

Supporting Information

High-Resolution Live-Cell Imaging and Analysis by Laser Desorption/Ionization Droplet Delivery Mass Spectrometry

Jae Kyoo Lee,¹ Erik T. Jansson,^{1,2} Hong Gil Nam,^{3,4,} Richard N. Zare^{1,*}*

¹Department of Chemistry, Stanford University, Stanford, California, 94305 USA

²Department of Chemistry – BMC, Uppsala University, SE-75124 Uppsala, Sweden

³Center for Plant Aging Research, Institute for Basic Science, Daegu 711-873, Republic of Korea

⁴Department of New Biology, DGIST, Daegu 711-873, Republic of Korea

*To whom correspondence should be addressed:

Prof. Richard N. Zare
Department of Chemistry
Stanford University
333 Campus Drive
Stanford, CA 94305-5080, USA
Tel: +1-650-723-3062
Email: zare@stanford.edu

Prof. Hong Gil Nam
Department of New Biology
DGIST, Daegu 711-873, Republic of Korea
Tel: +82-53-785-1800
Email: nam@dgist.ac.kr

Validation of analyte identities secreted from PC12 cells with LC-MS MRM

LC-MS analyses with MRM of the biological and control samples were performed on an Agilent 1100 LC (Agilent, Santa Clara, CA) equipped with an Eclipse Plus 5 μm phenyl-hexyl column, 100 \AA pore size, 150 mm x 2.1 mm (Agilent, Santa Clara, CA) and coupled to an ESI Quattro Premier triple quadrupole MS (Waters, Milford, MA). Separation of the analytes was performed by injection of 10 μL of sample solution. A binary solvent system was used on the LC, with buffer A (H_2O with 0.1% formic acid) and buffer B (acetonitrile with 0.1% formic acid). The column temperature was kept at 25°C. A 250 $\mu\text{L}/\text{min}$ flow rate was used with gradient elution where buffer B increased linearly over the course of a 10-minute run as follows: 0–2 min 0% B, 2–5 min 0–25% B, 5–6 min 25–70% B, 6–7 min 70% B, 7–8 min 70–0%B, 8–10 min 0% B. MS parameters used for MRM were: capillary voltage 5 kV, source temperature 110°C, desolvation temperature 350°C. Cone voltage was parent-ion dependent and collision energy was daughter-ion dependent. For phenethylamine (m/z 122), the cone voltage was set to 18 V; collision energy 25 eV (to m/z 76.8), 19 eV (to m/z 78.7), 21 eV (to m/z 102.7) and 12 eV (to m/z 104.8). For tyramine (m/z 138), the cone voltage was set to 15 V; 25 eV (to m/z 76.7) and 9 eV (to m/z 120.8). Dwell times were 50 ms per transition.

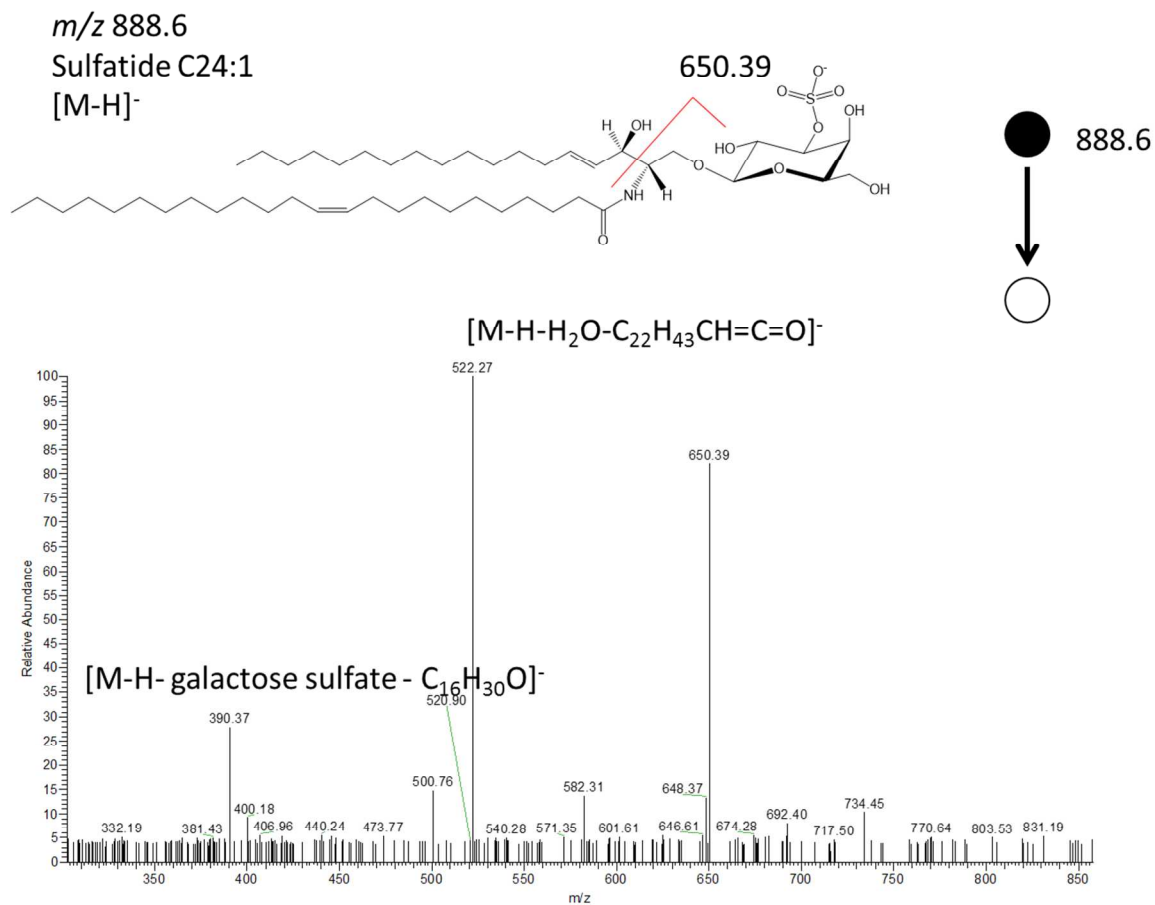


Figure S1. Negative ion mode tandem mass spectrometry analysis of the precursor ion at m/z 888.6

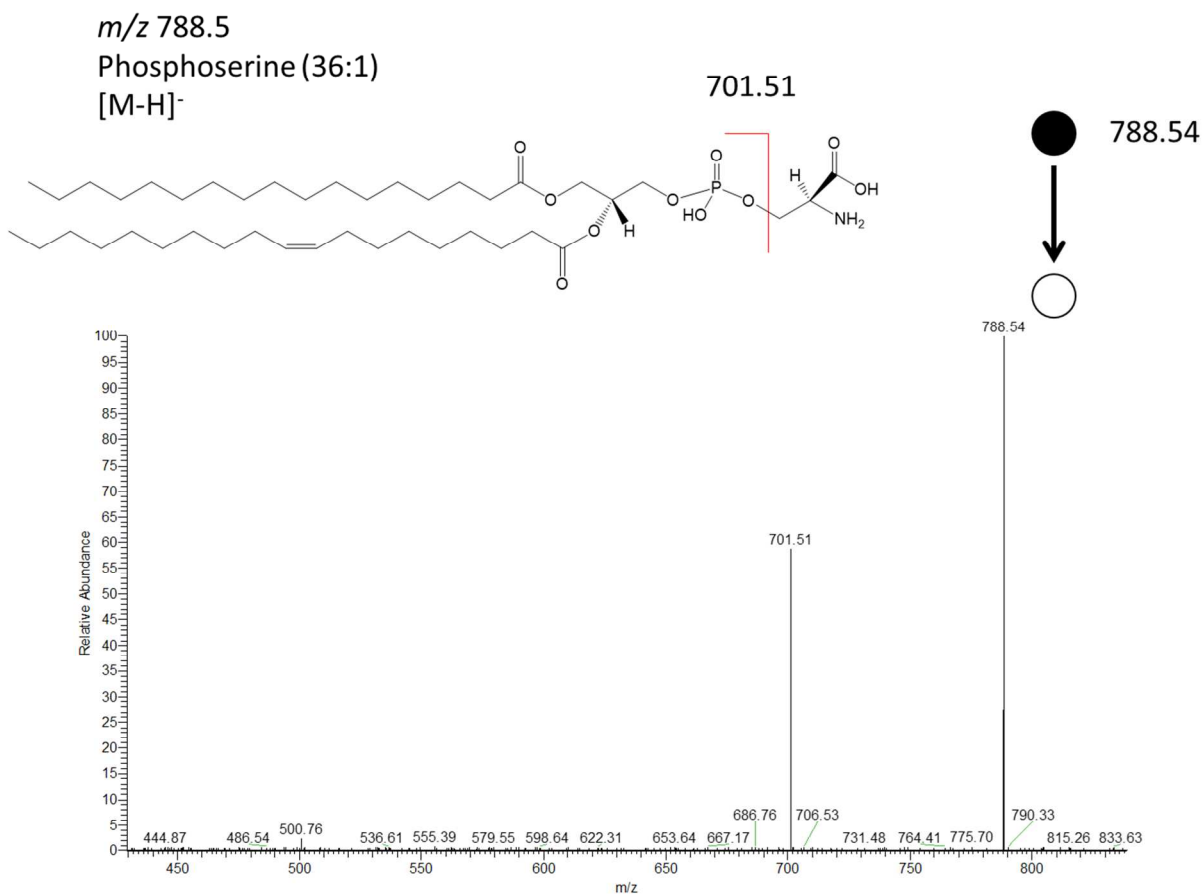


Figure S2. Negative ion mode tandem mass spectrometry analysis of the precursor ion at m/z 788.54

m/z 885.6
Phosphatidylinositol (38:4)
[M-H]⁻

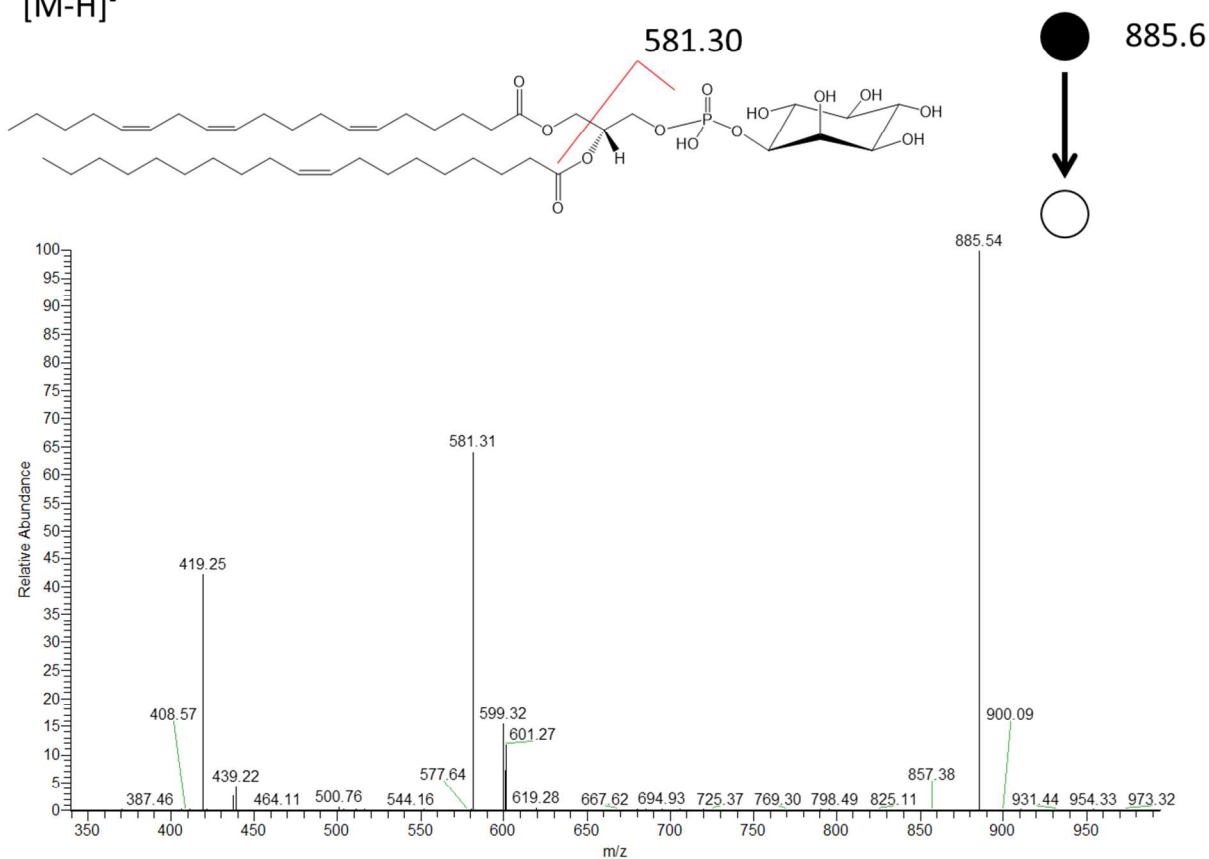


Figure S3. Negative ion mode tandem mass spectrometry analysis of the precursor ion at m/z 885.6

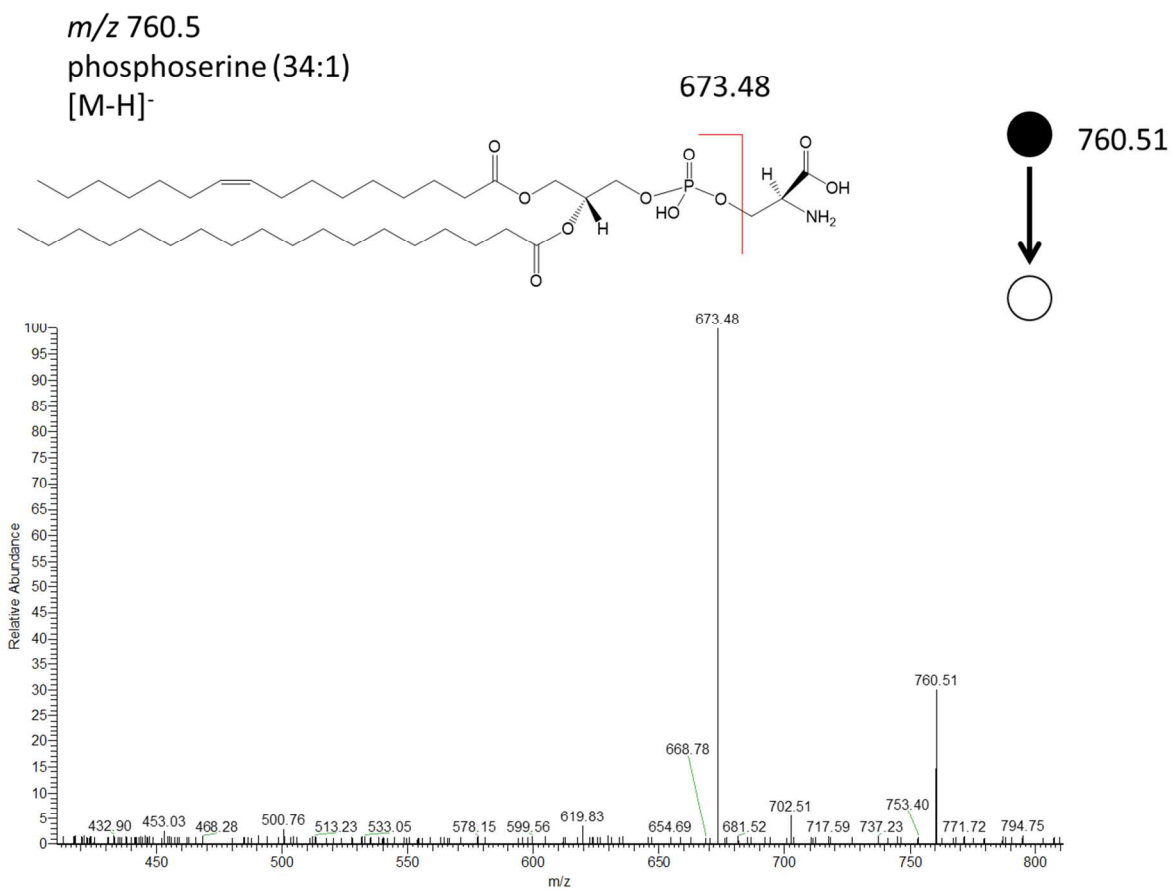


Figure S4. Negative ion mode tandem mass spectrometry analysis of the precursor ion at m/z

760.51

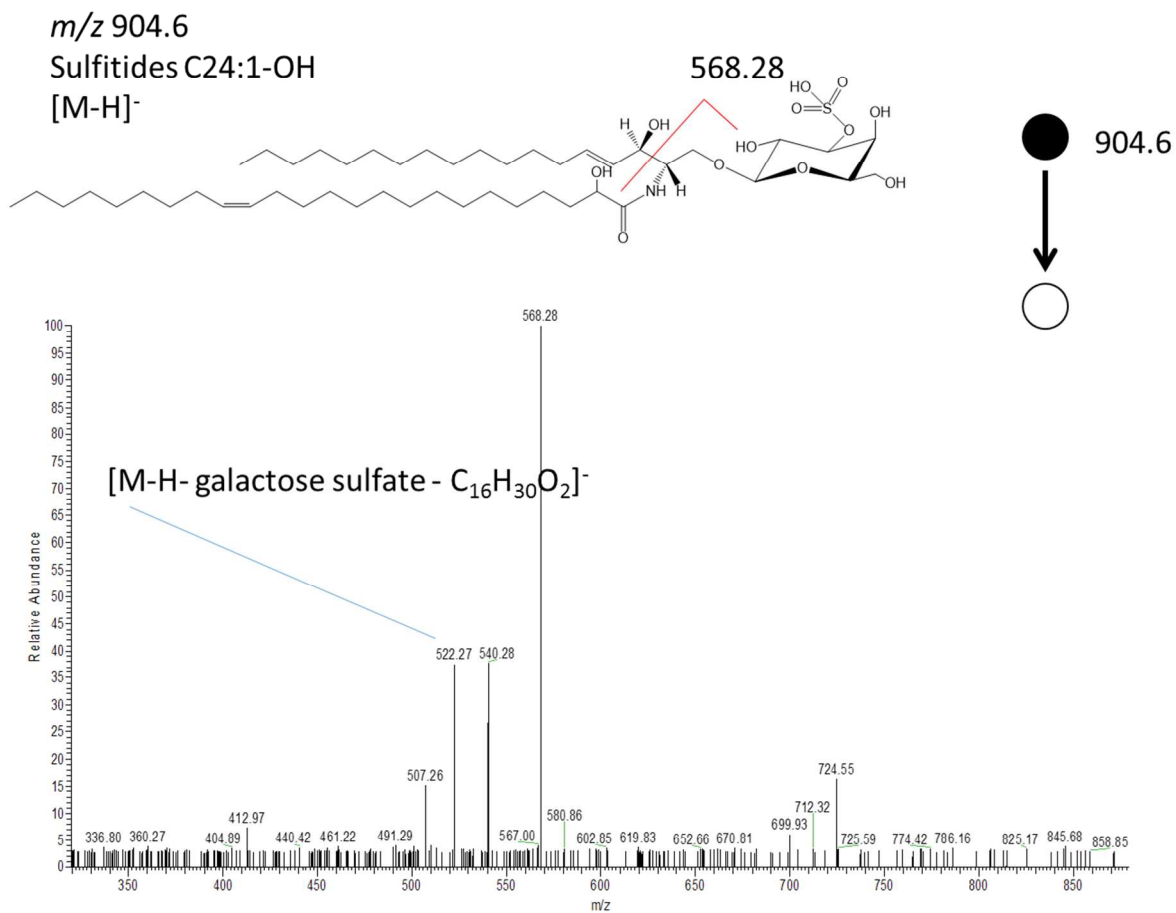


Figure S5. Negative ion mode tandem mass spectrometry analysis of the precursor ion at m/z

904.6

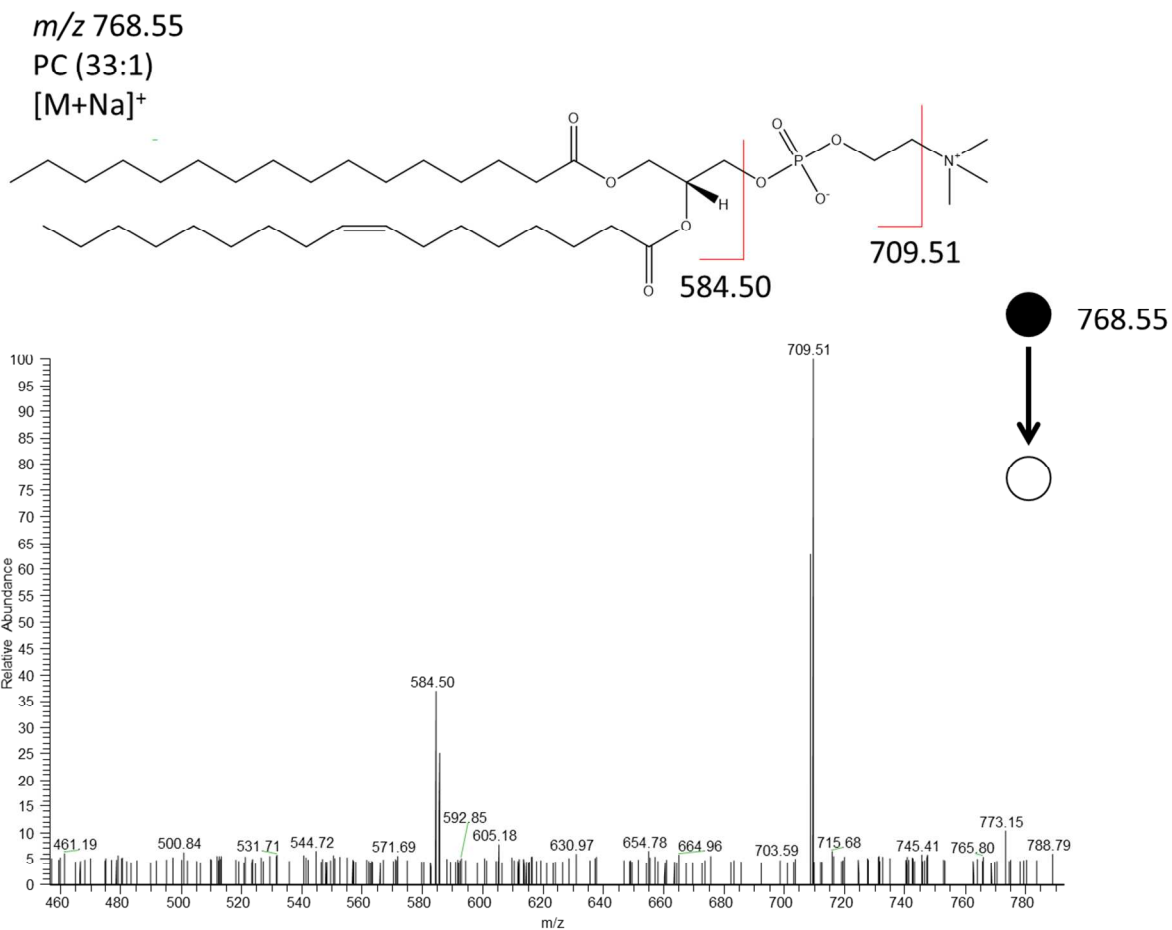


Figure S6. Positive ion mode tandem mass spectrometry analysis of the precursor ion at m/z 768.55

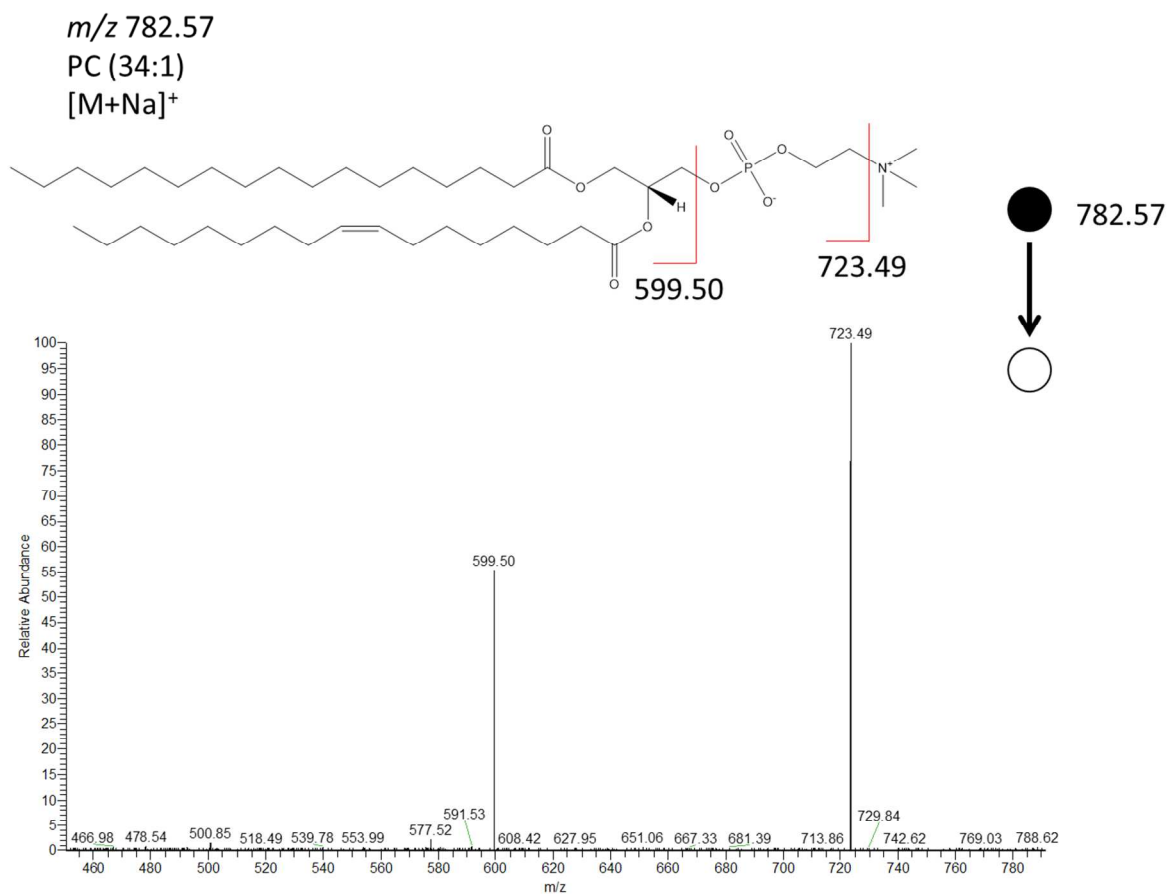


Figure S7. Positive ion mode tandem mass spectrometry analysis of the precursor ion at m/z 782.57

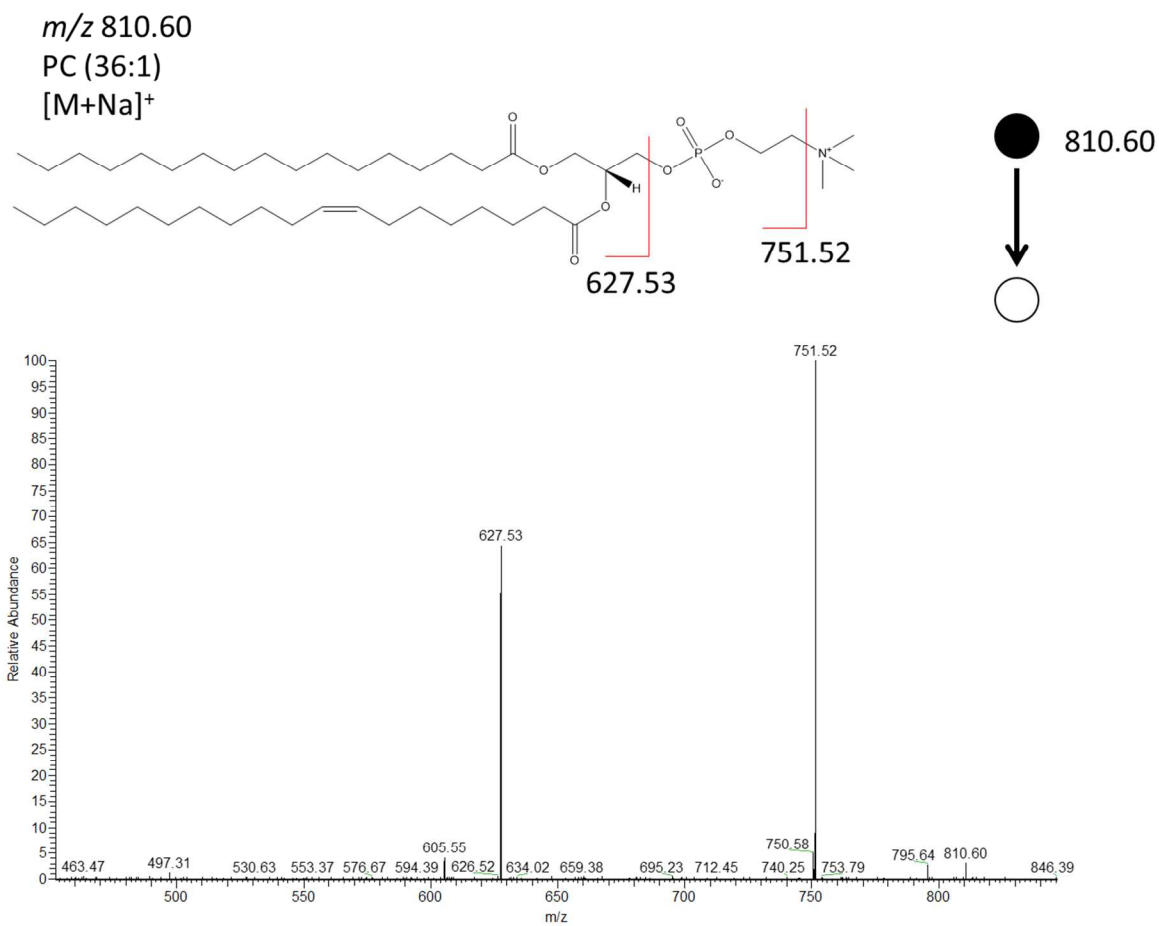


Figure S8. Positive ion mode tandem mass spectrometry analysis of the precursor ion at m/z 810.60

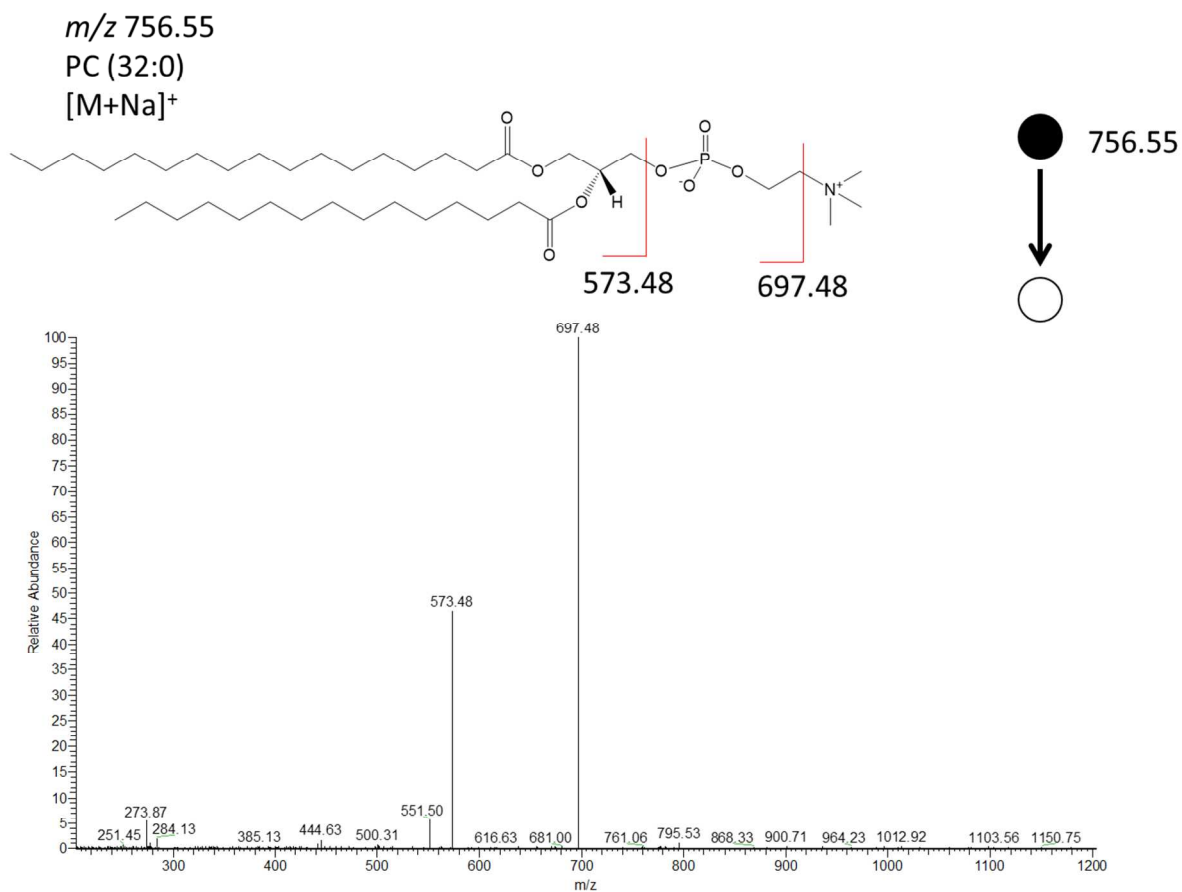


Figure S9. Positive ion mode tandem mass spectrometry analysis of the precursor ion at m/z

756.55

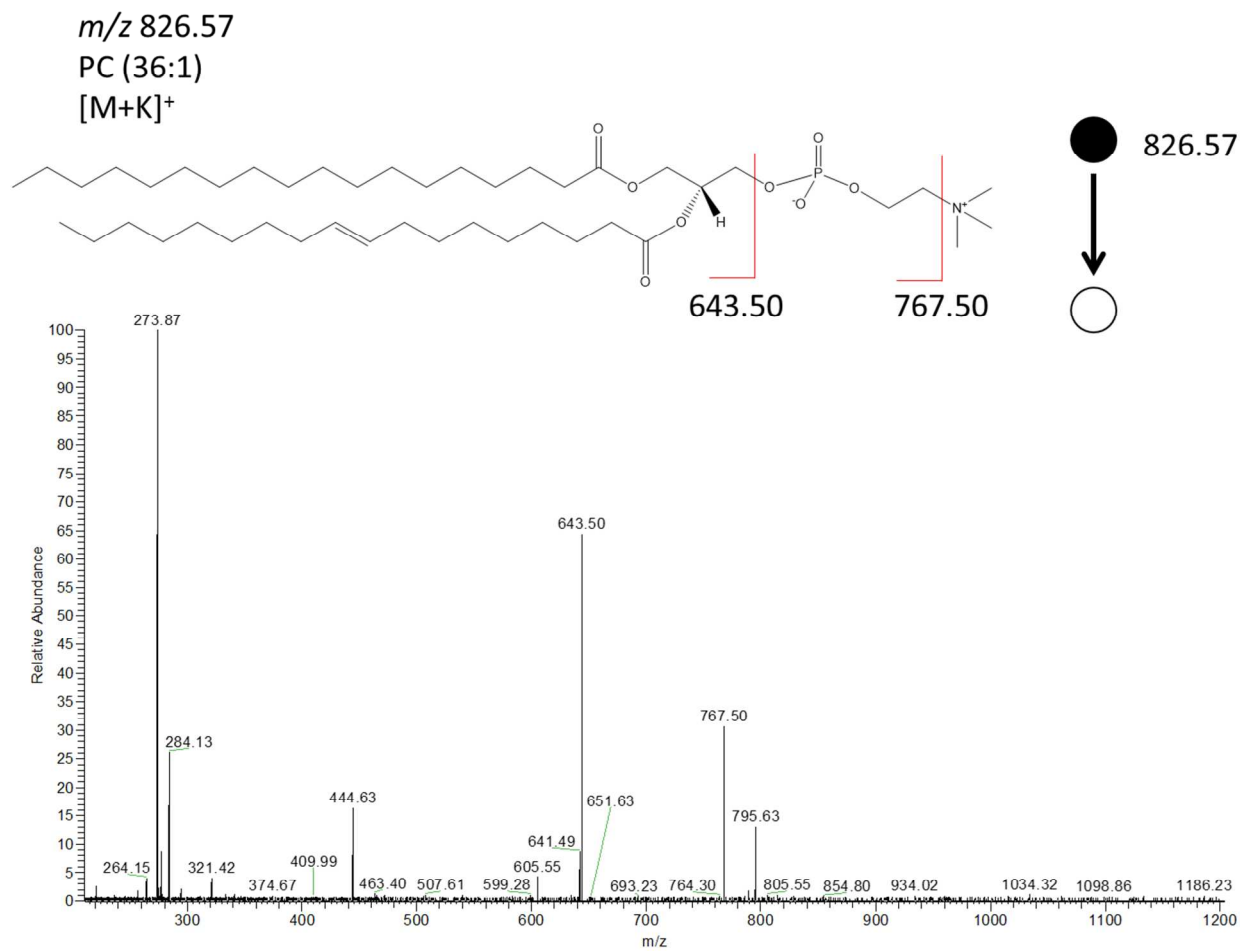


Figure S10. Positive ion mode tandem mass spectrometry analysis of the precursor ion at m/z 826.57

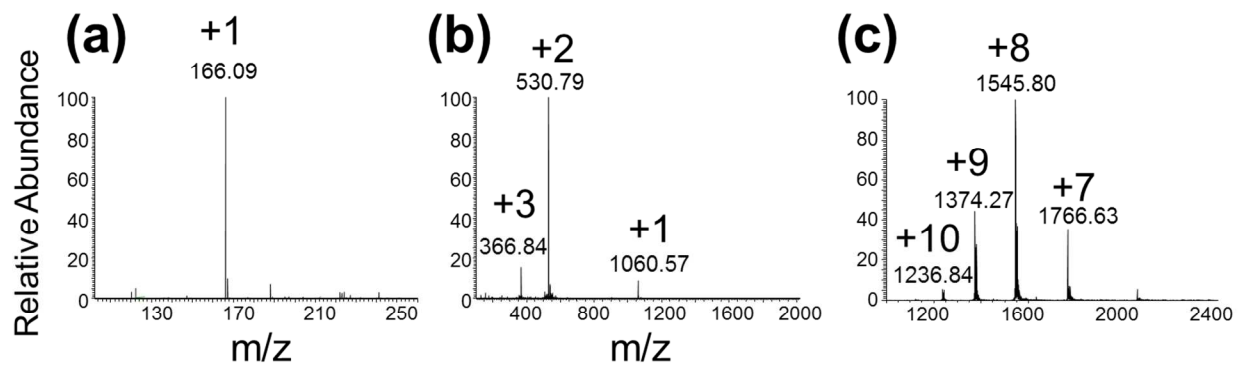


Figure S11. Mass spectra of the samples in liquid phase including (a) 100 μ M phenylalanine, (b) 100 μ M bradykinin, and (c) 100 μ M cytochrome *c*. The charge state of the ions is indicated by +z.

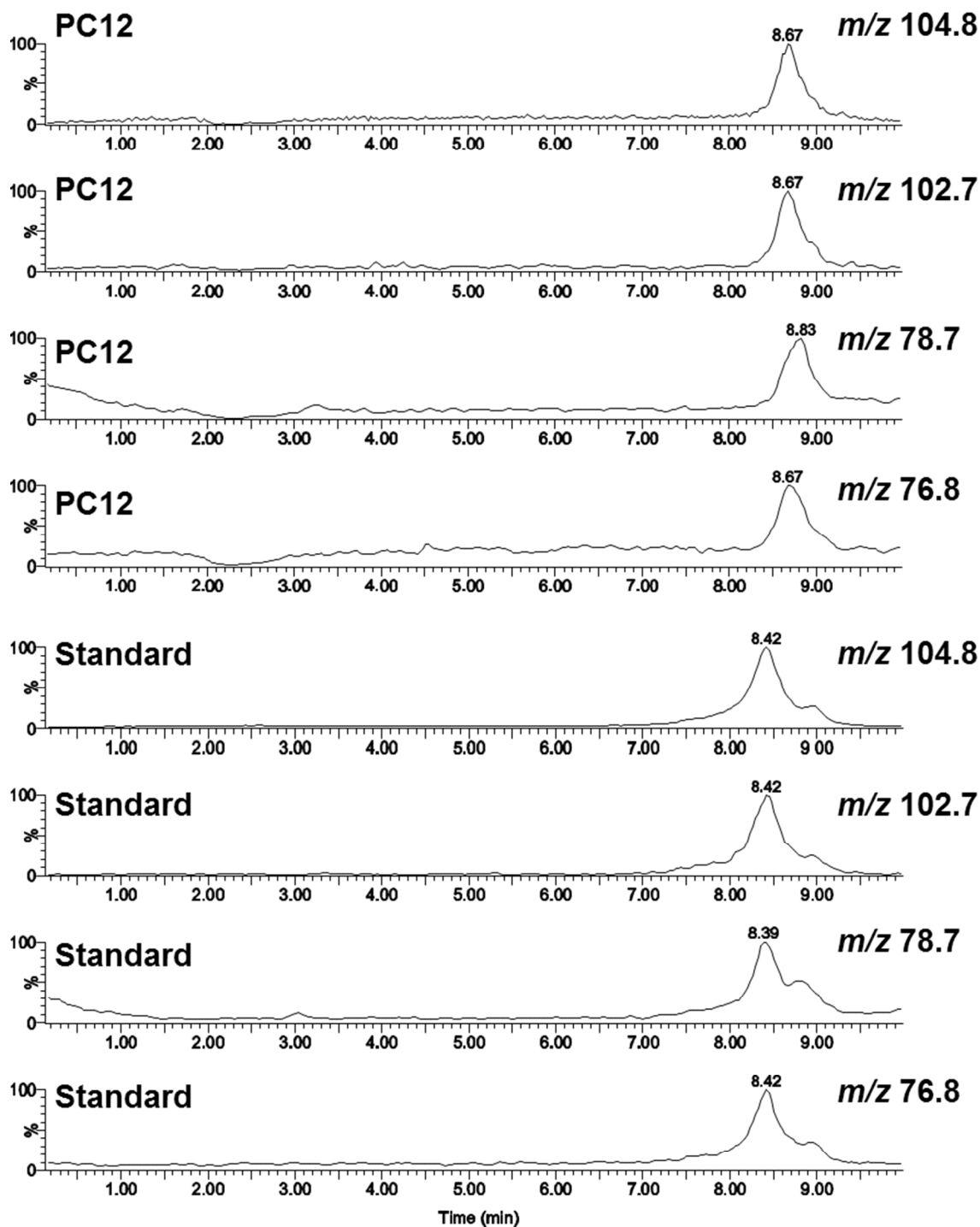


Figure S12. Multiple reaction monitoring LC-MS for phenethylamine showing a comparison of the biological sample with analytical standard sample (0.2 μ M phenethylamine). The matching of four unique fragments of phenethylamine (m/z 122) at m/z 104.8, m/z 102.7, m/z 78.7 and m/z 76.8, between the releasates from PC12 cells and the analytical standard confirmed the identity of the releasate as phenethylamine.

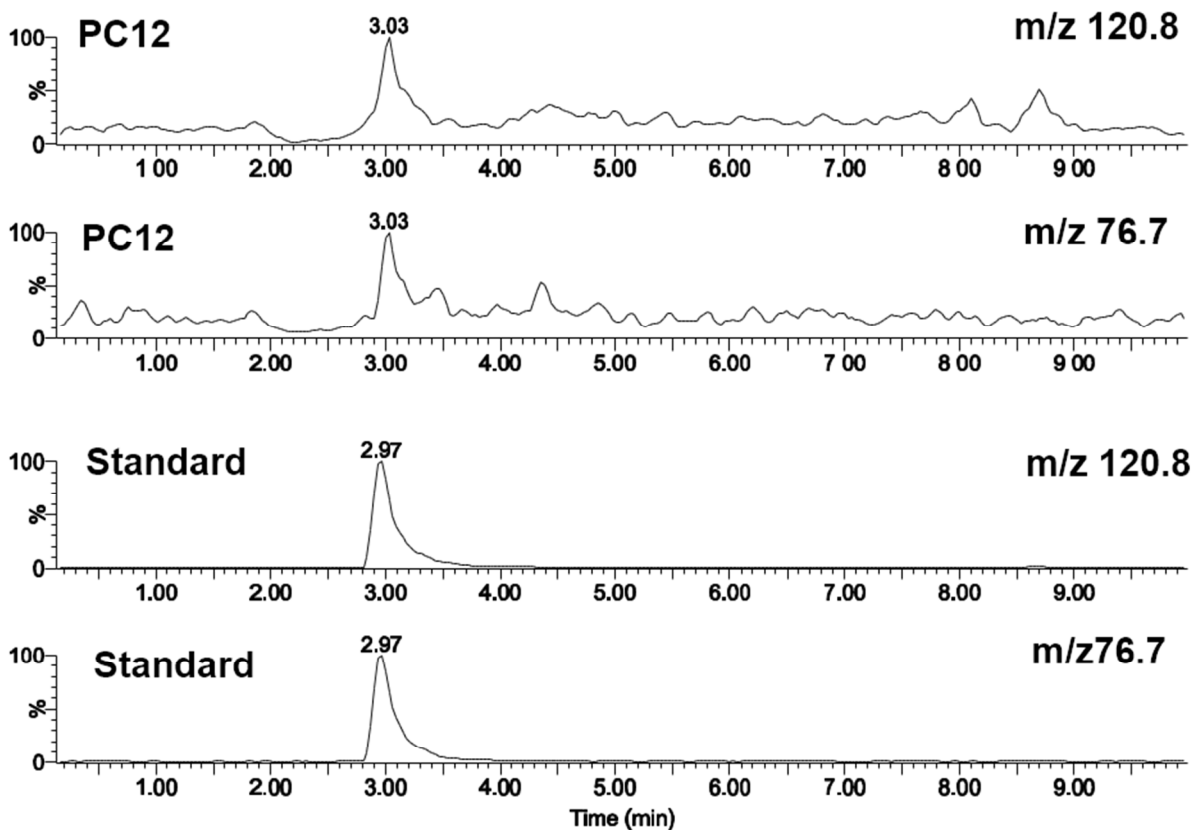


Figure S13. Multiple reaction monitoring LC–MS for tyramine showing a comparison of biological sample with analytical standard sample (0.2 μ M tyramine). The matching of two unique fragments from tyramine (m/z 138) at m/z 120.8 and m/z 76.3, between the releases from PC12 cells and the analytical standard confirmed the identity of the releasate as tyramine.