

## Kit Insert

### Relevant Products

- #U0900G Green Cell Stress Sensor (CMV)
- #U0901G Ratiometric Cell Stress Sensor (CMV)
- #U0915G Ratiometric Cell Stress Sensor (CAG)
- #U0921G XBP1-IRE1 Ratiometric Cell Stress Sensor (CMV)

### Materials Included

- Cell Stress sensor in BacMam vector:  $\sim 2 \times 10^{10}$  VG/mL in ESF 921 Insect Cell Culture Medium (Expression Systems #96-001-01).
- Sodium Butyrate (Sigma Aldrich product # B5887) 500 mM in H<sub>2</sub>O.

Add sodium butyrate to cultured cells to maintain BacMam expression as needed. Other HDAC inhibitors may work as well or even better in certain cell types. For expression in CHO cells, we recommend valproic acid instead of sodium butyrate. Many cell types maintain expression without an HDAC inhibitor.

- Thapsigargin (Cayman Chemical product # 10522) dissolved in DMSO at 1mM.

Thapsigargin is a SERCA pump inhibitor that induces high levels of ER stress. It is used as the positive control when conducting the cell stress assay. **Thapsigargin is supplied in 20  $\mu$ L aliquots in airtight tubes purged with nitrogen. Thapsigargin will arrive on ice. It should immediately be aliquoted and stored at -20°C. On the day of the experiment an aliquot may be thawed and diluted into H<sub>2</sub>O or DPBS buffer and used immediately. Discard any unused thapsigargin aliquot.**

### Storage

BacMam vectors and HDAC Inhibitors should be stored at 4°C and protected from light in the original package. Avoid repeated freeze/thaw cycles. We recommend retesting BacMam stock after storing for more than 12 months at 4°C, or after any freeze-thaw cycle. Store HDAC inhibitor at 4°C. Store control agonist at -20°C.

### QA/QC

BacMam stocks are tested for sterility. Samples are added to rich cell culture media without antibiotic and incubated at 37°C, 5% CO<sub>2</sub>, and checked for bacterial or fungal growth after 5 days. Viral genes (VG) per milliliter (mL) are measured by qPCR with primers specific to VSVG. Check tube label for exact titer. Viral genomic DNA at multiple dilutions are run in qPCR against a standard curve to generate an average titer for each BacMam stock. Each tube of stock is labeled with VG/mL and a stock keeping unit (SKU) identifier. To test efficacy, serial dilutions are added to cultured HEK 293 cells. After 24 hours, fluorescent cells are counted to establish transducing units per mL of stock.

### Biosafety

BacMam is the modified baculovirus, *Autographa californica*, AcMNPV. Baculovirus is pseudotyped to infect mammalian cells, but it does not replicate and its genome is silent in mammalian cells. While it should be handled carefully, in a sterile environment, it is classified as a BSL-1 reagent.

For Research Use Only. Not recommended for use or sale in human or animal diagnostic or therapeutic products. This product contains no substances which at their given concentration are considered to be hazardous to health, however we recommend handling with care. Wear impervious gloves and eye protection when handling. Do not ingest.

Review the protocols on Montana Molecular's Website: [www.montanamolecular.com](http://www.montanamolecular.com) before using these products.

**Materials are provided without warranty, express or implied. End user is responsible for making sure product use complies with applicable regulations. No right to resell any components of these products is conveyed. Reverse engineering or modification of these products is not permitted.**

Made in the USA