

## New Leading Light™ Wnt Reporter Assay with the Highest Sensitivity

Leading Light™ Wnt Reporter Assay (ENZ-61001) **New**  
 Screen-Well® Wnt Pathway Compound Library (BML-2838)

Recombinant Wnt-3a (mouse) (ENZ-60001)  
 Recombinant DKK-1 (mouse) (ENZ-60002) **New**

You asked. We delivered. Enzo's new Leading Light™ Wnt Reporter Assay was developed to meet the market need for a **high sensitivity system** that accelerates screening analysis of Wnt pathway modulators. This product is a luciferase-based assay for the analysis of canonical Wnt signaling.

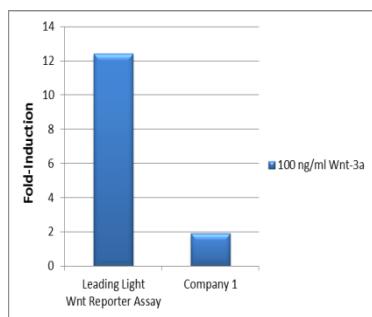
- **Highest Sensitivity in response to Wnt effectors without the use of lithium chloride**
- **Highly active Wnt-3a protein included to ensure robust signal**
- **96- or 384-well format allows high throughput analysis of Wnt pathway effectors**
- **Validated kit provides cells, media, reagents and controls ensuring reproducible results**

### Enzo Life Sciences: A Leader in Providing Enhanced Sensitivity Detection Platforms to Scientists

The addition of our new **Leading Light™ Wnt Reporter Assay** continues this mission of providing high-value tools to the research community to enable discovery for a variety of clinically relevant disease states, including cancer, osteoporosis, cardiovascular disease, diabetes, and neurodegeneration. Discover more with this pioneering, sensitive assay for screening Wnt modulators, as well as a supporting portfolio of ELISA kits and reagents to help you monitor Wnt signaling from cell surface to nucleus.

### Greater Sensitivity with a Superior Signal-to-noise Ratio

The Leading Light™ Reporter Assay yields a greater than 12-fold increase in signal over background, surpassing the signal intensity of other commercially available reporter assays. A robust signal-to-noise ratio allows for enhanced detection of Wnt pathway modulators.

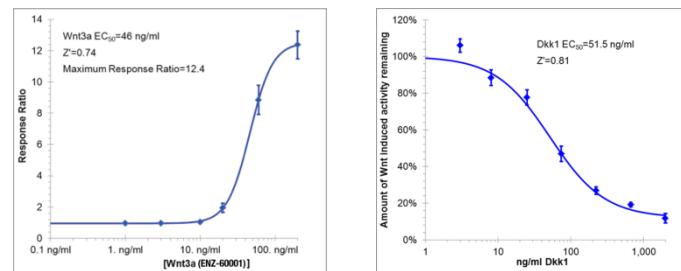


**Figure 1: Signal intensity of Leading Light™ versus competitor.**

When treated with 100 ng/mL of Wnt-3a (ENZ-60001), the Leading Light Reporter Assay yielded greater than 12-fold increase in signal. The fold-induction for Company 1 was determined using published product information for their Wnt-3a protein and reporter assay.

### Dynamic range of signal with No Artificial Signal Enhancement

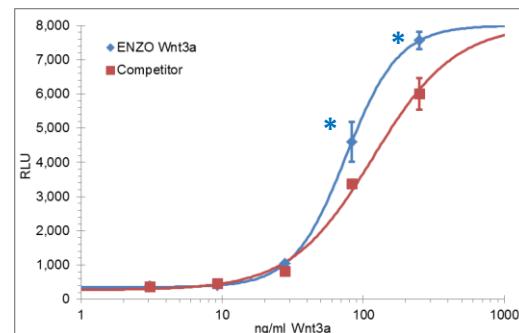
The Leading Light™ Wnt Reporter Assay includes an engineered 3T3 mouse fibroblast cell line with a luciferase reporter gene under the control of a Wnt responsive promoter. This optimized system requires no lithium chloride to boost signal, ensuring confidence in Wnt pathway screening results.



**Figure 2: Dose response of Leading Light™ Wnt Reporter Cells to Wnt-3a and Dkk-1 proteins.** (A) Leading Light™ Wnt Reporter Cells were treated with indicated doses of Wnt-3a protein (ENZ-60001) for six hours. The chemiluminescence in the Wnt-3a-treated cells increased in a dose-dependent manner. (B) Cells were treated with the indicated doses of Dkk-1 in the presence of 200 ng/mL Wnt-3a. Dkk-1 inhibits Wnt-3a-elevated luciferase levels in a dose dependent manner.

### Enzo Highly Active Proteins Outperform the Competition

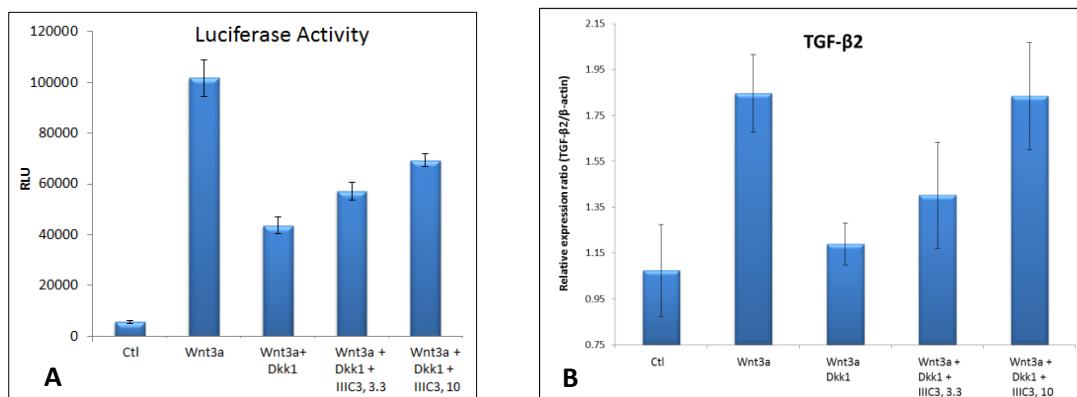
The Leading Light™ Wnt Reporter Assay includes cells, media, and detection reagents required for testing. Additionally, active Wnt-3a and Dkk-1 are provided as controls. In a head-to-head comparison, Enzo recombinant Wnt-3a protein had greater activity versus an identical recombinant protein from a competitor.



**Figure 3: Comparison of Wnt-3a protein activity.** Leading Light™ Wnt Reporter Cells were treated with indicated doses of Wnt-3a protein (ENZ-60001 or competitor product). The chemiluminescence in the Wnt-3a-treated cells increased in a dose-dependent manner.\* denotes significant different as measured by a student t-test.

## Reproducible Results from a Validated System

The Leading Light™ Wnt Reporter Assay was validated by detecting Wnt activation of an endogenous Wnt target gene, TGF- $\beta$ 2. Pathway activation or inhibition was modulated using a small molecule control, IIIC3, which inhibits Dkk-1 binding, and Wnt ligands (Wnt-3a and Dkk-1).



**Figure 4. Validated system for screening small molecules.** Leading Light Reporter Cells were seeded and incubated at 37°C overnight. After 24h, cells were treated with Wnt-3a, Wnt-3a + DKK-1, and Wnt-3a + DKK-1 + (3.3 or 10  $\mu$ M) IIIC3 (DKK-1 inhibitor) for 6 hours at 37°C. Untreated cells were used as a background control. After 6h treatment, the chemiluminescence in each conditioned sample was measured using the detection reagents (A). In an identical experiment following treatment, cells in each well were harvested and total RNA was isolated (B). Real-time RT-PCR was performed to detect mRNA level of TGF- $\beta$ 2 (Wnt target gene) in each sample. TGF- $\beta$ 2 mRNA levels in each sample were normalized against  $\beta$ -actin levels of mRNA.

## Let Enzo Provide Your Wnt Research Solution

Enzo has provided reliable, consistent, and innovative products to researchers for almost 40 years. We are dedicated to expanding the field of Wnt research by offering novel products, thoughtful application data and peer-reviewed publications. The Leading Light™ Wnt Reporter Assay along with the Screen-Well® Wnt Pathway Library and numerous ELISAs, antibodies and proteins combine to provide a total solution for Wnt pathway-related researchers.

With our broad array of research tools, Enzo provides high-purity, high-sensitivity reagents applicable to the following Wnt Pathway areas of interest:

- Assessing functions and activities of various Wnt-related ligands
- Compound screening of small molecules and antibodies for identification of Wnt inhibitors or agonist
- Drug screen of small molecules that modulate the Wnt signaling pathway
- Analysis of RNAi or gene expression phenotypes

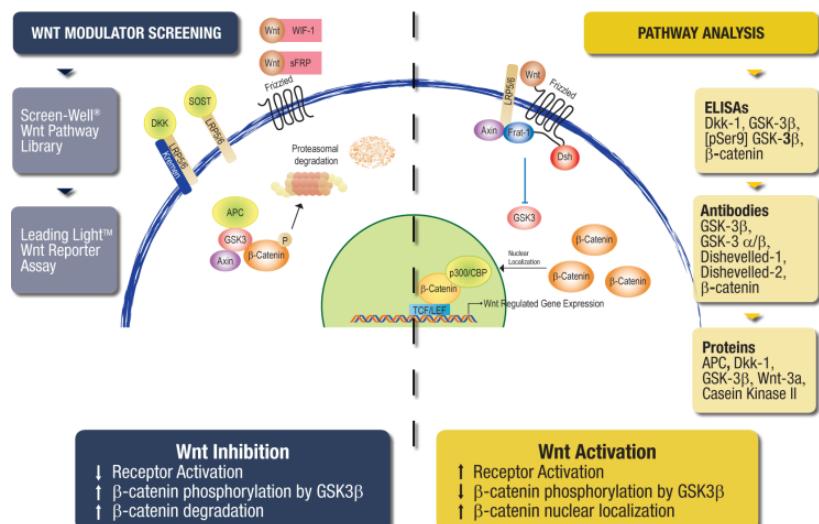
### Application Note:

*Cell-Based Screening of Focused Bioactive Compound Libraries: Assessing Small Molecule Modulators of the Canonical Wnt Signaling and Autophagy-Lysosome Pathways*

### Article:

*Li, X et al. Chemical and genetic evidence for the involvement of Wnt antagonist Dickkopf2 in regulation of glucose metabolism. Proc Natl Acad Sci U S A. 2012 Jul 10;109(28):11402-7.*

**Enzo is continually adding new products to our Wnt portfolio. Check back with us soon to learn about our next generation binding assays and compounds to support Wnt pathway analysis!**



Life Sciences **scientists enabling scientists.**

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