

## ERAI ER Stress Detector

Cellular stresses, which are implicated in various diseases, include stresses caused by intra-tissue factors such as low oxygen status and intracellular stresses such as endoplasmic reticulum (ER) stress and oxidant stress. ER stress is caused by the accumulation of unfolded proteins in the ER lumen and associated with neurodegenerative diseases, metabolic syndrome, and various cancers.

**ERAI ER Stress Detector HD** utilizes the expression regulation system of ER stress factor XBP1 protein. *XBPI* mRNA is transcribed with intron sequence, which is translated as inactive XBP1 protein. However, under the ER stress condition, intron is spliced out, and active XBP1 protein is produced. ERAI ER Stress detector is a reporter gene in which luciferase gene is connected to downstream of intron region of *XBPI* cDNA. In cells introduced with this detector gene, ER stress induces splicing of the *XBPI* intron sequence, and fusion protein of XBP1 N terminal region and luciferase is translated.

ERAI ER Stress Detector can be used to detect ER stress with luciferase activity in high S/N ration.. If high sensitivity is required for your assay, **ERAI ER Stress Detector** is recommended.

For detection of ER stress in living mouse, Tg type ERAI-Luc mouse which is introduced with ERAI ER Stress Detector is suitable.

|          |   |
|----------|---|
| Material | Plasmid DNA   |
| Quantity | 5 µg DNA/vial, 20 µL TE (sterilized)                |
| Storage  | - 20°C  |
|          | Plasmid is not treated in endotoxin free condition. |



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**【An example of experimental procedure】**

Cultured cells are transfected with ERAI ER Stress Detector and stimulated with ER stress inducer tunicamycin.

- 1) One day prior to transfection, cells are plated in 35 mm culture dish. Cell number should be 50 - 80% confluent after overnight culture (approximately  $1 - 3 \times 10^5$  cells/dish for adherent cells).
- 2) ERAI ER Stress Detector HD plasmid is transfected. Detail of transfection method follows the manufacturer's instruction of transfection reagent.
- 3) After 24 - 48 hours, culture medium is exchanged to new one with 5  $\mu\text{g/ml}$  tunicamycin. Note that the optimum concentration of tunicamycin depends on each cell line.
- 4) After 6 hours incubation with tunicamycin, cells are harvested and assayed for luciferase activity by Luciferase Assay System (Promega). Please see detailed method in instruction of product.

**【References】**

Hosoda A., Tokuda M., Akai R., Kohono K., Iwawaki T. Biochem J., 425, 117-125 (2010)

**【License statements】**

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