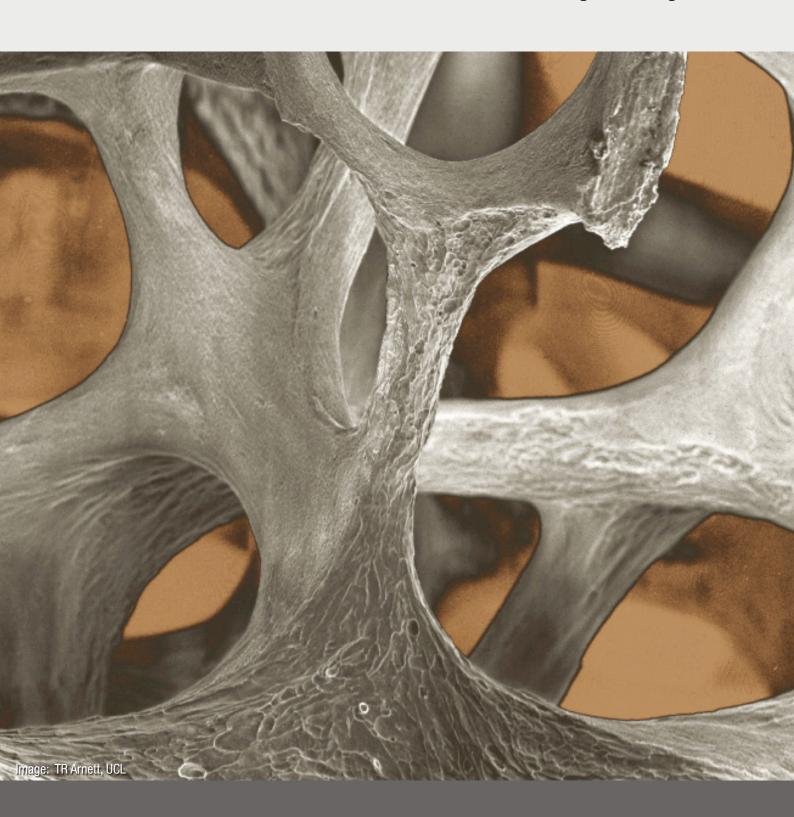


Wnt Pathway
Integrated Solutions for
Screening Wnt Regulators

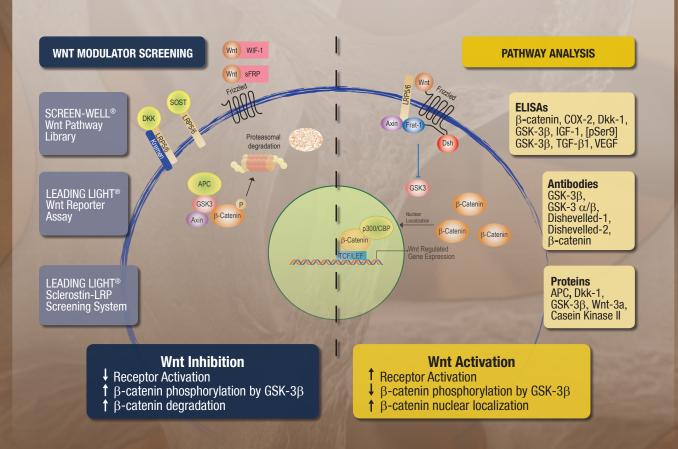


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# **SOLUTIONS FOR WNT PATHWAY SCREENING**

### High-throughput Assays for Canonical Wnt Pathway Analysis

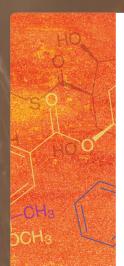
Since the discovery of the Wnt gene over 30 years ago as the *Int1* proto-oncogene involved in the pathology of Mouse Mammary Tumor Virus (MMTV), Wnt expression and signal transduction have been identified as critical components in regulating embryogenesis, cell proliferation, and bone, glucose, and lipid metabolism. Perturbations in Wnt expression, surface receptors, signaling partners (including  $\beta$ -catenin and GSK-3 $\beta$ ), and the battery of Wnt-regulated genes (collectively the canonical Wnt pathway) are known to manifest in a variety of clinically relevant disease states, including cancer, osteoporosis, cardiovascular disease, diabetes, and neurodegeneration.



# **Cell-Based Assays & Biomarker Detection**

Enzo's panel of fluorescence-based live cell assays are designed to assess the impact of toxic agents on overall cell function, with particular emphasis on the plasma membrane, lysosomal, mitochondrial and nuclear compartments. Many of the assays have been optimized for analysis by flow cytometry, while others are geared towards microplate-based cytometry.

Our portfolio of over 300 ELISA & detection kits includes sensitive, specific assays for relevant markers of cell viability, signaling pathways, and organ-specific toxicity. Over 40 years of assay development experience and state-of-the-art manufacturing facilities ensure time-tested reproducibility.



### **Small Molecule Chemistry**

Our long-standing, flagship SCREEN-WELL® Compound Library product family offers an easy, ready-to-use method to streamline compound screening. We have a unique offering of focused compound libraries for organ-specific toxicology, as well as FDA approved drugs, natural products, chemical genomics, pathway targeting, and more.

Our in-house chemical production capabilities, combined with our supplier network built over a 40 year history, gives us the ability to rapidly and inexpensively source traditional, custom, and bulk compounds in the gram to kilogram scale. We complete our sourcing using stringent quality control standards with state-of-the-art methods.

# HTS ASSAYS FOR CANONICAL WNT PATHWAY ANALYSIS

Development of Wnt pathway modulating drugs requires the ability to rapidly screen the effects of novel or repurposed compounds and biological therapeutics on the expression of Wnt-regulated genes, and confirmation of such effects by related changes in key signaling components. Together, the LEADING LIGHT® Wnt Reporter Assay, LEADING LIGHT® Sclerostin-LRP Interaction Screening System, and SCREEN-WELL® Wnt Pathway Library facilitate high-throughput analysis of Wnt-related signaling.

### LEADING LIGHT® Wnt Reporter Assay (ENZ-61001)

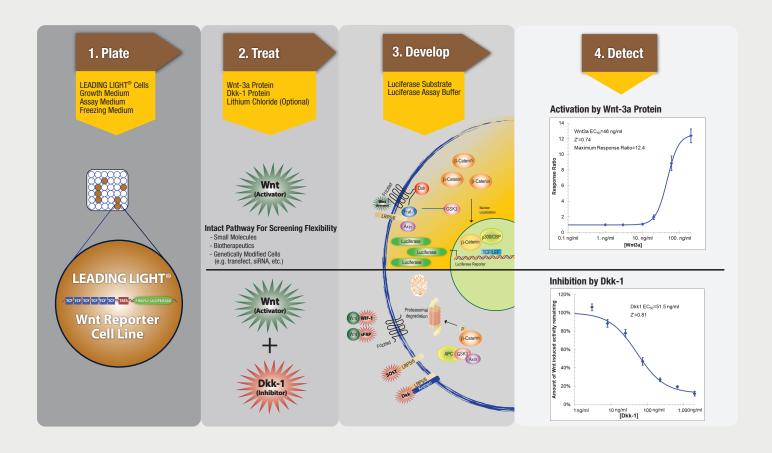
### Reliable, All-inclusive Screening of Wnt Pathway Modulators

The LEADING LIGHT® Wnt Reporter Assay Starter Kit is a high-sensitivity, luciferase-based assay for the analysis of canonical Wnt signaling. The assay uses a 3T3 mouse fibroblast cell line modified with a luciferase reporter gene under the control of Wnt-responsive promoters (TCF/LEF). The chemiluminescent readout is amenable to both 96-well and 384-well plate formats, facilitating high-throughput analysis of Wnt pathway activators and inhibitors.

### Screen Your Wnt Pathway In Only 4 Steps!

- A true end-point detection system for screening compounds
- . Greater sensitivity than other whole cell assay kits on the market
- Reproducibility that comes from using a validated system
- . Time-saving, no-transfection kit includes cells, media, detection reagents and controls

| RELATED PRODUCTS                               |           |  |  |  |  |
|--|-----------|--|--|--|--|
| Product Name                                   | Product # |  |  |  |  |
| Wnt-3a (mouse),<br>(recombinant)               | ENZ-60001 |  |  |  |  |
| Dkk-1 (mouse),<br>(recombinant) (active)       | ENZ-60002 |  |  |  |  |
| LEADING LIGHT® Wnt<br>Reporter Cell Line       | ENZ-61002 |  |  |  |  |
| LEADING LIGHT® Wnt Cell<br>Line Medium Pack    | ENZ-60003 |  |  |  |  |
| LEADING LIGHT® Wnt Cell<br>Line Assay Reagents | ENZ-60004 |  |  |  |  |



## SCREEN WNT PATHWAY ACTIVATORS & INHIBITORS

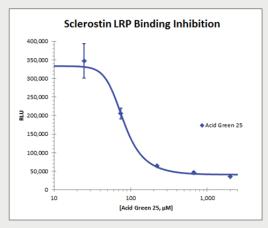
### LEADING LIGHT® Sclerostin-LRP Interaction Screening System (ENZ-61003)

### Novel Cell-free Sclerostin/LRP Binding Assay Ideal for Primary Inhibitor Screens

Sclerostin is encoded by the SOST gene and serves as a negative regulator of bone formation. Wnt ligands bind to Frizzled (Fz) and LRP5/6 receptors to trigger the canonical Wnt signaling cascade, which has been reported to play an important role in bone development. Sclerostin can bind to LRP5/6 to inhibit the canonical Wnt signaling. The LEADING LIGHT® Sclerostin-LRP Screen System is a simple and rapid biochemical assay which does not require cell lines or transfection reagents, making it a convenient platform for initial screening of inhibitors of the Wnt antagonist Sclerostin.

- High-sensitivity chemiluminescent readout (signal-to-noise ratio of  $\geq 10$ )
- Rapid protocol yields results in just 2.5 hours
- No cell lines or transfection reagents required
- Reproducible, high-throughput amenable assay (Z'-factor 0.871)
- Amenable for high-throughput screening applications

# Measurement of Standard Curve of Sclerostin-LRP



Inhibition of Sclerostin binding by Acid Green 25. The LRP5-ALP was bound to sclerostin in the presence of the indicated concentration of Acid Green 25 used to inhibit this interaction.



# SCREEN-WELL® Wnt Pathway Compound Library (BML-2838)

The SCREEN-WELL® Wnt Pathway Library offers a convenient solution for benchmarking compounds against a collection of 75 known Wnt modulators. The library is available in 100µL and 500µL formats (dissolved in DMSO) for use as a companion to the LEADING LIGHT® Wnt Reporter Assay or individually as a general screening tool for chemical genomics or Wnt assays.

### SCREEN-WELL® Wnt Pathway Library Targets Include:

| Wnt5a | Dkk         | Axin   | Porcupine         | β-catenin | LRP5/6 |
|-------|-------------|--------|-------------------|-----------|--------|
| TCF   | Dishevelled | GSK-3β | Adenylate Cyclase | COX-II    | sFRP-1 |

# QUANTIFY CRITICAL WNT PATHWAY PROTEINS

### Sensitive Wnt Pathway ELISA Kits

Obtain fully quantitative data with immunometric detection of  $\beta$ -Catenin, total and phosphorylated GSK-3 $\beta$ , and Dkk-1 in human, mouse and rat samples.

- Sensitive assays measure picogram levels compared to microgram levels in Western blot
- Suitable for cell lysates, culture supernates, plasma or serum samples (see table)
- High-throughput format allows analysis of 40 samples in duplicate in less than 3 hours
- Simple protocols and liquid color-coded reagents save time and reduce error

| PRODUCT                  | PRODUCT #    | SENSITIVITY | SPECIFICITY |       | SAMPLE TYPES |              |                    |       |        |
|--------------------------|--------------|-------------|-------------|-------|--------------|--------------|--------------------|-------|--------|
|                          |              |             | Human       | Mouse | Rat          | Cell Lysates | Culture Supernates | Serum | Plasma |
| β-catenin                | ADI-900-135  | 27.0 pg/mL  | •           | •     | •            | •            |                    |       |        |
| COX-2 (human) ELISA Kit  | ADI-900-094  | 0.25 ng/mL  | •           |       |              | •            |                    |       |        |
| Dkk-1                    | ADI-900-151  | 0.98 pg/mL  | •           |       |              |              | •                  | •     | •      |
|                          | ADI-900-172  | 116.7 pg/mL |             | •     |              |              | •                  | •     | •      |
|                          | ADI-900-172  | 26.1 pg/mL  |             |       | •            |              | •                  | •     | •      |
| GSK-3β                   | ADI-900-144  | 74.4 pg/mL  | •           |       | •            | •            |                    |       |        |
| IGF-1 (human), ELISA Kit | ADI-900-150  | 48.5 pg/mL  | •           |       |              |              |                    | •     | •      |
| [pSer9] GSK-3β           | ADI-900-123A | 9.0 pg/mL   | •           | •     | •            | •            |                    |       |        |
| TGF-β1 ELISA Kit         | ADI-900-155  | 3.3 pg/mL   | •           | •     | •            |              | •                  | •     | •      |
| VEGF (human), ELISA Kit  | ENZ-KIT156   | 3.979 pg/mL | •           |       |              |              | •                  | •     | •      |

# Highly Characterized Wnt-Related Proteins & Antibodies

Enzo offers a wide selection of high-quality reagents for analysis of Wnt related signal transduction, including purified proteins and thoroughly validated monoclonal and polyclonal antibodies.

| SELECT WNT PATHWAY PROTEINS & ANTIBODIES             |               |                                    |                |  |  |
|--|---------------|------------------------------------|----------------|--|--|
| Product Name   | Product #     | Product Name                       | Product #      |  |  |
| Agrin, mAb (Agr-247)                                 | ADI-AGR-540   | GSK-3β, pAb                        | ADI-KAP-ST002  |  |  |
| Axin-1 (human), (recombinant)                        | ENZ-PRT130    | Laminin (human fibroblasts)        | ENZ-PRT159     |  |  |
| Axin-2 (human), (recombinant)                        | ENZ-PRT131    | Laminin-2, mAb (4H8-2)             | ALX-804-190    |  |  |
| β-Catenin, mAb (12F7)                                | ADI-KAM-ST001 | MMP-9 (catalytic domain) (human)   | BML-SE360      |  |  |
| BMP-4 (human), (recombinant)                         | ENZ-PRT146    | NGAL (rat), mAb (8)                | BPD-ABS-039-08 |  |  |
| BMP-4, mAb (26H16)                                   | ADI-905-757   | Noggin (human), (recombinant)      | ENZ-PRT127     |  |  |
| BMP-4, mAb (6B7)                                     | ADI-905-834   | Osteocalcin (mouse), pAb           | ALX-210-333    |  |  |
| Casein Kinase II (human), (recombinant)              | BML-SE124     | Osteopontin, mAb (53)              | ADI-905-629    |  |  |
| Collagen I, rat tail                                 | ALX-522-435   | Osteoprotegerin (human), mAb       | ALX-804-813B   |  |  |
| Dishevelled-1, pAb                                   | BML-DA4170    | VE-cadherin, pAb                   | ALX-210-232    |  |  |
| Dishevelled-2, pAb                                   | BML-DA4270    | VEGF, mAb (6B7)                    | ADI-905-164    |  |  |
| Dkk-1 (mouse), (recombinant) (His-tag)               | ADI-KPR-CP100 | VEGF-121 (human), (recombinant)    | ENZ-PRT128     |  |  |
| EGF (human), (recombinant)                           | ENZ-PRT154    | VEGF-121 (human), (recombinant)    | ENZ-PRT129     |  |  |
| Fibronectin (human plasma)                           | ENZ-PRT158    | Vitronectin (human), (recombinant) | ENZ-PRT179     |  |  |
| GSK-3 $\alpha$ / $\beta$ , mAb (1H8)                 | ADI-KAM-ST002 | Wnt-3a (mouse), (recombinant)      | ENZ-60001      |  |  |
| $GSK-3\beta \; (human),  (recombinant) \; (GST-tag)$ | BML-SE355     | Wnt-1, pAb                         | ADI-905-617    |  |  |

#### You may also be interested in:













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