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eIF3 δ shRNA (h) Lentiviral Particles: sc-43749-V

BACKGROUND

The initiation of protein synthesis in eukaryotic cells is regulated by interactions between protein initiation factors and RNA molecules. Eukaryotic initiation factors (eIFs) are utilized in a sequence of reactions that lead to 80S ribosomal assembly and, ultimately, translation. The eukaryotic initiation factor-3 (eIF3) scaffolding structure is the largest of the eIF complexes and includes eIF3 α , eIF3 β , eIF3 γ , eIF3 δ , eIF3 ϵ , eIF3 ζ , eIF3 η and eIF3 θ all of which function to control the assembly of the 40S ribosomal subunit. Association of eIF3 proteins with the 40S ribosomal subunit stabilizes eIF2-GTP-Met-tRNA^{iMet} complex association and mRNA binding, and promotes dissociation of 80S ribosomes into 40S and 60S subunits, thereby promoting the assembly of the pre-initiation complex. Overexpression of eIF3 proteins is common in several cancers, suggesting a role for eIF3 proteins in tumorigenesis.

REFERENCES

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CHROMOSOMAL LOCATION

Genetic locus: EIF3G (human) mapping to 19p13.2.

PRODUCT

eIF3 δ shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see eIF3 δ siRNA (h): sc-43749 and eIF3 δ shRNA Plasmid (h): sc-43749-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

eIF3 δ shRNA (h) Lentiviral Particles is recommended for the inhibition of eIF3 δ expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

eIF3 δ (R-20): sc-16362 is recommended as a control antibody for monitoring of eIF3 δ gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz MarkerTM compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz MarkerTM Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruzTM Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor eIF3 δ gene expression knockdown using RT-PCR Primer: eIF3 δ (h)-PR: sc-43749-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

PROTOCOLS

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