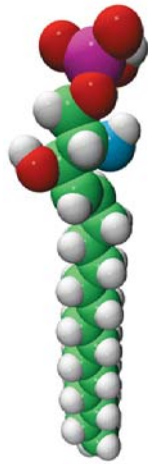
www.lipidmaps.org

LIPID MAPS Lipidomics Workshop

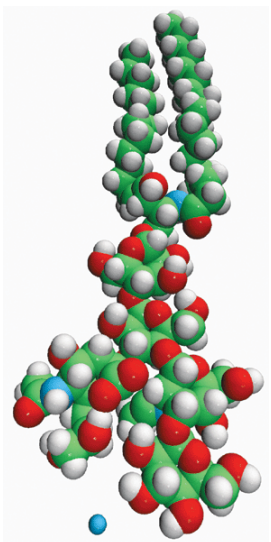
April 28, 2007



Novel Lipid Analysis

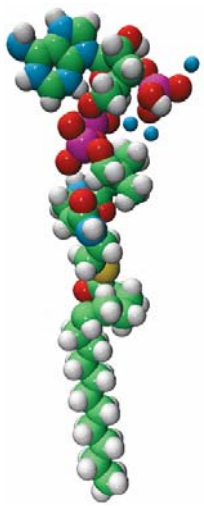
Teresa A. Garrett

Department of Biochemistry
Duke University School of Medicine
Durham, NC



Other LIPID MAPS Core K members:

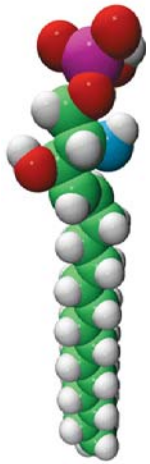
Chris Raetz
Ziaqiang Guan
Reza Kordestani
Andrea Ryan



www.lipidmaps.org

LIPID MAPS Lipidomics Workshop

April 28, 2007



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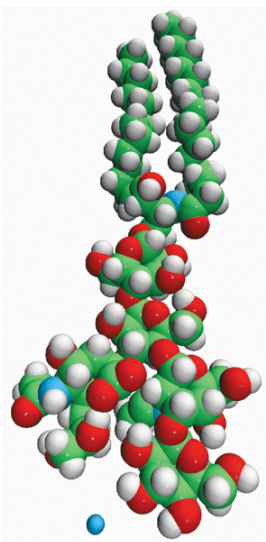
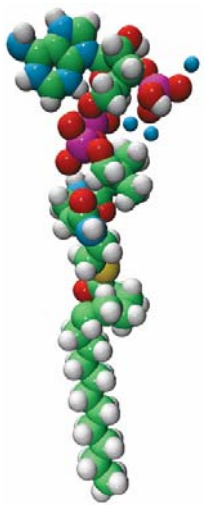


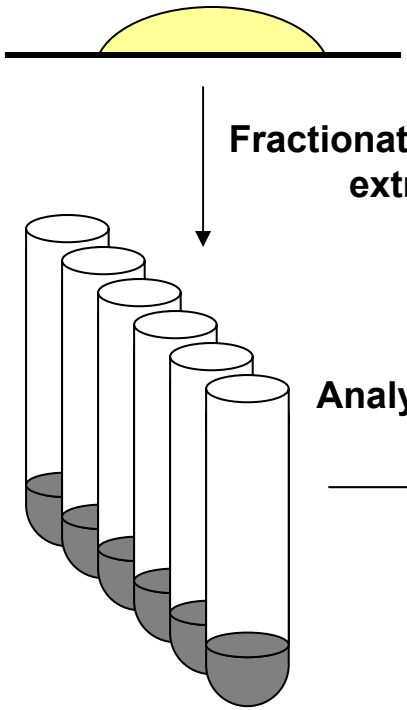
Other Core K Objectives

Quantification of cardiolipin, dolichols, and coenzyme Q

Methods in Enzymology. 2007. Volume 424.
"Lipidomics and Bioactive Lipids".
Edited by H. Alex Brown.

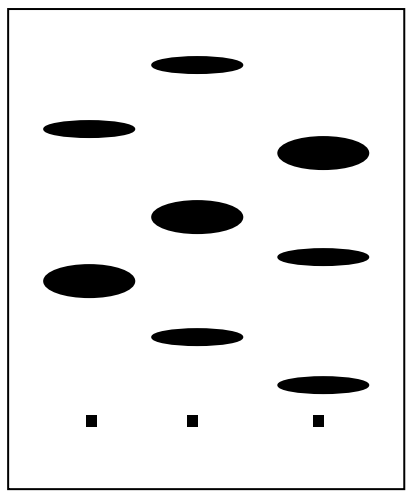


Identifying Novel Lipids

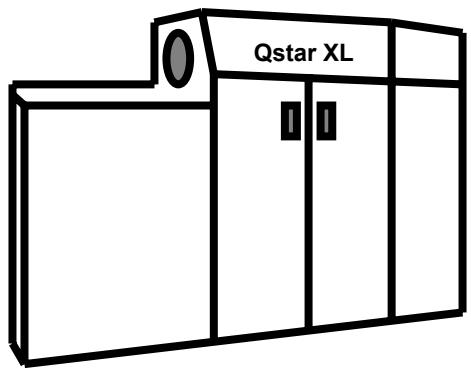


Fractionate gram quantities of total lipid extract on DEAE Cellulose

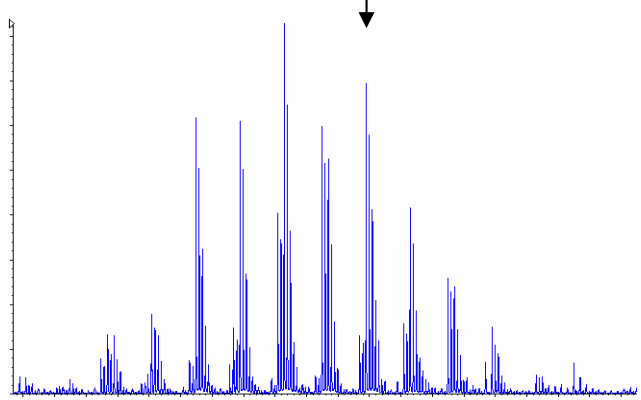
Analyze by TLC



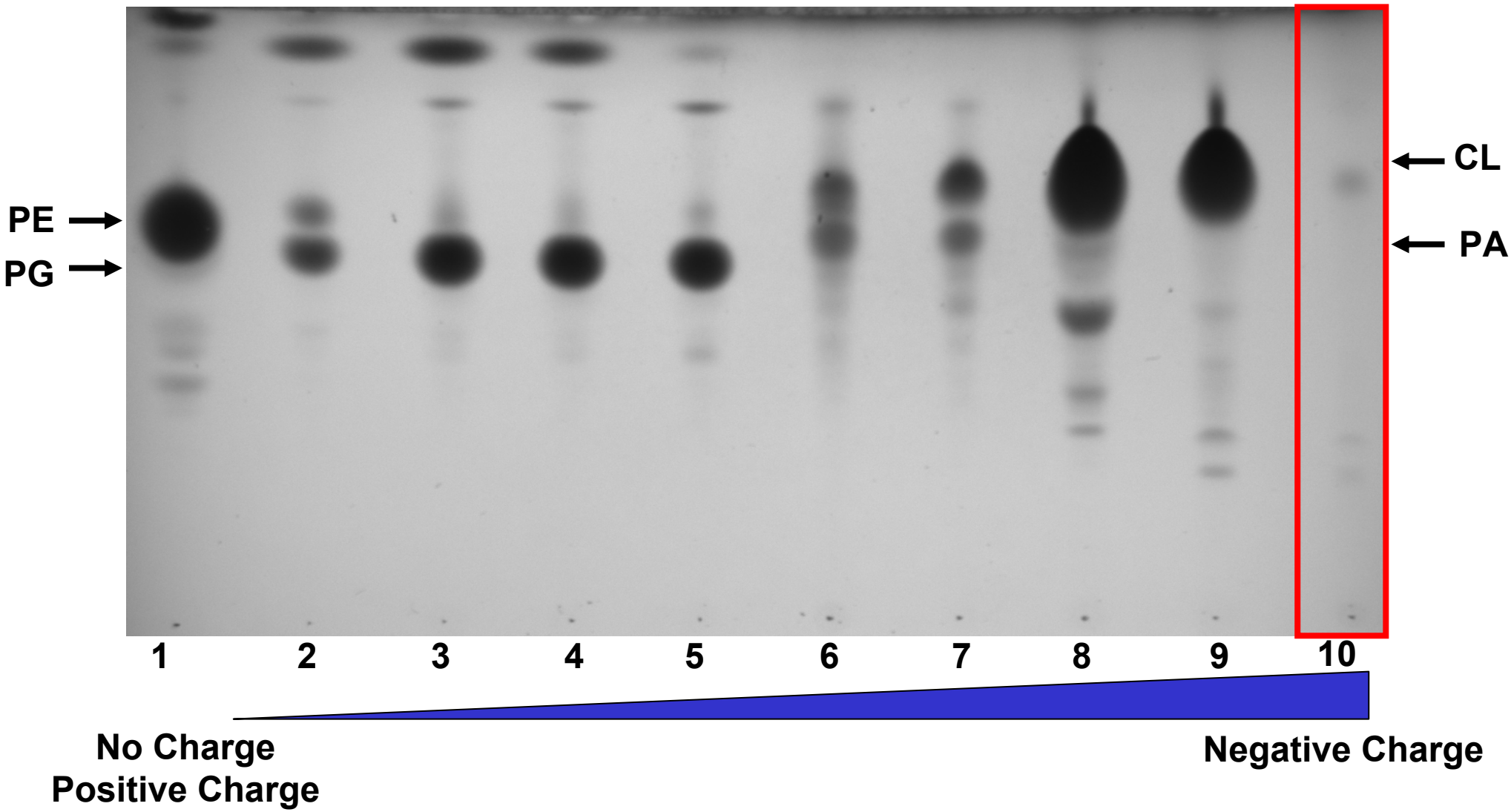
Prep TLC and mass spec



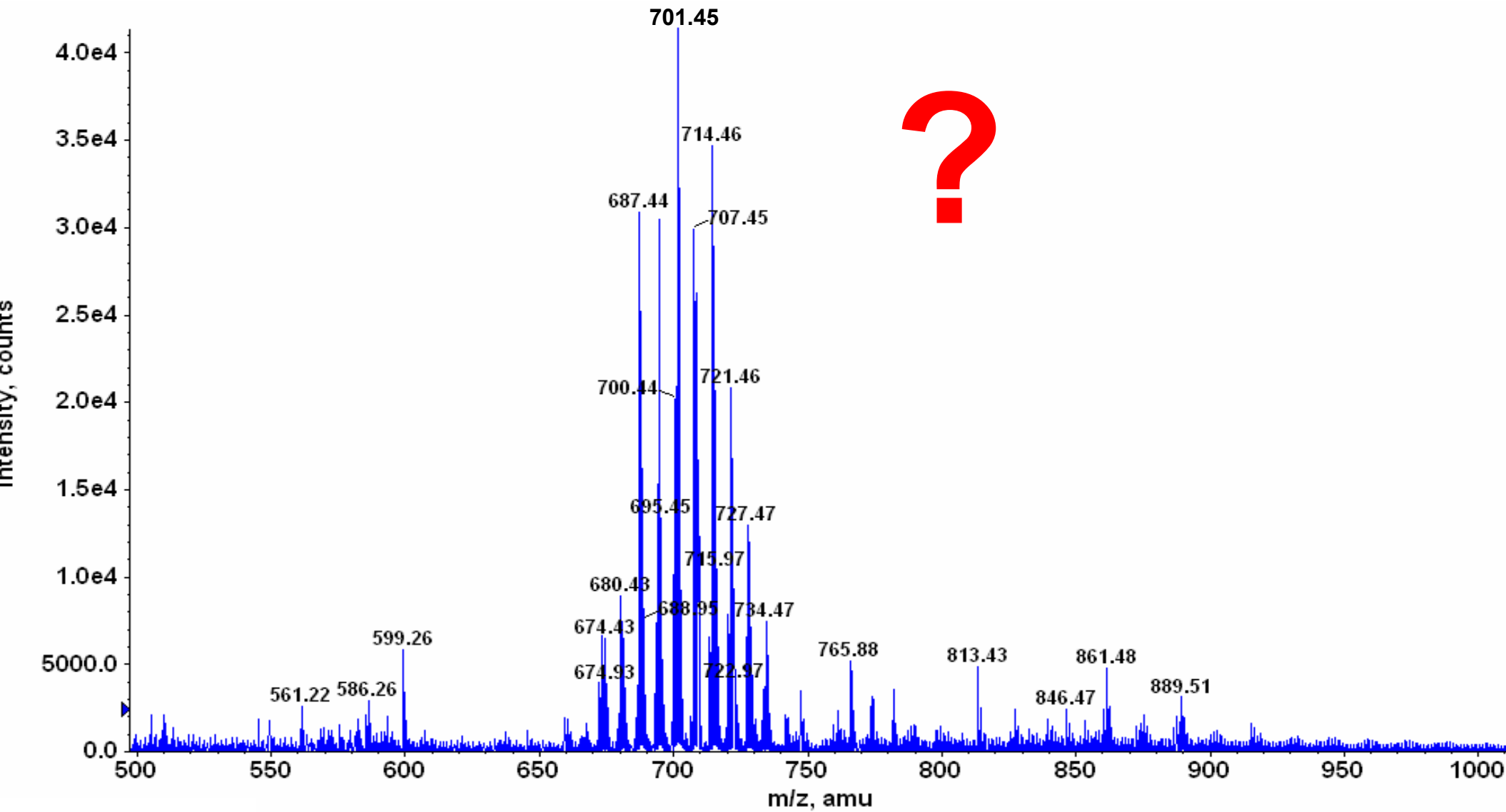
Analyze by MS, MSMS, and/or LCMS



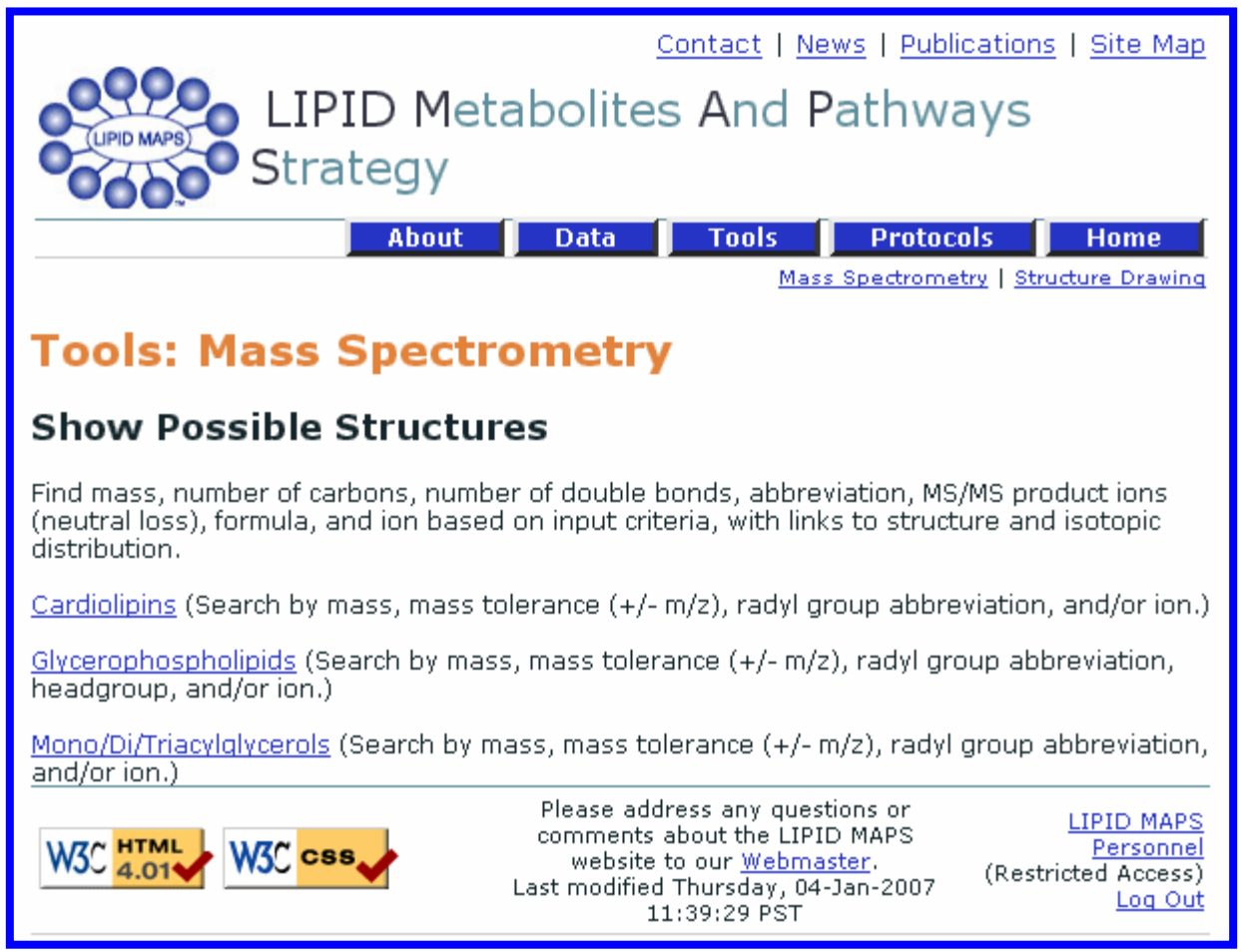
Anion Exchange Chromatography of *E. coli* Total Lipids



ESI-MS of Fraction #10

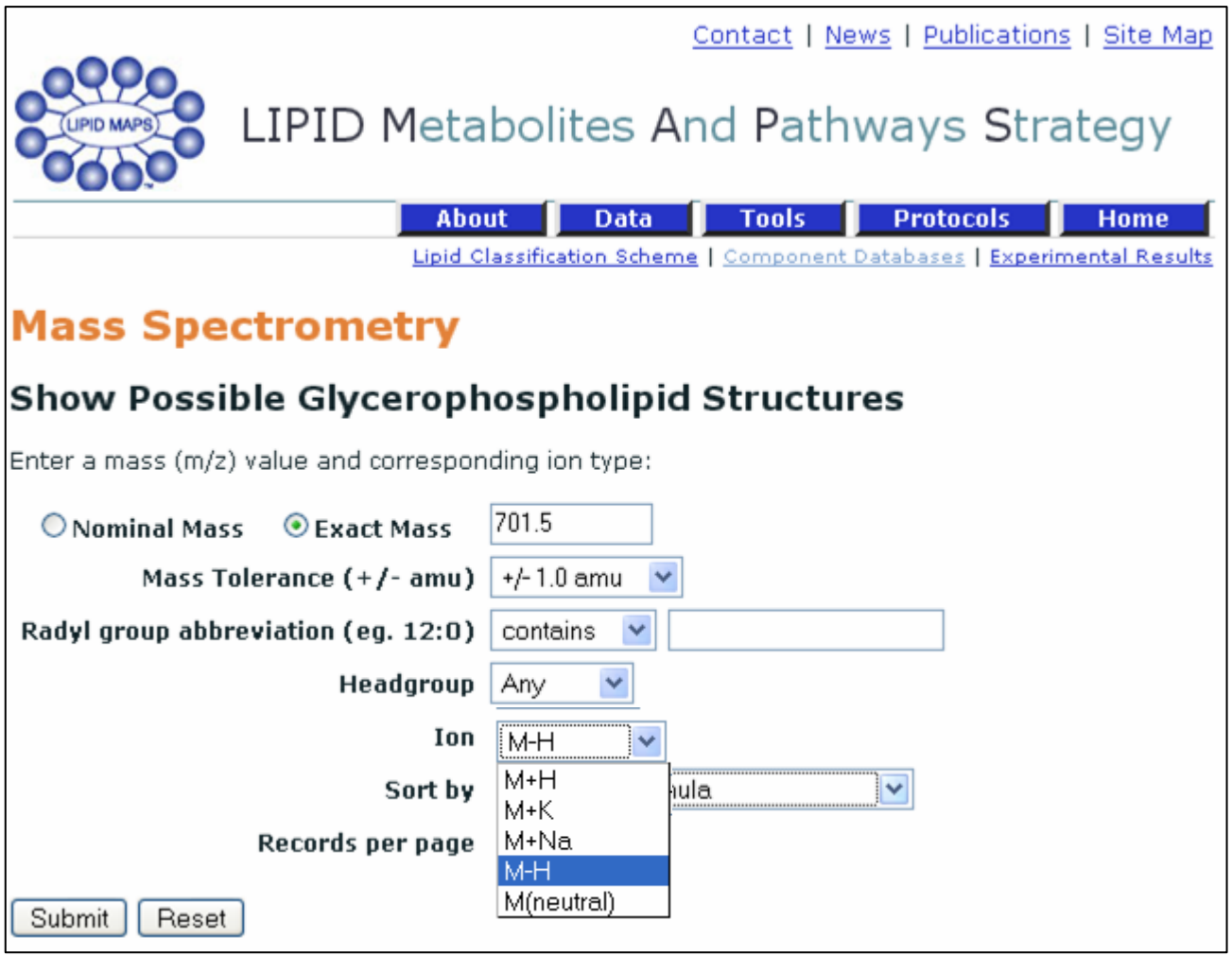


Lipid MAPS Mass Spectrometry Tools

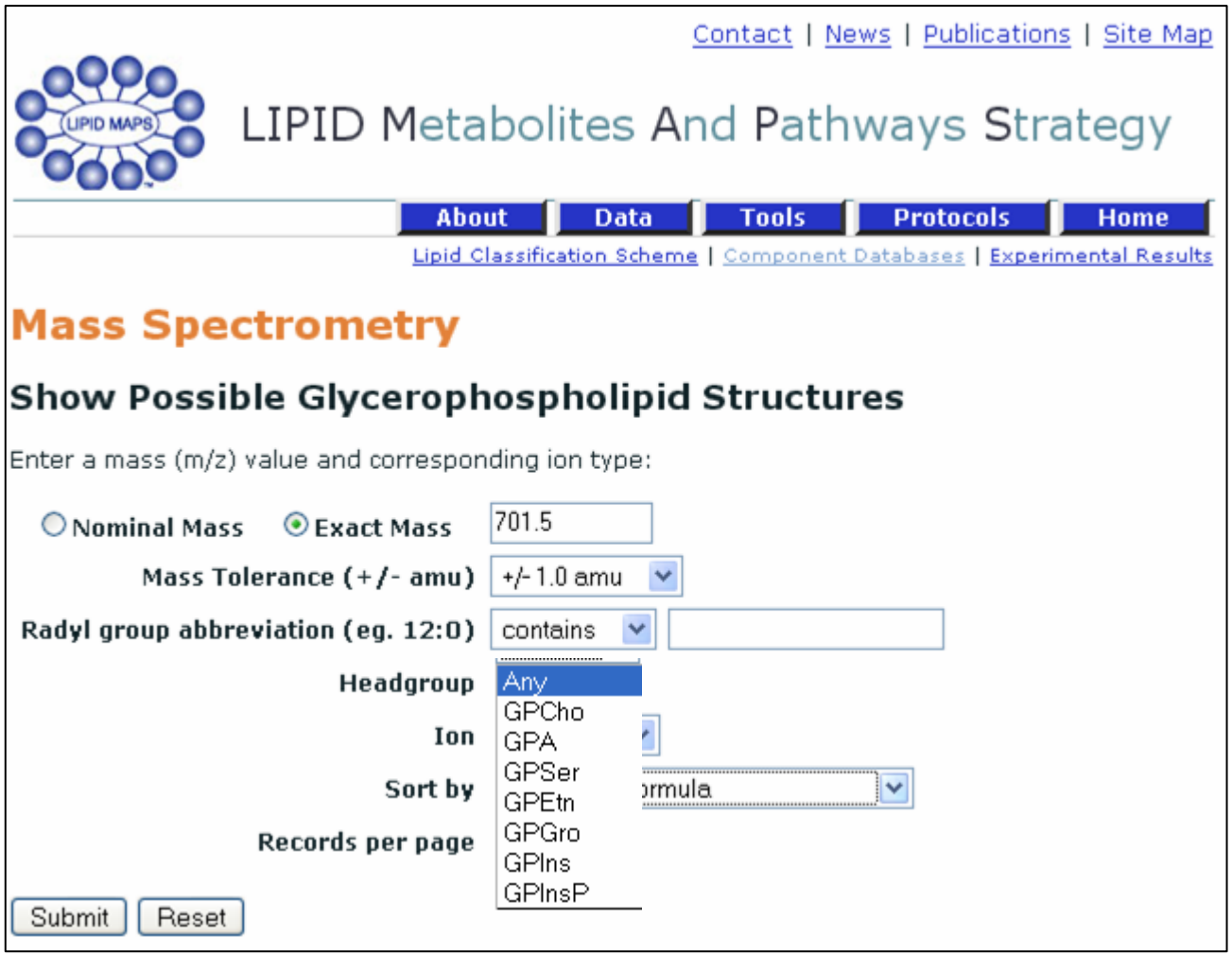


http://www.lipidmaps.org/tools/ms

Lipid MAPS Mass Spectrometry Tools



Lipid MAPS Mass Spectrometry Tools



Lipid MAPS Mass Spectrometry Tools

LIPID Metabolites And Pathways Strategy

Mass Spectrometry

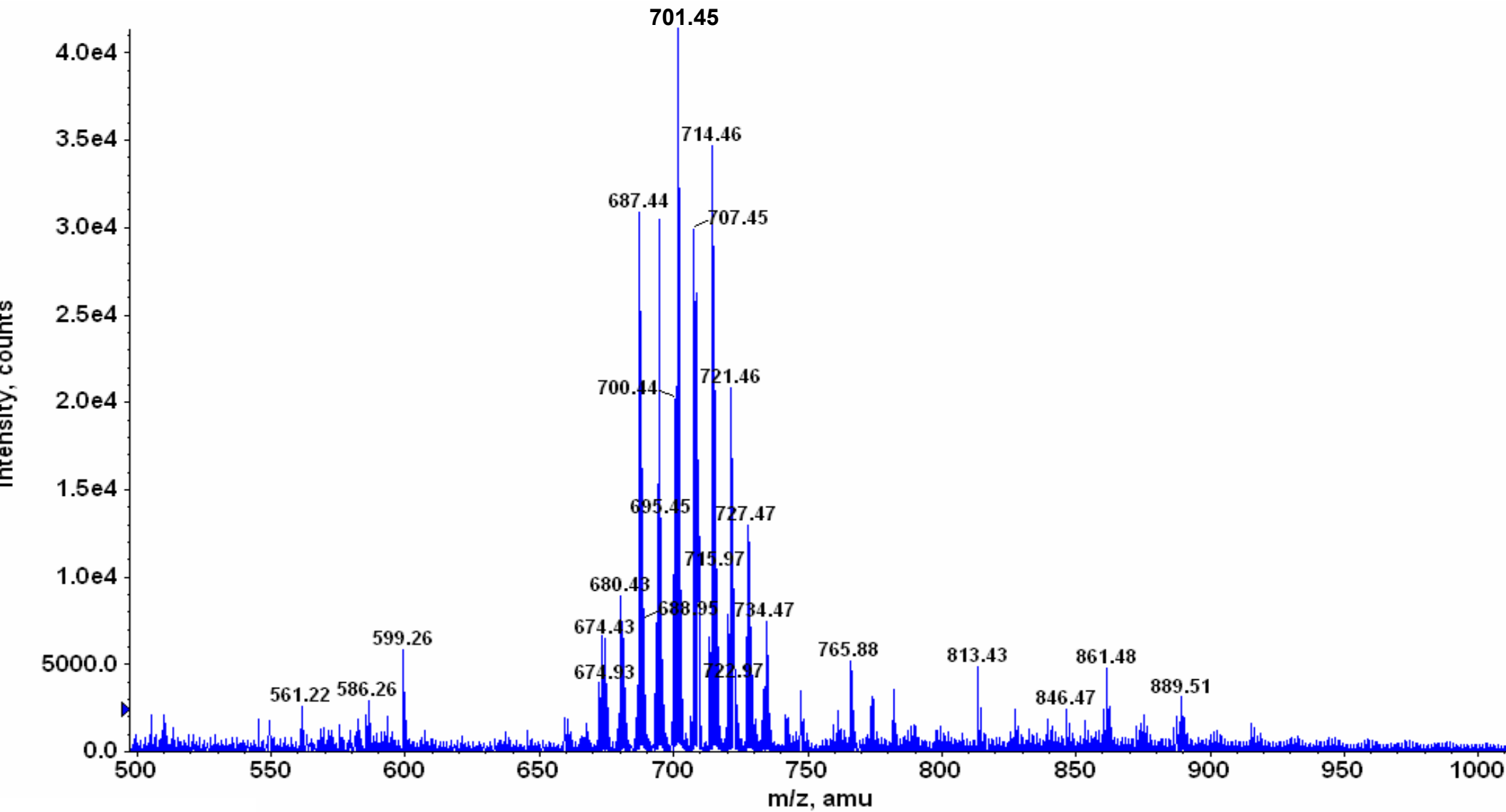
Possible Glycerophospholipid Structures

Exact Mass: 701.5 Mass Tolerance: +/- 1.0 amu Refine Search New Search

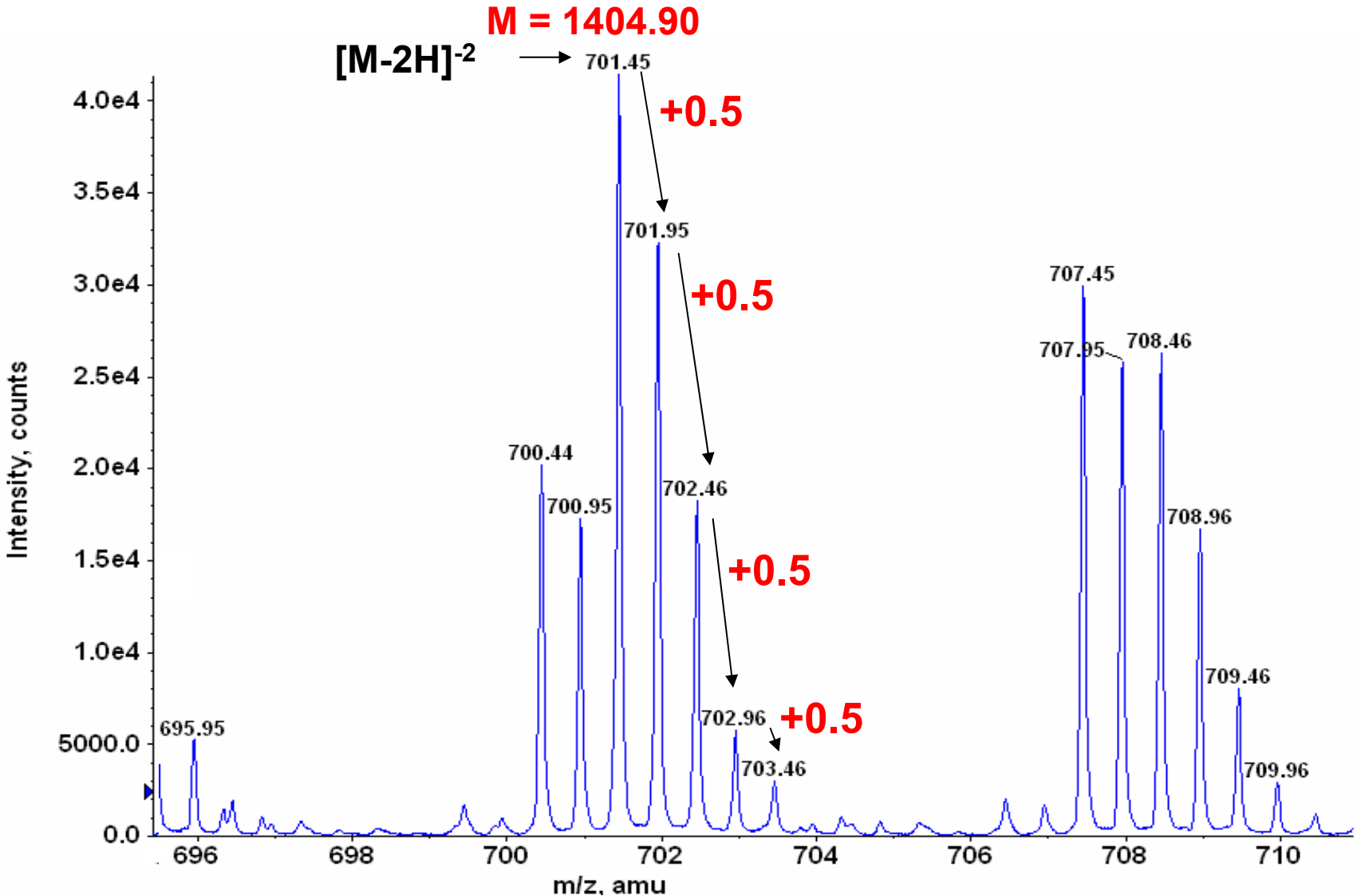
C=Number of Carbons; DB=Number of double bonds; sn1('1),sn2...=MS/MS Product Ions (neutral loss)

Mass	C	DB	Abbrev.	M-sn1-H	M-sn1-H2O-H	M-sn2-H	M-sn2-H2O-H	sn1 acid(-)	sn2 acid(-)	HG	Formula
701.2704	20	3	20:3(8Z,11Z,14Z)/0:0	413.0251	395.0145	687.2911	669.2805	305.2481	30.982	GPInsP	C29H52O15P2
702.4346	30	2	10:0/20:2(11Z,14Z)	548.2988	530.2882	412.1736	394.163	171.1385	307.2637	GPSer	C36H66NO10P
702.4346	30	2	12:0/18:2(9Z,12Z)	520.2675	502.2569	440.2049	422.1943	199.1698	279.2324	GPSer	C36H66NO10P
702.4346	30	2	13:0/17:2(9Z,12Z)	506.2519	488.2413	454.2206	436.21	213.1855	265.2168	GPSer	C36H66NO10P
702.4346	30	2	17:2(9Z,12Z)/13:0	454.2206	436.21	506.2519	488.2413	265.2168	213.1855	GPSer	C36H66NO10P
702.4346	30	2	14:1(9Z)/16:1(9Z)	494.2519	476.2413	466.2206	448.21	225.1855	253.2168	GPSer	C36H66NO10P
702.4346	30	2	16:1(9Z)/14:1(9Z)	466.2206	448.21	494.2519	476.2413	253.2168	225.1855	GPSer	C36H66NO10P
702.4346	30	2	18:2(9Z,12Z)/12:0	440.2049	422.1943	520.2675	502.2569	279.2324	199.1698	GPSer	C36H66NO10P
702.4346	30	2	20:2(11Z,14Z)/10:0	412.1736	394.163	548.2988	530.2882	307.2637	171.1385	GPSer	C36H66NO10P
701.4394	31	3	13:0/18:3(9Z,12Z,15Z)	505.2567	487.2461	441.2254	423.2148	213.1855	277.2168	GPGro	C37H67O10P
701.4394	31	3	18:3(9Z,12Z,15Z)/13:0	441.2254	423.2148	505.2567	487.2461	277.2168	213.1855	GPGro	C37H67O10P
701.5122	36	1	14:0/22:1(13Z)	491.3138	473.3032	381.2043	363.1937	227.2011	337.3107	GPA	C39H75O8P
701.5122	36	1	19:0/17:1(9Z)	421.2356	403.225	451.2825	433.2719	297.2794	267.2324	GPA	C39H75O8P
701.5122	36	1	17:1(9Z)/19:0	451.2825	433.2719	421.2356	403.225	267.2324	297.2794	GPA	C39H75O8P
701.5122	36	1	14:1(9Z)/22:0	493.3295	475.3189	379.1886	361.178	225.1855	339.3263	GPA	C39H75O8P

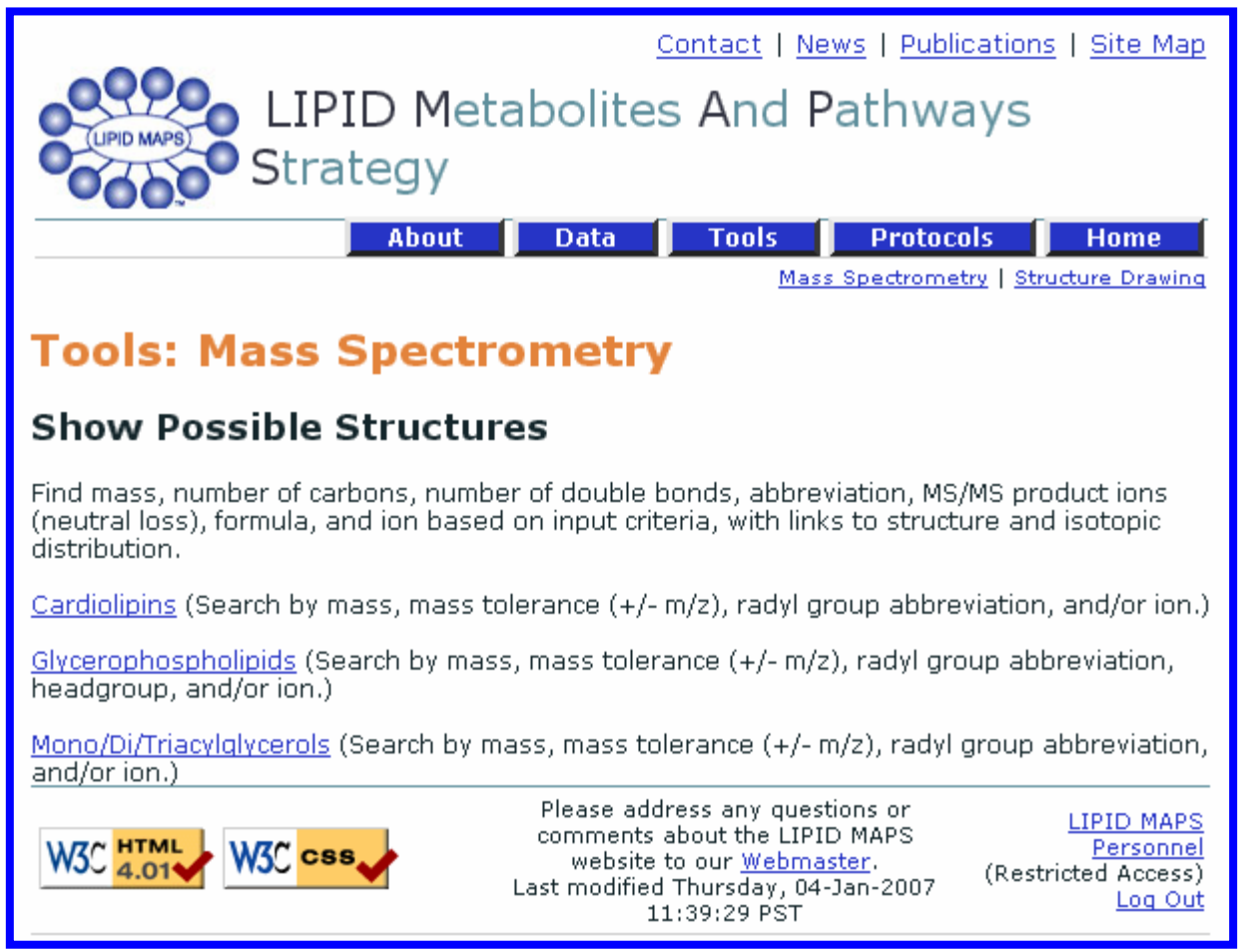
ESI-MS of Fraction #10



Major Ions of Fraction #10

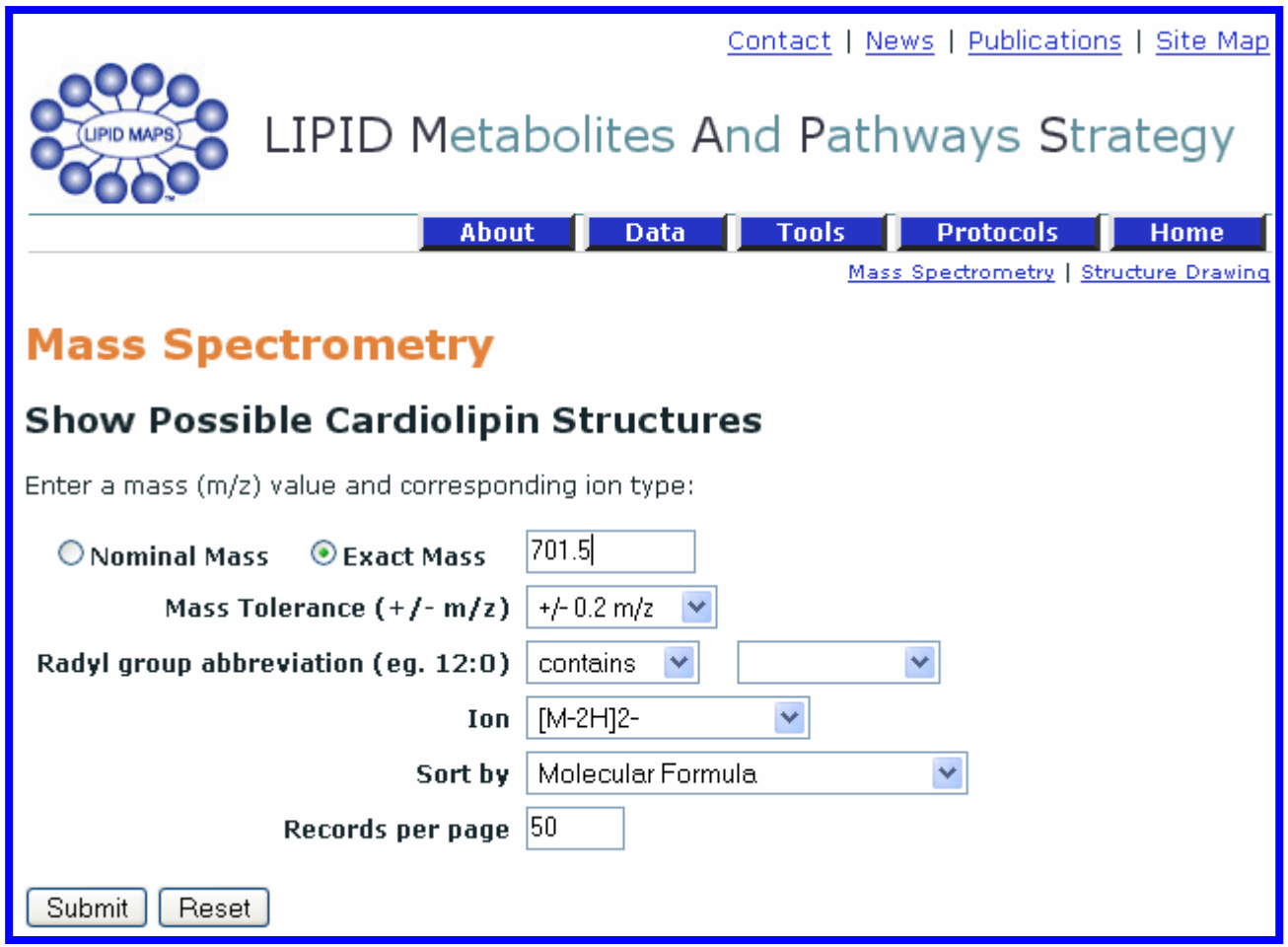


Lipid MAPS Mass Spectrometry Tools

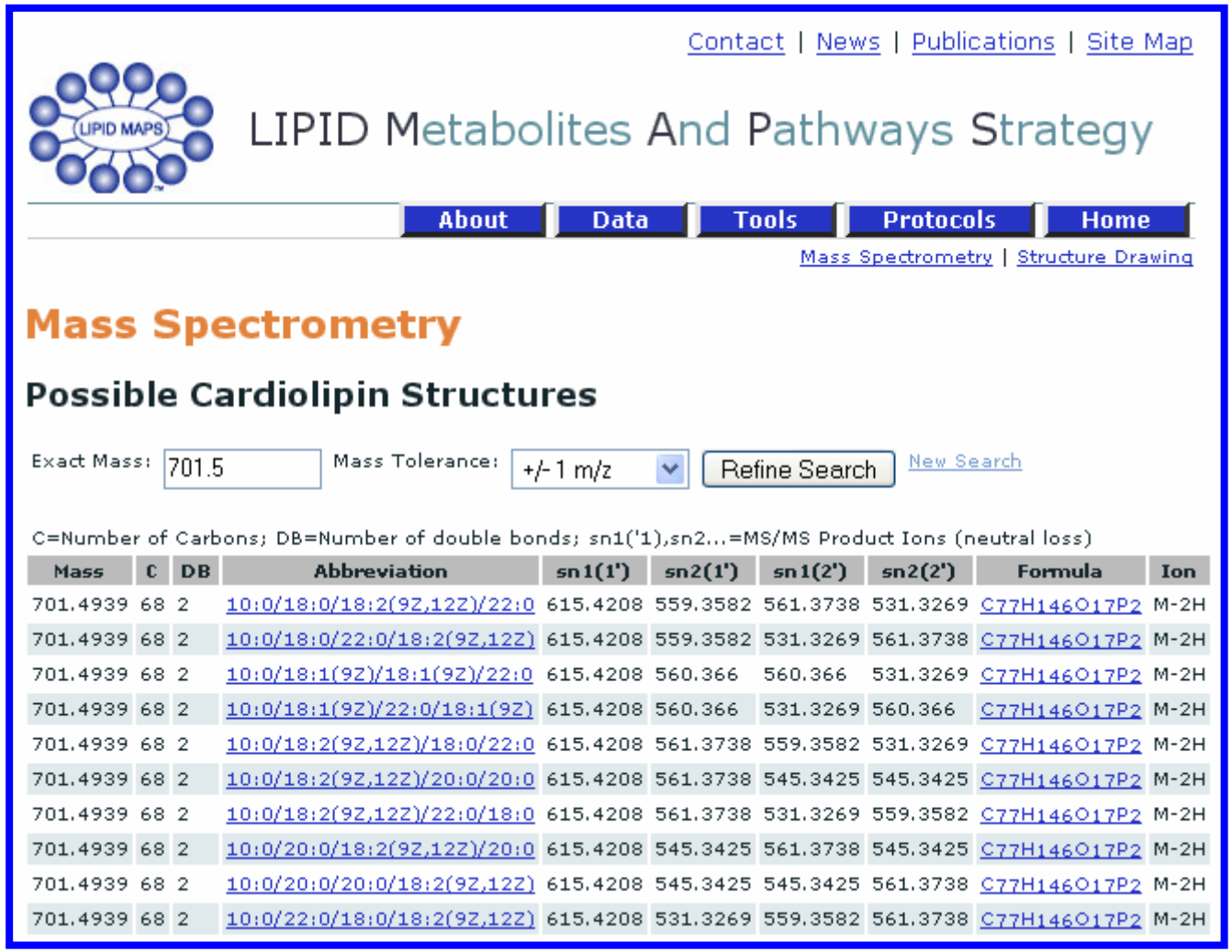


http://www.lipidmaps.org/tools/ms

Lipid MAPS Mass Spectrometry Tools



Lipid MAPS Mass Spectrometry Tools



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LIPID Metabolites And Pathways Strategy

About | Data | Tools | Protocols | Home

Mass Spectrometry | Structure Drawing

Mass Spectrometry

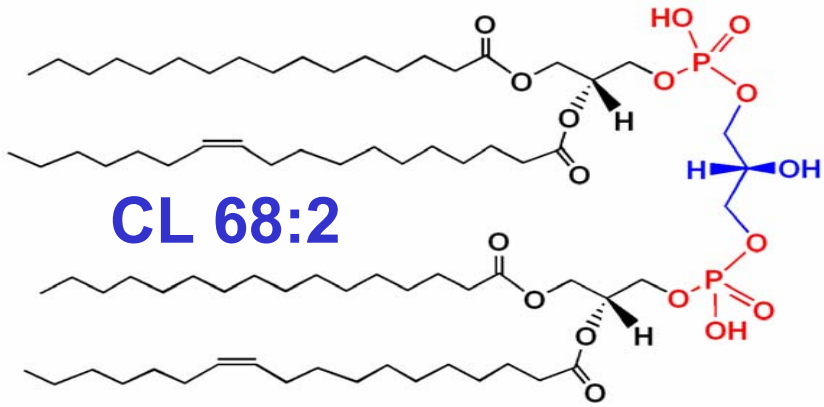
Possible Cardiolipin Structures

Exact Mass: 701.5 Mass Tolerance: +/- 1 m/z Refine Search New Search

C=Number of Carbons; DB=Number of double bonds; sn1('1),sn2...=MS/MS Product Ions (neutral loss)

Mass	C	DB	Abbreviation	sn1(1')	sn2(1')	sn1(2')	sn2(2')	Formula	Ion
701.4939	68	2	10:0/18:0/18:2(9Z,12Z)/22:0	615.4208	559.3582	561.3738	531.3269	C77H146O17P2	M-2H
701.4939	68	2	10:0/18:0/22:0/18:2(9Z,12Z)	615.4208	559.3582	531.3269	561.3738	C77H146O17P2	M-2H
701.4939	68	2	10:0/18:1(9Z)/18:1(9Z)/22:0	615.4208	560.366	560.366	531.3269	C77H146O17P2	M-2H
701.4939	68	2	10:0/18:1(9Z)/22:0/18:1(9Z)	615.4208	560.366	531.3269	560.366	C77H146O17P2	M-2H
701.4939	68	2	10:0/18:2(9Z,12Z)/18:0/22:0	615.4208	561.3738	559.3582	531.3269	C77H146O17P2	M-2H
701.4939	68	2	10:0/18:2(9Z,12Z)/20:0/20:0	615.4208	561.3738	545.3425	545.3425	C77H146O17P2	M-2H
701.4939	68	2	10:0/18:2(9Z,12Z)/22:0/18:0	615.4208	561.3738	531.3269	559.3582	C77H146O17P2	M-2H
701.4939	68	2	10:0/20:0/18:2(9Z,12Z)/20:0	615.4208	545.3425	561.3738	545.3425	C77H146O17P2	M-2H
701.4939	68	2	10:0/20:0/20:0/18:2(9Z,12Z)	615.4208	545.3425	545.3425	561.3738	C77H146O17P2	M-2H
701.4939	68	2	10:0/22:0/18:0/18:2(9Z,12Z)	615.4208	531.3269	559.3582	561.3738	C77H146O17P2	M-2H

Cardiolipin Ions

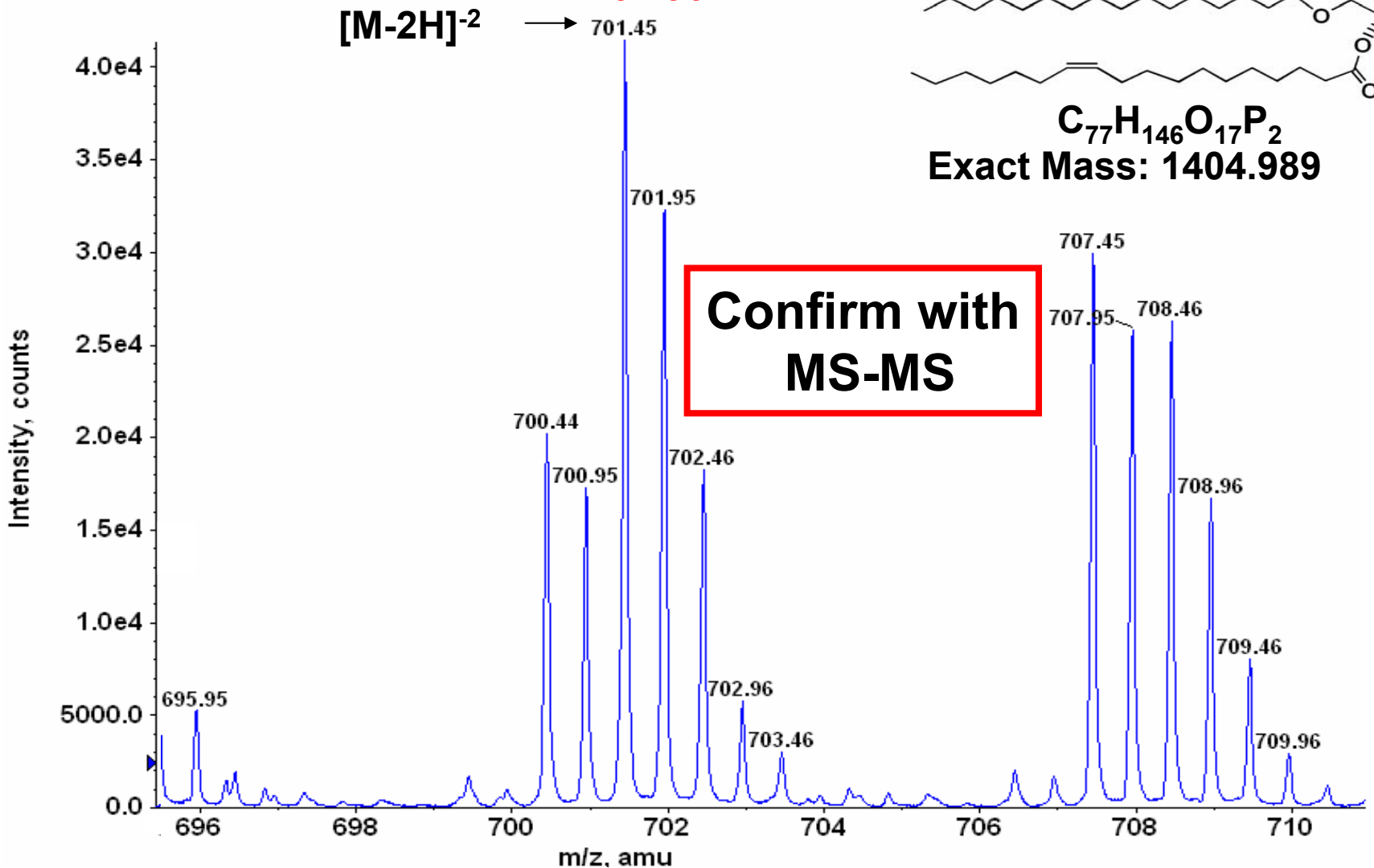


$C_{77}H_{146}O_{17}P_2$

Exact Mass: 1404.989

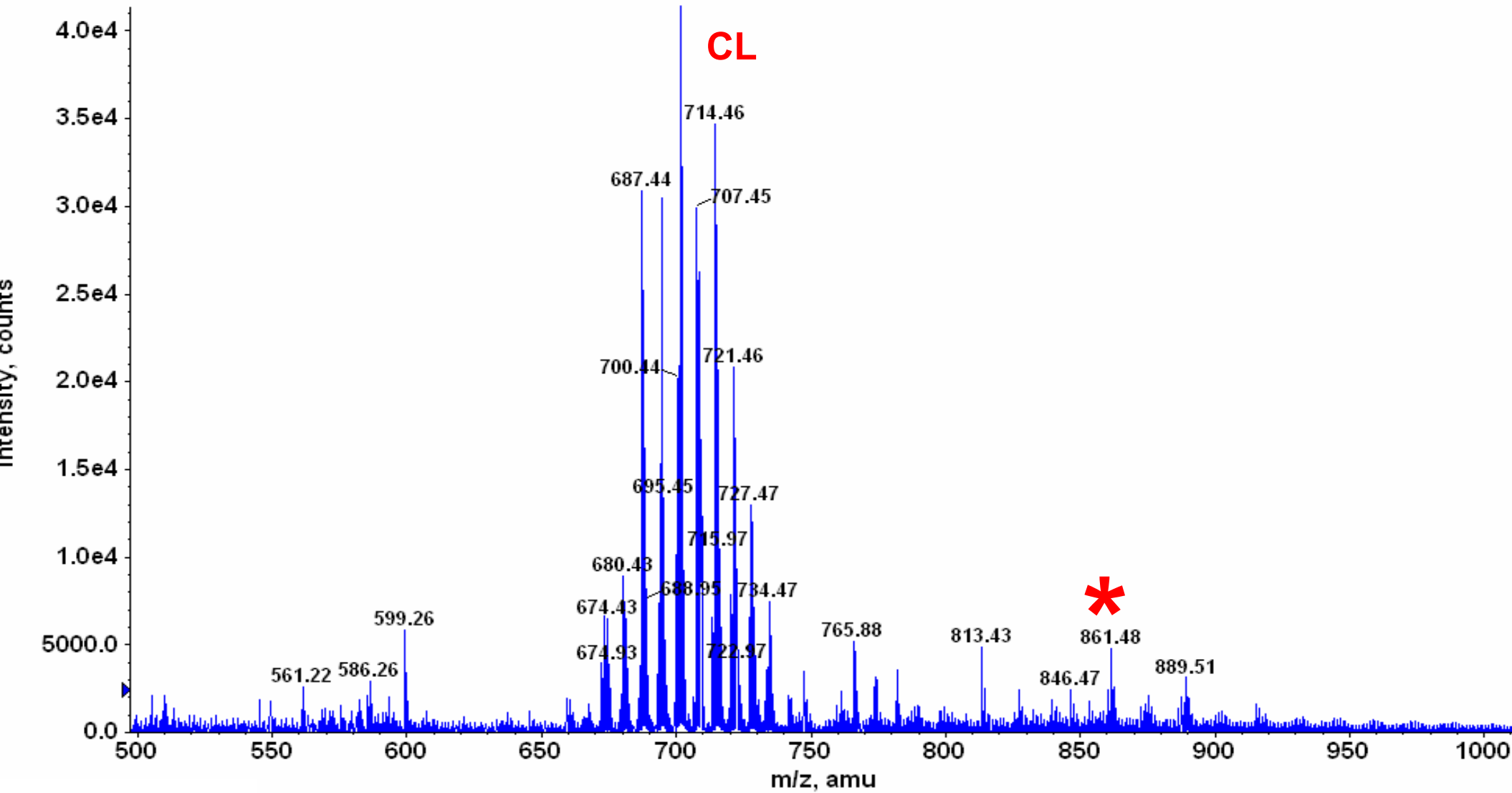
M = 1404.90

$[M-2H]^{-2}$ → 701.45

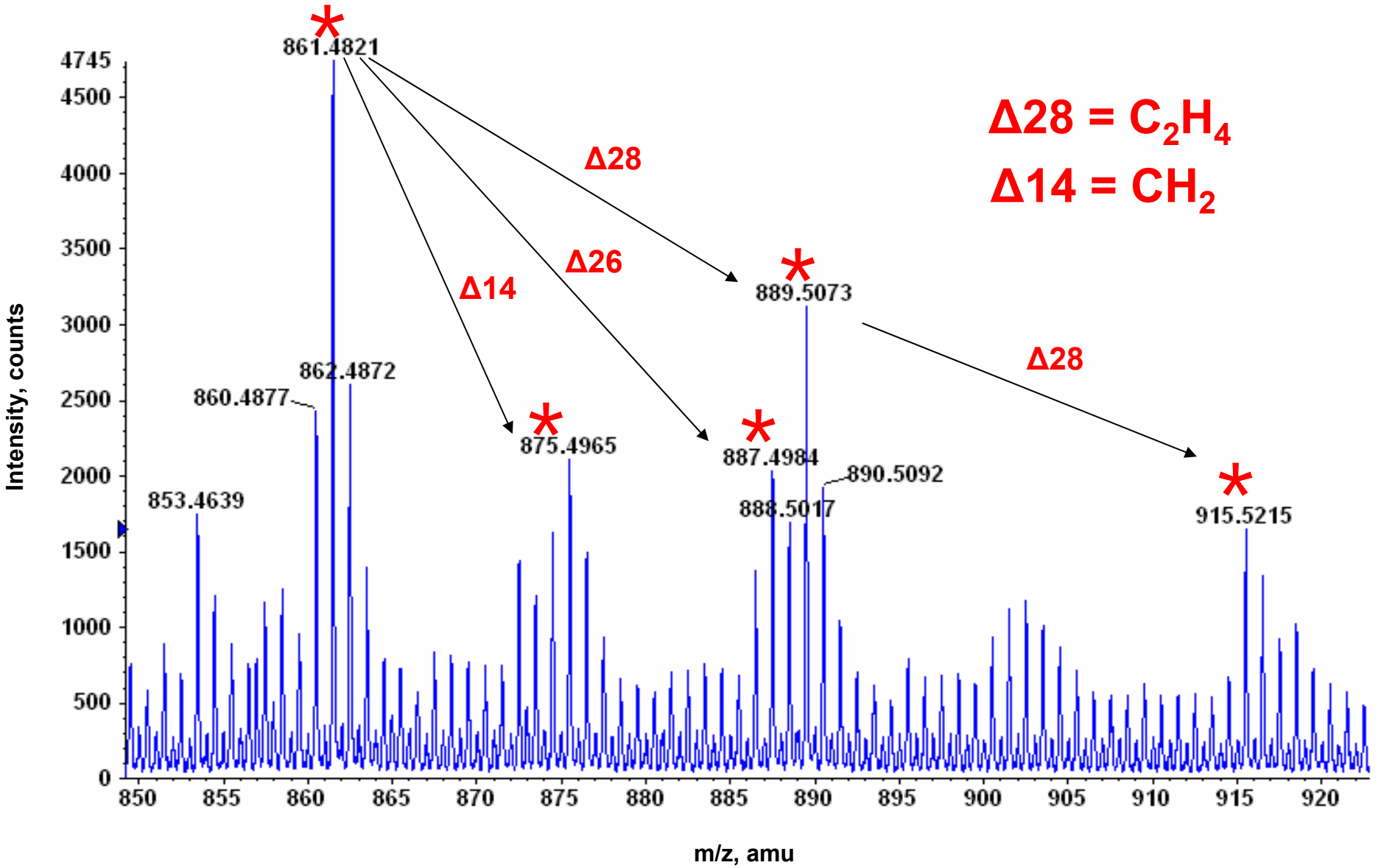


Confirm with MS-MS

ESI-MS of Fraction #10



ESI-MS of Unknown Ions



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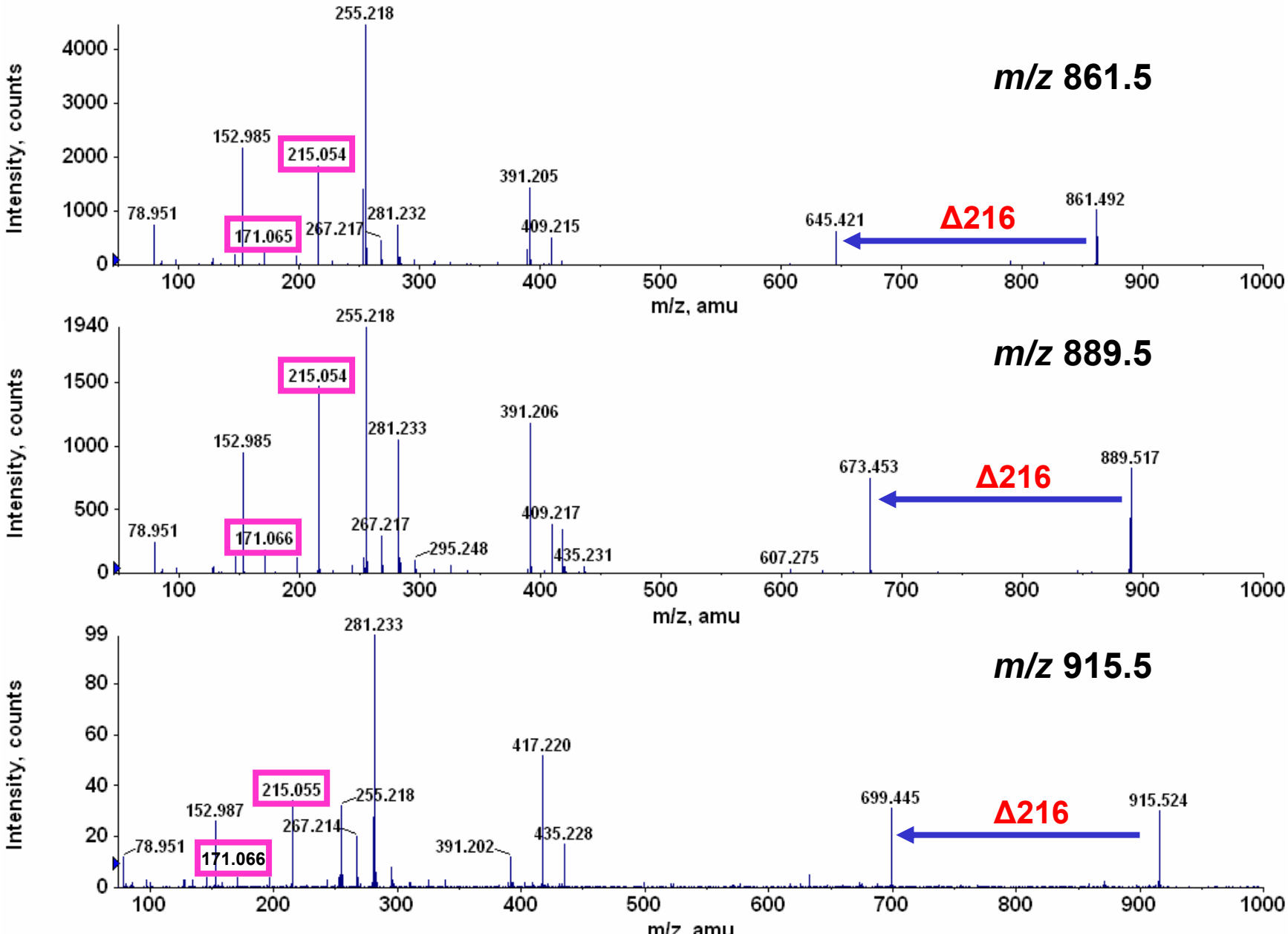
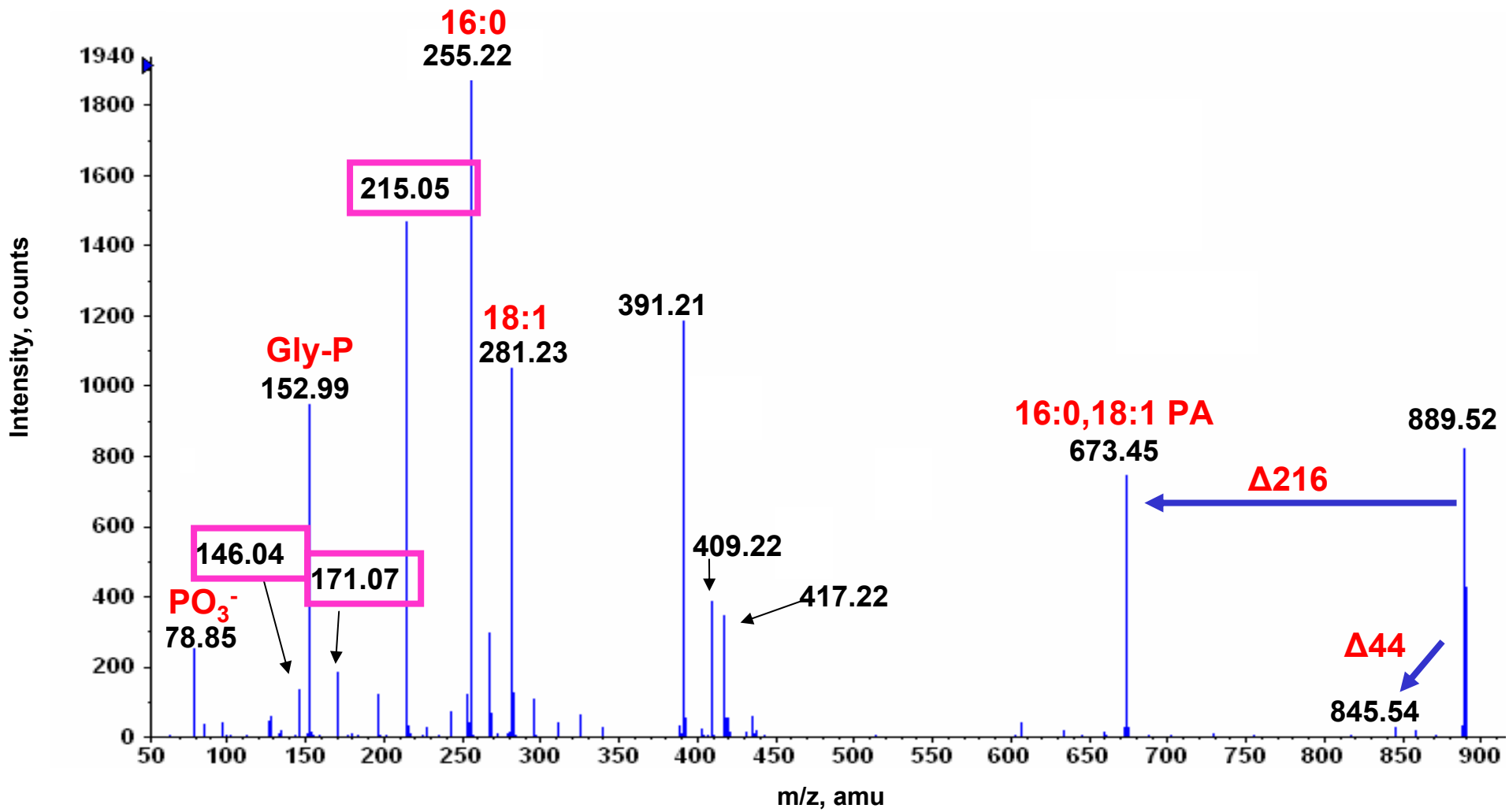


MSMS of Unknown Ion 889.52



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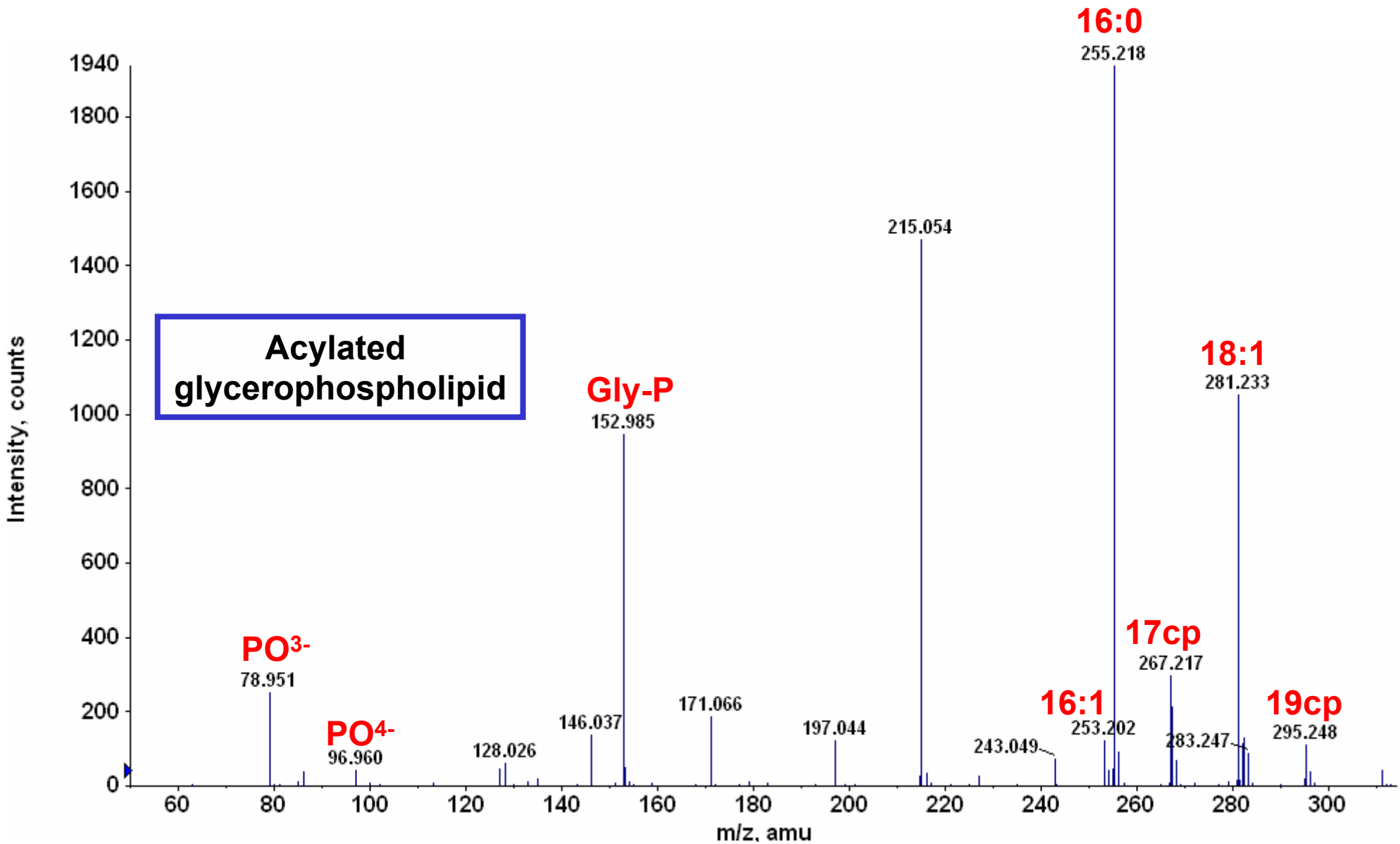

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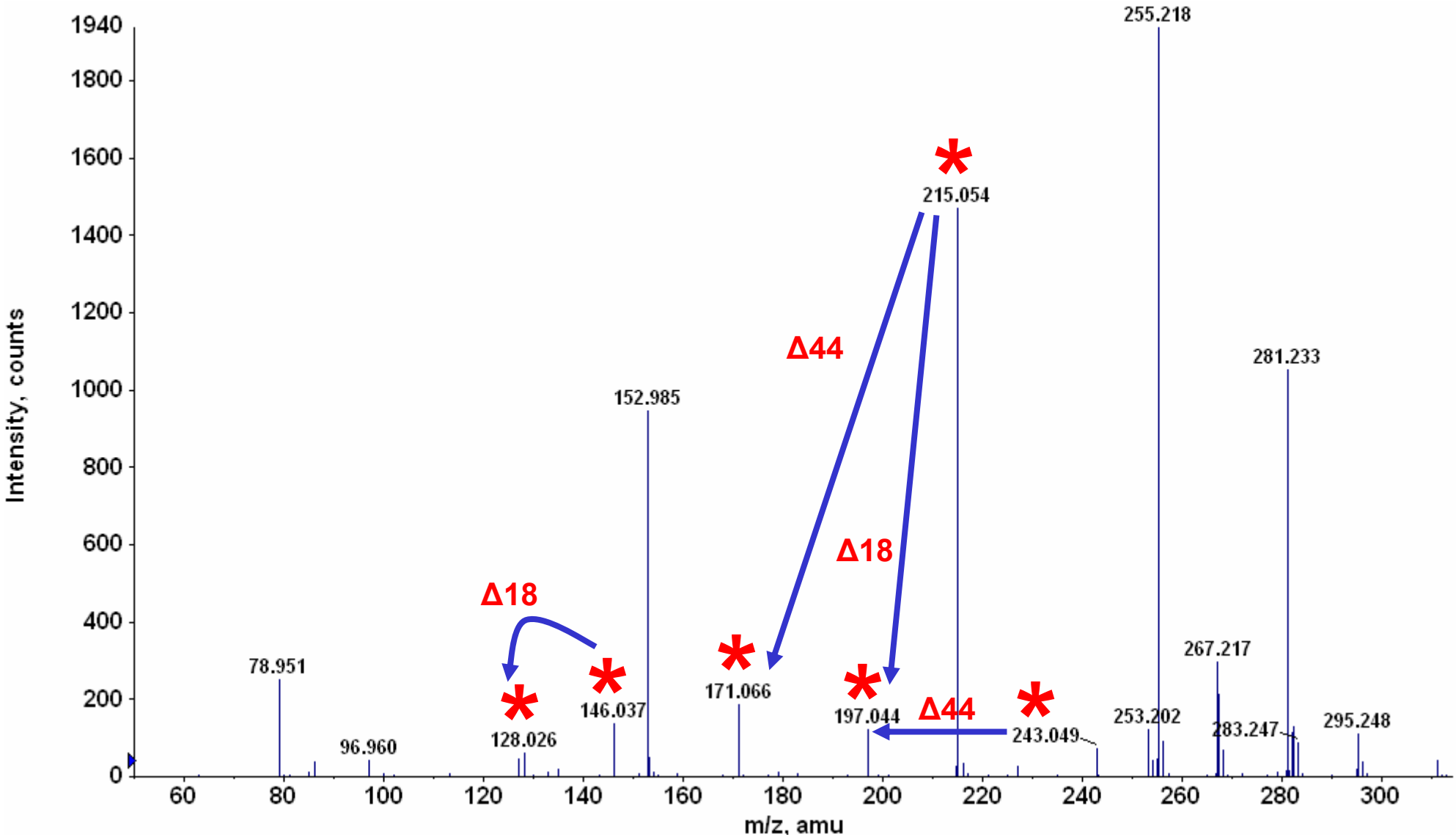

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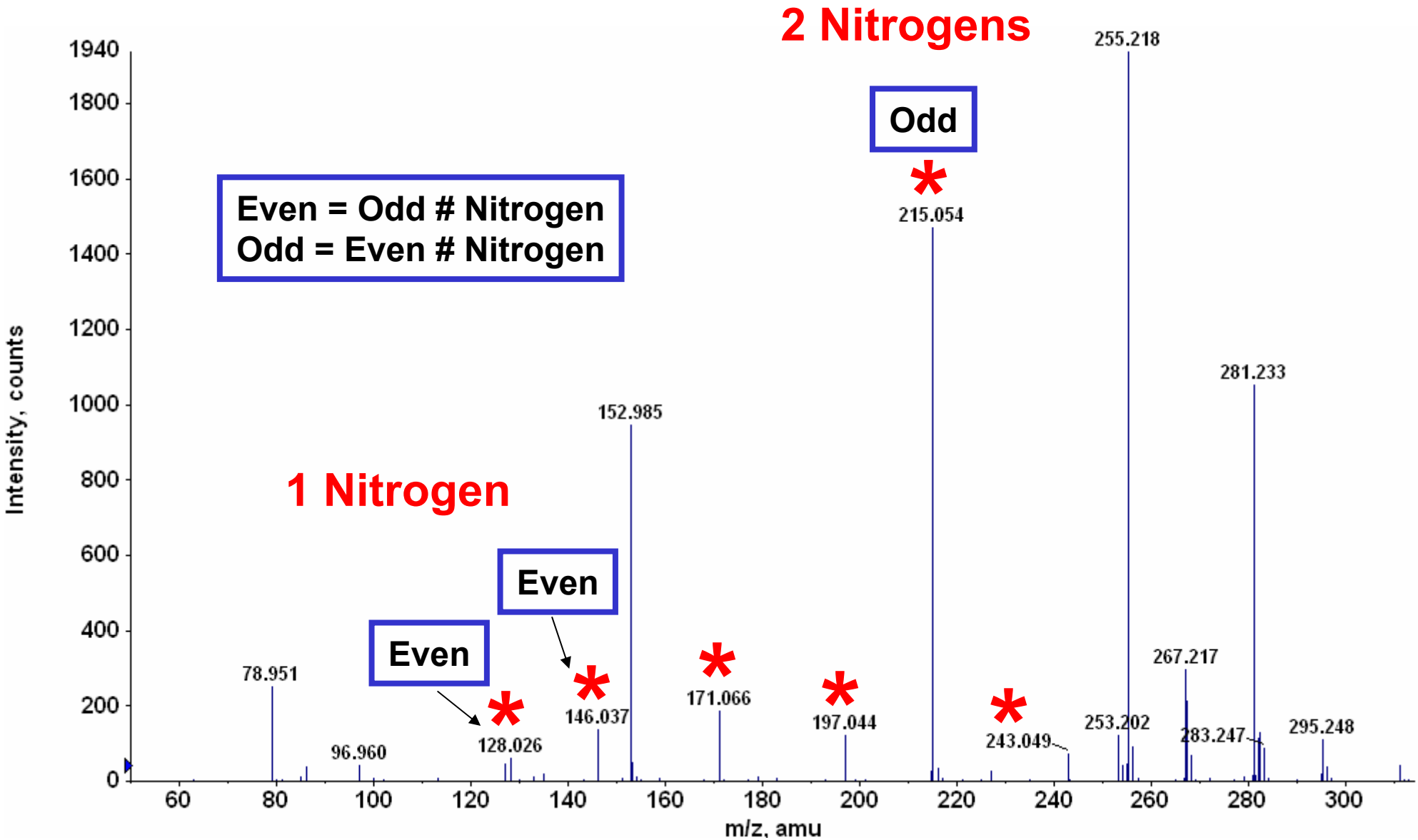

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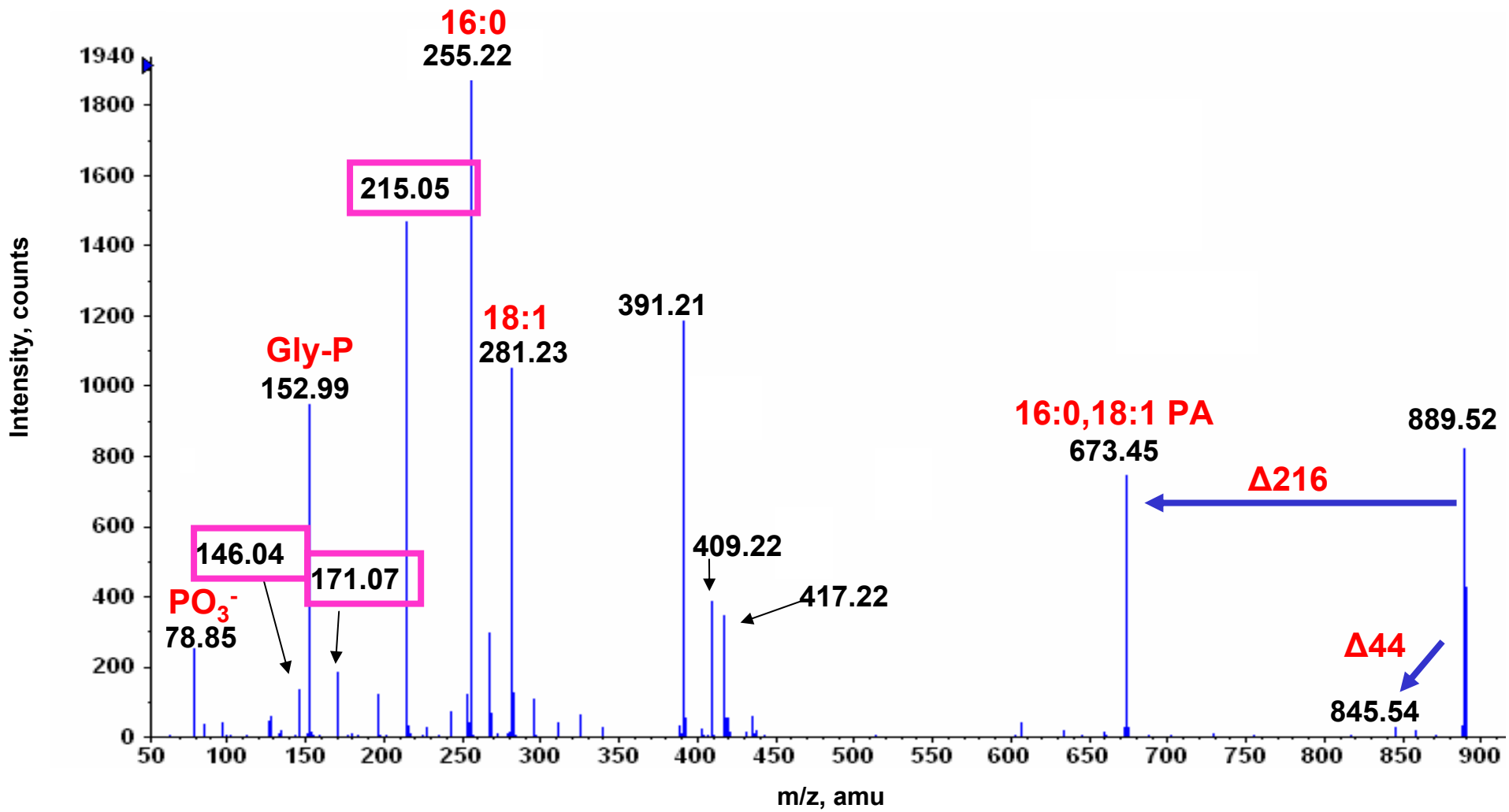
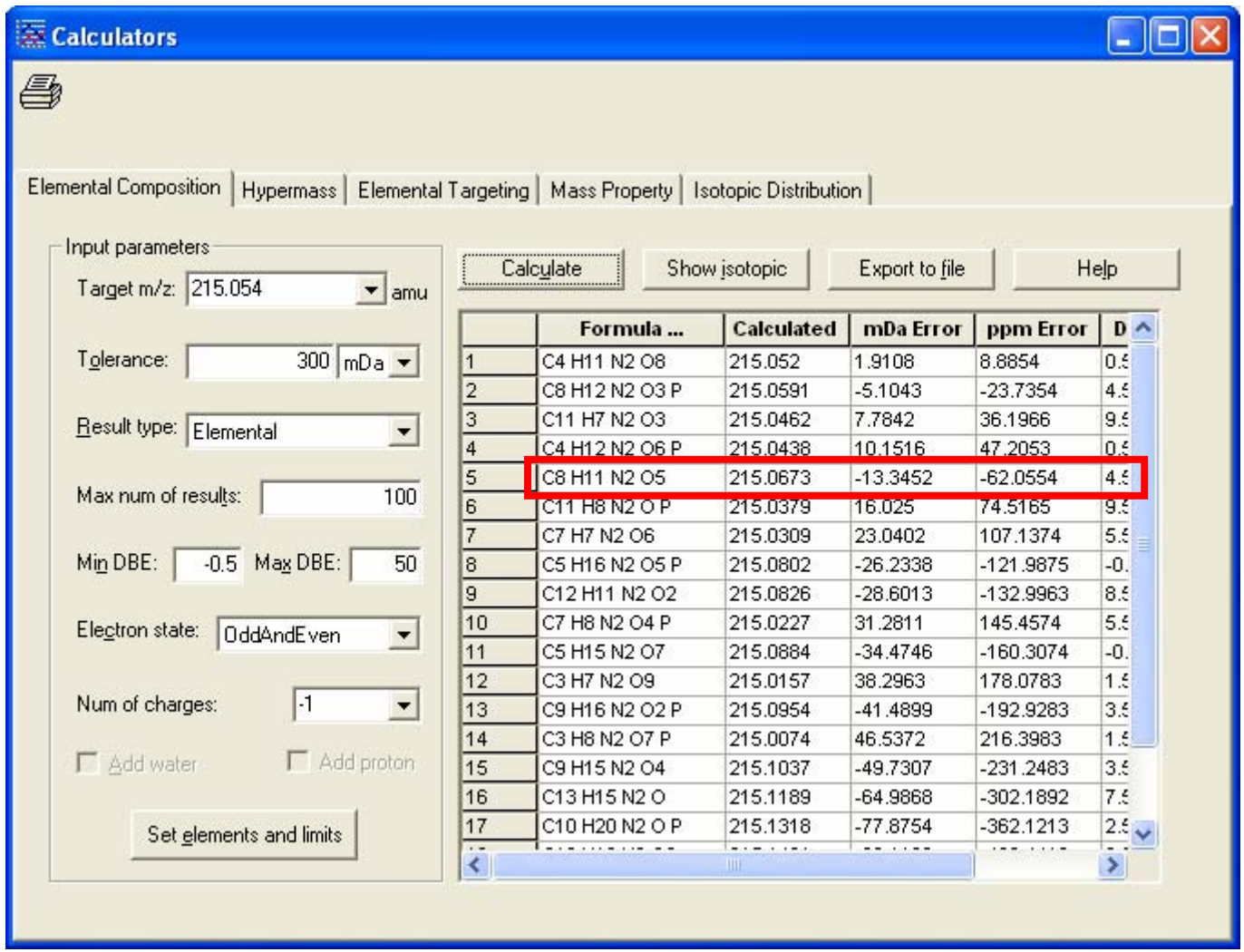


Elemental composition Calculator



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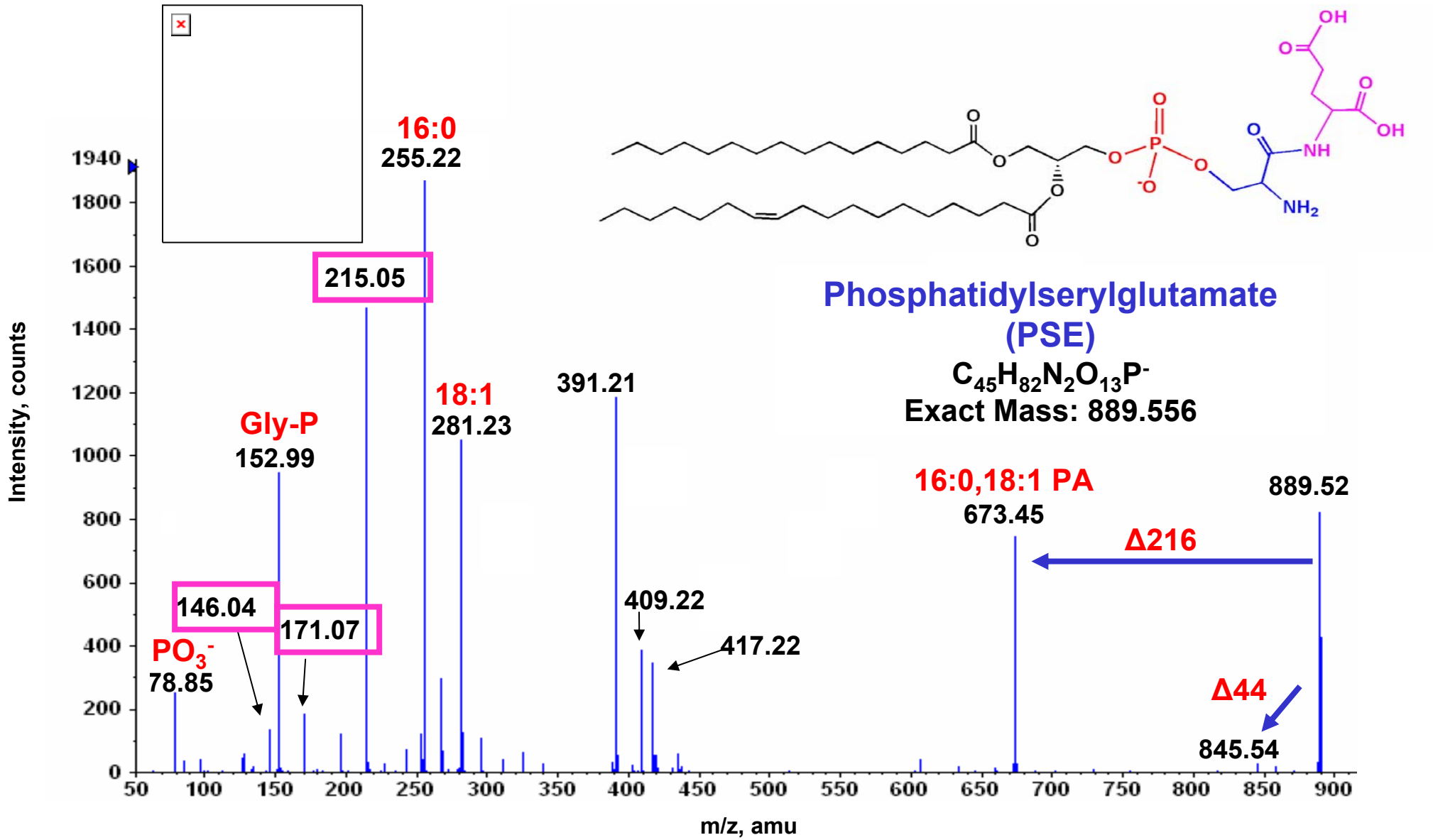
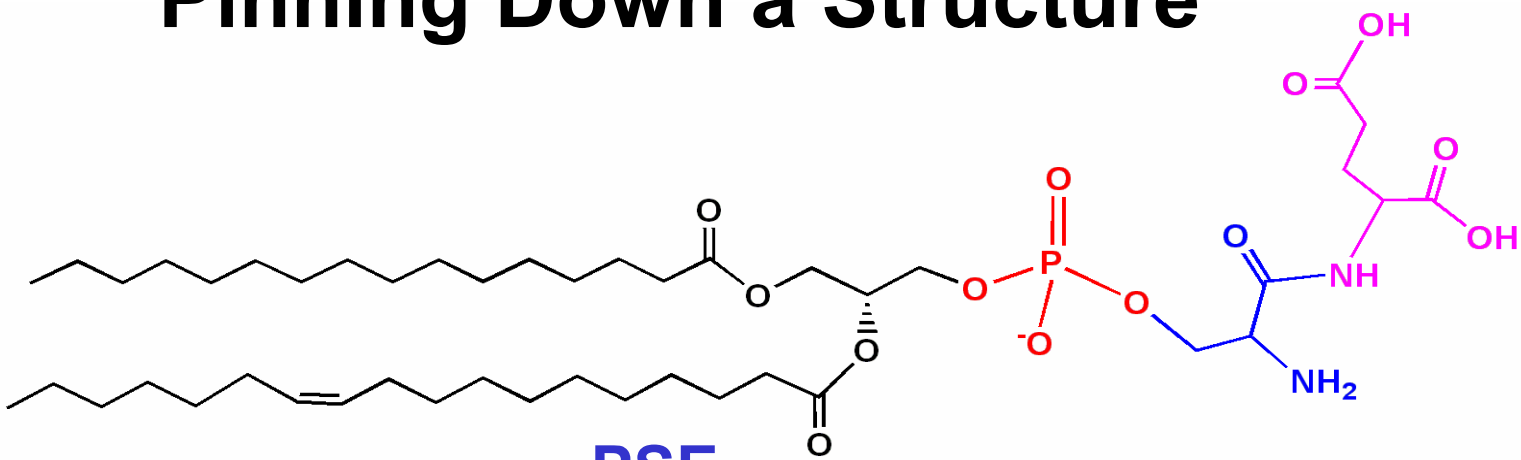


Pinning Down a Structure



PSE

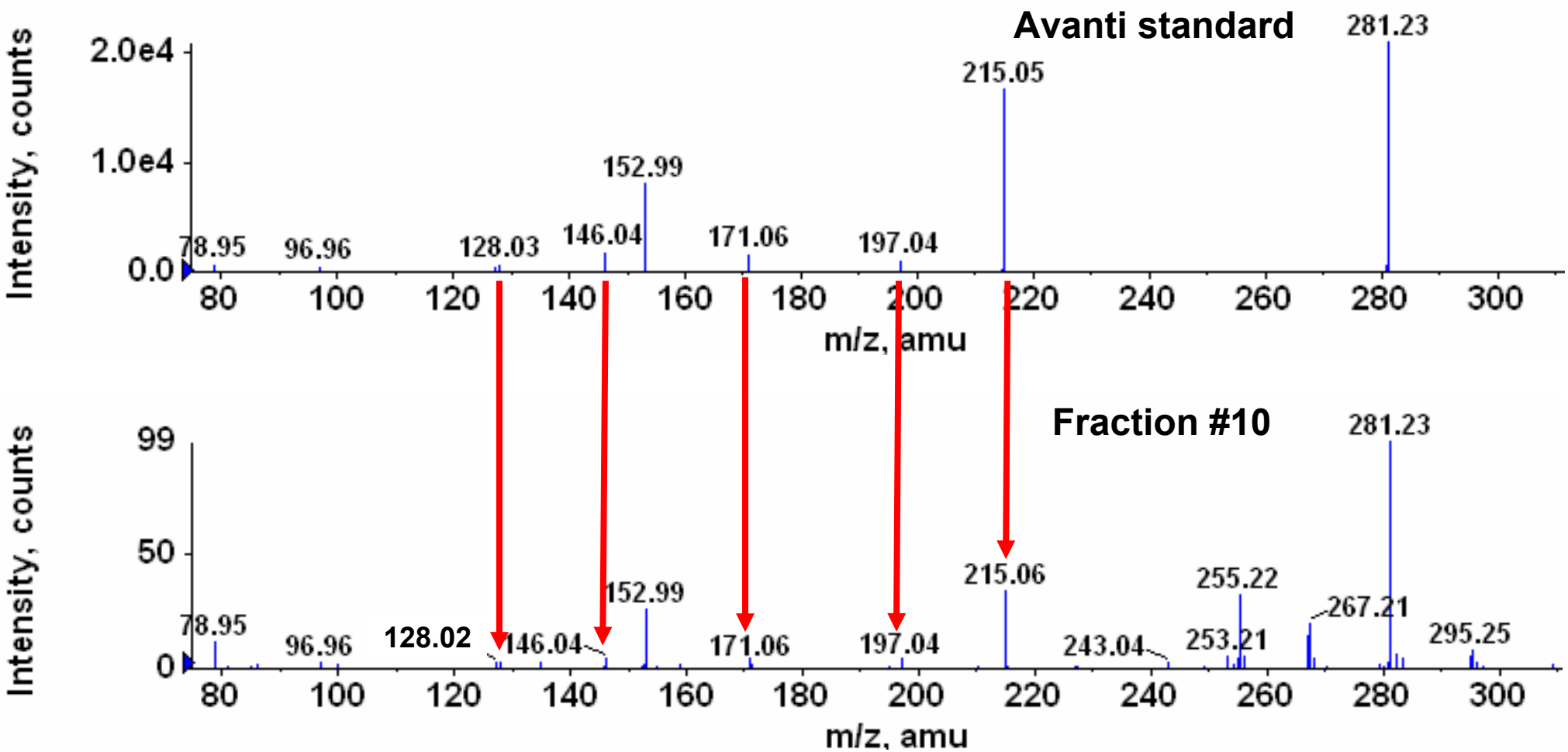
Isotopic labeling

Purify and characterize by NMR and GC-MS

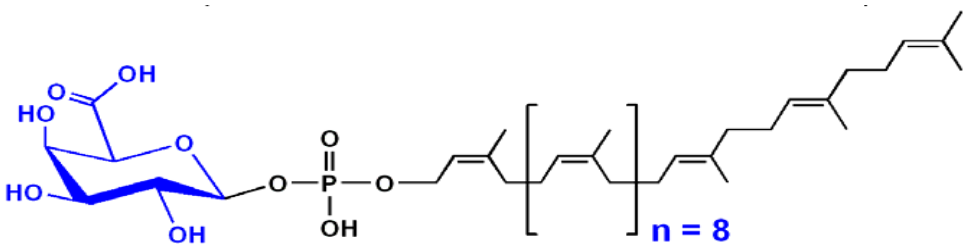
Compare MS, MSMS and LC-MS with synthetic standard

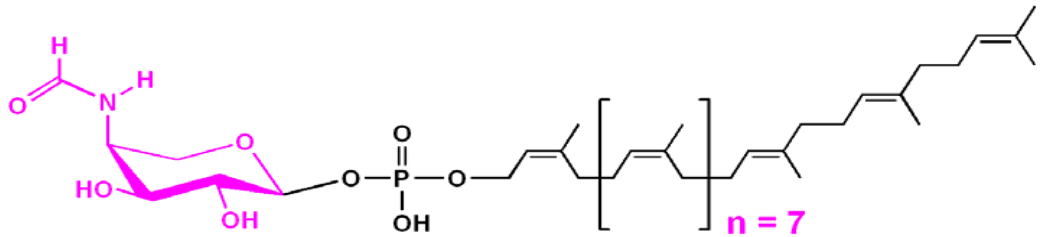
Develop an *in vitro* biosynthetic assay

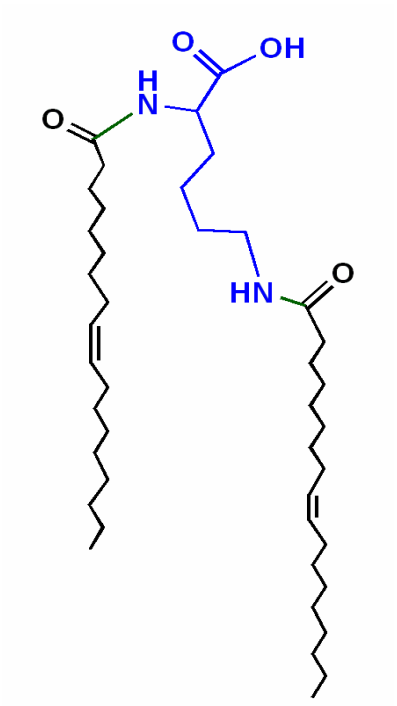
MSMS of PSE
Standard v. Natural Product

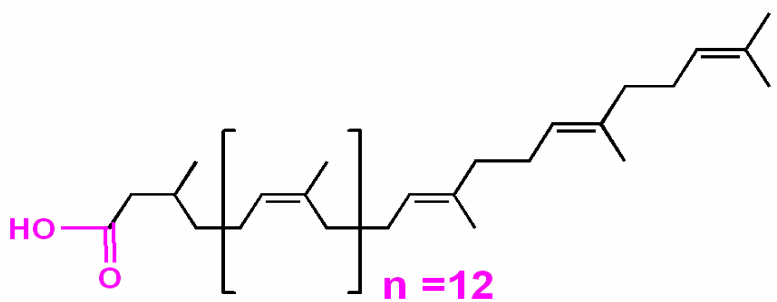


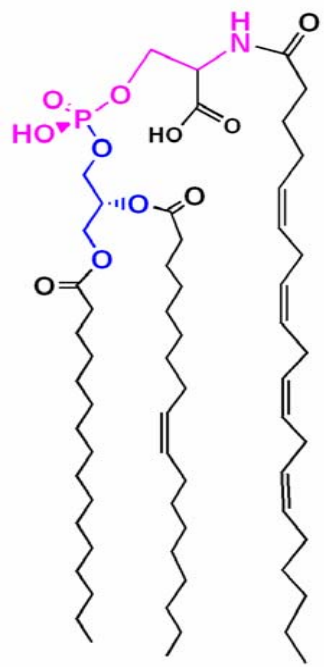
New Structures Identified Using ESI-MS



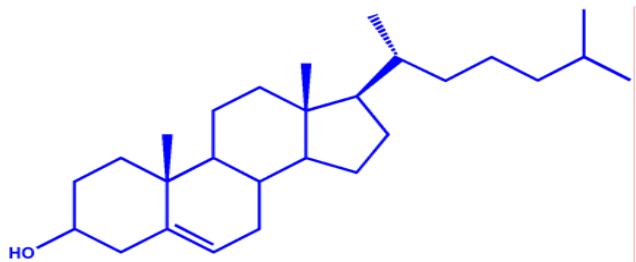








Metabolite Identification



New pathways

Enzyme identification

>40 enzymes

Drug Targets

Genome annotation

Bob Murphy – UCHSC
Walt Shaw – Avanti Polar Lipids

LIPID MAPS

NIH
1U54GM069338
Lipidmaps.org