Supplementary Information (SI)

1,3,5-Triazine as Branching Connector for the Construction of Novel Antimicrobial Peptide Dendrimers. Synthesis and Biological Characterization

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Figure S1: Truncated dendrimers found during the synthesis of G2 dendrimer containing PHBA.

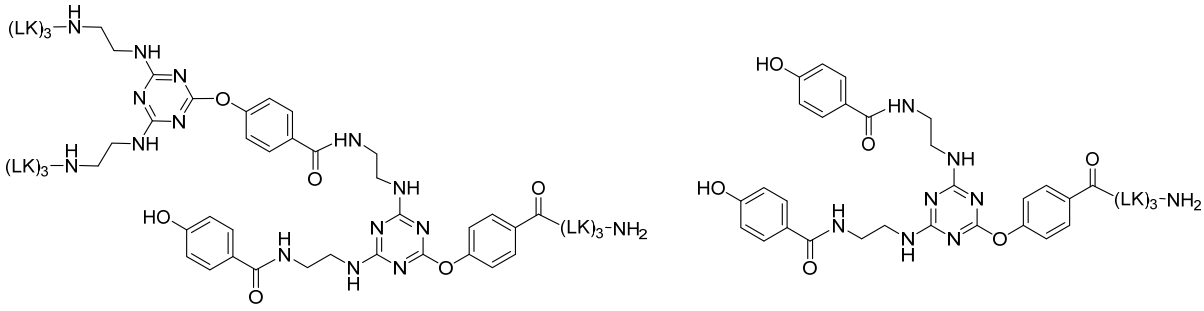


Figure S2: HPLC trace of purified **1** (H-[(LK)$_3$]$_2$-(EDA)$_2$-TA-PHBA-(LK)$_3$-NH$_2$).
Method: 15-50% B into A in 15 min

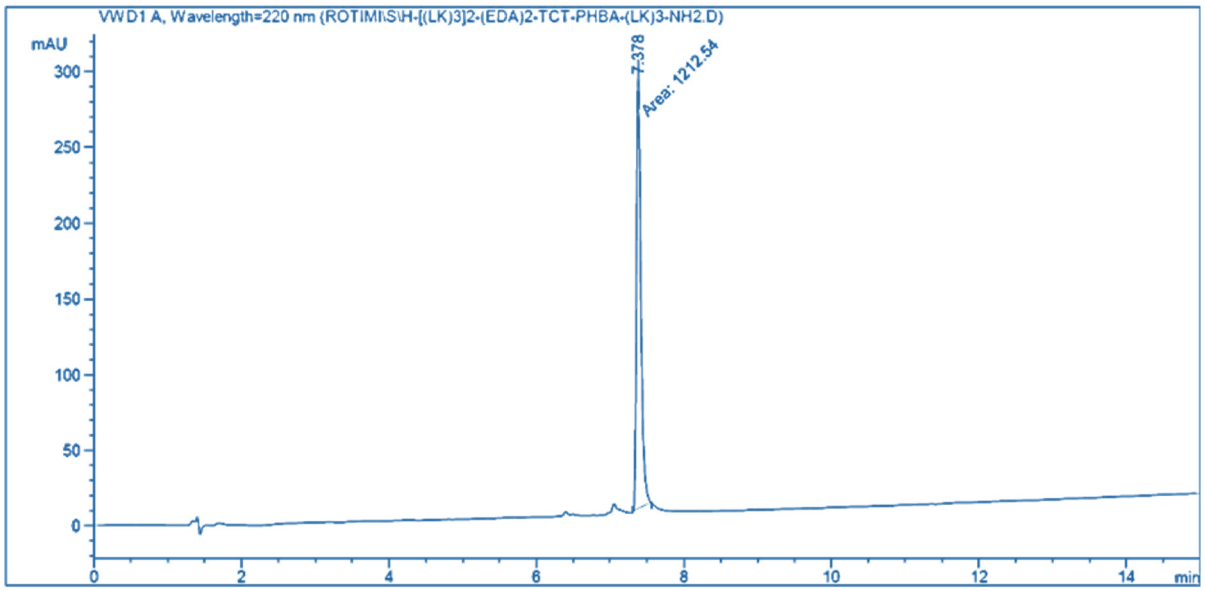


Figure S3: ESI-MS of **1** (H-[(LK)$_3$]$_2$-(EDA)$_2$-TA-PHBA-(LK)$_3$-NH$_2$).
Expected mass= 2,505.37
Observed mass = (627.21 $[M+4H]^{+}/4$, 501.81 $[M+5H]^{+}/5$, 418.57 $[M+6H]^{+}/6$)

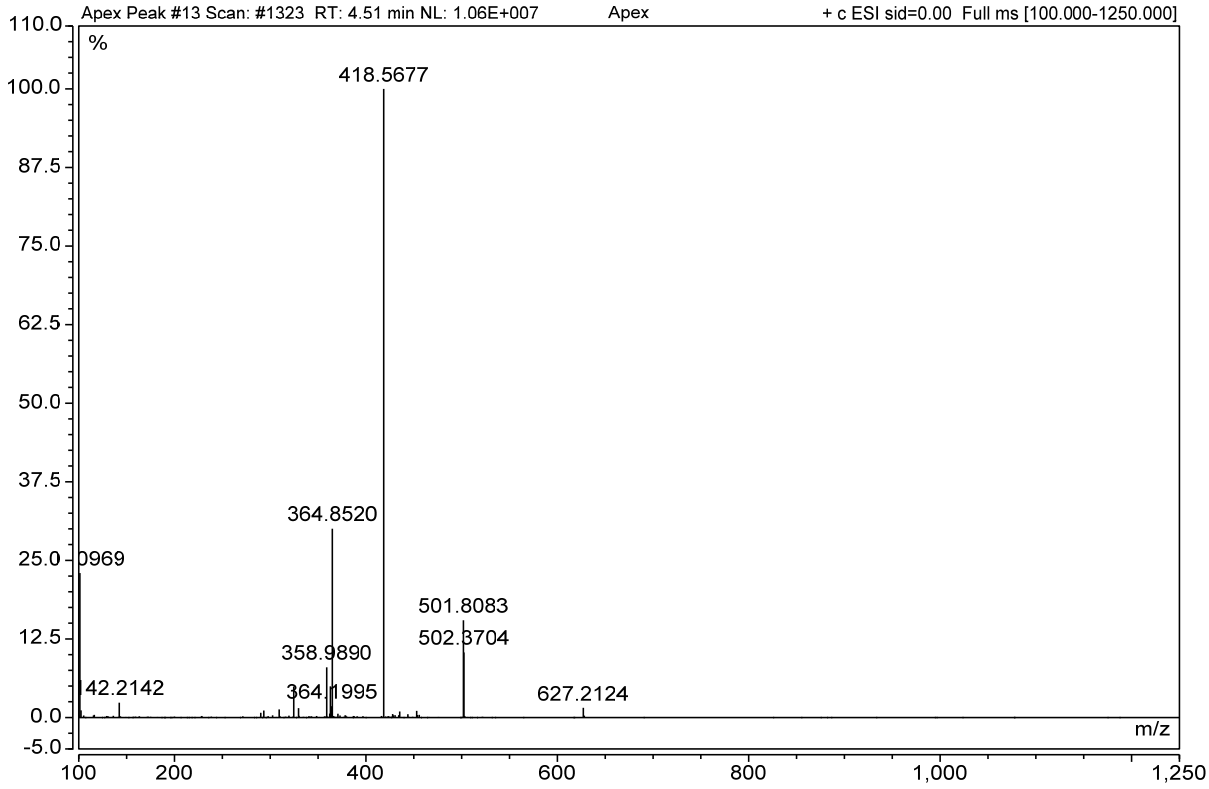


Figure S4: HPLC trace of purified **2** (H-[(LK)$_3$]$_2$-(EDA)$_2$-TA-(LK)$_3$-NH$_2$).
Method 15-50% B into A in 15 min

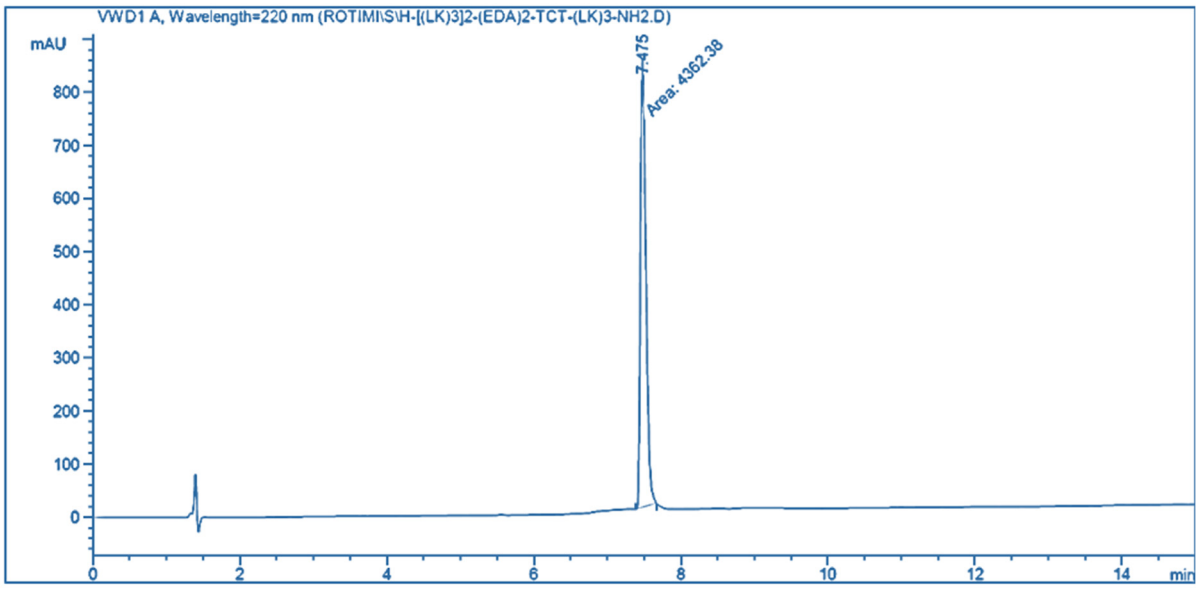


Figure S5: ESI-MS of **2**: (H-[(LK)$_3$]$_2$-(EDA)$_2$-TA-(LK)$_3$-NH$_2$)
Expected mass= 2385.26
Observed mass = (597.27 $[M+4H]^{+}/4$, 478.07 $[M+5H]^{+}/5$, 398.41 $[M+6H]^{+}/6$)

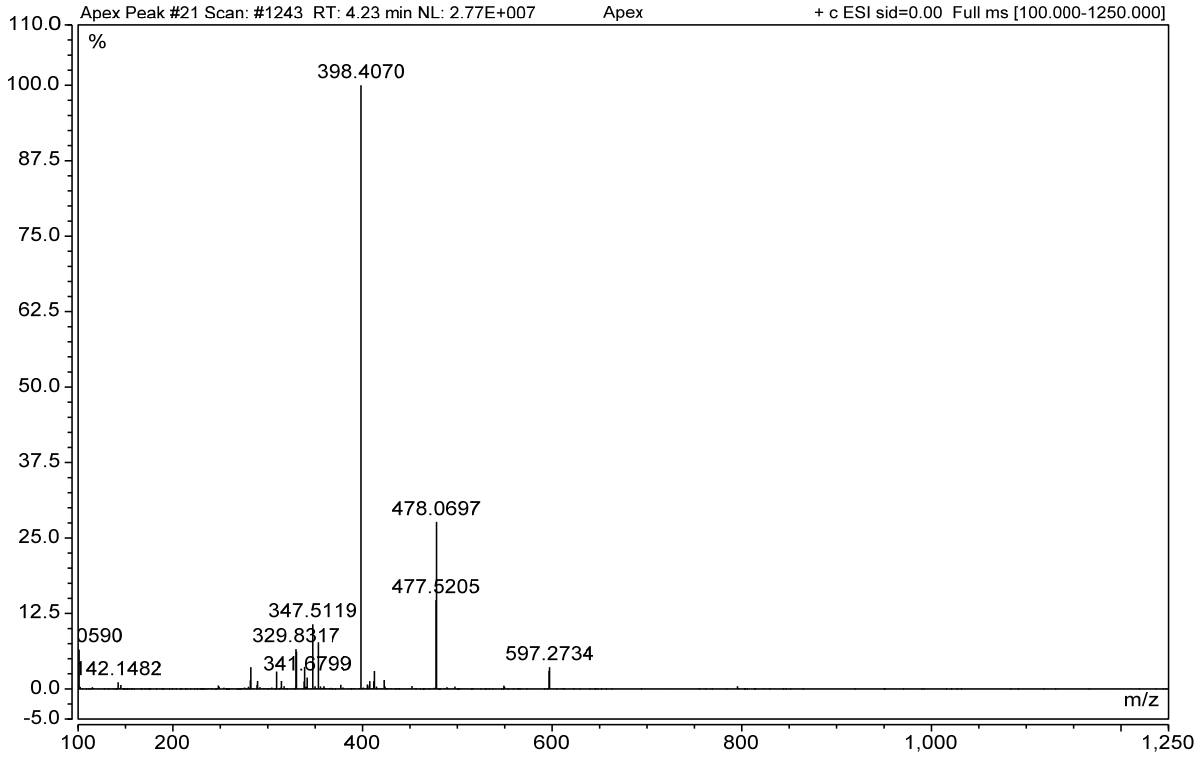


Figure S6: HPLC trace of purified **3** (H-[(LK)$_3$]$_4$-(EDA)$_4$-(TA)$_2$-(EDA)$_2$-TA-(LK)$_3$-NH$_2$).
Method 20-40% B into A in 15 min

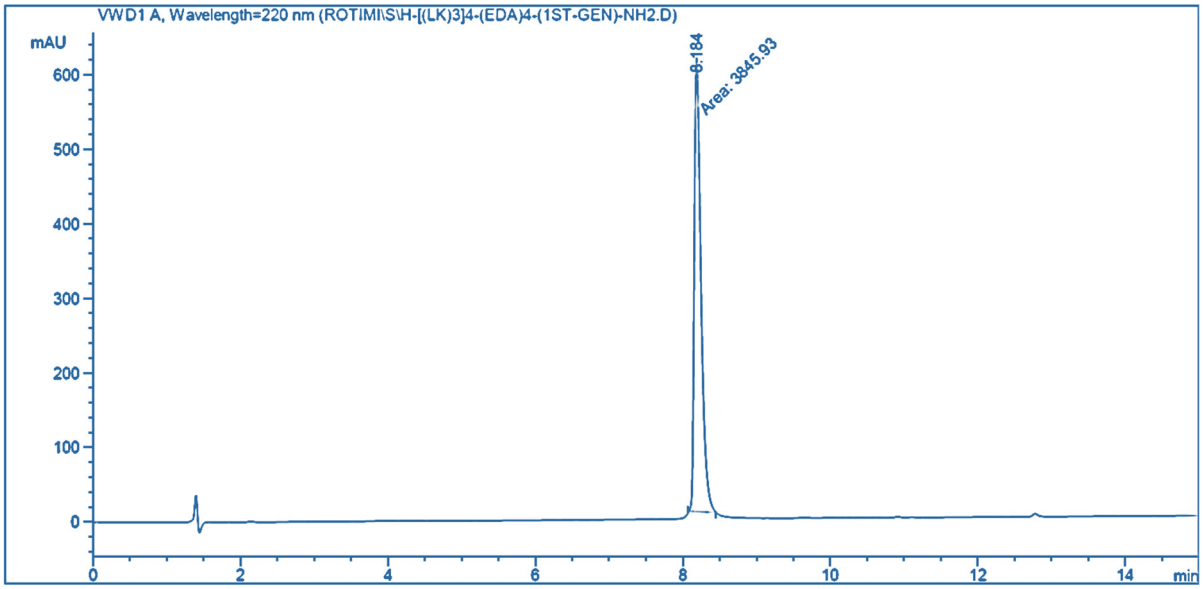


Figure S7: ESI-MS of **3**: (H-[(LK)$_3$]$_4$-(EDA)$_4$-(TCT)$_2$-(EDA)$_2$-TCT-(LK)$_3$-NH$_2$).
Expected mass= 4223.73
Observed mass = (705.04 [M+6H] +/6, 604.42 [M+7H] +/7, 528.99 [M+8H] +/8)

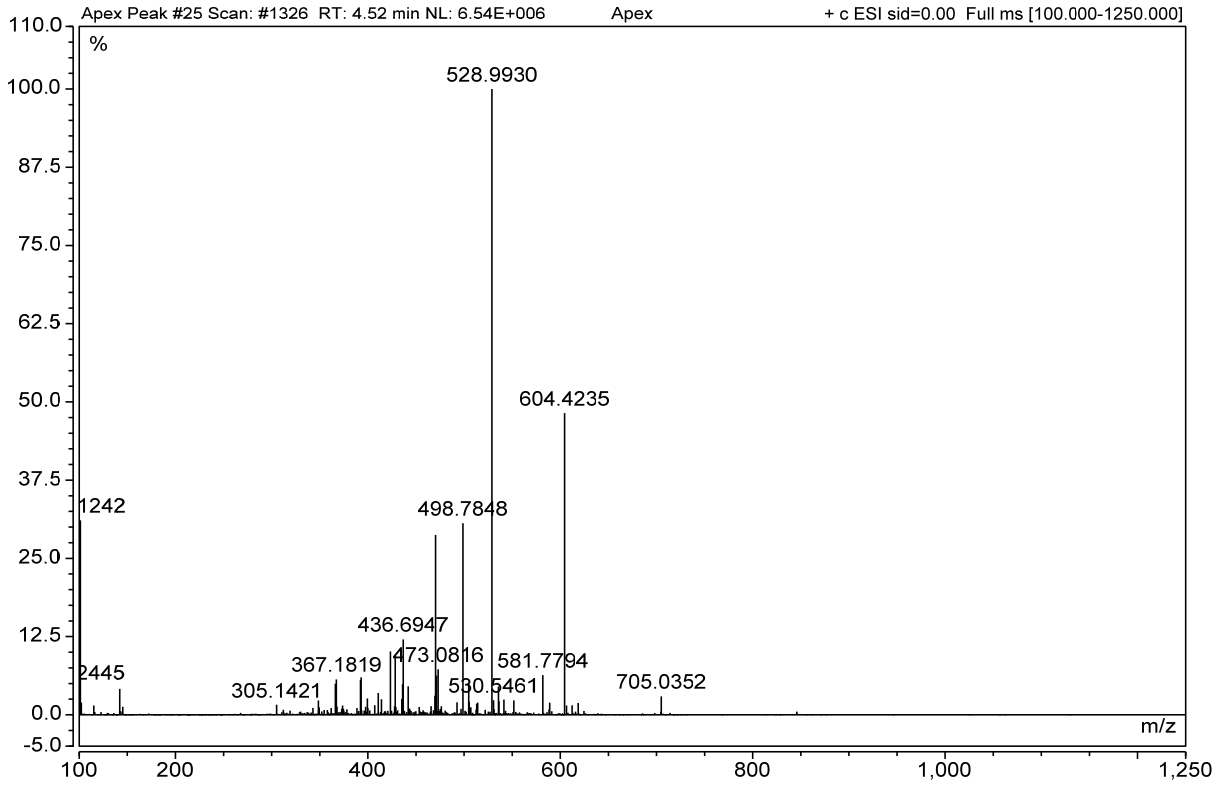


Figure S8: HPLC trace of purified **A** (H-(LK)$_3$-NH$_2$).
Method 5-95% B into A in 15 min

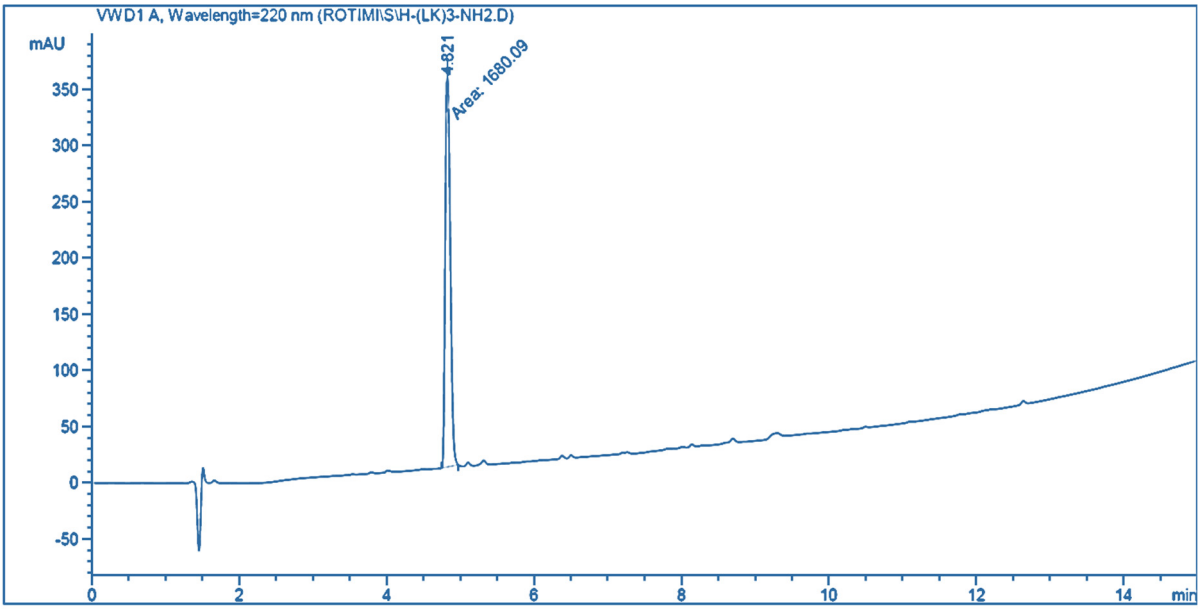


Figure S9: ESI-MS of **A**: (H-(LK)$_3$-NH$_2$).
Expected mass= 741.04
Observed mass = (741.62 $[M+H]^+$, 371.44 $[M+2H]^+/2$, 248.24 $[M+3H]^+/3$)

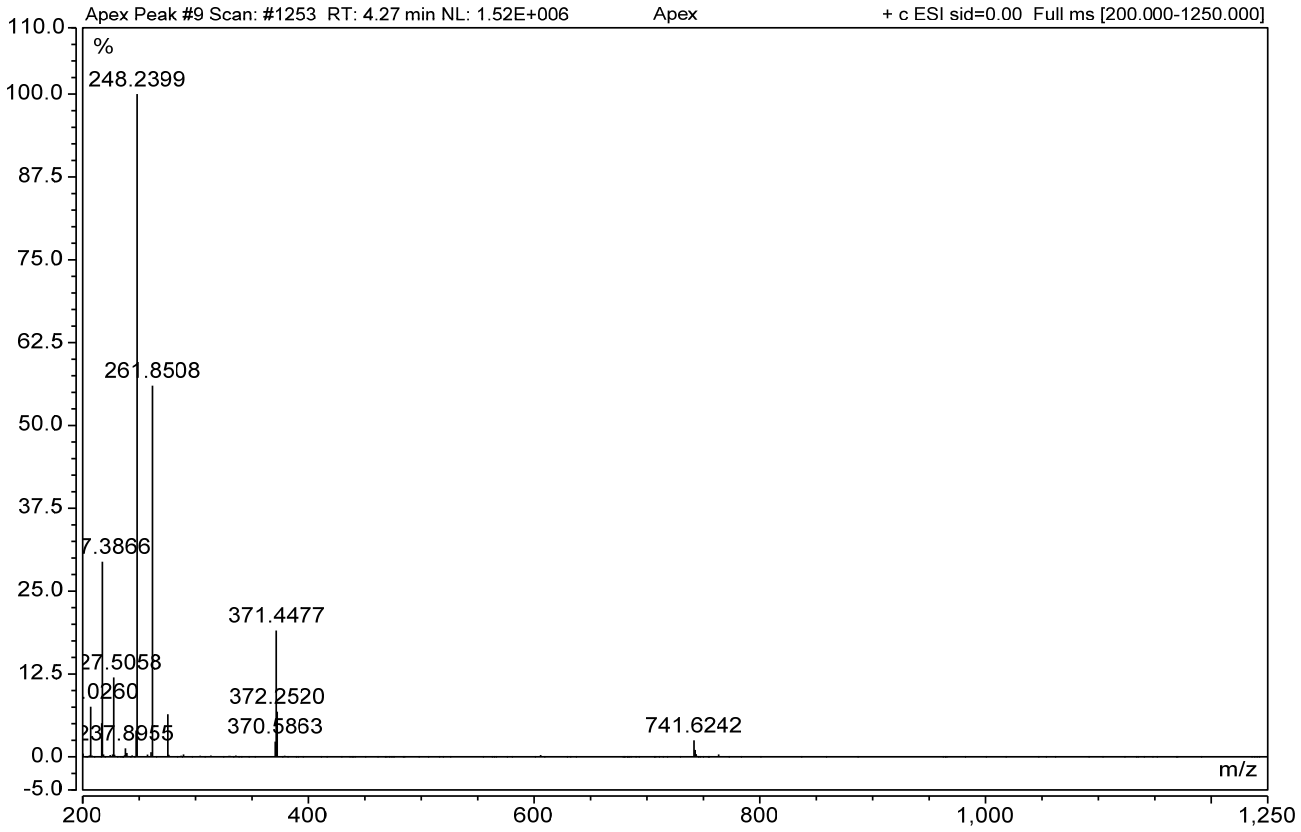


Figure S10: HPLC trace of purified **B** (H-[(LK)$_3$]$_2$-(EDA)$_2$-TA-PHBA-GFL-NH$_2$).
Method 15-70% B into A in 15 min

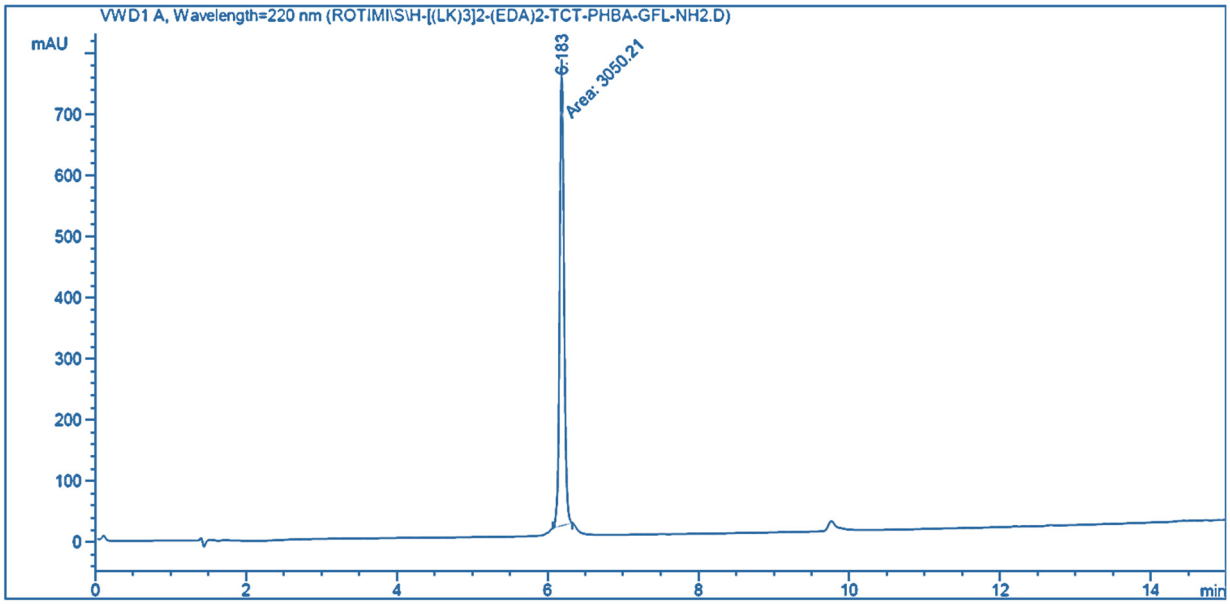


Figure S11: ESI-MS of **B**: (H-[(LK)$_3$]$_2$-(EDA)$_2$-TA-PHBA-GFL-NH$_2$).
Expected mass= 2097.77
Observed mass = (700.14 $[M+3H]^+/3$, 525.57 $[M+4H]^+/4$)

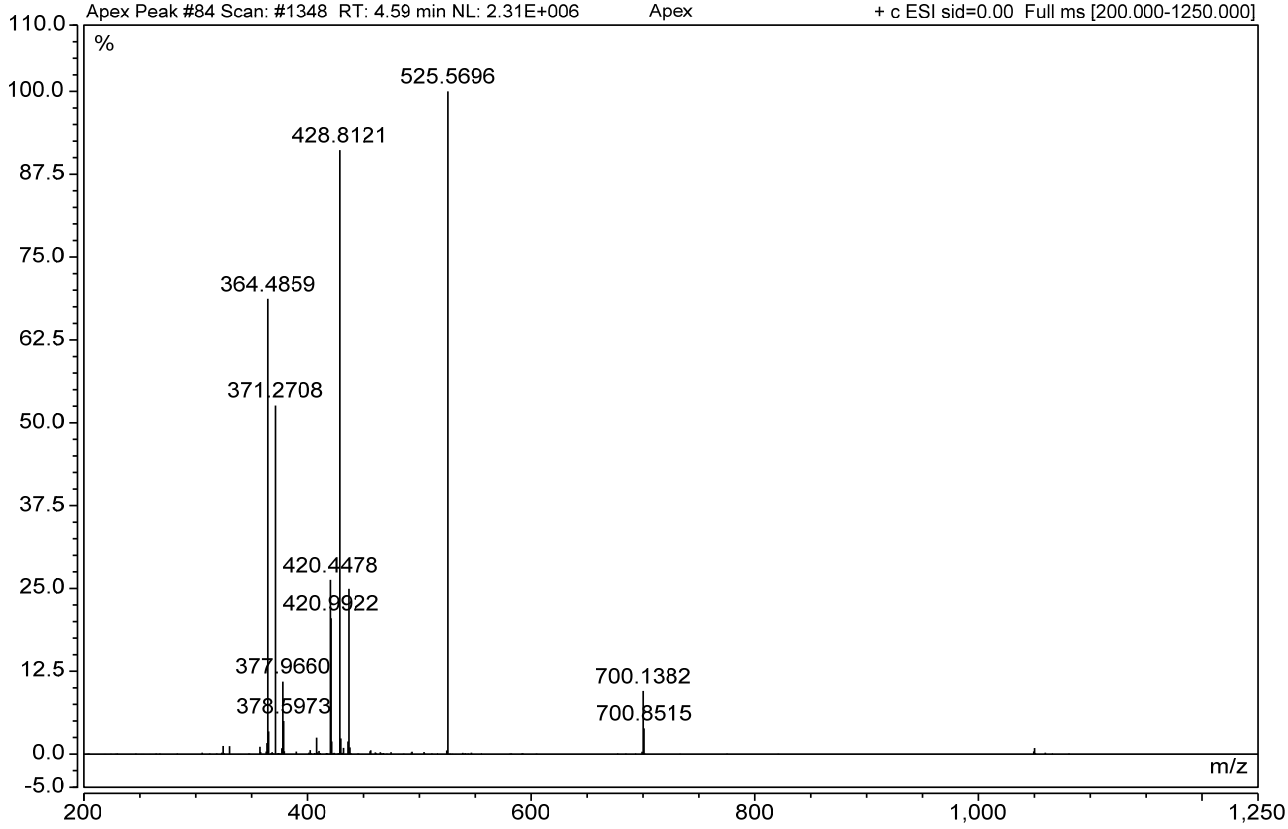


Figure S12: HPLC trace of purified **C** ((Ac-EDA)$_2$-TA-PHBA-GFL-NH$_2$).
Method 15-40% B into A in 15 min

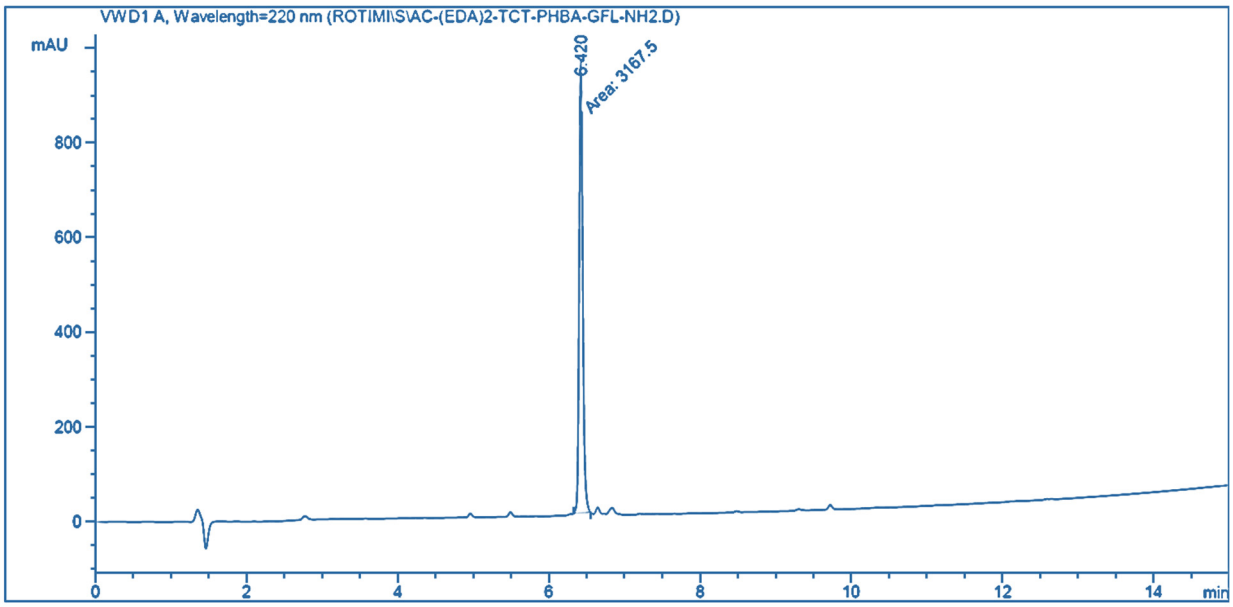


Figure S13: ESI-MS of **C**: ((Ac-EDA)$_2$-TA-PHBA-GFL-NH$_2$).
Expected mass= 733.83
Observed mass = (734.30 $[M+H]^+$, 368.07 $[M+2H]^{+}/2$)

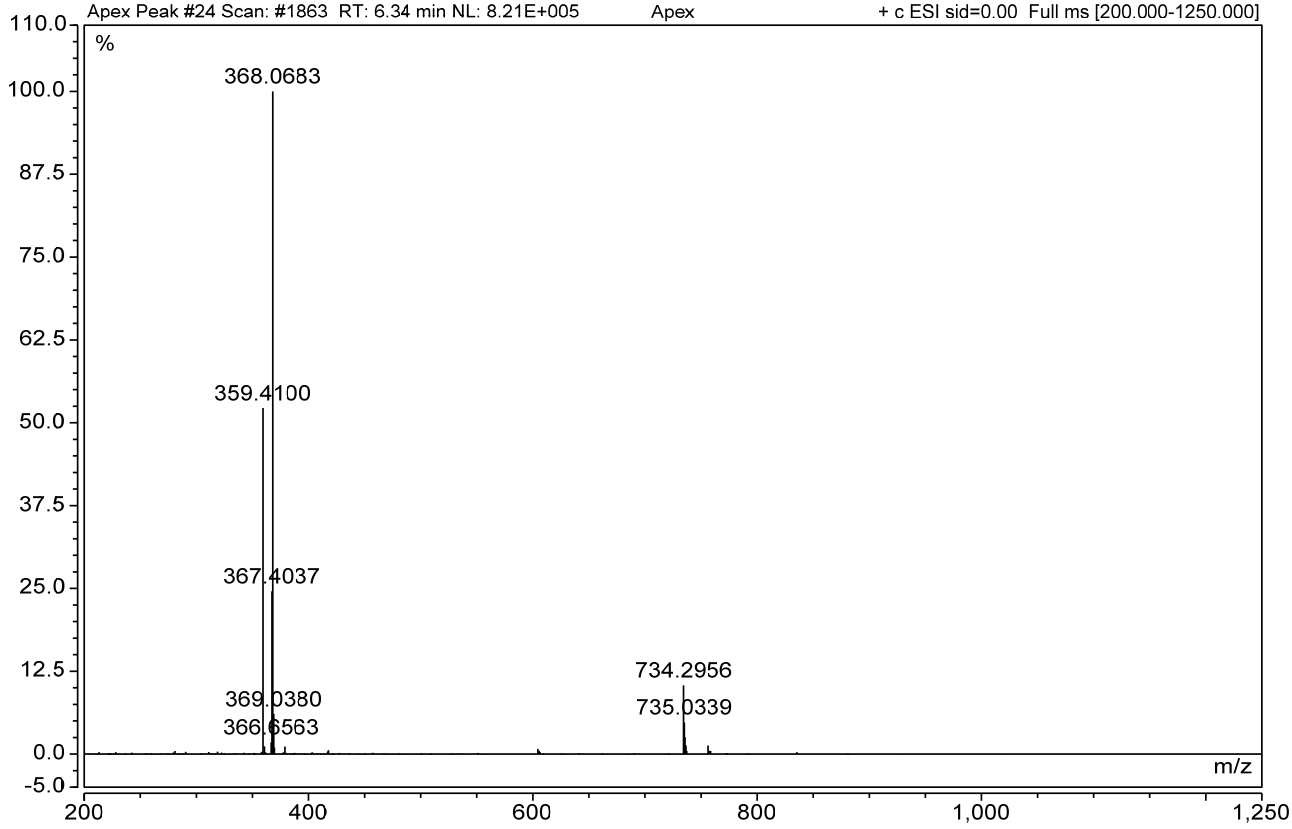


Figure S14: HPLC trace of purified **D** ((Ac-EDA)$_2$-TA-PHBA-(LK)$_3$-NH$_2$).
Method 15-50% B into A in 15 min

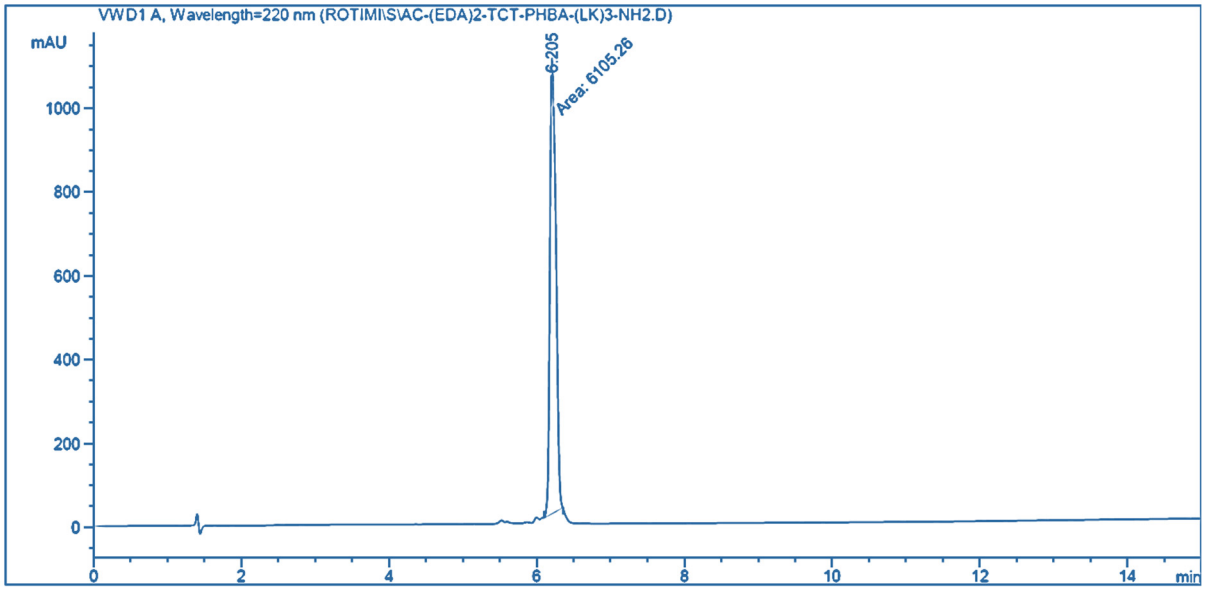


Figure S15: ESI-MS of **D**: ((Ac-EDA)$_2$-TA-PHBA-(LK)$_3$-NH$_2$).
Expected mass= 1141.43
Observed mass = (570.91 $[M+2H]^+/2$, 381.19 $[M+3H]^+/3$, 286.14 $[M+4H]^+/4$)

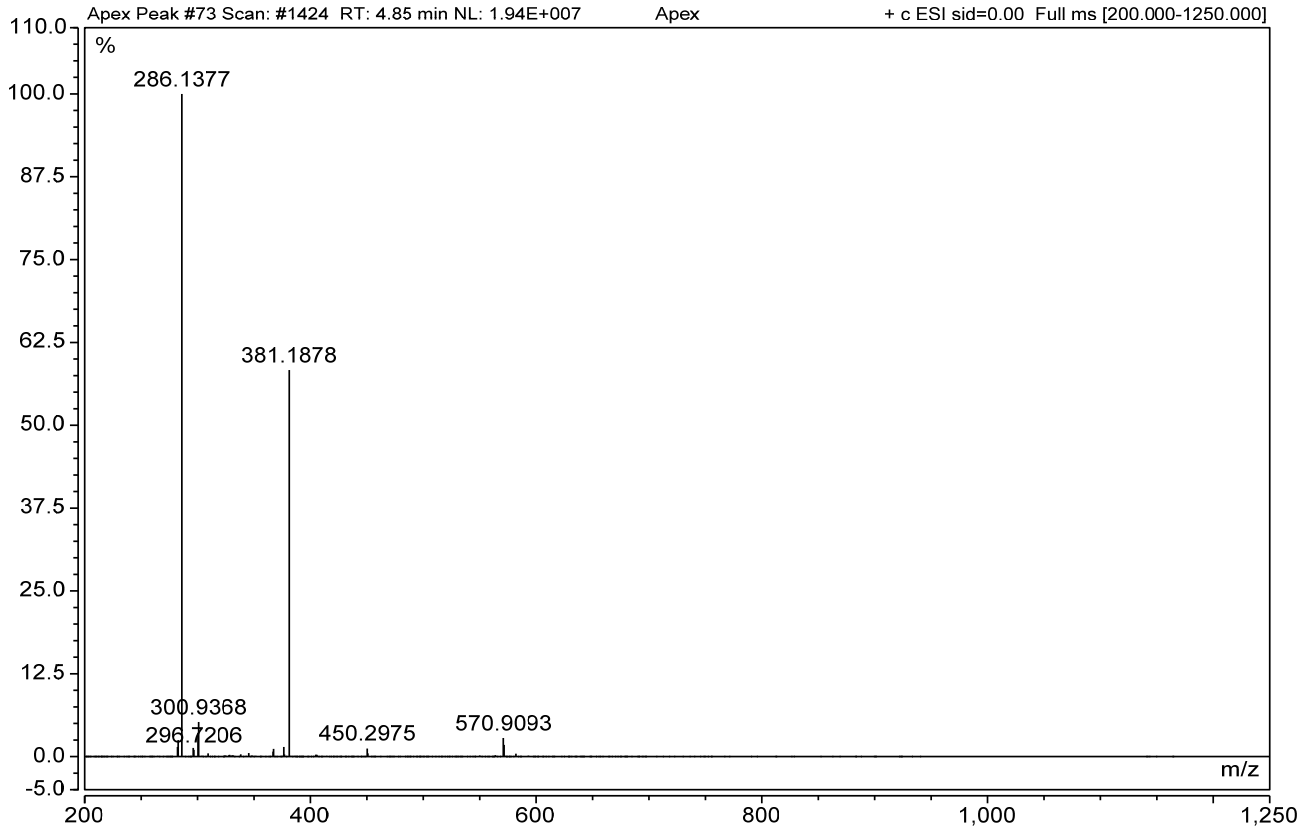


Figure S16: HPLC trace of purified **E** ((Ac-EDA)$_2$-TA-(LK)$_3$-NH$_2$).
Method 15-50% B into A in 15 min

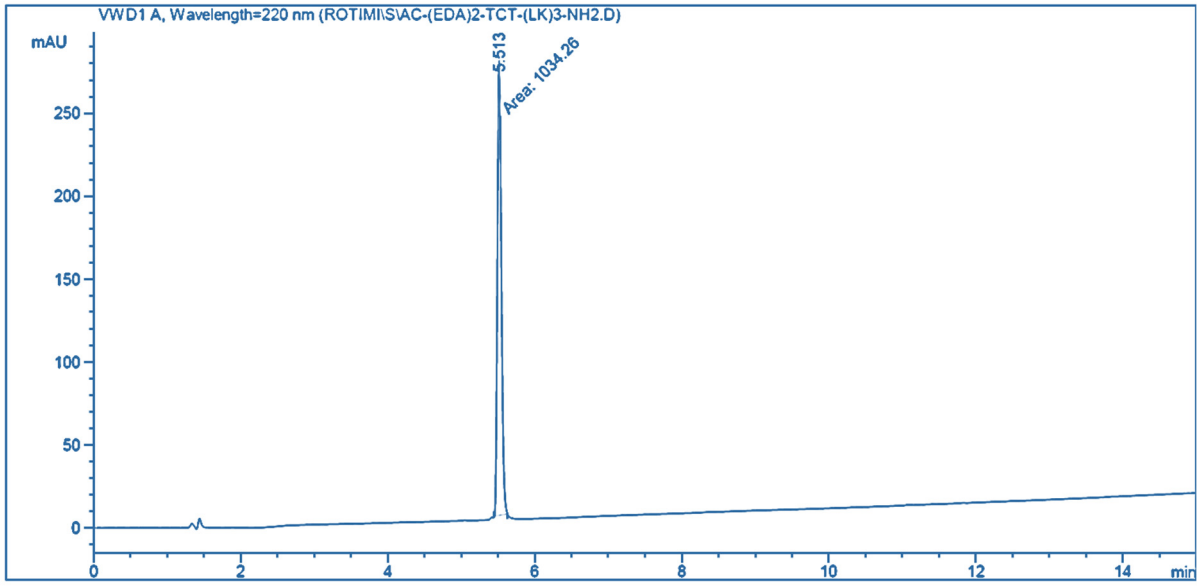


Figure S17: ESI-MS of **E**: ((Ac-EDA)$_2$-TA-(LK)$_3$-NH$_2$).
Expected mass= 1021.32
Observed mass = (511.30 [M+2H] +/2, 341.11 [M+3H] +/3, 256.12 [M+4H] +/4)

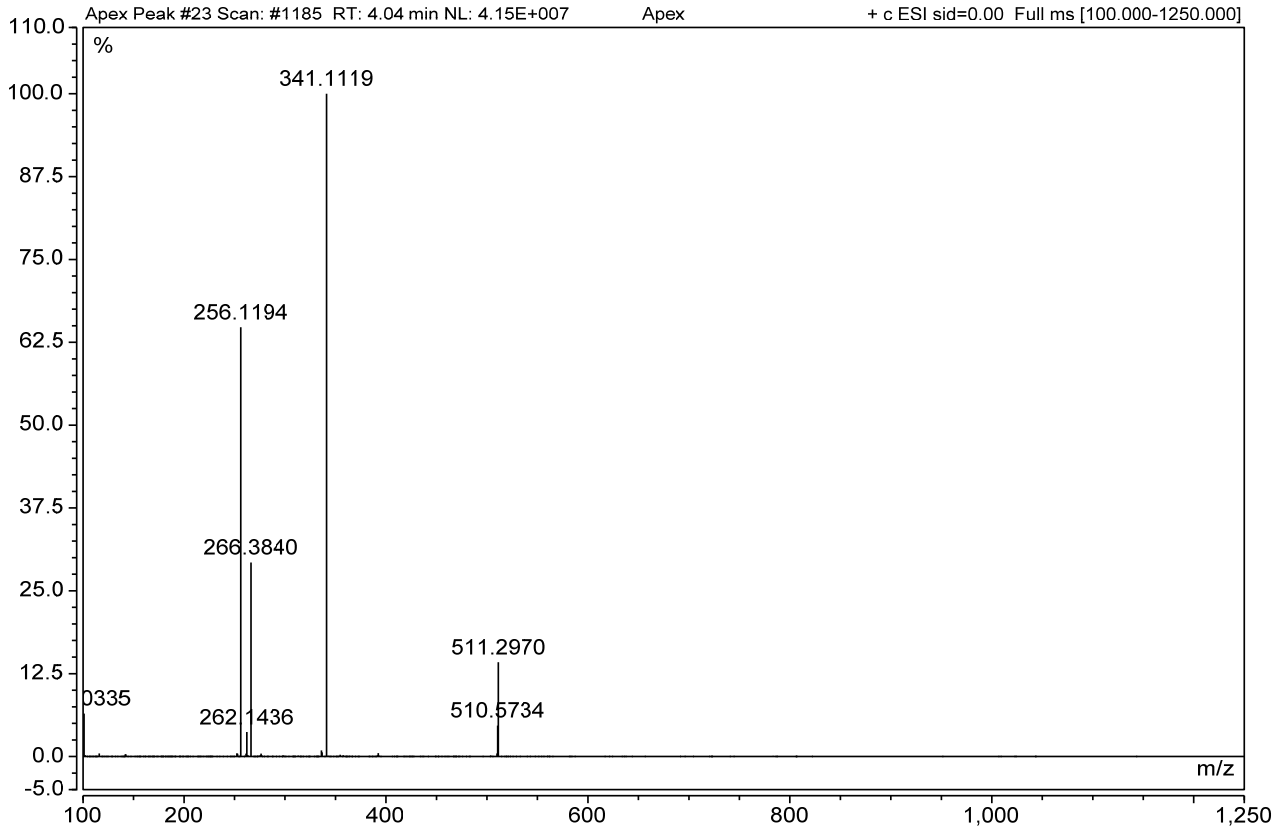


Figure S18: HPLC trace of purified **F** (H-[(LK)$_3$]$_2$-K-(LK)$_3$-NH$_2$).
Method 15-50% B into A in 15 min

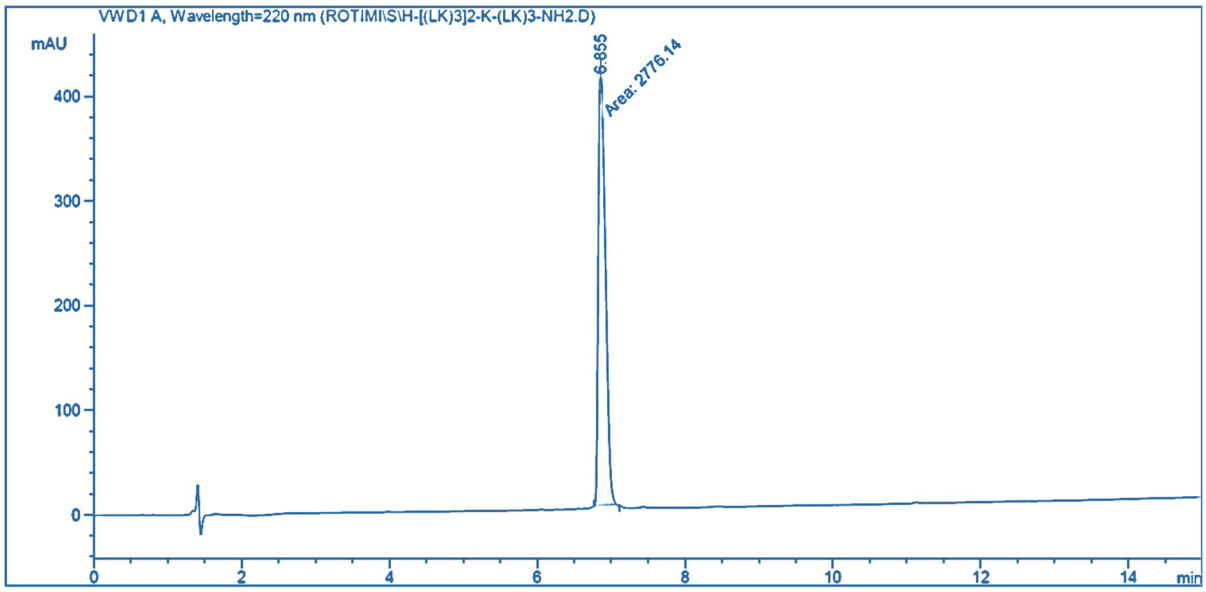


Figure S19: ESI-MS of **F**: (H-[(LK)$_3$]$_2$-K-(LK)$_3$-NH$_2$).
Expected mass= 2317.22
Observed mass = (580.38 $[M+4H]^+/4$, 464.49 $[M+5H]^+/5$, 387.24 $[M+6H]^+/6$)

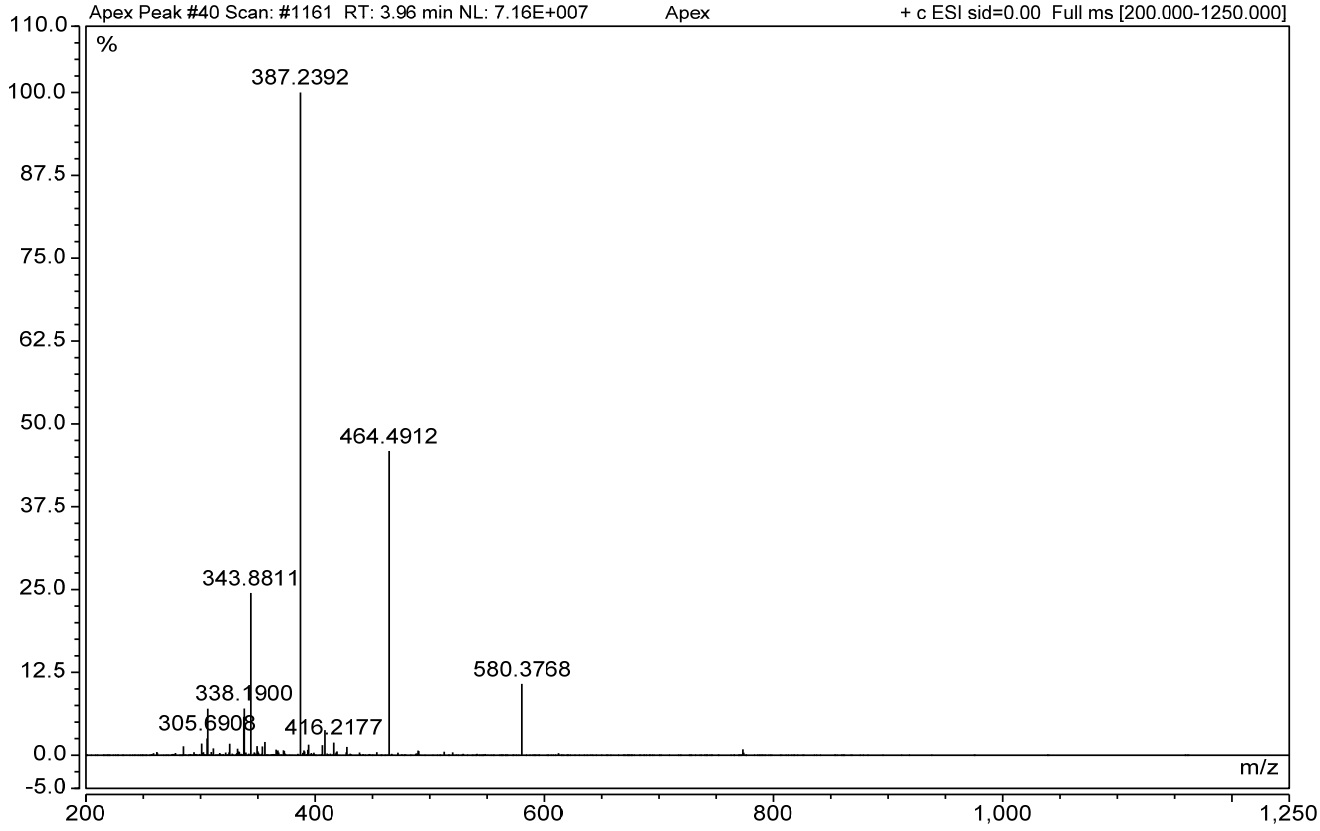


Figure S20: HPLC trace of purified **G** (H-(LK)$_9$-NH$_2$).
Method 15-50% B into A in 15 min

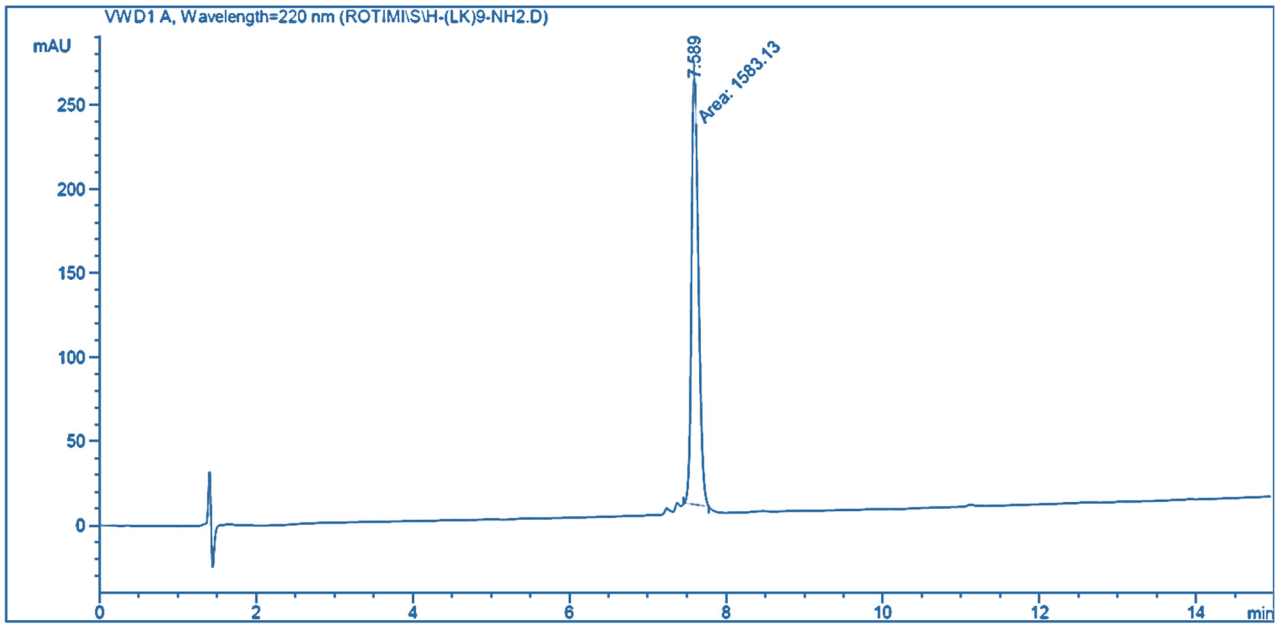


Figure S21: ESI-MS of **G**: (H-(LK)$_9$-NH$_2$).
Expected mass= 2189.05
Observed mass = (730.53 [M+3H] +/3, 548.33 [M+4H] +/4, 438.87 [M+5H] +/5 365.90 [M+6H] +/6)

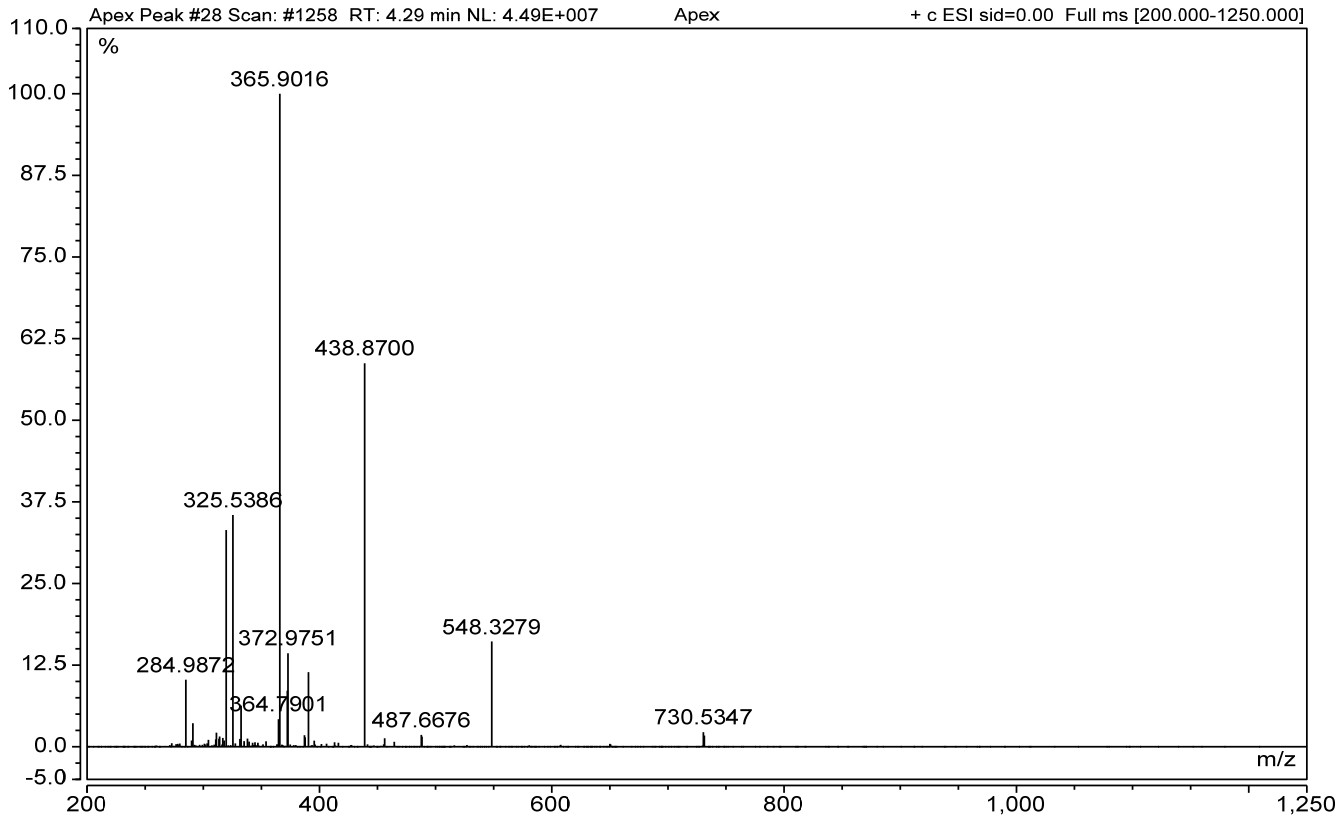
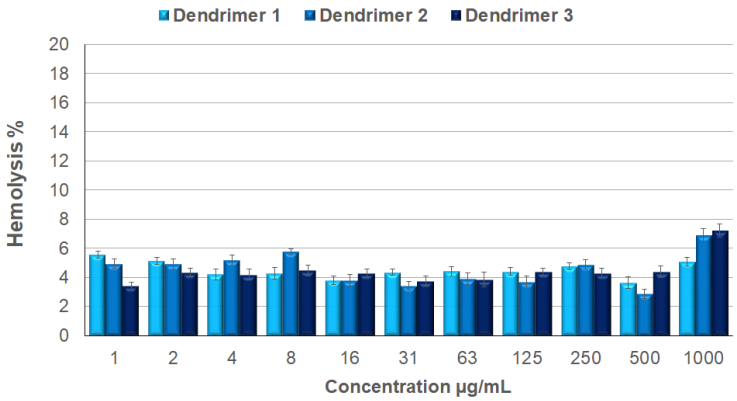


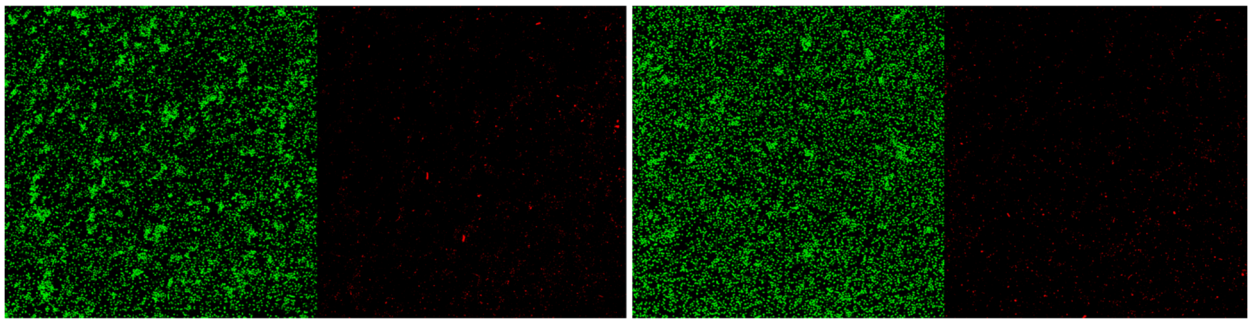
Figure S22: Hemolysis and cytotoxicity at 64µg/mL of dendrimers 1,2, and 3.



	% Cytotoxicity at 64µg/mL	
	L-929	HepG2
Dendrimer 1	2.8	0
Dendrimer 2	0	0
Dendrimer 3	0	0

Figure S23: Confocal light microscopy imaging after Live/Dead staining of *E. coli* 208691.

(A) **(B)**



(A) Untreated cells (*E. coli)* ; (B) *E. coli* treated with 0.25 µg/mL of AO and dendrimer 2 at MIC. Similar images were obtained for all the isolates and dendrimers included in the intracellular accumulation of acridine orange assay.