

	Edison Room	Boardroom 2	Woodbridge room
4:00-4:30 p.m.			LabVIEW Workshop by Kapeeshwar Krishana (Princeton University and LRIG)
4:30-5:00 p.m.			
5-5:30 p.m.	IDBS: Improving the Quality of Valuable screening data	Thermo: High Throughput Dispensing Using Thermo Electron's Multidrop Combi Bulk Reagent Dispenser and RapidStak Microplate Stacker	Chemspeed: Accelerating Chemistry Workflows by automated High Output Technologies
5:30-6:00 p.m.	MDS Sciex: CellKey™ System- A Powerful New Automated Approach to Cellular Analysis Presented by Kathy Zhao, MDS Sciex	Merck: Evaluation of a Novel Cleaning Technology for Simplifying and Increasing the Efficiency of siRNA Screening	Agilent: Automated Protein and DNA Analysis: The Agilent 5100 Automated Lab-on-a-Chip Platform
6:00-6:30 p.m.	Global Array: Continuous Tape Based Automation as a Replacement for Microplates	Artel and Caliper workshop. "Method Validation for Caliper Liquid Handlers using the Artel MVS"	Kardex Engineering: Materials Management Principles and ASRS for Benchmark HTS
6:30-7 p.m.	Biacore: Combining in-silico techniques and selectivity based label free screening as a powerful tool for drug discovery		Sirius Analytical: pKa measurement in four minutes – the key to measuring drug solubilities at 37°C
7-7:30 p.m.	Beckman Coulter: Introducing New Biomek Liquid Handlers with software and hardware advancements that help achieve reliable and accurate results for laboratory automation	FOCUS Biology	

IDBS: Improving the Quality of Valuable screening data

Abstract: Today's scientists face increasing demands to handle, analyze and validate vast amounts of screening data. The XE module for ActivityBase™ provides specific functionality to address these demands by providing a single environment that is easy to use, flexible and intuitive. **ActivityBase™ XE** facilitates data visualization, analysis, QA and QC, and verification, resulting in improved workflow and productivity.

This workshop will address how to achieve ultra high performance screening and streamline workflow for the screening scientists by providing enhanced data visualization, plate format flexibility, sophisticated curve fitting, data selection, multi-level annotation and automatic plate exclusion. Join this workshop to learn more about **ActivityBase™ XE**, a comprehensive, flexible and dynamic tool to manage and drive your screening research.

Accelerating Chemistry Workflows by automated High Output Technologies

Lei Xiao, Ph.D.

Support Chemist

Chemspeed Inc., 7 Deer Park Dr.; Suite L, Monmouth Junction, NJ 08852, USA
Phone: 732 329 1225, Fax: 732 329 1225, e-mail: josef.schroer@chemspeed.com

The use of automated High Output Technologies has expanded from its origins in Biotech and CombiChem into laboratories of many other areas in chemistry and related science. Highly innovative new fields like Catalyst-, Polymer- and Material Science successfully have implemented High Output Technologies to significantly increase their workflow efficiency.

For this reason equipment that was originally designed to accelerate the process of Drug Discovery, is now common in all laboratories where multiple dispensing of liquids and solids represents one of the core requirements.

Further needs strongly depend on particular workflows and are governed by the characteristics of individual applications. Those needs might cover a very broad range starting from highly sophisticated conditions, like the use of reactive gases under elevated pressure, to relative “simple” techniques like extraction, filtration or evaporation.

For this reason only flexible, highly modular, and scalable equipment has the chance of covering the diverse needs of these industries.

The general requirement for increased output and efficiency has boosted the continued evolution of automated laboratory equipment.

Herein, we will describe an innovative robotic platform that will not only allow fully automated dispensing of solid and liquids, but also enables the user complete flexibility to configure the platform with the appropriate hardware for the application of his choice.

Using selected examples, from a number of industries, this presentation will show how a variety of challenging parallel workflows fully have been automated on Chemspeed instrumentation.

CellKey™ System- A Powerful New Automated Approach to Cellular Analysis Presented by Kathy Zhao, MDS Sciex

Cell surface receptors and their associated signaling pathways are attractive drug targets. Drug discovery programs for GPCRs and TKRs rely on a variety of assays that traditionally involve radioactive, fluorescence and bioluminescence-based detection. MDS Sciex will highlight a revolutionary label-free assay platform called the CellKey™ system, which is based on cellular dielectric spectroscopy (CDS). We present data that illustrates the universal ability of the system to measure, in real-time, endogenous cell surface receptor activation in live cells. We compare the CellKey™ platform to other cell-based assays and show its use in analyzing pharmacology in cell lines and primary cells. Through dissection of signal transduction pathways, we correlate the CellKey™ response to cellular physiology and identify the G-protein coupling mechanism of uncharacterized GPCRs. Lastly, we show examples of how the CellKey™ system's high information capabilities and its ease of use can significantly reduce assay development and costs in drug discovery.

Evaluation of a Novel Cleaning Technology for Simplifying and Increasing the Efficiency of siRNA Screening

Erica Stec, Merck's Automated Biotechnology Lab

Plasma technology has been shown to be a reliable alternative to the more typical tip wash or replacement techniques currently used with automated liquid handling devices throughout the biopharmaceutical and diagnostic industries. The TipCharger System utilizes cold plasma with disposable polypropylene micropipette tips to clean without the use of traditional wash solvents or waste generation.

The use of a dry cleaning method eliminates the problems associated with contamination in wash solutions and delivers results comparable to or better than existing washes in less than half the time. Evaluation goals in the Automated Biotechnology Lab in North Wales included simplification of tip wash processes, reduction/elimination of biological waste generation, and cost savings through reduction in assay time and consumable usage.

Comparative analyses of tips cleaned with conventional DNase/RNase free water and TipCharger generated plasma in siRNA assay protocols will be discussed.

Automated Protein and DNA Analysis: The Agilent 5100 Automated Lab-on-a-Chip Platform

Phillip M. Douglass, Ph.D.
Senior Applications Scientist, Agilent Technologies

This seminar will describe the Agilent 5100 Automated Lab-on-a-Chip Platform (5100 ALP), newly introduced by Agilent. This state-of-the-art instrument, incorporating advances in microfluidic and automation technologies, is ideal for high throughput sizing and quantitation of DNA and proteins traditionally done on Agarose slab gels or with SDS-PAGE. The Agilent 5100 ALP provides the speed and precision needed by labs in the pharmaceutical and biotech industries as well as academia and government institutions. Processing 50 to 2,000 samples per day with ease, the 5100 ALP replaces the tedious and time-consuming sample handling common of slab gels with fully automated, unattended operation, 24 hours a day. An integrated Oracle® database opens the door to previously unavailable opportunities for storage, queries and filtering of large data sets which result from high throughput analysis.

Continuous Tape Based Automation as a Replacement for Microplates

Increased throughput in screening operations has driven the industry towards complex robotics systems to handle the station-to-station processing of microplates. The requirement for individual handling of microplates can be eliminated by implementation of instrumentation and materials utilizing a continuous tape based process instead of microplates. Advantages of this technology include streamlining of automation, savings on consumables, and an ideal format for miniaturization of assay reaction volumes. This presentation will outline how a continuous tape based automation system has replaced a traditional microplate based system for a genomics application at Prevention Genetics in Marshfield Wisconsin, with a resulting cost savings of 90% on materials and reagents.

Materials Management Principles and ASRS for Benchmark HTS

Mel Reichman, Ph.D.
PharmDC, West Chester, PA
mreichman@pharmdc.com

Management of large compound libraries for successful drug screening requires focus on minute details and operational discipline. The origins of the field hail to natural products screening, wherein a core team managed large libraries of samples having poorly characterized properties, sometimes prepared under less than rigorous conditions. The history of drug screening reveals interesting similarities to modern HTS operations, including managing troublesome solubility issues, deployment of novel replication tools and parallel pipetters, clever bioassay design, and accelerated drug screening with innovative plate matrices and rapid plate scanning (with sophisticated 'retinal detectors') for binary hit selection.

Unfortunately, a poor confirmation rate for actives observed in the primary screen remains the norm rather than the exception in HTS through today, which some ascribe in part to compound management issues. Starting in the mid-90s, pharma began to rapidly expand their compound libraries and made huge capital investments in their compound repository infrastructure by acquiring installing Automated sample retrieval systems (ASRS).

We will assess the key issues and solutions critical to benchmark sample management for valid drug screening and review 'State-of-the-Art' and the impact of ASRS-Driven Materials Management.

pKa measurement in four minutes – the key to measuring drug solubilities at 37°C

John Comer

Sirius Analytical Inc. Suite S120, South Tower, 255 Old New Brunswick Road, Piscataway, NJ 08854. www.sirius-ai.com

Most published solubility values have been measured at 25°C. The human body is at 37°C, but it is difficult to make traditional shake-flask measurements at this temperature; unless the entire apparatus is at 37°C, problems occur such as precipitation of sample from saturated solution as it cools inside pipettes or instruments. We have developed a way to measure aqueous solubility of drugs at 37°C using CheqSol. This new technique of chasing equilibrium solubility during a UV-assisted pH-metric titration is done in a temperature controlled glass vial maintained at 37°C. The volumes of acid and base added during the titration are small, and temperature equilibrates rapidly after each addition.

The CheqSol technique requires accurate pKa values for the drug, measured at the same temperature as the solubility. pKas of poorly water-soluble samples must be measured in water-solvent mixtures (e.g. water-methanol), but methanol evaporates quickly at 37°C, and volume change during experiment will affect the result in traditional pKa measurement techniques.. We have developed a new pH-UV method called Fast D-PAS to measure pKa in four minutes at 37°C. Acid-base titrations are done in a mixture of buffer solutions, which stabilises the pH electrode reading, allowing data points to be collected rapidly. Very little solvent evaporates in four minutes, providing accurate measured pKa values at 37°C.

Using this method, we measured the solubility of a number of drugs at 37°C. We observed that solubility of some drugs barely changed between the two temperatures, while others were significantly more soluble at higher temperature. For example, we have noted no significant change in the solubility of chlorpromazine between 25°C and 37°C. However, sulfamerazine was 50% more soluble at 37°C than at 25°C, while diclofenac was 100% more soluble at 37°C than at 25°C. Solubility differences like these could affect bioavailability, as drugs need to be in solution before they can permeate through membranes in the body.

Combining in-silico techniques and selectivity based label free screening as a powerful tool for drug discovery

Pär Säfsten, Technical Project Manager

Dept. Systems and Applications, R&D

Biacore AB, Uppsala, Sweden

Candidate drug compounds are commonly screened by inhibition assays that reflect their affinity for the target protein. Binding selectivity is also a critical property, however, and assays providing both affinity and selectivity data at a relatively early stage may greatly facilitate the drug discovery process. Small molecules with good affinity and selectivity properties for the targeted subunit of a heterotrimeric protein were identified using in-silico modelling of the natural antagonist. The selected compounds were assayed for binding to one subunit and the full-length protein using a new protein interaction array system, Biacore[®] A100 (Biacore AB). 60 Compounds with desired selectivity were then re-analyzed against a more extensive protein panel. Parallel analysis against full-length isomers, wild-type and mutant subunits and unrelated control proteins provided comprehensive affinity and selectivity information enabling well-informed selection of candidates. These findings could not have been obtained by conventional single-target analysis, demonstrating the value of a multi-protein panel approach. Data will be presented to show that compounds selected using such a strategy express strong potency in a cell-based activity assay. These results and others will be used to demonstrate the use of Biacore A100 in the drug discovery process.

Introducing New Biomek Liquid Handlers with software and hardware advancements that help achieve reliable and accurate results for laboratory automation.

Beckman Coulter

Beckman Coulter will introduce the new Biomek FX^P and Biomek NX^P Liquid Handlers. These new systems have many hardware and software enhancements that improve performance and reliability. A new version of Beckman Coulter's Biomek software - v3.3- is also being introduced, with improvements that include enhanced password protocols on methods and improved tools for method organization and retrieval. This presentation will highlight their capabilities in a series of case studies on cell-based assays, high-density microarray target prep, and ADMETox applications.

Artel Caliper Workshop

All users of automated liquid handling equipment are familiar with the need to ensure proper performance of their equipment. But for most users, calibration and performance testing is time consuming and burdensome. The Artel MVS Multichannel Verification System can help reduce the burden of calibration testing and also significantly reduce the time needed to optimize performance or validate test methods. Caliper Life Sciences Liquid Handling Equipment allows for easy optimization of aspirate/dispense protocols, ensuring precise and accurate instrument performance. This demonstration will highlight not only the ease of use of the Artel MVS, but also the performance capabilities of Caliper equipment, and the benefit of validating critical test protocols.

High Throughput Dispensing Using Thermo Electron's Multidrop Combi Bulk Reagent Dispenser and RapidStak Microplate Stacker

Osama El-Badry, Ph.D.
Field technical sales specialist
Mid Atlantic and South East regions

- Microplate Instrumentation
- Spectrophotometry

Laboratory Equipment Division
Thermo Electron Corporation

Accurate, fast, high throughput dispensing has become essential in today's laboratories. To address this need Thermo Electron introduces the Multidrop Combi, a bulk reagent dispenser that combines all the great features of previous Multidrops microplate dispensers, and adds several new features that make it very attractive to end-users. The Multidrop Combi offers:

- Wider volume range: 0.5ul to 2500ul.
- Greater plate flexibility: can dispense into 6-, 12-, 24-, 48-, 96-, 384- and 1536-well plates
- Ability to dispense into plates of varying heights: 5-50mm
- Ability to dispense to selected columns
- Ability to dispense different volumes into different columns
- Very easy to use interface and programming
- Performance, as measured by CV, is exceptional
- Finally, the Multidrop Combi may be operated independently, or with our RapidStak microplate stacker, or other robots.

FOCUS Research (FR) is a multi-factor analysis tool which can take advantage of your investment in LIMS, and help lower production costs and improve troubleshooting. FR can automatically identify factors and factor sets (where factors work together, but not necessary alone) that contribute to specific outcomes. This is particularly useful in complex, high throughput environments where increasing efficiency and decreasing downtime is very important.

Posters

1) A Novel Screening Platform for Transporter Proteins

IonGate Biosciences

2) Achieving Ultra High Performance Screening

IDBS

3) A fast method for measuring solubility of ionizable molecules

Sirius Analytical Inc

4) Inert Sample Processing - Solutions for Combating Sample Precipitation in Compound Libraries

Authors: Mel Reichman, PhD (PharmDC), and Sandesh Gowda (KEMS)

5) Expanding the Suite of Assays Technologies That Can Be Reliably Run in 1536 Well Format

Rose Hughes[1]; Robert Bukar[1]; Ben Baumann[1]; Nathan Heinrich[2]; Paul Queeney[1]

¹ Kalypsys, Inc., 10420 Wateridge Circle, San Diego, CA 92123

² Remmele Engineering, 677 Transfer Road, St. Paul, MN 55114

6) Vari Syringe

John Brohan, Traders Micro

7) Micro-High Speed Liquid Chromatography Coupled to an Evaporative Light Scattering Detector for the Rapid Analysis of Compounds of Pharmaceutical Interest

Tovatech LLC

8) A novel generic system for kinase assays and real-time enzyme and inhibitor kinetics

Nanostream

9) Applied Biosystems (Joseph Frank)

10) High-throughput automation for RNA isolation from blood stabilized in PAXgene™ Blood RNA Tubes

Qiagen

11) The BioRobot® Universal System – Multiple Applications on One Instrument

Thomas Weierstall; Anja Schultz; Martin Heller and Carola Schade

QIAGEN GmbH, 40724 Hilden, Germany

12) Securely Publishing Screening Data to Multiple User Groups with a Lightweight Apache Web-server Application

Ken S. Stoney and Frank D. Mentch, Ph.D.
Edge4 Technologies

13) Centralized vs. Multi-Site Compound Management REMP, Donat Elsener (REMP AG) and Michael Girardi (REMP USA)

14) Kalypsys: Identifying hits and elucidating targets from a Neutrophil Oxidative Burst uHTS in 1536-well microplates

15) Non-Contact Dispensing of Yttrium Oxide Beads in 1536: Justin Murray, MERCK

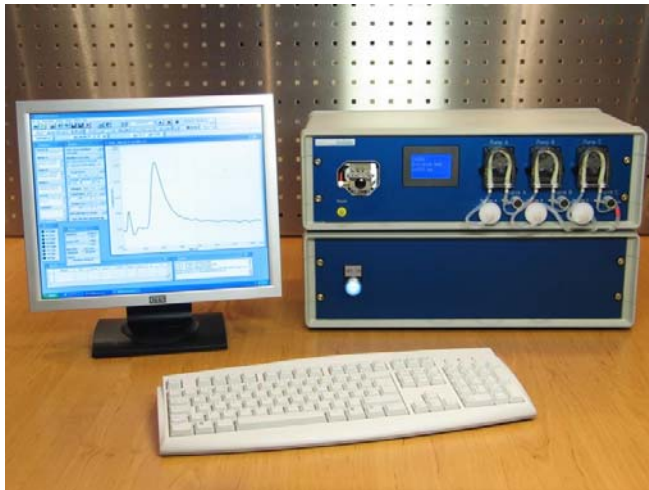
A Novel Screening Platform for Transporter Proteins

Carsten Haber, Wolfgang Doerner, Jim Richey
IonGate Biosciences

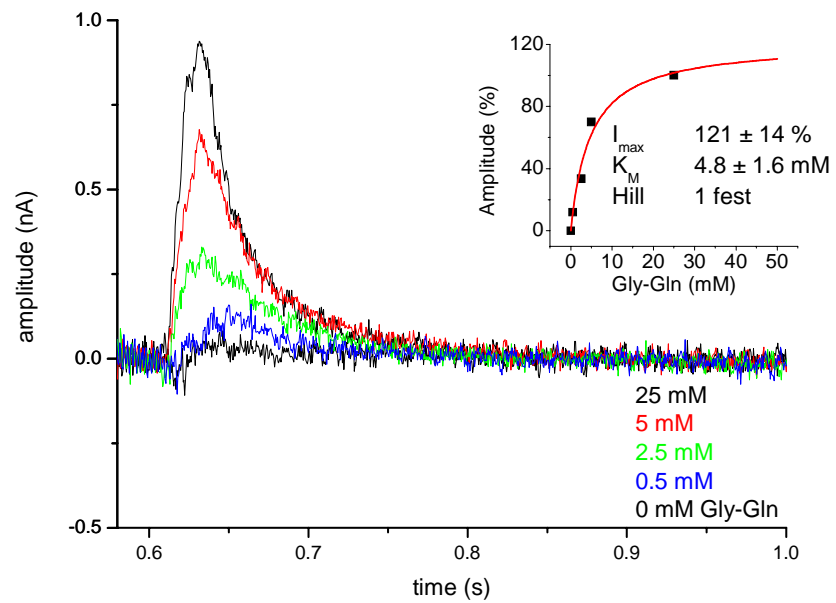
IonGate Biosciences (Frankfurt/Germany – www.iongate.de) has developed an electrochemical biosensor which permits robust and label-free screening of electrogenic membrane proteins on solid supported membranes (SSMs).

The central element of the instrument is a specially treated gold surface which is designed to specifically adsorb transporter-containing membrane targets. The membrane components self-assemble on the gold surface to form a large number of small vesicles doped with transporter molecules. These solid supported membranes are highly stable and enclose isolated small volumes of buffer from the bulk solution. The SSM acts as a carrier for the membrane fragments, and in parallel as a high-capacitance, low-conductance electrode. Via rapid solution exchange, substrate ions are moved by energy (ATP)- or gradient driven transport processes out or into the vesicle cell and trigger a capacitive charging current on the underlying sensor surface.

The sensitivity of the sensor is sufficient to detect electrogenic binding of substrates or single turnover reactions within the protein. To date, seventeen different transporter proteins have been investigated using IonGate's biosensor. The technology is highly scaleable and a workstation operating on a 96 well plate will be available later this year.



SURFE2R benchtop unit with software interface



Electrogenic transfer currents for different substrate concentration levels of Gly-Gln

Achieving Ultra High Performance Screening

High throughput (HT) and Ultra High Throughput (UHT) screening remains a valuable and necessary approach to modern drug discovery. With the use of numerous standardized biochemical assays now commonplace, successful application of HTS requires accurate and meaningful handling of the experimental results. Specifically, this means capturing vast data volumes generated by HTS and UHTS programs and subsequent validation and verification. Decision-making regarding HTS data can be supported via comprehensive and intuitive visualization of results. Also important is the establishment and maintenance of analysis templates for this type of experiment. In order for HTS and UHTS to be successfully implemented in drug discovery, all the aforementioned issues must be addressed.

A fast method for measuring solubility of ionizable molecules

Karl Box, John Comer, Martin Stuart
Sirius Analytical Inc. Suite S120, South Tower, 255 Old New
Brunswick Road, Piscataway, NJ 08854. www.sirius-ai.com

Some ionizable molecules form supersaturated solutions; others do not. We have developed CheqSol, an experimental method that first distinguishes between molecules in these two classes, and continues by rapidly measuring the equilibrium and kinetic solubility of compounds that do not form supersaturated solutions in water; these assays take 15 – 20 minutes per sample. If the software detects that the sample does become supersaturated, it controls the assay in a different way, and obtains the result by chasing equilibrium; these assays take 30-45 minutes per sample. When titrating solutions of samples in their ionized form towards a pH where they are unionized, precipitate of unionized species (detected by light scattering) will suddenly appear at a certain pH, if the sample is poorly soluble. The concentration of unionized sample at this pH is equivalent to the kinetic solubility. Samples that do not form supersaturated solutions will precipitate rapidly and quantitatively in response to further additions of titrant. If the pH is recorded throughout the titration, the solubility may be derived from the shape of the Bjerrum curve after precipitation, following wellunderstood principles of mass balance and charge balance. The kinetic solubility is found to be equal to the intrinsic solubility for this class of molecule. On the other hand, samples that supersaturate undergo a process of chasing equilibrium, and the solubility is derived in a different way. The poster will clearly explain each type of assay. Solubility measurements for nine samples that do and nine that do not form supersaturated solutions will be presented, showing excellent agreement with values obtained by the shake-flask method.

Inert Sample Processing - Solutions for Combating Sample Precipitation in Compound Libraries

Authors: Mel Reichman, PhD (PharmDC), and Sandesh Gowda (KEMS)

Management of large compound libraries for successful drug screening requires focus on minute details and operational discipline. Infuriatingly, a poor confirmation rate for actives observed in the primary screen remains the norm rather than the exception in HTS, which some ascribe in part to perturbation of compound libraries consequent to their exposure to ambient humidity and freeze-thawing. KEMS and PharmDC have developed a family of inert enclosures compliant with stacker-equipped, automated pipetting stations. They allow sample processing under an ideal, controlled environment with limited human intervention (i.e. "hubotic"). Our automated Mini-Store (AMS) integrated with our Plate Processing Workcell (PPW) is an ideal robotic solution for unattended compound library formatting with the highest QC/QA in a cost effective, state-of-the-art system.

Expanding the Suite of Assays Technologies That Can Be Reliably Run in 1536 Well Format

Rose Hughes[1]; Robert Bukar[1]; Ben Baumann[1]; Nathan Heinrich[2]; Paul Queeney[1]

¹ Kalypsys, Inc., 10420 Wateridge Circle, San Diego, CA 92123

² Remmele Engineering, 677 Transfer Road, St. Paul, MN 55114

Kalypsys Systems has produced a 1536 well washer/dispenser workstation with unparalleled flexibility and reliability. Kalypsys' washer/dispenser has all the standard features of a 1536 well dispenser and more. The flexibility of the system is exemplified by the fact that it can dispense any volume of up to eight different reagents, including cells, into any well on a plate, in volumes as low as 200nl. The washer/dispenser is unique in that it brings ELISAs into the world of 1536 through the utility of its best in class 1536 well plate washing abilities. In addition, the system's ability in dispensing SPA beads is unmatched. YSi and YOx SPA beads often stick to a dispenser's tubing during the pause in between plate dispenses and a prime is typically required between every plate to ensure even delivery of the beads. Kalypsys' engineers have invented a unique SPA dispense option that keeps the beads re-circulating at all times, significantly reducing sticking. The ability to recover any primed SPA beads has eliminated waste as well as reduced the screening costs and the preparation of excess SPA beads. Any type of SPA bead can be dispensed efficiently and cost effectively with this system. In this poster we will present data from a number of biochemical and cell based assays, including ELISAs and SPA to demonstrate the rapid generation of robust and reliable data from this unique and flexible instrument.

Vari Syringe

The Vari-Syringe will dispense the volumes from an Excel spreadsheet column into the vials in a rack. It uses the Hamilton Microlab 500 series dual syringe pump

The different volumes feature is needed for example when adding D6DMSO to recently purified compounds, or adding Acetonitrile to weighed tissue samples before grinding. The software is designed to be easy to use with different shaped racks. There is even a wireless option to let you use it inside a fume hood or glove bag.

<http://www.tradersmicro.com/Projects/Vari-Syringe/VMainPage.html>

Micro-High Speed Liquid Chromatography Coupled to an Evaporative Light Scattering Detector for the Rapid Analysis of Compounds of Pharmaceutical Interest

Rachel KOHN, Davy GUILLARME, Serge RUDAZ, Cedric SCHELLING, Jean Luc VEUTHEY
University of Geneva (Switzerland)
Michel DREUX
SEDERE Inc

The recent developments in liquid chromatography (LC) are mainly related to both system miniaturization (micro liquid chromatography, μ -LC) and analysis time decrease. For the latter, several solutions could be implemented, such as the use of monolithic stationary phases; high temperature (High Temperature Liquid Chromatography, HTLC) or low particle size (below 2 μ m) at pressures reaching 1000 bar (Ultra Performance Liquid Chromatography, UPLC). The main goal of these techniques is the use of high linear velocity compared to conventional LC without loss of efficiency. For the analysis of compounds with and without chromophores, the evaporative light scattering detector (ELSD) is an attractive alternative to conventional detection such as UV due to its low cost, good sensitivity and quasi-universality. The coupling of ELSD with μ -fast-LC was investigated for several pharmaceutical compounds of interest. An experimental design was used to show the influence of LC parameters (flow rate, temperature or mobile phase composition) and ELSD parameters (nebulization pressure and evaporation temperature) on band broadening and sensitivity. Nebulization step appears to be the most critical parameter for dispersion associated with ELSD in these conditions. Appropriate modifications in nebulization chamber geometry were brought to make ELSD fully compatible with μ -fast LC without compromising sensitivity.

A novel generic system for kinase assays and real-time enzyme and inhibitor kinetics

Jun Wu, Surekha Vajjhala, and Steve O'Connor
Nanostream, Inc.

Protein kinases control many cellular activities through protein phosphorylation and dephosphorylation in response to extra cellular signals. Abnormality in the activities of the kinases in these regulation pathways have been linked to many human diseases. Classical screening platforms, e.g., radioactive methods and ELISA-based assay methods, generally require multiple labor-intensive and time-consuming steps. To address these limitations, we have developed a generic high-throughput screening assay platform. In this assay, nonphosphorylated peptide was used as a substrate, and phosphorylated peptide was resolved from nonphosphorylated peptide via micro parallel chromatography. In contrast to a traditional plate reader, the Nanostream LD system detected less than 1% conversion of enzymatic reactions and yielded Z' factors ranging from 0.6 to 0.85. In addition, the system provided real time monitoring of kinase and inhibitor kinetics. From a single experiment, the optimal enzyme, ATP, and substrate concentrations were determined. Furthermore, the compiled data allowed us to determine K_m for ATP and for substrate and also to perform time course studies. Compared with conventional HPLC and LC-MS, our system enables parallel chromatographic analysis of 24 samples. The system significantly reduced the time required to develop the assay and to implement the assay in a screening mode. The integrated system software permits fast and accurate data analysis. The system can be used to facilitate target identification and assay development in drug discovery.

High-throughput automation for RNA isolation from blood stabilized in PAXgene™ Blood RNA Tubes

Thorsten Voss¹⁾, Ralf Wyrich¹⁾, Daniel Langendörfer¹⁾, Thomas Rothmann²⁾, Uwe
Oelmüller¹⁾

¹⁾ PreAnalytiX, ²⁾ QIAGEN

Artificial modifications of the RNA content and profile blood samples post-phlebotomy caused by degradation and gene induction is well documented. Doubtful or inconsistent results are the consequence, especially for quantitative or semi-quantitative analytical methods such as qRT-PCR assays and microarrays. There is a need, therefore, for stabilization of cellular RNA species to freeze the gene expression profile at the time of blood collection. The manual version of the PAXgene Blood RNA System is commonly used to address this problem. To allow a higher sample throughput combined with a controlled and completely documented process (e.g. for clinical trials), we have designed protocols and kits for two state-of-the-art robotic platforms. This study was conducted to show the performance of these robotic solutions. The prepared RNA was tested in several qRT-PCR assays and on Affymetrix GeneChips.

Blood was collected in PAXgene Blood RNA Tubes. For automated cellular RNA isolation, the PAXgene Blood RNA MDx Kit was used on both the BioRobot® MDx and the BioRobot® Universal System (QIAGEN). The quantity and quality of the RNA samples were analysed by spectrophotometric analysis and on an Agilent 2100 Bioanalyser. Downstream gene expression analyses were performed by real-time qRT-PCR assays and Affymetrix GeneChip® arrays. For the array analysis, a new PNA-based GeneChip Globin Reduction Protocol combined with the GeneChip Blood RNA Concentration Kit (Affymetrix) was performed prior to labeling.

The mean RNA yield from more than 2100 blood samples was 6.9 µg per tube, with a standard deviation of 0.96 and a CV of 14.4% between tubes of a single donor. The yields showed a good correlation to the white blood cell count of the samples. A_{260}/A_{280} for purified RNA was between 1.8 and 2.2. The average residual gDNA content of the same RNA was very low with 0.02% (w/w). No RT-PCR inhibition and no cross contamination of microtiter wells could be detected with qRT-PCR. With two validated qRT-PCR assays, the variability of the Ct values were within the assay specific failure rates. The performance on the GeneChips was comparable to RNA isolated by the PAXgene Blood RNA manual procedure.

In conclusion, the PAXgene automated system provides fully automated RNA isolation from PAXgene Blood RNA Tubes and is easy to implement for gene expression analysis in situations where high throughput RNA extraction is required.

The BioRobot® Universal System – Multiple Applications on One Instrument

Thomas Weierstall; Anja Schultz; Martin Heller and Carola Schade

QIAGEN GmbH, 40724 Hilden, Germany

During recent years many researchers in the field of molecular biology have turned from a discovery type of research, studying individual elements such as DNA sequences, RNA expression levels, or individual proteins, to a more integrated approach in order to understand the functioning of whole systems and their reaction to any perturbation. Using a broad range of high-throughput technologies, these studies often involve a global network of researchers and laboratories contributing to the project. Therefore, standardization of procedures is a major focus in order to ensure comparability of data being generated.

We developed an automation solution that allows users to perform a wide range of standardized high-throughput applications using pre-tested, optimized protocols on a single platform, the BioRobot Universal System.

These protocols include purification of DNA and RNA from a variety of sample types such as cells, tissue, blood, and buccal swabs, as well as RT-PCR and PCR setup in 96-well format.

Total RNA can be purified from up to 192 cell culture samples per run. The efficiency of the purification procedure is demonstrated by linear CT values over a wide range of starting template amounts. Downstream analysis of replicate samples using quantitative RT-PCR show a %CV of CT values <3%, demonstrating the high reproducibility of the purification and reaction setup procedure. The system also allows high-throughput total RNA purification from tissue samples, including difficult-to-lyse fatty and fibrous tissues, and PAXgene blood RNA tubes.

Users can easily switch between applications since no hardware configuration changes are required making the BioRobot Universal System the ideal solution for laboratories performing high-throughput systems biology applications.

Securely Publishing Screening Data to Multiple User Groups with a Lightweight Apache Web-server Application

Ken S. Stoney and Frank D. Mentch, Ph.D.
Edge4 Technologies, www.edge4tech.com

Contract research organizations, government labs and internal screening facilities often need to provide protected access to proprietary data for multiple customer groups. This can be a bottle-neck, especially when dealing with diverse data formats and more than a few customers. The method described here combines the security and accessibility features of the Apache web server with an easily implemented and maintained data federation tool that allows data to reside in any combination of relational databases, files, and spreadsheets.

Complex commercial LIMS databases, custom databases and simple one-off flat-files and spreadsheets are equally accommodated. The application can also function "in reverse", taking a hodge-podge of files, tables, data marts and data warehouses and combining them into one or more focused "channels" based on user group interests and requirements.

Centralized vs. Multi-Site Compound Management REMP

The cost of drug development has soared over the last decades, as well as, the efforts into sales and marketing of the end-product drugs. Companies realize that the larger they are the easier it is to bare large investments and leverage their marketing activities. In order to maximize return-on-investment, the companies are engaging in continuous merging activities.

Life Science Research companies, nowadays, consist of multiple R&D sites distributed all around the world. Individual sites often work on dedicated targets or on specific therapeutic areas with synergies generated through access to each others research results. In order to maximize global research effectiveness, all research scientists need an overview and physical access to the entire compound collection of independently built up libraries.

Identifying hits and elucidating targets from a Neutrophil Oxidative Burst uHTS in 1536-well microplates

Rose Hughes; Robert Bukar; Jeff Davis; Jessica Cramp; Elizabeth Gardiner

Kalypsys, Inc., 10420 Wateridge Circle, San Diego, CA 92123

The open-ended use of the proprietary uHTS System in combination with the dedicated efforts of the scientists at Kalypsys, Inc. has enabled the Neutrophil Oxidative Burst Program to continue to push the boundaries of compatibility of assays in 1536-well format. The search for a small molecule that is a selective oxidative burst inhibitor for Chronic Obstructive Pulmonary Disease as well as other chronic inflammatory disease modification has shown that 1536-well screening is not limiting but actually holds the key to several assay capabilities that were not thought to be made easily transferable to the smaller format. The small volumes used and minimal effort by personnel has been made possible by the enabling technologies of this cost effective platform. Assessment of human neutrophil oxidative burst has traditionally been done in cuvettes or 96 well formats. Kalypsys has brought this assay into two flexible 1536 well formats utilizing an inexpensive luminescent or fluorescent 1536-well microplate. Target elucidation from a dozen other assays also miniaturized into the 1536-well format were run using human neutrophils and allowed for kinetic data collection from these high throughput screens to be run in a matter of days. The ability to easily screen primary human cells in 1536 well format and quickly elucidate hits against specific targets with minimal input from scientists and screeners is an enormous feat made possible by use of the Kalypsys Screening System.

Non-Contact Dispensing of Yttrium Oxide Beads in 1536

Justin Murray

Merck Research Labs, NW-3

Department of Automated Biotechnology

140 Wissahickon Avenue

North Wales, PA 19454

Currently there are two types of imaging scintillation proximity assay (SPA) beads, yttrium oxide (YOx) and polystyrene (PS). PS beads are easier to dispense, but they do not have the enhanced sensitivity that YOx beads possess, which depending on the assay can increase the signal to background (S/B) ratio. Both bead types are similar in physical size, but YOx beads are four times more dense and therefore heavier than PS beads which makes it difficult to keep the beads in suspension and difficult to dispense without variance in well-to-well bead concentration. Using the intrinsic optical properties of the beads we have developed a dispense quality control (QC) method that is sensitive to both bead concentration and well volume. Using this method, we eliminate the need for radioactive materials for dispense QC purposes. We have also developed a technique to keep the beads in suspension for the extended periods of time necessary for high throughput screening. Using these techniques, we have optimized two dispensers for YOx SPA bead non-contact dispense into 1536 well format - the Kalypsys SPA Bead Dispenser (KSPA) and the CartesianX (CarX). The data shown is a comparison of the two dispensers including both dispense QC and enzymatic binding data.