Tandem MS Analysis and An Emerging Genome: The Sea Urchin Sperm Plasma Membrane Proteome

Anna T. Neill[1], Terry Gaasterland[2], John R. Yates III[3], Victor D. Vacquier[4]

Keywords: sea urchin, fertilization, membrane protein, tandem MS, whole genome sequence

1 Introduction.

Fertilization is a fundamental cellular process involving signaling and recognition at cell surfaces. Sperm plasma membrane proteins play key roles by mediating sperm motility, egg recognition, and gamete fusion. Phylogenetically, sea urchins lay at the base of the deuterostome lineage which leads to the vertebrates. Pragmatically, sea urchins provide an excellent model for the study of fertilization because large quantities of gametes are readily available. Moreover, sperm plasma membrane proteins are easy to isolate from the rest of the cell, and can be obtained separately from the head and the tail (flagellum). Thus it is straightforward to study proteins in the context of their subcellular location.

2 Approach.

We use micro liquid chromatography (μ-LC) tandem mass spectrometry (MS/MS) to characterize the sea urchin sperm plasma membrane proteome. One way to interpret this type of MS data relies on prior knowledge of target protein sequences. Sea urchin genome sequencing is currently underway but in early stages. Consequently, the interpretation of sea urchin MS data requires methods to extract protein information from unassembled or partially assembled genomic sequence. Our approach is to translate protein sequence fragments from all open reading frames in the raw genome sequence data, and then to reduce and merge the fragments into a minimally redundant protein database, which is then searched with the collected spectra using mass-to-peptide assignment software.

[1] Scripps Institution of Oceanography, University of California, San Diego, La Jolla, CA 92093-0202. E-mail: aneill@ucsd.edu

[2] The Rockefeller University, New York, NY 10021. E-mail: gaasterl@genomes.rockefeller.edu

[3] Department of Cell Biology, The Scripps Research Institute, La Jolla, CA 92037. E-mail: jyates@scripps.edu

[4] Scripps Institution of Oceanography, University of California, San Diego, La Jolla, CA 92093-0202. E-mail: vvacquier@ucsd.edu