



The Handy Little Book of Proteomics Protocols: Methods for Biomarker Identification and Validation

Benjamin C Orsburn Ph.D.

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This text is exactly what it claims to be, a handy little book for people performing proteomic analysis. The 32 methods described in this book are essential to any proteomics lab. They include tried-and-true methods such as the Bradford protein assay, as well as how to set-up and perform SDS-PAGE. Cutting-edge techniques are described here as well, including Filter Aided Sample Preparation (FASP) and Filter Aided Capture and Elution (FACE). Several methods also detailed for the enrichment of peptides for phosphoproteomic analysis. A number of techniques have not been previously published elsewhere, as they were created or modified by the author. Methods detailed in this text include: Lysis of: prokaryotic, mammalian, and plant cells, including subcellular fractionation. Protein determination by Lowry and Bradford Dialysis based buffer exchange One- and two- dimensional gels In-gel, in-solution and filter-aided protein digestion methods with trypsin and other, less common, proteolytic enzymes. Peptide cleanup with silica tips and spin cartridges. Silica tip-based fractionation. Three separate, complementary phosphopeptide enrichment strategies and how to detect other post-translational modifications including ubiquitination. General tips and a robust protocol for setting up LC-MS. Validation of biomarkers by antibody-based assays such as immunocytochemistry and blotting, as well as the targeted ion MS-based methods MRM and HR-SIM.

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