

DESCRIPTION

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|---------------------------|---|
| Species Reactivity | Human |
| Specificity | Detects human MANF in direct ELISAs and Western blots. |
| Source | Polyclonal Goat IgG |
| Purification | Antigen Affinity-purified |
| Immunogen | Mouse myeloma cell line NS0-derived recombinant human MANF Leu22-Leu179 Accession # P55145 |
| Formulation | Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS. |

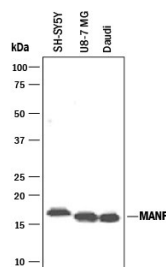
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. [General Protocols](#) are available in the Technical Information section on our website.

| | Recommended Concentration | Sample |
|---------------------------|--|---------------|
| Western Blot | 1 µg/mL | See Below |
| Simple Western | 10 µg/mL | See Below |
| Knockout Validated | MANF is specifically detected in HEK293T human embryonic kidney parental cell line but is not detectable in MANF knockout HEK293T cell line. | |

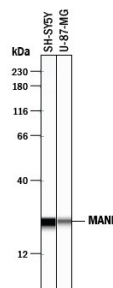
DATA

Western Blot



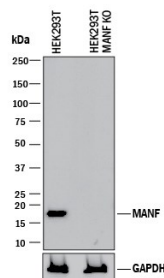
Detection of Human MANF by Western Blot. Western blot shows lysates of SH-SY5Y human neuroblastoma cell line, U-87 MG human glioblastoma/astrocytoma cell line, and Daudi human Burkitt's lymphoma cell line. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human MANF Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3748) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for MANF at approximately 17 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Simple Western



Detection of Human MANF by Simple Western™. Simple Western lane view shows lysates of SH-SY5Y human neuroblastoma cell line and U-87 MG human glioblastoma/astrocytoma cell line, loaded at 0.2 mg/mL. A specific band was detected for MANF at approximately 24 kDa (as indicated) using 10 µg/mL of Goat Anti-Human MANF Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3748) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

Knockout Validated



Western Blot Shows Human MANF Specificity by Using Knockout Cell Line. Western blot shows lysates of HEK293T human embryonic kidney parental cell line and MANF knockout HEK293T cell line (KO). PVDF membrane was probed with 1 µg/mL of Goat Anti-Human MANF Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3748) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for MANF at approximately 17 kDa (as indicated) in the parental HEK293T cell line, but is not detectable in knockout HEK293T cell line. GAPDH (Catalog # AF5718) is shown as a loading control. This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

PREPARATION AND STORAGE

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|--------------------------------|--|
| Reconstitution | Reconstitute at 0.2 mg/mL in sterile PBS. |
| Shipping | The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C |
| Stability & Storage | Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution. |

BACKGROUND

Mesencephalic astrocyte-derived neurotrophic factor (MANF), also known as arginine-rich, mutated in early stage tumors (ARMET) and arginine-rich protein (ARP), is a 20 kDa member of the ARMET family of proteins (1). The name ARMET comes from the fact that the protein was initially thought to be 50 aa longer at the N-terminus and to contain an arginine-rich region (2-5). The presence of a specific mutation changing the previously numbered codon 50 from ATG to AGG, or deletion of that codon, has been reported in a variety of solid tumors (2-4). Human MANF is synthesized as a 179 amino acid (aa) precursor that contains a 21 aa signal sequence and a 158 aa mature chain. Mature human MANF is 99%, 98%, and 96% aa identical to mature rat, mouse and bovine MANF, respectively. MANF is localized to the endoplasmic reticulum (ER) and Golgi apparatus, and is also secreted (5). In the CNS, MANF selectively protects nigral dopaminergic neurons, versus GABAergic or serotonergic neurons, which suggests that MANF may be indicated for the treatment of Parkinson's disease (1). MANF is also one of the 12 commonly unfolded protein response (UPR)-upregulated genes (5). One study showed that MANF plays an important role in protecting cells against tunicamycin and thapsigargin-induced cell death (5). Loss of MANF renders cells more susceptible to those drugs, but also increases cell proliferation and decreases cell size (5). Another study showed that MANF is an endoplasmic reticulum stress response (ERSR) gene in the heart that can be induced and secreted in response to ER stresses, including ischemia, and that extracellular MANF may protect cardiac myocytes in an autocrine and paracrine manner (6).

References:

1. Petrova, P.S. *et al.* (2003) J. Mol. Neurosci. **20**:173.
2. Shridhar, V. *et al.* (1996) Oncogene **12**:1931.
3. Shridhar, R. *et al.* (1996) Cancer Res. **56**:5576.
4. Shridhar, V. *et al.* (1997) Oncogene **14**:2213.
5. Apostolou, A. *et al.* (2008) Exp. Cell Res. **314**:2454.
6. Tadimalla, A. *et al.* (2008) Circ. Res. **103**:1249.