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DEPARTMENT OF CHEMISTRY

Capillary Electrophoresis-Electrospray Ionization-Mass Spectrometry Analyses of Single Cells

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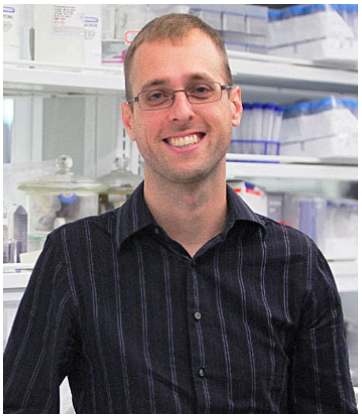

Label-free single-cell proteomics by mass spectrometry (MS) is currently incompatible with complex tissues without requiring cell culturing, single-cell dissection, or tissue dissociation. We here report the first example of label-free single-cell MS-based proteomics directly in single cells in live vertebrate embryos. Our approach integrates optically guided in situ subcellular capillary microsampling, one-pot extraction–digestion of the collected proteins, peptide separation by capillary electrophoresis, ionization by an ultrasensitive electrokinetically pumped nanoelectrospray, and detection by high-resolution MS (Orbitrap). With a 700 zmol (420 000 copies) lower limit of detection, this trace-sensitive technology confidently identified and quantified ~750–800 protein groups (<1% false-discovery rate) by analyzing just ~5 ng of protein digest, viz. < 0.05% of the total protein content from individual cells in a 16-cell *Xenopus laevis* (frog) embryo. After validating the approach by recovering animal–vegetal-pole proteomic asymmetry in the frog zygote, the technology was applied to uncover proteomic reorganization as the animal–dorsal (D11) cell of the 16-cell embryo gave rise to its neural-tissue-fated clone in the embryo developing to the 32-, 64-, and 128-cell stages. In addition to enabling proteomics on smaller cells in *X. laevis*, we also demonstrated this technology to be scalable to single cells in live zebrafish embryos. Microsampling single-cell MS-based proteomics raises exciting opportunities to study cell and developmental processes directly in complex tissues and whole organisms at the level of the building block of life: the cell.

Students, meet the speaker after the seminar in a student/postdoc session from 5:45-6:15 pm

Date: Wed, Jan. 27, 2020
Time: 4:30-5:30 pm
Location: Virtual Seminar (Zoom)

https://wvu.zoom.us/j/98754582111?pwd=YjlHVzkyMlV3VlNIT2FIR2RNVDIwQT09