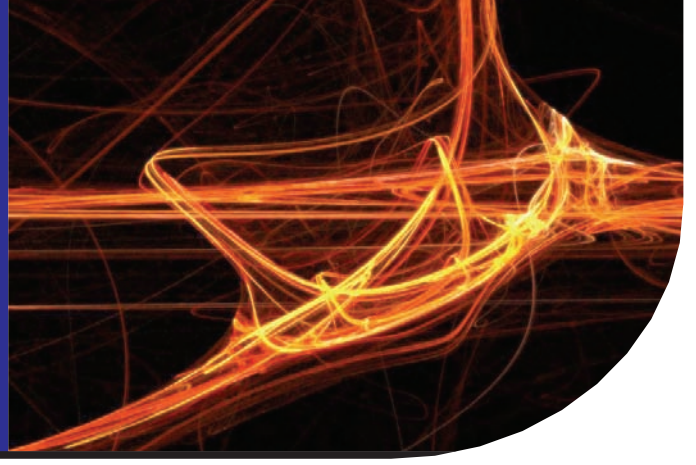

Glycan Analysis Opens Up New Possibilities For Mass Spectrometry

Report on the 58th ASMS Conference, Salt Lake City, Utah, USA May 23rd – 27th

Once again the mighty ASMS Conference flexes its muscles and heads into town, on this occasion Salt Lake City, to allow practitioners of the technique to meet, network, hear presentations from the great and the good, view poster sessions and see the latest offerings from the manufacturers and suppliers. The scale and credibility of this scientific meeting is certainly having an effect on the attendance on more generalist scientific meetings in North America such as the Pittsburgh Conference on Applied Spectroscopy and the HPLC series of shows held in the US every two years. Its organisation is something that other meeting organisers could take some points from, especially the rates charged to attendees. Having noted that point it is also in danger of becoming a victim of its own success and scale if it continues to expand at the current rate. Clear, selective planning was required by attendees prior to even heading out to the venue as to which oral and poster sessions they should be attending. The event this year had in excess of 6,000 attendees, over 160 exhibitors and over 2,700 posters. In addition to that also on offer were over 50 oral sessions, countless workshops, short courses and Tutorial sessions and a spare mental hard drive was a necessity.

> ***"The UPLC Glycan Solution coupled with Waters mass spectrometers provides additional structural confirmation of the glycans..."***

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AWARDS

Recognition of one's contribution for the advancement of science in general should be the aim of all scientists. In this case the topic is Mass Spectroscopy. An award from the ASMS committee for Distinguished Contribution in Mass Spectroscopy was made to Dr Martin L. Vestal for his work in the development of the first commercial MALD-TOF Instrument - the Voyager series. Such is the importance of Dr Vestals work and its relevance today is the fact that over half of the commercially available Instruments utilise elements of his design.

The Biemann Medal was presented to Dr David C. Muddiman from North Carolina State University for his work on 'hydrophobic tagging', which not only resulted in a sensitivity gain of one order of magnitude on ESI (electrospray ionisation) by the addition to samples of a hydrophobic alkyl chain molecule, but also was utilised to improve the response of peptides in ESI systems.

Finally the Ron Hites award for outstanding Research Publication in JAMAS (Journal of the American Society for Mass Spectrometry) went to Professor Facuundo Fernandez from the Georgia Institute of Technology for his article on 'Direct Quantitaion of Active Ingredients in Solid Artesunate Antimalarials by Noncovalent Complex Forming Reactive Desorption Electrospray Ionisation Mass Spectroscopy' [1]

APPLICATION AREAS

No real surprises in that the 'usual suspects' from previous years were to the forefront and in all likely hood will continue to be the main stay for the pure mass spec and hybrid, LC/MS/MS and GC/MS/MS markets. Proteomics (indeed most – omics sub groups) and Bio-markers tended to predominate although Forensics, Clinical and Food and Beverage are also increasing. And with 2010 being a Football World Cup year Drug Testing applications will also have a high profile ensuring we have a level playing field on that front for all competing teams.

One exception to that generalisation which saw both Agilent and Waters launching packages is the area of Glycan research, highly important in the growing biopharmaceutical market place. Glycans are branching polymeric sugar molecules that are attached to biopharmaceuticals – proteins, peptides, monoclonal antibodies – following protein translation. The final result of glycan addition and maturation reactions within the cell is a heterogeneously modified biotherapeutic where the various glycosylated forms can differentially affect the efficacy and safety profile of the biopharmaceutical product. Determination of the sites of glycan attachment on the protein and the population of glycans that are present at each site are required for both therapeutic and safety assessments. The complexity of all possible glycan structures places tremendous demands on the analytical techniques used for their analysis. Yet, consistency of glycosylation is routinely used as a sensitive marker for demonstrating control of the biotherapeutic production process. The U.S. Food and Drug Administration, and other regulatory agencies, have handed down guidance requesting stricter control of product glycosylation, leading to greater industry investment in analytical techniques that promise to facilitate a better understanding of these complex biotherapeutic drug products.

The Waters UPLC Glycan Analysis Solution comprises an ACQUITY UPLC BEH Glycan Separation Technology column optimized for use with the ACQUITY UPLC® System together with fluorescence detection (FLR) to separate the released glycans of biopharmaceuticals as their 2-aminobenzamide (2-AB) or other fluorescent derivatives. The UPLC Glycan Separation Solution is designed to provide robust, high-resolving, reproducible, and rapid methods that out-perform analyses by HPLC. The high resolving power and sensitivity of the UPLC Glycan Solution allows laboratories to separate and detect isobaric isomers (same mass, different retention time), a wide range of glycans - high mannose, neutral and sialylated - and low abundant glycans – those present in very small concentrations relative to other glycans.

The UPLC Glycan Solution coupled with Waters mass spectrometers provides additional structural confirmation of the glycans.

As an MS/MS data analysis tool, SimGlycan software predicts the glycan structure on a protein molecule scores it and generates a list of probable glycans that closely match the given MS profile. The SimGlycan database is a large relational database containing 8,553 glycans and the database is continuously updated as information on additional glycans is published. Both glycopeptide and released glycan profiles are supported.

The Agilent approach resolves around its Chip technology and is associated with monoclonal antibody molecules. N-glycans and monoclonal antibody molecules are a critical part of biopharmaceutical research and analysis. A unique product for a specific requirement, the mAb-Glyco-Chip improves the productivity of biopharmaceutical analysis at least 20 times when compared to the current CE-FLD method, and at least five times when compared to the current MALDI-MS method. Conventional methods of N-glycan analysis, such as LC-FLD or CEFLD, can take two to three days for completing one single analysis. The new HPLC-Chip/MS workflow significantly reduces the total analysis (including on-chip enzymatic reaction, glycan separation and MS analysis) to tens of minutes. This creates a major productivity enhancement for biopharmaceutical analysis. Coupled with an Agilent Accurate Mass TOF or Q-TOF mass spectrometer, the combined system allows for complete, rapid characterisation and quantitative profiling of glycan structures on recombinant antibodies.

In terms of sample origins the Forensics application areas do appear to striking a cord with many of the young scientists, perhaps as a result of the plethora of TV CSI seemingly everywhere type programmes, resulting in blood spot and bio-analytical assays gaining more interest. Indeed Agilent Technologies launched a LC/MS Forensic Toxicology kit for use with its LC-QTOF MS. The application kit is designed to allow users to quickly and easily set up screening Experiments that produce exceptionally high-confidence results. The kit combines the specificity of an accurate mass database with the additional confidence of high-quality, accurate mass MS/MS spectra in a personal compound database and library (PCDL). The kit is a complete package with recommended methods and application note according to the company.

INSTRUMENTATION

As with most major International shows the 'big guns' choose the event to launch onto the market place this years efforts from their R+D Departments and announce any strategic business deals they have been working on since the last meeting. That is not to say that these offerings are the most technically innovative or advanced on the market nor offer the most benefits to the customer since smaller suppliers to the Mass Spectrum community also have specialised skills at their disposal which are not always matched by their marketing Budgets.

In addition to the Glycan orientated product line Waters also launched two new Mass spectrometers for its Xevo MS platform – the Xevo TQ-S and Xevo G2 QTof – 'that bring a step change in performance to benchtop mass

spectrometry.' The Waters Xevo TQ-S is a tandem mass spectrometer. It is capable of accurately measuring target compounds in complex samples at femtogram levels or below. The Xevo TQ-S is designed for UPLC/MS/MS applications such as biological and medical research, bioanalysis, food safety, environmental monitoring and forensics testing where speed, sensitivity, and accuracy are of primary importance.

The Xevo TQ-S features new StepWave™ ion-transfer technology, a revolutionary off-axis design for dramatically increasing the efficiency of ion transfer from the ion source to the quadrupole MS analyser while at the same time actively eliminating undesirable neutral contaminants. This gives the Xevo TQ-S the claimed increase in sensitivity. When compared to earlier generation mass spectrometer models the chromatographic peak areas are typically more than 30 times bigger and the signal-to-noise ratios are typically 5 to 10 times better.

The novel, high speed transfer optics and collision cell design allows the TQ-S system to acquire both full scan MS and MRM data simultaneously on narrow, 1-2 second wide UPLC peaks and still obtain >12 points across the peak, something that is particularly useful in methods development when dealing with a complex matrix, for monitoring for the presence of new components in the sample during routine analysis or troubleshooting an assay.

With the Xevo TQ-S mass spectrometer, scientists can now confidently quantify compounds at lower concentrations than previously possible. The instrument's sensitivity allows scientists to consider diluting samples in order to reduce matrix effects that interfere with compound measurements. In addition to this, the ability to work with smaller sample volumes, a big benefit when analysing precious biological samples taken from animals or humans, becomes possible.

Agilent launched their new highly sensitive (company claims to be 'the most sensitive in the world') triple quadrupole (TQ) MS. Claimed to be capable of working at the 10-21 (zeptomole) range it is aimed at pharmaceutical researchers looking at drug and metabolite components at previously levels of quantification.

Bruker launched several new products amongst which the new Fourier transform mass spectrometer (FTMS) appeared to raise the most interest with its application to MALDI imaging sources. Once again aimed at the Phamaceuticals research market the Instrument is claimed to 'provide unrivalled platform for high performance molecular imaging (HPMI?) of small molecules (drugs, metabolites lipids) in tissue.'

Thermo Fisher Scientific are still finding new avenues for their LTQ Orbitrap Velos® technology and they heavily promoted their 'metabolimic screening workflow solutions' which rely heavily on the technology. Proxean, recently acquired by the company, promoted their Easy –nLCTM II second generation LC-MS aimed at the Proteomics market and designed to allow scientists to improve productivity with improved new autosamplers and software.

Not forgetting that Thermo Fisher also have GC-MS platforms the company also launched two new instruments, the GC-QMS: the ISQ, and a GC-TQMS: the TSQ Quantum XLS. The ISQ is designed to offers rugged and reliable performance and nonstop productivity enhancements compared to previous single quadrupole models and the TSQ Quantum XLS 'delivers the highest sensitivity with femtogram-range limits of detection and unrivaled performance in GC-MS/MS structure selective detection for multi-component target compound quantitation.'

At the IMSC (International Mass Spec Conference) held in Bremen, Germany last September it was announced that ABI Sciex was being acquired by a company, Danaher, who had no prior involvement within the mass spec market and the community was awaiting what direction it would take one of its most innovative and largest companies in. The answer was to be found in the ABI Sciex Triple TOF 5600 series. Designed to offer quantitative and qualitative capabilities to scientists working on profiling biological samples the company summed up the concept behind the Instruments' development as ' being designed to integrate comprehensive qualitative exploration, to rapid and high resolution quantitation.'

Perkin Elmer launched their Flexar TM SQ 300 MS platform. The mass detection system provides a rugged ion source design and wide molecular weight detection range for HPLC and UHPLC applications. Suitable for use in the pharmaceutical and chemical industries, the Flexar SQ 300 MS platform enables efficient and reliable ionization of compounds in both positive and negative modes for the efficient analysis of a broad range of analytes. For food and environmental testing applications, a patented multi-stage ion path allows for multi-residue analysis with high limits of detection sensitivity. Moreover, advanced Collision Induced Dissociation (CID) technology enables fragmentation for further confirmation of molecular structure.

Flexar SQ 300 MS offers ease of switching between the Ultraspray ESI and Field-free APCI ion sources and fast interchangeability of probes. The probes can be specific to a user, application or sample type, helping to minimise the risk of cross contamination and instrument downtime. Its patented grounded ion source design allows for quick and safe probe installation. The probe position can be easily optimised for different flows and chemical environments to maintain excellent ionisation performance.

Shimadzu Scientific Instruments showed a recently developed platform, the Nanotrap® Biomarker Discovery Platform in partnership with Ceres Nanosciences and Nonlinear Dynamics giving researchers the ability to enrich, preserve and screen low level biomarkers, directly from complex biological samples. Nano traps are core shell hydrogel particles with porous interiors and size exclusion shells. Affinity and size exclusion are delivered in one step allowing rapid amplification from small sample quantities.

SOFTWARE

Since the science of mass spectrometry depends upon the availability of large easily searchable databases and the requirement for the ability to quickly deconvulute data there were many new software offerings from specialist companies. Instrument companies who did not acquire these specialist companies relied on their own existing systems and usually offered an upgrade to previous versions with commensurate customer benefits.

FOLLOW THAT

Dates and venues for the forthcoming ASMS meetings have been announced as follows:

June 5-9th 2011 Denver, CO
May 20-24th 2012 Vancouver, BC, Canada
June 9-13th 2013 Minneapolis, MN

REFERENCES

1. Leonard Nyadong, Sameer Late, Michael D. Green, Anjay Banga and Facundo Fernandez; JASMS, 2008, Vol. 19, 380-388

FINAL THOUGHT

With all the claims from the manufacturers of having the 'most sensitive, fastest, most accurate, highest resolution etc. (strike out whichever are not of interest) Instruments when are we going to see a neutral evaluation of the market leaders offerings to decide upon the Gold, Silver and Bronze recipients of those titles, until the next ASMS anyway?

South African Doping Control for 2010 World Cup

Agilent Technologies, Inc instruments were selected by the South African Doping Control Laboratory as the primary provider of gas-phase testing instruments for the 2010 World Cup. The South African Doping Control Laboratory is certified by the World Anti-Doping Agency and will be responsible for all doping testing during the competition.

The South African Doping Control Laboratory is equipped with state-of-the-art Agilent gas chromatography (GC) and mass spectrometry (MS) instruments to confirm the chemical identity of suspected banned substances found in testing samples.

Agilent provided five GC/MSD systems and an Agilent 7000 Series Triple Quadrupole GC/MS system. Agilent technicians provided technical support throughout the event.

"The use of the 7000 Series Triple Quadrupole GC/MS system from Agilent has raised the standard of doping testing to a higher level," said Dr Pieter J. van der Merwe, Director, South African Doping Control Laboratory. "This instrument is significantly increasing the sensitivity and specificity of detection."

"Agilent is proud of our leadership role in anti-doping testing instruments, which dates back to the 1970s," said Mike McMullen, President of Agilent's Chemical Analysis Group. "We are honored that the South African Doping Control Laboratory selected Agilent to help ensure a level playing field and fair competition at the 2010 World Cup."

Circle no. 27

The Highest Performance in its Class

Shimadzu has launched a new GCMS-QP2010 Ultra gas chromatograph mass spectrometer (GC/MS) offering the highest performance in its class. It provides fast data measurement speed with five times higher sensitivity than previous models.

In addition to improving laboratory productivity for high-speed analysis, it also offers improved applicability for comprehensive two-dimensional gas chromatography (GC×GC), which can achieve ultra high separation. It is also the first GC/MS system to have environmentally friendly features for saving power consumption and carrier gas.

The GCMS-QP-2010 quadrupole series have been presented at the 34th ISCC event (International Symposium on Capillary Chromatography) in Riva del Garda, Italy. In the same event, the world premiere of the new ChromSquare software took place, a software for Comprehensive Chromatography and providing of in-depth insights at any point in the process.

The GCMS-QP2010 Ultra incorporates the new GC-2010 Plus setting new standards in speed, efficiency, precision, sensitivity and ease of operation. Featuring the specially designed AFT (Advanced Flow Technology), the GC-2010 Plus combines highest separation efficiency with increased productivity and reduced analysis time.

The GCMS-QP2010 Ultra's most important features: significantly improved scan speed and sensitivity in fast analysis; high-speed oven cooling for shorter analysis time; twin line MS system eliminates the need to swap columns; and the first environmentally friendly GC-MS.

The new GCMS-QP2010 Ultra fulfills laboratory demands for enhanced productivity through various technologies, which improve throughput, and helps to reduce operating costs while minimising environmental impact with its Ecology mode. With the introduction of this product, Shimadzu aims to increase its market share of gas chromatograph mass spectrometers worldwide.

Circle no. 28