



INDIGO Biosciences, Inc.

The Nuclear Receptor Company™

New Robust, Ready-to-Use Cell-Based Nuclear Receptor Reporter Assay Products from INDIGO Biosciences for use in High-Throughput Screening (HTS), Off-Target Screening, and Toxicology.
PPAR γ , LXR β , ER α and MR
Sensitive, Cost-effective Nuclear Receptor Reporter Assay Systems for Biotech & Pharmaceutical Discovery

INDIGO Biosciences, Inc., The Nuclear Receptor Company™, announces the launch of a unique portfolio of cell-based, Nuclear Receptor Reporter Assay products for on-demand use in drug discovery and development. INDIGO Biosciences' assays enable sensitive screening of test compounds for agonist or antagonist activities against human nuclear receptors expressed within the cytoplasm and nuclear environments of healthy, dividing mammalian cells.



Figure 1. Human Peroxisome Proliferator-Activated Receptor Gamma (PPAR γ) Reporter Assay System

Robust Assays Ready When You Are

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INDIGO Biosciences delivers all-inclusive, sensitive and cost-effective Cell-Based Nuclear Receptor Assay Systems. They are the perfect research solution for high-throughput on-target drug discovery campaigns, as well as low and medium throughput screens to assess their potentially toxic or adverse off-target interactions with nuclear receptors.

INDIGO's Assay Systems provide value to the Pharma and Biotech industries by allowing them to focus on their primary goals: drug discovery and development. INDIGO's products alleviate the significant money, time and human resource expenditures otherwise required for advanced in-house assay development, optimization, scale-up and QA. All of INDIGO's assay products are shipped frozen for over-night delivery, and may be further stored at customers' facilities for their on-demand use. Our assays are ready when you are!

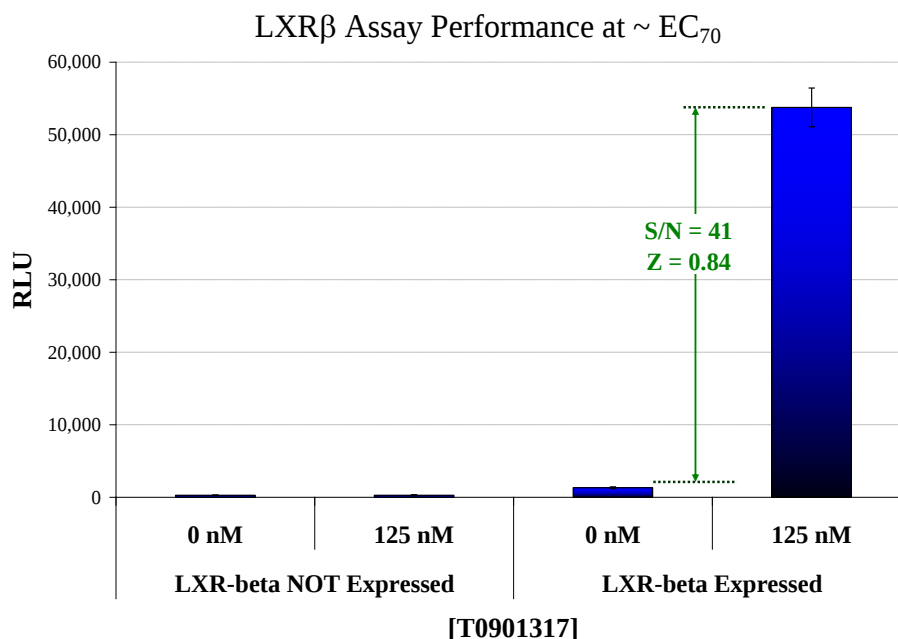


Figure 2. Validation of the LXR β reporter assay for screening applications.

LXR β reporter cells were treated with media alone, or media containing 125 nM ($\sim$ EC₇₀) T0901317, a potent agonist of LXR β . To assess the amount of background signal contributed from endogenously expressed LXR's, and/or from any other factor(s) causing gratuitous activation of the luciferase reporter

gene, “mock” reporter cells were specially prepared to contain only the luciferase vector (ie., withOUT the LXR β expression vector). Mock reporter cells and the LXR β reporter cells were cultured, treated with T0901317, and processed in identical manner. Luminescence intensities were quantified using a plate-reading luminometer. Signal-to-noise and Z' values were calculated for the LXR β reporter cells treated with $\sim$ EC₇₀ concentration of control agonist.

RESULTS & CONCLUSIONS: LXR β reporter cells treated with 0 and 125 nM T0901317 yielded a Z' value of 0.84. Agonist treated mock reporter cells demonstrated insignificant levels of background luciferase activity (< 0.5% of the EC₇₀ value). Therefore, ligand-induced luminescence results strictly through activation of the exogenous human LXR β protein expressed in these reporter cells. These data confirm the robust performance of the LXR β Reporter Assay System, and demonstrate its suitability for use in high-throughput screening applications.

INDIGO Biosciences' optimized Nuclear Receptor Reporter Assay Systems yield excellent Z' scores and high signal-to-noise ratios. In addition, the reporter cells require no cumbersome post-thaw handling, and yield greater than 95% viability.

THAW. FEED. DOSE. READ.™

INDIGO Nuclear Receptor Assay Systems are easy to use. Just THAW. FEED. DOSE. READ. The Protocol Quick Guide, included in each kit, outlines the simple 14 Step protocol that takes the kit from Freezer to Data Analysis. All in about 24 hours. Hands-on time is very minimal throughout the process.

Without the need to create assays in-house, a user can be ready to launch an HTS campaign or a small screening batch in about 24 hours. Each kit also contains a positive-control agonist to verify assay performance, and to benchmark the results or test compounds.

Alternative protocols outlined in the Technical Manual allow users to perform agonist dose-curves, antagonist dose-curves, or high-throughput single-point screening of test compounds for agonist activities or antagonist activities.

INDIGO's Nuclear Receptor Reporter Assay Systems utilize stable-glow luciferase chemistry, thus allowing plates to be processed in batch, and dramatically reducing the start-to-finish read-time of assay plates.

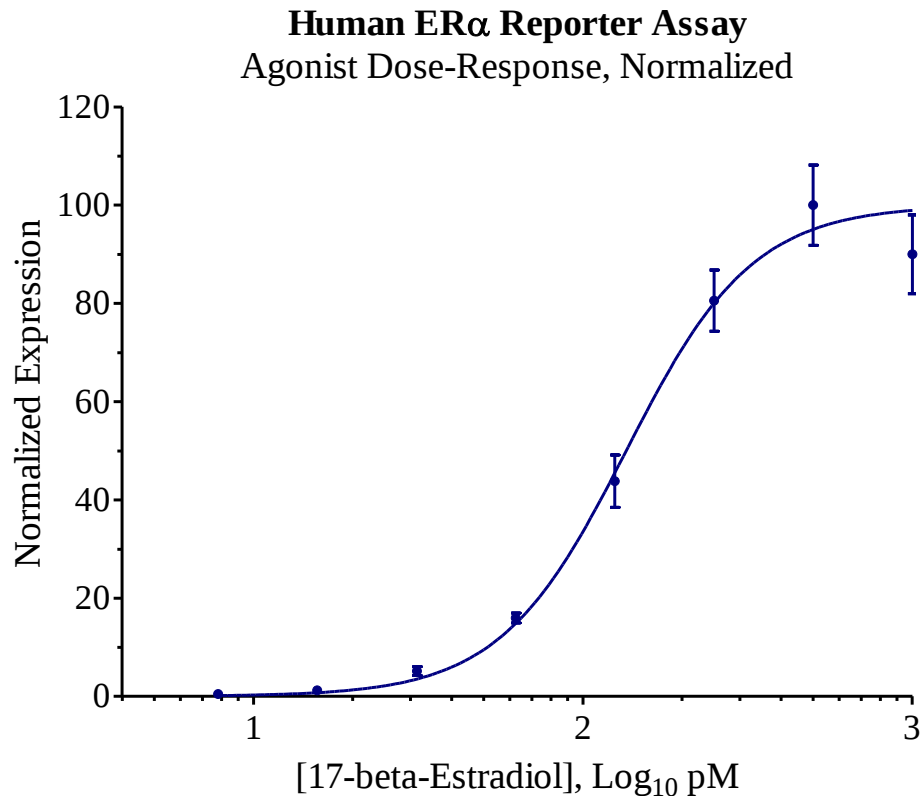


Figure 3. Agonist dose-response of the ER α Reporter Assay System.

The ER α dose-response assay was performed according to the protocol provided in the Technical Manual. Plated reporter cells were treated for 20 hours in screening media supplemented with diminishing pM concentrations of 17- β -estradiol, the positive-control agonist for ER α . Treatment media were removed and 100 μ l of luciferase detection reagent was applied directly to the cells, and luminescence intensities were quantified. 17- β -estradiol concentrations were transformed to log₁₀ pM, luminescence values were normalized to 100%, and the agonist dose-response was plotted.

INDIGO's Proprietary Cryomite™ Process Allows for JIT Delivery

INDIGO Biosciences developed a proprietary cryopreservation process called **CryoMite™** that enables long-term preservation of the company's unique reporter cells. Now The Company's live-cell assays can ship to customers via overnight delivery, providing users the convenience of readily available, robust cell-based Nuclear Receptor Reporter Assays.

INDIGO Biosciences' Nuclear Receptor Assay System kits are all-inclusive and easy-to-use, saving customers time and money. The Company's current portfolio of reporter assay product families includes: the Human Peroxisome Proliferator-Activated Receptor-Gamma (PPARG), Liver X Receptor-Beta (LXR β), Estrogen Receptor alpha (ER α), and Mineralocorticoid Receptor (MR). New offerings of human Nuclear Receptor Assay Systems will be added to the INDIGO Biosciences product portfolio on an ongoing basis. Please visit INDIGO's website www.indigobiosciences.com for the latest offerings of our Nuclear Receptor assay products.

INDIGO Biosciences' nuclear receptor reporter assays are optimally configured for processing in 3x 32-, 96- and 384-well plate formats, which are offered as all-inclusive assay systems. Each kit includes nuclear receptor reporter cells, an assay plate, optimized culture media, positive-control agonist, and detection reagent, as well as a detailed Technical Manual and Protocol Quick Guide.

Custom manufactured Bulk Reagent Packs are also offered to accommodate the needs of high-throughput screening (HTS) applications. INDIGO can also offer custom screening panels, so multiple Nuclear Receptors can be studied on a single screening.

INDIGO Biosciences currently offers Nuclear Receptor Reporter Assay Systems for these nuclear receptor targets:

PPAR γ , LXR β , ER α and MR

All Products Available in Multiple Formats for Flexible Use:

Product Format	Description	Format	Kit Type	Configuration	Plate Included
01-32	Kit designed to perform 3 distinct groups of 32 assays, utilizing four 8-well strips affixed to a 96-well frame (included)	3 x 32 Assays in 96-Well	Flexible Use	Low Throughput	96-Well Frame and 8-Well Strips
01	Kit format is intended for users wishing to dedicate one entire 96-well plate to performing a nuclear receptor assay.	1 x 96-Well	Single Use	Medium Throughput	96-Well
01B	Kit format is intended to accommodate HTS applications in which a minimum of ten (10) 96-well plates will be dedicated to a single screening run.	10 x 96-Well	Bulk	HTS	None
02	Kit format is intended for users wishing to dedicate one entire 384-well plate to performing a nuclear receptor assay.	1 x 384-Well	Single Use	Medium Throughput	384-Well
02B	Kit format is intended to accommodate HTS applications in which a minimum of ten (10) 384-well plates will be dedicated to a single screening run.	10 x 384-Well	Bulk	HTS	None

For more information about INDIGO Biosciences, Inc., the company's new single-use nuclear receptor reporter assay products or its proprietary CryoMite™ technology, please contact Mark Capriani, Vice President of Sales & Marketing at INDIGO Biosciences, Inc. by calling (814) 234-1919 or by e-mailing to mark@indigobiosciences.com. You may also visit INDIGO Biosciences, Inc. on the worldwide web at www.indigobiosciences.com.

INDIGO BIOSCIENCES
The Nuclear Receptor Company

Founded in 2005, privately-held INDIGO Biosciences, Inc. provides unique, cost-effective, reliable and user-friendly target-specific nuclear receptor assay products and contract screening services to drug developers, toxicologists and researchers within the pharmaceutical, biotechnology, chemical and nutraceutical industries.

INDIGO Biosciences developed a unique and superior processing method called CryoMite™ which provides long-term cryo-preservation of cells for shipping and storage, enabling The Company to provide its unique Nuclear Receptor Assays for third party use via overnight delivery.



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