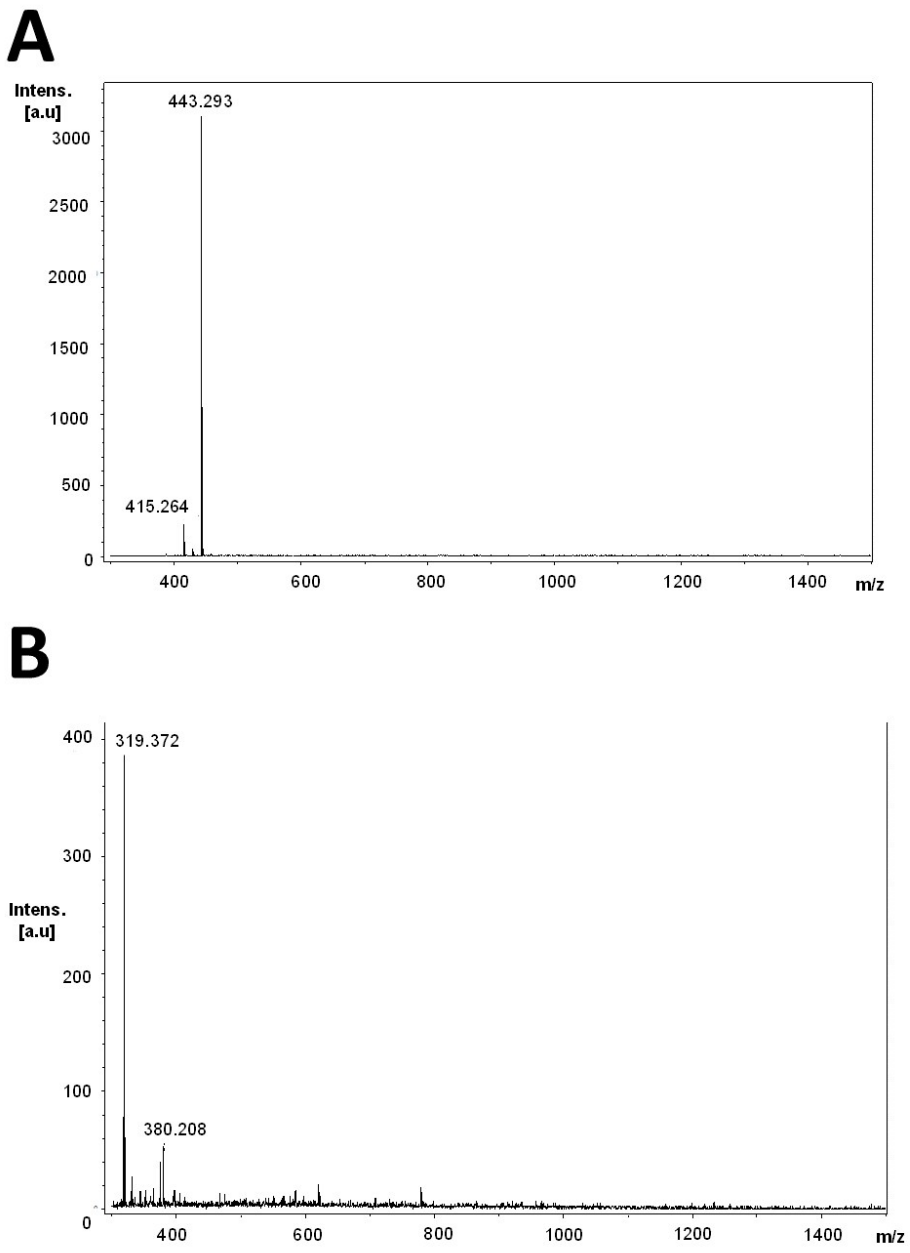

Figure S6. [R6G][TPB] nanoGUMBOS control in **(A)** positive mode and **(b)** negative mode MALDI mass spectrometry.

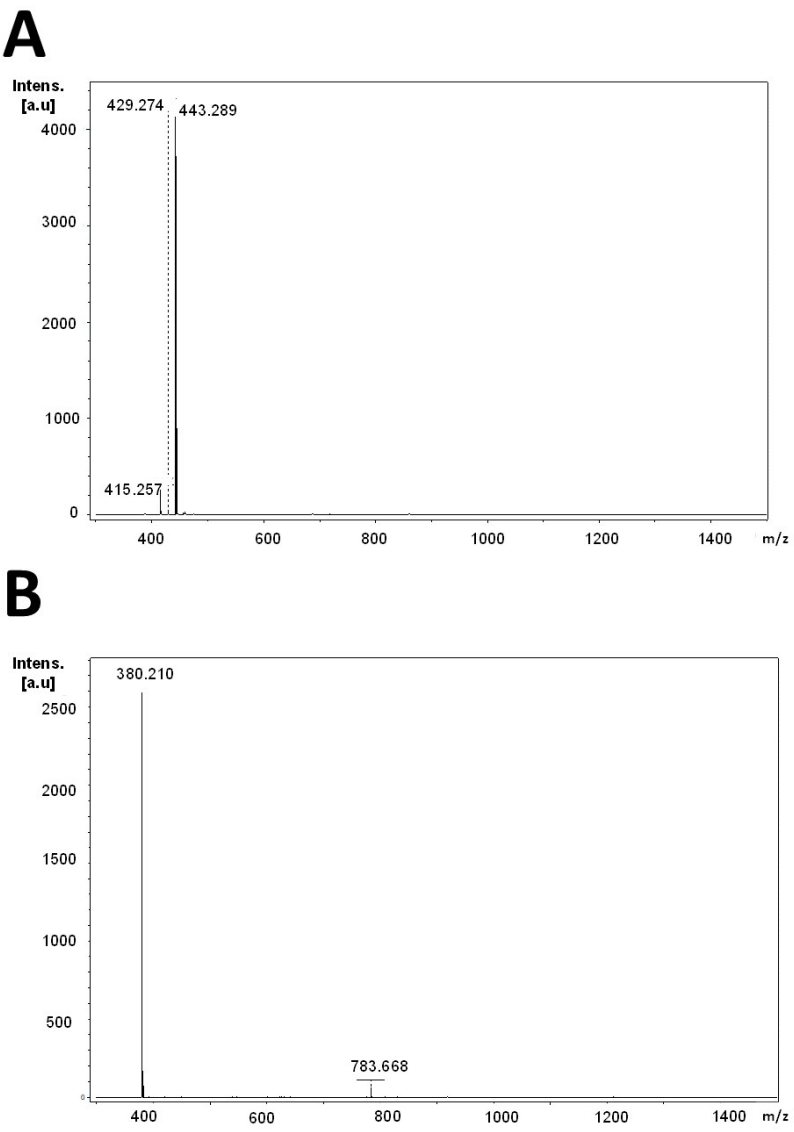

Figure S7. [R6G][BETI] nanoGUMBOS control in **(A)** positive mode and **(b)** negative mode MALDI mass spectrometry.

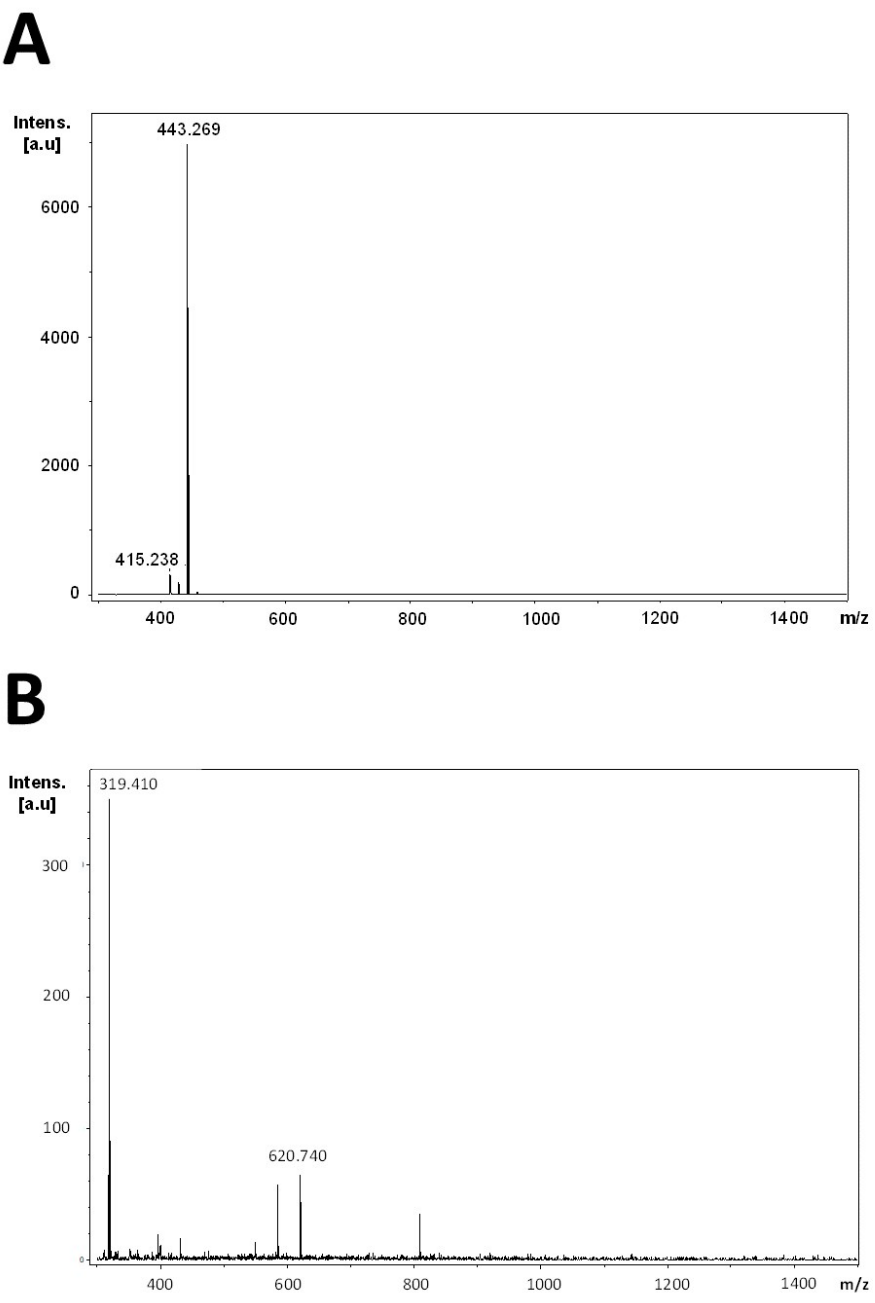

Figure S8. [R6G][TPB] nanoGUMBOS templated with 2HP-α-CD in **(A)** positive mode and **(b)** negative mode MALDI mass spectrometry.

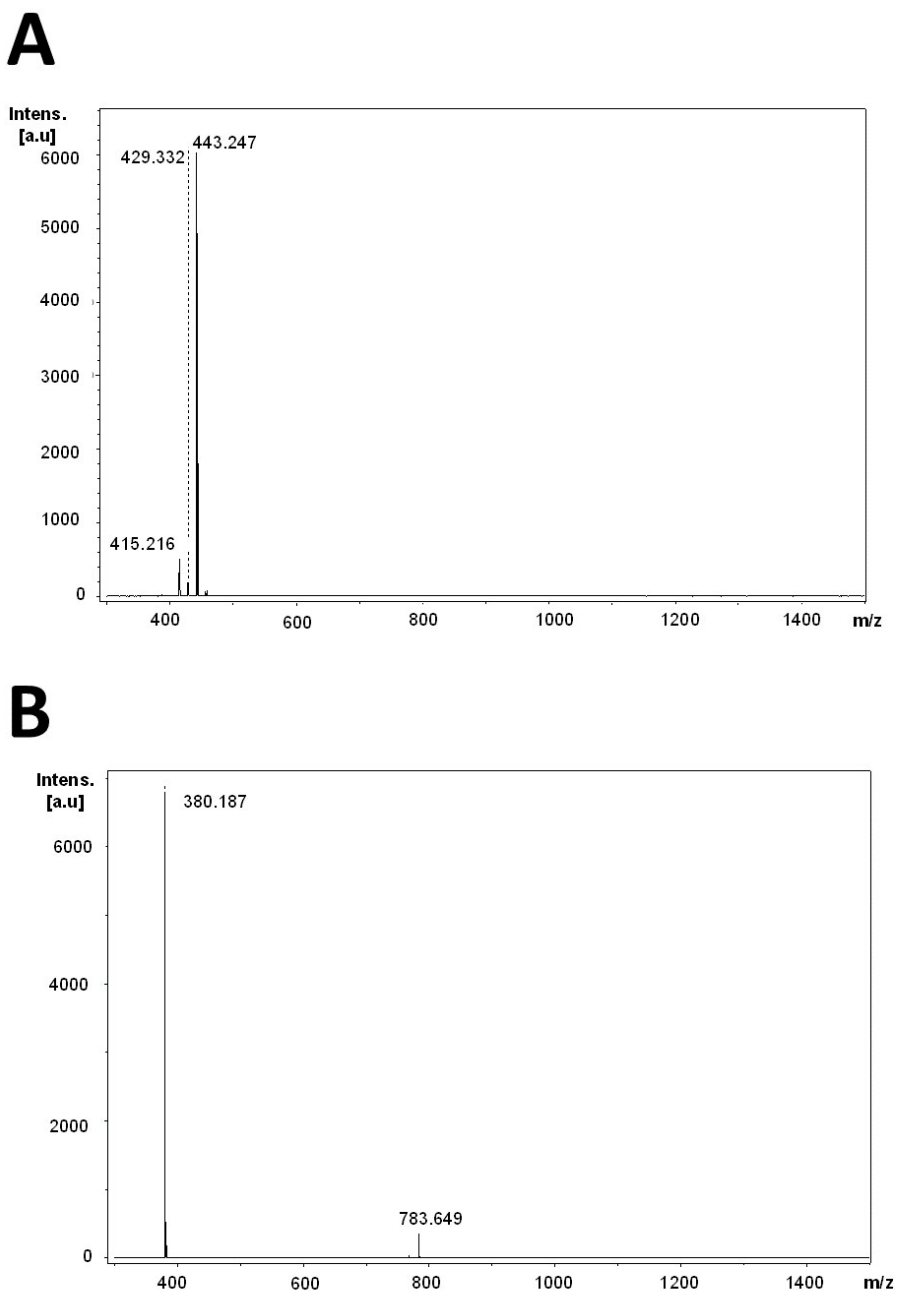

Figure S9. [R6G][BETI] nanoGUMBOS templated with 2HP-α-CD in **(A)** positive mode and **(b)** negative mode MALDI mass spectrometry.

The observed masses (m/z) for cation and anion mode are 443 and 319 respectively, which correspond to the masses of R6G and TPB (Figure S6). The peaks observed in Figure S7 correspond to the mass of R6G and BETI. Similarly, for figure S8 and S9, the masses (m/z) observed for cation and anion mode also correspond to the mass of the R6G cation and TPB or BETI anion. The lack of CD peaks observed in the MALDI mass spectrum of the 2HP-α-CD templated [R6G][TPB] and [R6G][BETI] nanoGUMBOS in figure S8 and S9 respectively suggest that CD was primarily employed as a template and washed away during the synthesis. Similar results were observed for all CD-templated nanoGUMBOS.

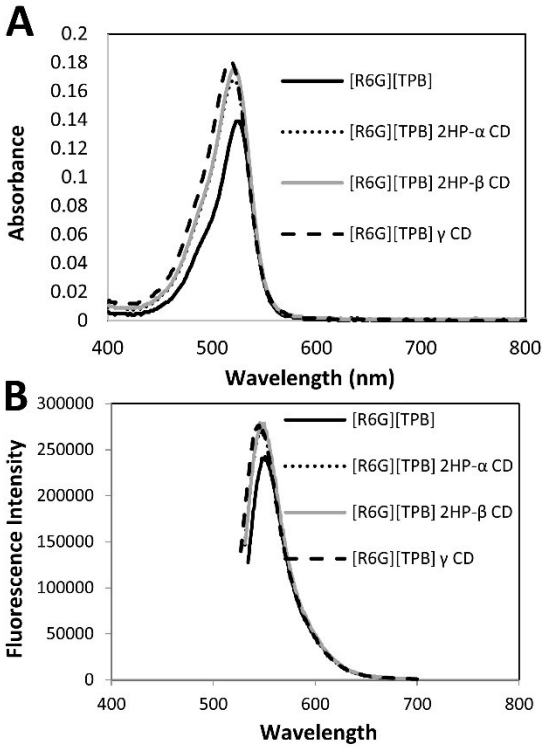

Figure S10. (A) Absorbance and **(B)** Fluorescence of [R6G][TPB] nanoGUMBOS with and without CD-template.

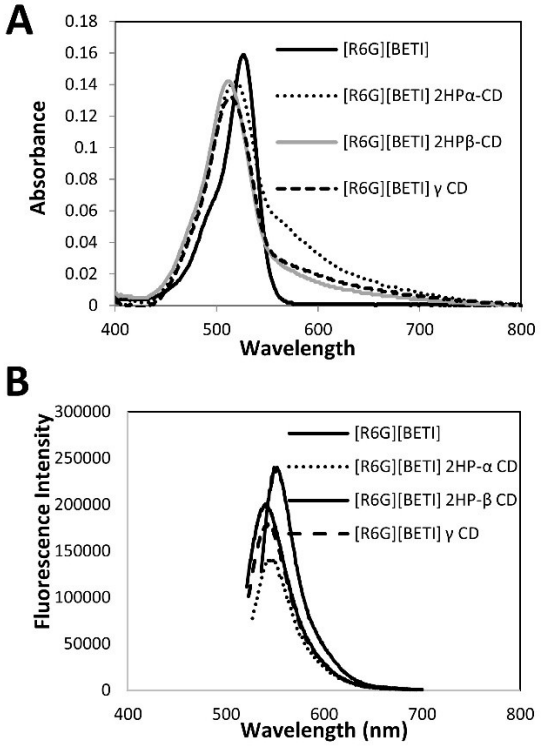

Figure S11. (A) Absorbance and **(B)** Fluorescence of [R6G][BETI] nanoGUMBOS with and without CD-template.

As shown in Figure S10, [R6G][TPB] nanoGUMBOS without CD templating displayed absorbance (λ_{abs}) and fluorescence emission peaks (λ_{em}) at 524 and 555 nm, respectively. CD-templated [R6G][TPB] nanoGUMBOS displayed a slight blue shift in absorbance maxima at 521, 521, 517 and emission maxima at 549, 549 and 546 nm, respectively, for nanoGUMBOS templated with 2HP-α-CD, 2HP-β-CD and γ-CD, respectively. As shown in Figure S11, [R6G][BETI] nanoGUMBOS without CD template displayed an absorbance (λ_{abs}) and fluorescence emission peak (λ_{em}) of 526 and 555 nm, respectively. All CD-templated [R6G][TPB] nanoGUMBOS displayed a slight blue shift in absorbance maxima at 517, 511 and 513 nm and emission maxima at 546, 541 and 544 nm for 2HP-α-CD, 2HP-β-CD and γ-CD templated [R6G][BETI] nanoGUMBOS, respectively.

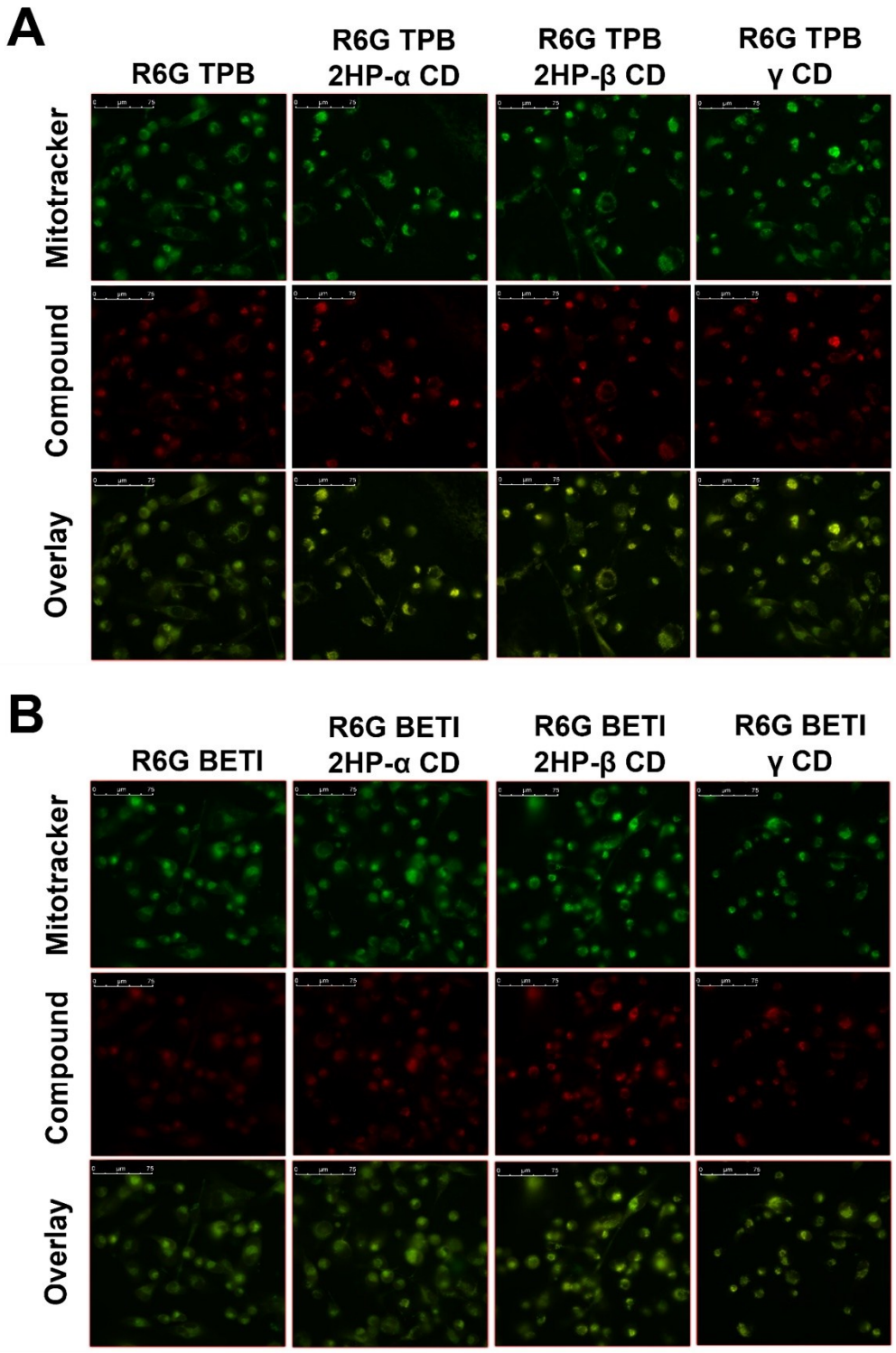

Figure S12. Microscopy images of **(A)** [R6G][TPB] and **(B)** [R6G][BETI] nanoGUMBOS with and without CD templating (shown as red fluorescence) in the presence of 10 nM mitotracker green (shown as green fluorescence) in MDA-MB-231 breast cancer cells.

Compound	Colocalization Coefficient
[R6G][TPB]	0.97
[R6G][TPB] 2HP-α-CD	0.97
[R6G][TPB] 2HP-β-CD	0.97
[R6G][TPB] γ-CD	0.96
[R6G][BETI]	0.96
[R6G][BETI] 2HP-α-CD	0.97
[R6G][BETI] 2HP-β-CD	0.98
[R6G][BETI] γ-CD	0.97

Table S1: Colocalization coefficients for R6G nanoGUMBOS