



Increase your assay productivity through innovative technologies, a series of case studies – presented by experts from GSK, Pfizer, Cephalon and Lundbeck, with opening remarks from Dr. Richard Eglén.

Sunday, April 6th
9:30 a.m. – 12:30 p.m.
Room 242

Learn how an innovative no wash ELISA alternative enables high throughput compound profiling and small molecule screening.

Understand how the combination of an innovative cell-based GPCR platform and cellular workstation provide robust, sensitive, cost effective HTS.

Welcome and Introduction.

9:30–9:55 [Accelerating drug discovery from Target Validation to Lead Optimization; the PerkinElmer Complete Solution](#)
Dr. Richard Eglén, President, PerkinElmer BioDiscovery, Waltham MA USA.

PART ONE: Conversion of traditional ELISA assays to the sensitive no-wash AlphaLISA™ technology.

10:00–10:30 [The Application of AlphaLISA for Compound Profiling and 1536-well format HTS.](#)
Stephen Rees, Director, Screening and Compound Profiling, GlaxoSmithKline, Stevenage UK.

ABSTRACT

For many years Enzyme Linked Immunosorbent Assays (ELISAs) have been employed for the measurement of peptide or protein production from mammalian cells in response to drug treatment. However the use of these assays in High Throughput Screening (HTS) or Compound Profiling has been limited due to the technical complexity of the assay protocol. In this talk I will describe the application of AlphaLISA technology to enable a high throughput 384-well compound profiling assay and a homogeneous 1536-well plate format HTS assay. The use of this assay to screen the GSK compound collection will be described. AlphaLISA enables the performance of traditional ELISA assays with the cost and simplicity of advanced cell based assay technologies.

10:35 – 11:05 [Development of a Cell-Based AlphaLISA Assay for the Evaluation of Native Kinases](#)
Robert Compton, Associate Research Fellow, Assay Development Group Leader, Primary Pharmacology, Pfizer Global Research and Development

ABSTRACT

Kinases are potentially one of the largest classes of drug targets. Analysis of the human genome suggests that it encodes 518 kinases. Much of the work to develop small molecule inhibitors of kinases has focused on ATP competitive inhibitors due primarily to limitations in approaches and available assays to evaluate inhibitor interactions with kinase targets. However, the high level of homology displayed in the ATP binding pocket across the kinome has resulted in selectivity issues as targets are prosecuted. Additional structural and functional complexity of kinases make drug discovery even more challenging.



We are currently pursuing a multi faceted approach to the therapeutic intervention of cytokine signaling in inflammatory disease using small molecule inhibitors. To this end, primary screening assays have been established to evaluate compound potency using recombinant kinase domain and when possible full length protein. It is of interest to establish high throughput cell based assays to explore lead compound potency and selectivity toward individual native kinases. AlphaLISA is an assay format that we have chosen to assess compound potency and selectivity versus selected kinase and related isoforms. Preliminary data suggests that AlphaLISA is a sensitive method that allows high throughput cell-based evaluation of potential small molecule inhibitors of kinase targets.

11:05 – 11:15 **COFFEE BREAK**

PART TWO: Advances in high-throughput GPCR cellular screening assays

11:15–11:45 [Screening With the Functional Cell Based Aequorin Platform for the Discovery of Novel GPCR Ligands](#)

Bruce E. Jones, Ph.D. Senior Scientist, Team Leader Aequorin Platform, Cephalon, Inc, West Chester, PA USA

In recent years, luminescence aequorin-based functional cellular assays for HTS have gained in interest. Much of this interest stems from the introduction of plate imagers compatible with the aequorin platform as well as an increase in the availability of cell lines expressing aequorin. This presentation provides a collective review of the aequorin platform. Discussed will be the basic principles of the aequorin system, cell line development and the process of assay development and validation in HTS environment. Additionally, assay-ready plate preparation, prototypical screening results and follow-on confirmation of active wells will be presented. The talk will conclude with an introduction into recent work using transient transfections with the aequorin platform.

11:50–12:20 [Development of an Aequorin Luminescence calcium assay for high-throughput screening using a the new plate reader platform, LumiLux®.](#)

Suresh B. Poda, PhD, Scientist-II, High Throughput Screening, Biological Research, Lundbeck Research USA, Inc., Paramus, NJ USA.

ABSTRACT

A luminescence assay using a new plate reader, the LumiLux has been validated for High Throughput Screening (HTS). In this study, we compared the aequorin luminescence based calcium mobilization assay to the fluorescence-based calcium assay. A cell line stably co-expressing apo-aequorin, a chimeric G-protein and a G-Protein Coupled Receptor (GPCR) was used to screen a collection of compounds using the Hamamatsu Photonics FDSS6000 and Perkin Elmer Lumilux as the plate readers. The assay parameters evaluated included: hit rate correlation, signal-to-noise ratio, false positives rates and overall assay performance calculated by Z' and standard deviation. The average Z' values and hit rates were comparable between assay platforms however the standard deviation for the agonist aequorin assay was significantly smaller. There was also a significant decrease in the number of false positives with the aequorin assay. These results suggest that the new plate reader Lumilux in combination with aequorin cells provides simple, cost effective, robust and sensitive assay for HTS.

12:20-12:30 **Q & A**