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Nota di contenuto	PROTEOMICS OF BIOLOGICAL SYSTEMS: Protein Phosphorylation Using Mass Spectrometry Techniques; CONTENTS; PREFACE; ACKNOWLEDGMENTS; ABOUT THE AUTHOR; 1: Posttranslational Modification (PTM) of Proteins; 1.1 OVER 200 FORMS OF PTM OF PROTEINS; 1.2 THREE MAIN TYPES OF PTM STUDIED BY MS; 1.3 OVERVIEW OF NANO-ELECTROSPRAY/NANOFLOW LC-MS; 1.3.1 Definition and Description of MS; 1.3.2 Basic Design of Mass Analyzer Instrumentation; 1.3.3 ESI; 1.3.4 Nano-ESI; 1.4 OVERVIEW OF NUCLEIC ACIDS; 1.5 PROTEINS AND PROTEOMICS; 1.5.1 Introduction to Proteomics; 1.5.2 Protein Structure and Chemistry

1.5.3 Bottom-Up Proteomics: MS of Peptides 1.5.3.1 History and Strategy; 1.5.3.2 Protein Identification through Product Ion Spectra; 1.5.3.3 High-Energy Product Ions; 1.5.3.4 De Novo Sequencing; 1.5.3.5 Electron Capture Dissociation (ECD); 1.5.4 Top-Down Proteomics: MS of Intact Proteins; 1.5.4.1 Background; 1.5.4.2 GP Basicity and Protein Charging; 1.5.4.3 Calculation of Charge State and Molecular Weight; 1.5.4.4 Top-Down Protein Sequencing; 1.5.5 Systems Biology and Bioinformatics; 1.5.6 Biomarkers in Cancer; REFERENCES; 2: Glycosylation of Proteins; 2.1 PRODUCTION OF A GLYCOPROTEIN 2.2 BIOLOGICAL PROCESSES OF PROTEIN GLYCOSYLATION 2.3 N-LINKED AND O-LINKED GLYCOSYLATION; 2.4 CARBOHYDRATES; 2.4.1 Ionization of Oligosaccharides; 2.4.2 Carbohydrate Fragmentation; 2.4.3 Complex Oligosaccharide Structural Elucidation; 2.5 THREE OBJECTIVES IN STUDYING GLYCOPROTEINS; 2.6 GLYCOSYLATION STUDY APPROACHES; 2.6.1 MS of Glycopeptides; 2.6.2 Mass Pattern Recognition; 2.6.2.1 High Galactose Glycosylation Pattern; 2.6.3 Charge State Determination; 2.6.4 Diagnostic Fragment Ions; 2.6.5 High-Resolution/High-Mass Accuracy Measurement and Identification; 2.6.6 Digested Bovine Fetuin REFERENCES 3: Sulfation of Proteins as Posttranslational Modification; 3.1 GLYCOSAMINOGLYCAN SULFATION; 3.2 CELLULAR PROCESSES INVOLVED IN SULFATION; 3.3 BRIEF EXAMPLE OF PHOSPHORYLATION; 3.4 SULFOTRANSFERASE CLASS OF ENZYMES; 3.5 FRAGMENTATION NOMENCLATURE FOR CARBOHYDRATES; 3.6 SULFATED MUCIN OLIGOSACCHARIDES; 3.7 TYROSINE SULFATION; 3.8 TYROSYLPROTEIN SULFOTRANSFERASES TPST1 AND TPST2; 3.9 O-SULFATED HUMAN PROTEINS; 3.10 SULFATED PEPTIDE PRODUCT ION SPECTRA; 3.11 USE OF HIGHER ENERGY COLLISIONS; 3.12 ELECTRON CAPTURE DISSOCIATION (ECD); 3.13 SULFATION VERSUS PHOSPHORYLATION; REFERENCES 4: Eukaryote PTM as Phosphorylation: Normal State Studies 4.1 MASS SPECTRAL MEASUREMENT WITH EXAMPLES OF HELA CELL PHOSPHOPROTEOME; 4.1.1 Introduction; 4.1.2 Protein Phosphatase and Kinase; 4.1.3 Hydroxy-Amino Acid Phosphorylation; 4.1.4 Traditional Phosphoproteomic Approaches; 4.1.5 Current Approaches; 4.1.5.1 Phosphoproteomic Enrichment Techniques; 4.1.5.2 IMAC; 4.1.5.3 MOAC; 4.1.5.4 Methylation of Peptides prior to IMAC or MOAC Enrichment; 4.1.6 The Ideal Approach; 4.1.7 One-Dimensional (1-D) Sodium Dodecyl Sulfate (SDS) PAGE; 4.1.8 Tandem MS Approach; 4.1.8.1 pS Loss of Phosphate Group 4.1.8.2 pT Loss of Phosphate Group

## Sommario/riassunto

Phosphorylation is the addition of a phosphate ( $\text{PO}_4$ ) group to a protein or other organic molecule. Phosphorylation activates or deactivates many protein enzymes, causing or preventing the mechanisms of diseases such as cancer and diabetes. This book shows how to use mass spectrometry to determine whether or not a protein has been correctly modified by the addition of a phosphate group. It also provides a combination of detailed, step-by-step methodology for phosphoproteomic sample preparation, mass spectral instrumental analysis, and data interpretation approaches. Furthermore, it i