

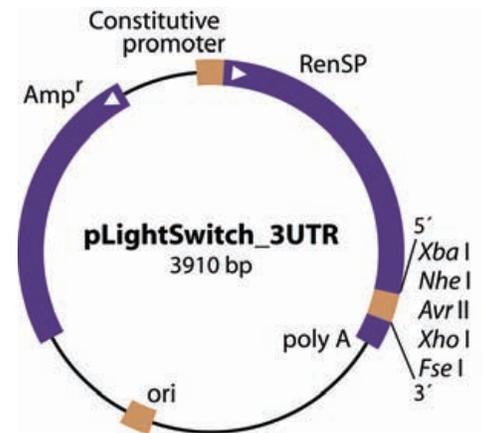
LightSwitch™ Synthetic miRNA Target Reporter

Catalog Nos: 26000-26949 – **Product IDs:** S880001-S880950

Concentration: ~30 ng/μl

Quality: OD 260/280 ratio ≥ 1.75

Clone & Vector Information: Each LightSwitch Synthetic miRNA Target Reporter construct contains an optimized synthetic target consisting of sequence repeats that are fully complementary to a variety of human and viral miRNAs, which are cloned downstream of the RenSP luciferase gene in the pLightSwitch_3UTR reporter vector. These vectors provide a highly sensitive readout for miRNA functional activity, making them ideal positive control targets in miRNA target validation experiments, or to determine the levels of natural miRNA expression in your cell line. LightSwitch vector maps, annotations, and sequence & primer information are available at www.activemotif.com/ls-vectors. For information about your specific clone, click its **miRBase Info** link in the table at www.activemotif.com/ls-synth-3utr.



LightSwitch Assays: Because all LightSwitch Reporter constructs utilize the RenSP luciferase reporter gene, you **MUST** use the LightSwitch Luciferase Assay Kit (Cat. Nos. 32031 & 32032) to perform luciferase assays with all LightSwitch vectors. This kit contains a proprietary substrate that was formulated specifically for use with our engineered RenSP gene. Other luciferase assay reagents are not compatible with RenSP. For more information, visit www.activemotif.com/ls-assay.

Transfection Reagents: We recommend DharmaFECT® Duo (Cat. Nos. 32044 & 32045) to co-transfect LightSwitch plasmids with miRNA mimics or inhibitors, as it efficiently delivers both plasmid DNA and small RNAs. If you are transfecting plasmid only, we recommend FuGENE® HD Transfection Reagent (Cat. Nos. 32042 & 32043) because it provides superior efficiency and low cytotoxicity across a wide variety of cell lines. For more information, please visit www.activemotif.com/transfect.

Positive & Negative Controls: We recommend that you include appropriate positive & negative control LightSwitch vectors when you perform your assays. We offer a panel of positive and negative 3'UTR control constructs. The empty pLightSwitch_3UTR reporter vector contains a constitutive promoter and RenSP with no 3'UTR, so it can serve as a high-expressing positive transfection control. The empty 3'UTR vector may also serve as a negative control for miRNA signaling because it contains no 3'UTR. We also offer additional control vectors that contain the 3'UTRs of common housekeeping genes, as well as random genomic sequences cloned downstream of RenSP.

Single vs. Dual Assay Design: Modern transfection reagents and optimized luciferase assay reagents such as LightSwitch have largely eliminated the need to do a co-transfection control. In most cases, using a dual assay format provides little benefit, while increasing costs and reducing assay sensitivity. Unless you are using a hard-to-transfect cell line, we recommend testing the variation between transfection replicates in a single transfection format. If you wish to do a co-transfection, we offer the LightSwitch Dual Assay Kit (Cat. No. 32035), which has been optimized for use with all LightSwitch reporter vectors. For more information on the pros and cons of co-transfection, please download our Technical Note by entering www.activemotif.com/ls-co-trans into your browser.