SREBP v1 Reporter Cell Line (HT1080)



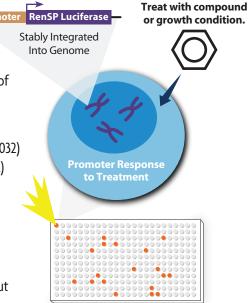
Catalog No.: 32221

The LightSwitch™ SREBP v1 Reporter Cell Line (HT1080) was designed for study of the cholesterol biosynthesis pathway. It contains a stably integrated LightSwitch Promoter Reporter GoClone™ construct (S711010), which is comprised of the promoter sequence of the LSS (lanosterol synthase or 2,3-oxidosqualene-lanosterol cyclase) gene cloned into the Promoter Reporter Vector, pLightSwitch_Prom, upstream of the RenSP luciferase gene. LSS catalyzes the first step in the biosynthesis of cholesterol.

IMPORTANT: Because all LightSwitch reporter cell lines contain the optimized RenSP luciferase gene, you MUST use our **LightSwitch Luciferase Assay Kit** (Cat. Nos. 32031 & 32032) to obtain optimal results. (Other luciferase assay reagents are not compatible with RenSP.)

Experimental Details:

- Assays were performed in triplicate. 10K cells per well were seeded in a 96-well white plate in standard media without antibiotic.
- 2. Cells were incubated at 37°C for 8 hours.
- 3. Standard media was removed and replaced with 100 μ l of low lipid media without antibiotic. The cells were then incubated at 37°C for 24 hours (serum starvation).
- 4. The cells were then induced with the treatment conditions indicated below. A 10X dose response series of Lovastatin was made in OptiMEM. Ten μl of the indicated 10X stock was added to the designated wells.
- 5. Cells were incubated at 37°C for 24 hours.
- 6. Plates were frozen at -80°C overnight. (This step is optional, but freezing ensures complete lysis of the cells prior to running the LightSwitch Assay.)
- 7. Plates were thawed to room temperature and LightSwitch Luciferase Assays were performed per the standard protocol.
- 8. The data was normalized to the Control 2 Reporter Cell Line (ACTB promoter, HT1080 cells; Cat. No. 32202); expression data for the control cell line was averaged across all doses. Experimental data points were then divided by this average value to normalize for non-specific effects.



Measure luciferase activity to determine effect of treatments.

