Development of MS-based analytical methods for glycomics & glycoproteomics

BACKGROUND OF RESEARCH

Dr Hyun Joo An is the director of the Asia Glycomics Reference Site (AGRS), which develops and validates new analytical platforms for glycomic and glycoproteomic analysis in collaboration with government agencies and regional industry. Since 2011, Dr An has been an associate professor at Chungnam National University. She received her BS and MS degrees from Inha University (Incheon, Korea) and her PhD from the University of California at Davis (United States).

Dr. An’s research focuses on bioanalytical mass spectrometry, with applications to glycomics, proteomics, and glycoproteomics. Her specific research interests include bioanalytical method development, biopharmaceutical characterization, disease biomarker discovery, clinical glycomics, and glycobiology. She has authored and co-authored over 60 peer-reviewed publications on these subjects, and holds multiple related patents.

The Asia Glycomics Reference Site (AGRS), under the directorship of Prof. Hyun Joo An, specializes in mass spectrometric characterization of glycans, glycoproteins, and proteins. The AGRS develops and validates new analytical platforms for glycomic and glycoproteomic analysis, using cutting-edge facilities and equipment. In addition to providing core glyco-analytical support to biopharmaceutical companies as well as governmental regulatory agencies, the AGRS conducts basic glycobiological research in collaboration with academic labs around the world.

OBJECTIVES OF RESEARCH

- Efficient and selective method to capture O-glycans from complex mixture
- Isomer-specific glycan separation and quantitation
- Selective enrichment of phosphoglycans using newly designed LC/MS chip
RESULT OF RESEARCH

Despite recent advances, site-specific profiling of protein glycosylation remains a significant analytical challenge for conventional proteomic methodology. To alleviate the issue, we propose glyco-analytical multispecific proteolysis (Glyco-AMP) as a strategy for glycoproteomic characterization. Glyco-AMP consists of rapid, in-solution digestion of an analyte glycoprotein (or glycoprotein mixture) by a multispecific protease (or protease cocktail). Resulting glycopeptides are chromatographically separated by isomer-specific porous graphitized carbon nano-LC, quantified by high-resolution MS, and structurally elucidated by MS/MS. To demonstrate the consistency and customizability of Glyco-AMP methodology, the glyco-analytical performances of multispecific proteases subtilisin, pronase, and proteinase K were characterized in terms of quantitative accuracy, sensitivity, and digestion kinetics. Glyco-AMP was shown to be effective on glycoprotein mixtures as well as glycoproteins with multiple glycosylation sites, providing detailed, quantitative, site- and structure-specific information about protein glycosylation.

The AGRS has developed new and improved analytical techniques for characterization of the glycoproteome, including (in order of increasing complexity and information content) preliminary site mapping, compositional glycan profiling, isomer-specific glycan profiling, glycosite-specific glycopeptide profiling, and glycoproteomic profiling using Agilent 6500 Series Accurate-Mass Quadrupole Time-of-Flight (Q-TOF) LC/MS interfaced with 1260 HPLC Chip cube system that can be applied in glycoproteomics and glycemic research. These new strategies will help us gain a greater understanding of an important yet largely unexplored portion of our biology: the glycome.

REFERENCE OF PUBLICATION


