

IRG-47 siRNA (m): sc-41793

BACKGROUND

A distinct family of interferon- γ (IFN- γ) inducible GTPases, belonging to the GTPase superfamily, are selectively induced by IFN- γ or bacterial lipopolysaccharide (LPS) stimulation. These putative GTPases include TGTP, IRG-47, LRG-47 and IGTP, and they are involved in mediating the cellular innate immune responses. Similar to other GTPases, they contain a characteristic nucleotide-binding domain for GTP and are functionally regulated by the binding and hydrolysis of GTP. In addition, these related proteins also contain significant sequence similarity among themselves, are largely similar in size, and yet they are differentially expressed. TGTP, or T cell specific GTPase, is preferentially expressed in T cells and is upregulated in response to TCR cross-linking. IGTP (inducibly expressed GTPase) is expressed predominantly in macrophages, whereas IRG-47 is primarily expressed in all cells derived from B cell lineages, and LRG-47 is highly expressed in macrophages following IFN- γ stimulation. Two additional proteins, ILGP and GTP1, are expressed in mouse embryonic fibroblasts and macrophages and are likewise upregulated by IFN- γ stimulation.

REFERENCES

1. Dever, T.E., et al. 1987. GTP-binding domain: three consensus sequence elements with distinct spacing. *Proc. Natl. Acad. Sci. USA* 84: 1814-1818.
2. Gilly, M. and Wall, R. 1992. The IRG-47 gene is IFN- γ induced in B cells and encodes a protein with GTP-binding motifs. *J. Immunol.* 148: 3275-3281.
3. Sorace, J.M., et al. 1995. Identification of an endotoxin and IFN-inducible cDNA: possible identification of a novel protein family. *J. Leukoc. Biol.* 58: 477-484.
4. Carlow, D.A., et al. 1995. Isolation of a gene encoding a developmentally regulated T cell-specific protein with a guanine nucleotide triphosphate-binding motif. *J. Immunol.* 154: 1724-34.
5. Taylor, G.A., et al. 1996. Identification of a novel GTPase, the inducibly expressed GTPase, that accumulates in response to IFN- γ . *J. Biol. Chem.* 271: 20399-20405.
6. Drysdale, B.E., et al. 1996. Identification of a lipopolysaccharide inducible transcription factor in murine macrophages. *Mol. Immunol.* 33: 989-998.

CHROMOSOMAL LOCATION

Genetic locus: Ifi47 (mouse) mapping to 11 B1.2.

PRODUCT

IRG-47 siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see IRG-47 shRNA Plasmid (m): sc-41793-SH and IRG-47 shRNA (m) Lentiviral Particles: sc-41793-V as alternate gene silencing products.

For independent verification of IRG-47 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-41793A, sc-41793B and sc-41793C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

IRG-47 siRNA (m) is recommended for the inhibition of IRG-47 expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

IRG-47 (G-3): sc-390264 is recommended as a control antibody for monitoring of IRG-47 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 reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor IRG-47 gene expression knockdown using RT-PCR Primer: IRG-47 (m)-PR: sc-41793-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.