11th Workshop on Recent Issues in Bioanalysis (11th WRIB) Poster Award winners 2017

The 11th WRIB held in Los Angeles, CA, USA in April 2017. It drew over 750 professionals representing large Pharmas, Biotechs, CROs and multiple regulatory agencies from around the world, from the global bioanalytical community. *Bioanalysis* and Bioanalysis Zone are very proud to be supporting the WRIB Poster Awards again this year, and we feature the profiles of the authors of the winning posters. Visit www.bioanalysis-zone.com to see the winning posters in full.

Winning Poster: Chiral derivatization of d- & l-Eflornithine for enantiomer identification from a racemate & chromatographic separation & chromatographic separation

Abram Brubaker, Neal Simmons, Ashley Brant, Ronald Shoup; AIT Bioscience, IN, USA

Neal Simmons, AIT Bioscience, IN, USA

Neal Simmons is a Senior Scientist II at AIT Bioscience (Indianapolis, IN, USA). He received his PhD in analytical chemistry from Iowa State University and has worked in bioanalytical laboratories at both large Pharma and CROs for 20 years. He joined AITB in 2016 in the LC–MS/MS group and focuses on bioanalytical assay development and validation supporting regulated as well as research projects. Prior to AITB, he spent 9 years in a global Pharma supporting clinical development programs through managing an in-house bioanalytical laboratory and outsourcing. His interests currently include anything unusual on the horizon plus tissues, derivatization, chiral assays and delving into hybrid LBA–LC–MS.

Q Please can you explain about your role at AIT Bioscience?

Working at a CRO requires wearing many hats as you can imagine. My primary role is to support research and method development of LC–MS/MS assays mainly focusing on small-molecule drugs and metabolites. In addition to the laboratory aspects, I also am a principal investigator/primary contact, which means working closely with sponsors on planning, conducting and final reporting of their projects.

Q What were your first thoughts on winning the WRIB Poster Award?

When the poster awards were announced, I was completely surprised. There were many

great posters and research topics presented so it was totally unexpected. I was thrilled to accept as the presenting author, but the work was very much a team effort. This was a project that could have gone in many directions and become extremely complicated, but instead had a very simple and elegant solution. We all appreciated the recognition from the panel as well as the comments and discussions with those who stopped by during the poster session earlier.

Q How did you carry out the work highlighted in this poster?

During the planning stage, we knew that the entire project hinged on one key question. How could a rugged method be devel-

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oped to measure the d- and l-enantiomers of eflo-rithine when enantiomeric reference standards are not currently available? To address the ruggedness issue, Abe decided to look at chiral derivatization, which would form a pair of diastereomers that could then be measured by reversed-phase LC–MS/MS. A similar approach was used in our laboratory a few years ago for chiral amphetamine analysis and it worked well. The final hurdle and heart of the poster, was assigning the proper identity, d- and l-, to the resulting chromatographic peaks without standards. This was solved by preparing our own standards using a published chiral eflo-rithine assay for fraction collection of the enantiomers from a racemic standard.

Q What were the key findings from your research?

Derivatization, chiral in this case, can sometimes provide a simple solution to a complex problem. The poster results and conclusions demonstrate that this approach worked well for measuring chiral eflo-rithine using standard reversed-phase LC–MS/MS conditions. After surveying derivatizing agents,

conditions, and pre-/post-derivatization cleanup, we found that the most effective processing was to perform a protein precipitation of plasma and then directly derivatize the supernatant. After a 5-min reaction, samples were evaporated to dryness then dissolved in an injection solvent. For confidentiality reasons, we could not present the validation details; however, the method demonstrated excellent statistics and chromatography. This was particularly important/surprising when you consider the simplicity of the final procedure. Finally, the assay was set up multiple times over the last 6 months with no difficulties, demonstrating overall ruggedness of this methodology.

Q What are you looking forward to working on over the next year?

We have quite a few unusual projects either queued up or under discussion which involve topics covered at WRIB this year. These include the challenges of assay development for ocular fluids and tissues as well as hybrid LBA–LC–MS. If there are suitable projects, I am also looking forward to expanding my derivatization experience.

Winning Poster: an innovative approach for detecting neutralizing antibodies directed to antibody-derived therapeutics based on the conventional bridging anti-drug antibody assay format

Veerle Snoeck, Carlo Boutton, Annelies Coddens, Benedicte Brackeva, Robbe D'Hondt, Gregory Daelman, Tom F Verhaeghe, Andreia Correia, Hans Ulrichs, Peter Casteels, Marie-Ange Buyse; Ablynx NV, Zwijnaarde, Belgium

Annelies Coddens, Ablynx NV, Zwijnaarde, Belgium

Annelies Coddens is a scientist in the Pharmacology Department of Ablynx (Belgium). She is a biotechnologist by training and obtained her PhD in 2008 in the Laboratory of Veterinary Immunology at the University of Ghent (Belgium). She continued her research as a postdoctoral researcher in the same laboratory and was author of multiple scientific publications and a patent application. She joined the Bioanalysis team of Ablynx in 2013, and was mainly involved in antidrug antibody (ADA) and NAb assay development.

Q What is your role at Ablynx?

In my current role, since 2016 as immunogenicity assay expert in the Pharmacology department of Ablynx, I am overseeing ADA and NAb assay development, transfer and validation, for in-house and outsourced assays in support of preclinical and clinical studies.

Q What were your first thoughts on winning the 11th WRIB Poster Award?

I was really surprised, but felt absolutely honored by the bioanalytical community. Knowing that we

have worked hard on it with the team, this is really rewarding.

Q How did you carry out the work highlighted in this poster?

The development of this novel approach for detection of NAb was a real team effort.

The concept was mainly driven by Veerle Snoeck and further worked out by a team of bioanalytical scientists and colleagues from the Discovery and CMC Department who made this new approach possible and designed and produced the null variant of our

drug molecule, which is the key in this novel concept for NAb testing. I contributed especially in the development and qualification of this assay, which was done by immunogenicity laboratory scientists under my supervision. I am privileged to work with highly talented and motivated assay developers at Ablynx. The development of this assay format required strong cross-departmental cooperation, which at Ablynx, a mid-size biotech company with about 400 employees, is easy to organize and runs very smoothly.

Q What were the key findings from this research?

We describe an innovative approach for detecting neutralizing antibodies directed to antibody-derived therapeutics based on the conventional bridging ADA assay format.

In this assay, we use a null variant of the drug molecule, which is a nonfunctional variant of the drug that is identical to the drug, except for altered CDRs, hence is nonfunctional for binding to the target under evaluation.

Signals generated in this assay reflect antibodies with neutralizing potential as non-neutralizing antibodies are complexed with the null variant, in a way comparable to the conventional drug displacement set-up (confirmatory assay). Main advantages of this method are the ease of development and the detection of neutralizing antibodies at the same sensitivity of the ADA assay, which allows unambiguous immunogenicity data interpretation, and allows the detection of the neutralizing fraction within the ADA sample.

Q What are you looking forward to working on over the next year?

I am looking forward to the start-up of new projects with new bioanalytical challenges and to the progress we will make in ongoing projects. Ablynx has more than 45 proprietary and partnered programs in development in various therapeutic areas (of which 8 are in the clinical development).

And I hope regulatory agencies will appreciate our novel approach for NAb detection and will endorse the use of this concept in our future programs.

Winning Poster: evaluation of potential biomarkers for anti-influenza A drug development programs

John Lowe, Jenny Li, Amy Noyes, Denise Nazzari, Jacqueline McBride, Eric Wakshull, Mauricio Maia; Development Sciences, Genentech, CA, USA

John Lowe, Genentech, CA, USA

I am currently a Principal Scientific Researcher working in the Bioanalytical Sciences Department at Genentech, Inc. (CA, USA). I received my BA in biological sciences from University of California, Berkeley and MS in clinical sciences from San Francisco State University. I joined Genentech, Inc. in 1990 after working for 4 years at University of California, San Francisco where I completed my Master

thesis. I specialize in developing PK, ADA and biomarker assays, to support preclinical and clinical studies for large molecules that are in late R&D stages. My research interests include evaluation of new technologies and application of novel bioanalytical methods in assay development to support preclinical and clinical studies.

Q Please can you explain about your role at Genentech?

I am currently a Principal Scientific Researcher working in the Bioanalytical Sciences Department at Genentech Inc. My main responsibility is to develop rugged analytical methods that include PK, ADA and biomarker assays to support preclinical and clinical studies for large molecules that are in late R&D stages.

Q How does it feel to win the 11th WRIB Poster Award?

I felt extremely privileged and thought it was a tremendous honor for our poster to be recognized in this

prestigious meeting. I would like to thank all of my co-authors and colleagues who had collaborated and offered support for this project.

Q Tell us a little about the work on which the Poster was based?

In conjunction with Genentech's clinical development of anti-Flu A therapies, we evaluated Simple Plex assay technology for the detection and measurement of eight inflammatory biomarkers of potential interest in the context of this disease. Assay performance was assessed in three matrices of pharmacological interest including serum, nasal swab (NS) and tracheal aspirate (TA) samples. Our assessments included analyte detectabil-

ity, assay reproducibility and sample dilution linearity. Sample analysis was performed in the Simple Plex 4-plex assay for a panel of eight biomarkers: IL-6, IL-8, IL-10, IP-10, MCP-1, IFN- γ , TNF- α and TNF- α -R1. Results for controls of all biomarkers were accurate and reproducible. The assay also showed acceptable detectability, reproducibility and linearity of dilution for serum and NS samples. In addition, Simple Plex 4-plex assay results showed good correlation with archival data for IL-8, MCP-1 and IP-10, when measured in serum as well as in NS samples.

Q What were the key conclusions from your research?

Various inflammatory biomarkers that may potentially be used during clinical development of anti-Flu A therapeutic molecules were evaluated. Our work demonstrated that the Simple Plex platform is sensitive, reproducible and multiplexable, with a simple assay procedure, limited hands-on requirements and short total assay time (1 h); however, our results also indicated that not all analytes meet our performance requirements and not all matrices are suitable for this technology. The Simple Plex 4-Plex assay performed adequately and we will further qualify the assay to support clinical studies.

Q What are you looking forward to working on over the next year?

We showed that with TA samples, detectability and dilution linearity were acceptable, whereas reproducibility was not. This lack of reproducibility with TA samples was likely due to the heterogeneous nature of the samples, which may be mitigated by implementing sample-pretreatment methods. Therefore, additional work is needed to improve reproducibility of biomarker quantitation in TA samples.

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Financial & competing interests disclosure

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No writing assistance was utilized in the production of this manuscript.