This is the supplementary material to the following article: Al-Dulaymi, M., and El-Aneed, A. (2017) Tandem mass spectrometric analysis of novel peptide-modified gemini surfactants used as gene delivery vectors. J. Mass Spectrom., 52: 353–366, which has been published in final form at doi: 10.1002/jms.3933.

Supplementary Material

Table S1. Mass accuracies of the triply charged ions $[M+H]^{3+}$ obtained during single stage ESI-QqToF-MS.

Gemini surfactants	Molecular formula	Theoretical, *m/z*	Observed, *m/z*	Mass accuracy (ppm)
12-7N(G-C_{11}-K_3)-12	$C_{65}H_{136}N_{11}O_5$	383.6903	383.6895	2.124
16-7N(G-C_{11}-K_3)-16	$C_{73}H_{152}N_{11}O_5$	421.0653	421.0633	4.945
16-7N(G-C_{11}-K)-16	$C_{61}H_{128}N_7O_3$	335.6687	335.6693	1.668
12-7N(G-C_6-K_3)-12	$C_{60}H_{126}N_{11}O_5$	360.3308	360.3344	5.555
16-7N(G-C_6-K_3)-16	$C_{68}H_{142}N_{11}O_5$	397.7059	397.7076	4.11
12-7N(G-C_6-K)-12	$C_{48}H_{101}N_7O_3$	274.9342	274.9356	4.887
16-7N(G-C_6-K)-16	$C_{56}H_{118}N_7O_3$	312.3093	312.3098	1.526
12-7N(C_{11}-K_3)-12	$C_{63}H_{133}N_{10}O_4$	364.6831	364.6828	0.988
16-7N(C_{11}-K_3)-16	$C_{71}H_{149}N_{10}O_4$	402.0582	402.0593	2.667
12-7N(C_6-K_3)-12	$C_{58}H_{123}N_{10}O_4$	341.3237	341.3257	5.73
16-7N(C_6-K_3)-16	$C_{66}H_{139}N_{10}O_4$	378.6988	378.7015	7.101

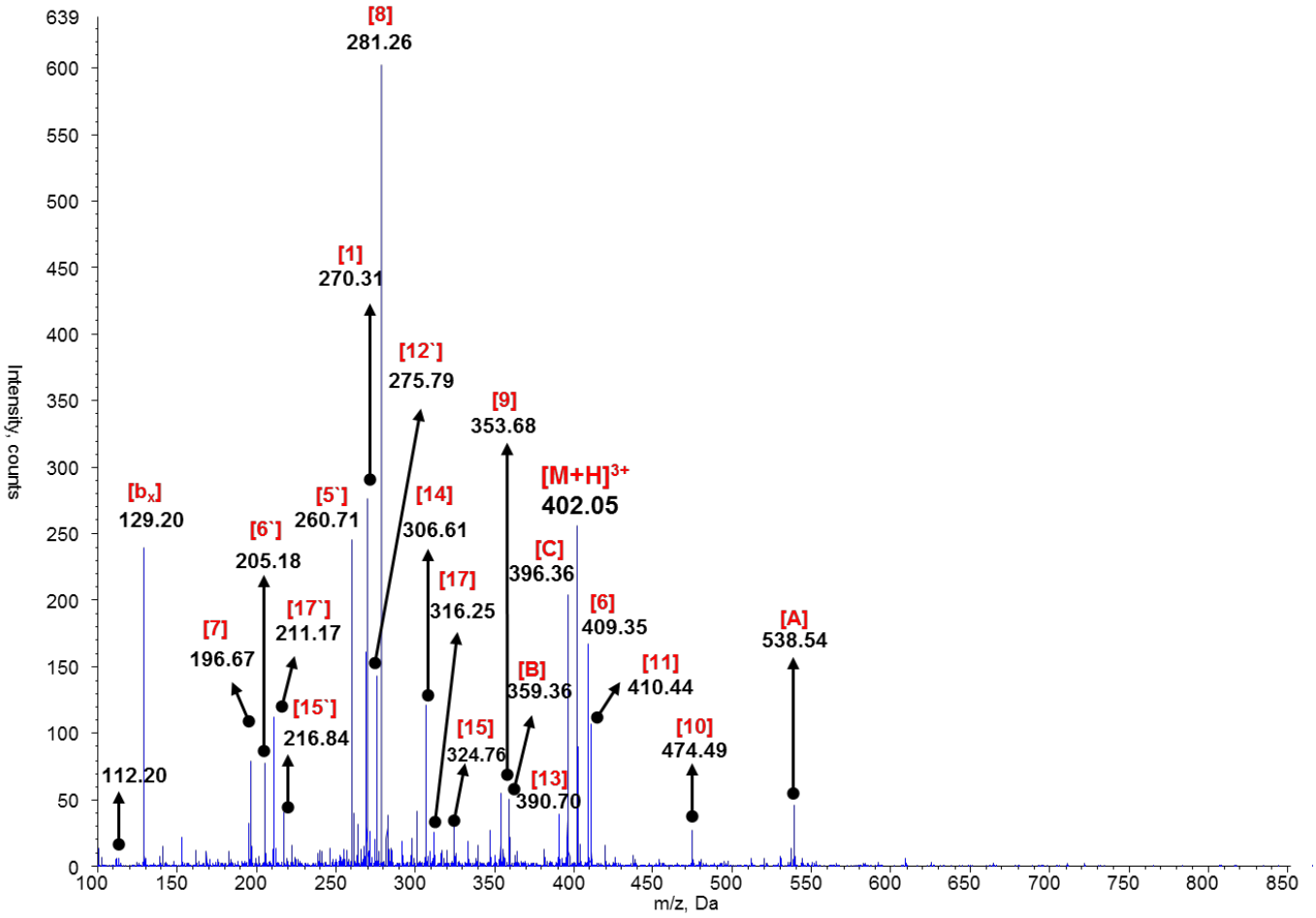

Figure S1. The ESI-QqToF MS/MS spectrum of 16-7N(C_{11}-K_3)-16 as a representative example of gemini surfactants with tri-terminal lysine moieties. Ions were labelled as designated in Figures 5-7.

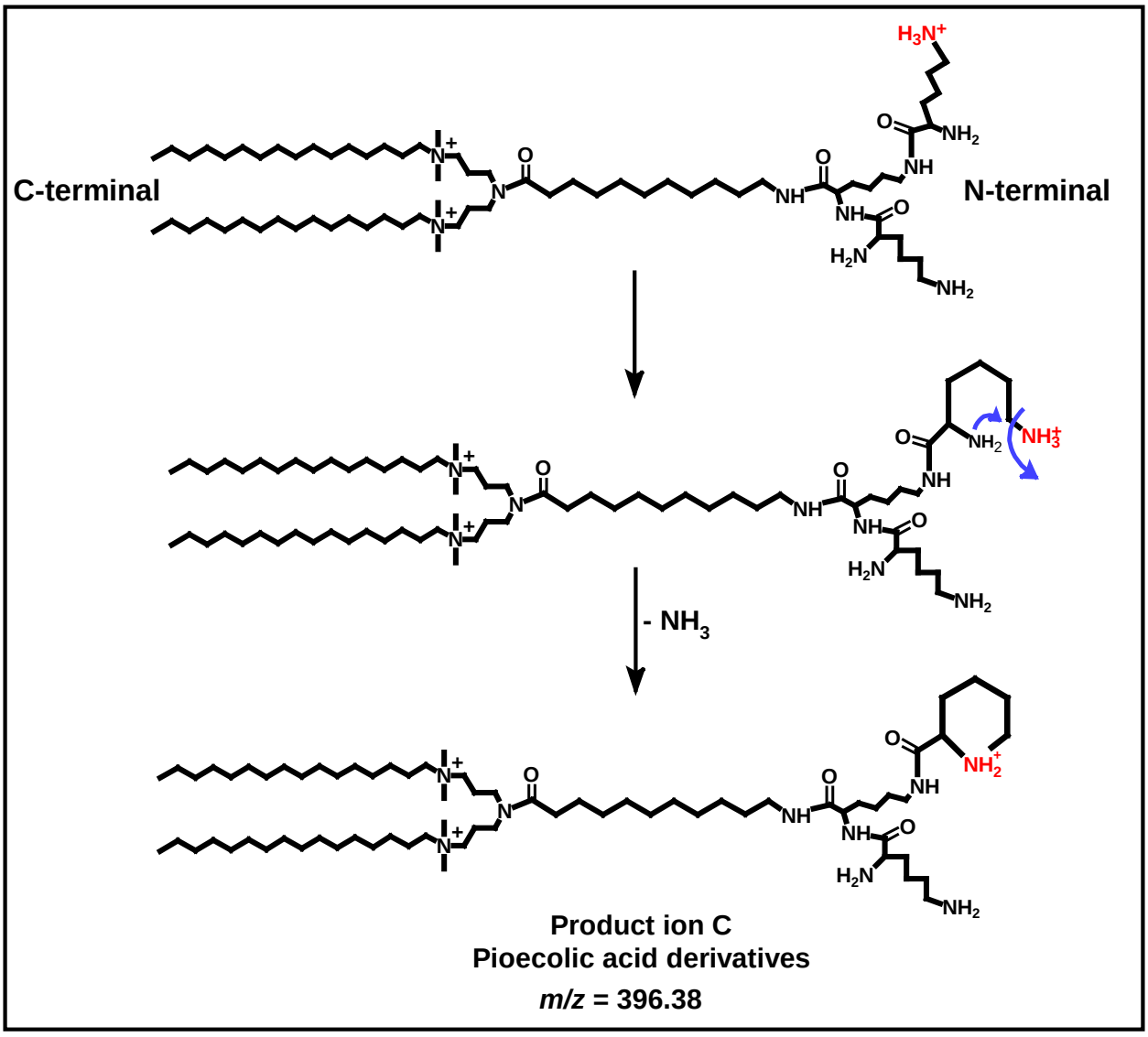

Figure S2. The proposed mechanism for the formation of product ion (C): loss of ammonia group from the lysine side chain.

Table S2. Summary of MS^3 analysis for 16-7N(C_{11}-K_3)-16 gemini surfactants.

Precursor ion	MS/MS product ions	Ms^3 product ions
402.05	538.54 [A]	806.76 [2], 403.88 [2`], 270.31 [1], 537.44 [3], 269.23 [3`], 129.10 [b_1], 678.66 [4], 339.83 [4`], 520.42 [5], 260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8]
	359.36 [B]	353.68 [9], 474.49 [10], 129.10 [b_1], 678.66 [4], 339.83 [4`], 270.31 [1], 410.44 [11], 550.57 [12], 275.79 [12`], 281.26 [8] , 270.31 [1]
	396.38 [C]	538.54 [A], 359.36 [B], 390.70 [13], 306.61 [14], 216.84 [15`], 324.76 [15], 300.94 [16], 316.25 [17], 211.17 [17`], 353.68 [9], 530.02 [9`], 789.73 [18], 395.37 [18`], 263.91 [18``], 474.49 [10], 537.44 [3], 269.23 [3`], 520.42 [5], 260.71 [5`], 270.31 [1], 129.10 [b_1]
	129.10 [b_1]	
	270.31 [1]	
	806.76 [2]	537.44 [3], 678.66 [4], 129.10 [b_1], 270.31 [1], 520.42 [5], 409.35 [6], 281.26 [8]
	403.88 [2`]	537.44 [3], 269.23 [3`],129.10 [$b_1$], 678.66 [4], 339.83 [4`], 270.31 [1], 520.42 [5], 260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8]
	537.44 [3]	520.42 [5], 409.35 [6], 281.26 [8]
	269.23 [3`]	260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1]
	678.66 [4]	409.35 [6], 281.26 [8], 129.10 [b_1], 270.31 [1]
	339.83 [4`]	409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1], 270.31 [1]
	520.42 [5]	409.35 [6], 281.26 [8]
	260.71 [5`]	409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1]
	409.35 [6]	281.26 [8]
	205.18 [6`]	196.67 [7], 281.26 [8], 129.10 [b_1]
	196.67 [7]	281.26 [8]
	281.26 [8]	

	353.68 [9]	474.49 [10], 678.66 [4], 339.83 [4`], 474.49 [10], 409.35 [6], 205.18 [6`], 129.10 [b_1], 270.31 [1], 196.67 [7], 281.26 [8], 395.37 [18`], 263.91 [18``], 520.42 [5], 260.71 [5`]
	530.02 [9`]	789.73 [18], 395.37 [18`], 520.42 [5], 260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8]
	474.49 [10]	678.66 [4], 339.83 [4`], 270.31 [1], 410.44 [11] , 409.35 [6], 205.18 [6`], 129.10 [b_1], 550.57 [12], 275.79 [12`], 281.26 [8]
	410.44 [11]	550.57 [12], 275.79 [12`], 270.31 [1], 281.26 [8]
	550.57 [12]	281.26 [8]
	275.79 [12`]	281.26 [8]
	390.70 [13]	300.94 [16], 316.25 [17], 211.17 [17`], 353.68 [9], 530.02 [9`], 789.73 [18], 395.37 [18`], 263.91 [18``], 474.49 [10], 520.42 [5], 260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1], 270.31 [1]
	306.61 [14]	216.84 [15`], 324.76 [15], 300.94 [16], 316.25 [17], 211.17 [17`], 395.37 [18`], 263.91 [18``], 520.42 [5], 260.71 [5`], 537.44 [3], 269.23 [3`],129.10 [b_1], 270.31 [1]
	324.76 [15]	316.25 [17], 537.44 [3], 520.42 [5], 409.35 [6], 281.26 [8] ,129.10 [b_1]
	216.84 [15`]	211.17 [17`], 269.23 [3`], 260.71 [5`], 129.10 [b_1]
	300.94 [16]	395.37 [18`], 263.91 [18``], 520.42 [5], 260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1], 270.31 [1]
	316.25 [17]	520.42 [5], 409.35 [6], 281.26 [8]
	211.17 [17`]	260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1]
	789.73 [18]	520.42 [5], 409.35 [6], 281.26 [8],
	395.37 [18`]	520.42 [5], 260.71 [5`], 409.35 [6], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1]
	263.91 [18``]	260.71 [5`], 205.18 [6`], 196.67 [7], 281.26 [8], 129.10 [b_1]

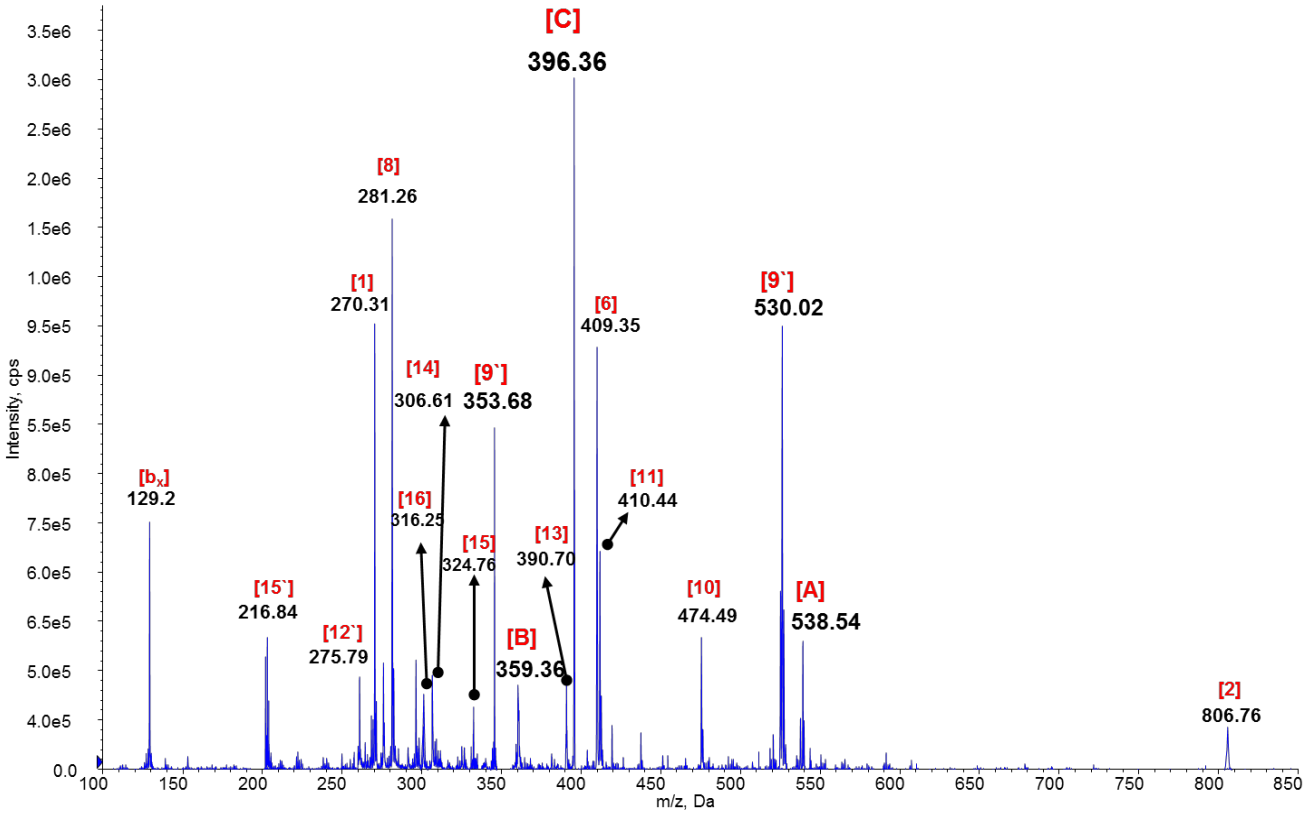

Figure S3. The ESI-QqLIT-MS^3 spectrum of product ion (C) at *m/z* 396.38 of 16-7N(C11-K3)-16 gemini surfactant. Ions were labelled as designated in Figures 7.

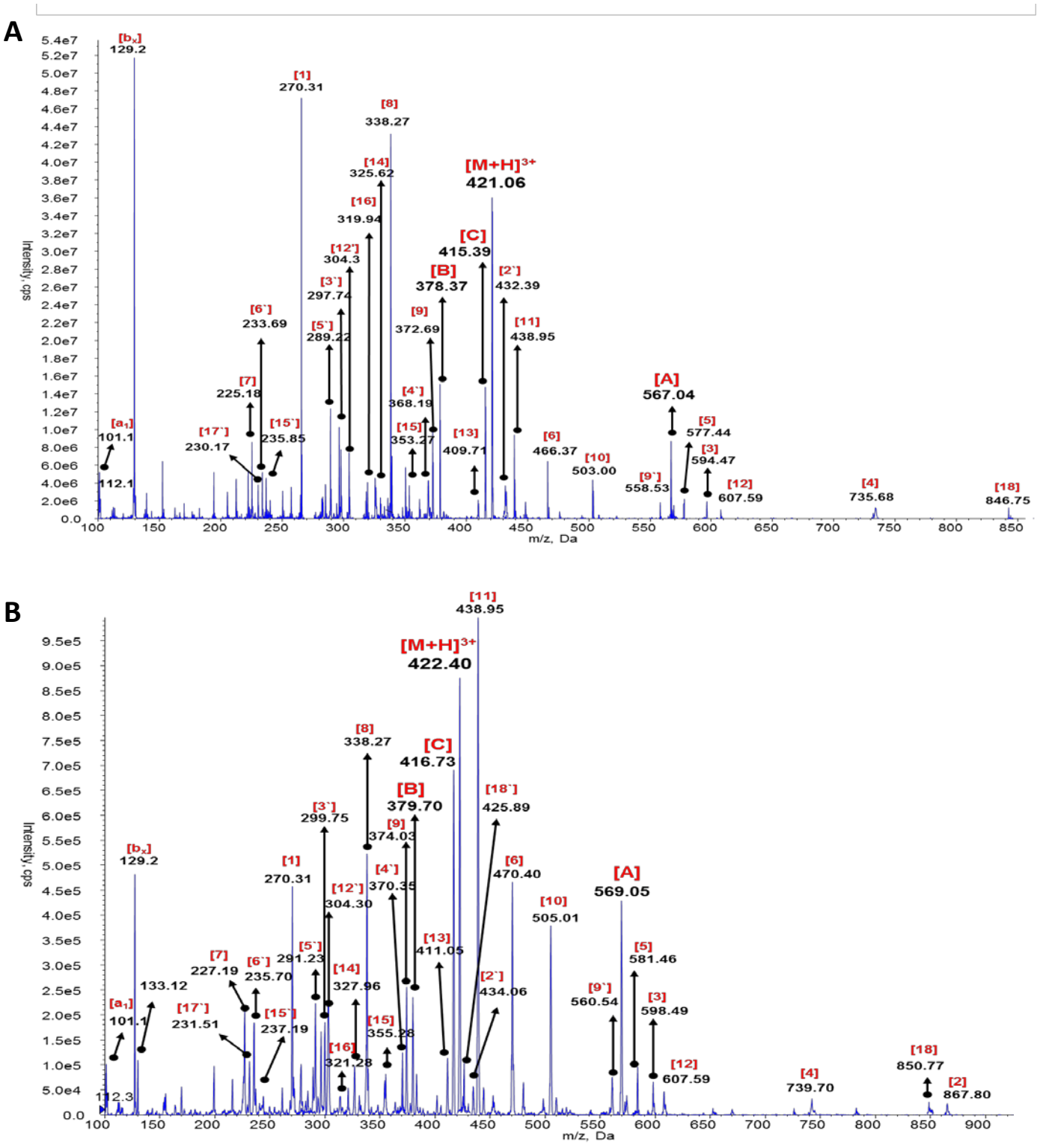

Figure S4. The ESI-QqLIT-MS/MS spectrum of (A) 16-7N(G-C_{11}-K_3)-16 and (B) its deuterated version 16-7N(G-C_{11}-K_D-K_2)-16 showing the similarities in the fragmentation patterns. Ions were labelled as designated in Figures 5-7.