



Product Information Sheet

Human MIF ELISA Kit

Catalog No.	EK0813
Size	96T
Range	156pg/ml-10,000pg/ml
Sensitivity	< 20pg/ml

Specificity

No detectable cross-reactivity with any other cytokine.

Storage

Store at 4°C for frequent use, at -20°C for infrequent use.

Avoid multiple freeze-thaw cycles (Shipped with wet ice.)

Expiration

Two months at 4°C and four months at -20°C.

Application

For quantitative detection of human MIF in sera, plasma, body fluids, tissue lysates or cell culture supernates.

Principle

Human MIF ELISA Kit was based on standard sandwich enzyme-linked immune-sorbent assay technology. Human MIF specific polyclonal antibodies were precoated onto 96-well plates. The human specific detection polyclonal antibodies were biotinylated. The test samples and biotinylated detection antibodies were added to the wells subsequently and then followed by washing with PBS or TBS buffer. Avidin-Biotin-Peroxidase Complex was added and unbound conjugates were washed away with PBS or TBS buffer. HRP substrate TMB was used to visualize HRP enzymatic reaction. TMB was catalyzed by HRP to produce a blue color product that changed into yellow after adding acidic stop solution. The density of yellow is proportional to the human MIF amount of sample captured in plate.

Kit Components

1. Lyophilized recombinant human MIF standard: 10ng/tubex2.
2. One 96-well plate precoated with anti- human MIF antibody.
3. Sample diluent buffer: 30 ml
4. Biotinylated anti- human MIF antibody: 130µl, dilution 1:100.
5. Antibody diluent buffer: 12ml.
6. Avidin-Biotin-Peroxidase Complex (ABC): 130µl, dilution 1:100.
7. ABC diluent buffer: 12ml.
8. TMB color developing agent: 10ml.
9. TMB stop solution: 10ml.

Material Required But Not Provided

1. Microplate reader in standard size and Automated plate washer.
2. Adjustable pipettes and pipette tips. Multichannel pipettes are recommended if there is a large amount of samples.
3. Clean tubes and Eppendorf tubes.
4. Washing buffer (neutral PBS or TBS).

Preparation of 0.01M **TBS**: Add 1.2g Tris, 8.5g NaCl; 450µl of purified acetic acid or 700µl of concentrated hydrochloric acid to 1000ml H₂O and adjust pH to 7.2-7.6. Finally, adjust the total volume to 1L.

Preparation of 0.01 M **PBS**: Add 8.5g sodium chloride, 1.4g Na₂HPO₄ and 0.2g NaH₂PO₄ to 1000ml distilled water and adjust pH to 7.2-7.6. Finally, adjust the total volume to 1L.

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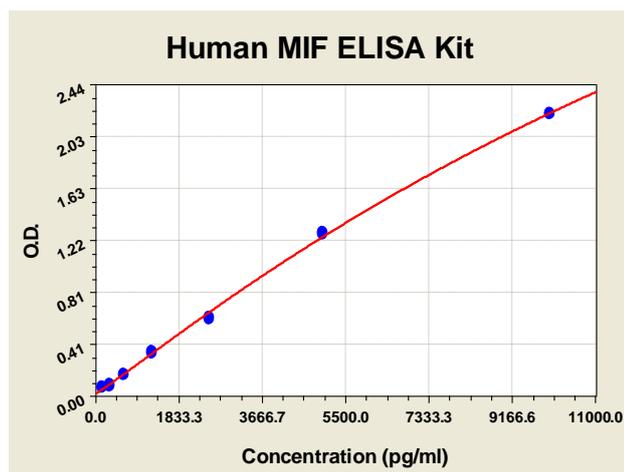
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Notice for Application of Kit

1. Before using Kit, spin tubes and bring down all components to bottom of tube.
2. Duplicate well assay was recommended for both standard and sample testing.
3. Don't let 96-well plate dry, dry plate will inactivate active components on plate.
4. In order to avoid marginal effect of plate incubation due to temperature difference (reaction may be stronger in the marginal wells), it is suggested that the diluted ABC and TMB solution will be pre-warmed in 37°C for 30 min before using.

Human MIF ELISA Kit-1X96 Well Plate Image



Background

Macrophage migration inhibitory factor (MIF) is a protein which in humans is encoded by the *MIF* gene.^{1,2} This gene is located to human chromosome 22q11.2.³ It is remarkably small; it has 3 exons separated by introns of only 189 and 95 bp, and covers less than 1 kb.⁴ This gene encodes a lymphokine that may be involved in cell-mediated immunity, immunoregulation, and inflammation.⁵ MIF plays a role in the regulation of macrophage function in host defense through the suppression of anti-inflammatory effects of glucocorticoids.⁶ This lymphokine and the JAB1 protein might form a complex in the cytosol near the peripheral plasma membrane, which may possibly indicate a role in integrin signaling pathways. MIF also plays a central role in the toxic response to endotoxemia and possibly septic shock.⁷ Macrophage migration inhibitory factor has been reported to interact with COP9 constitutive photomorphogenic homolog subunit 5, CD74, BNIPL, and CXCR4. The standard product used in this kit is recombinant human MIF, consisting of 115 amino acids with the molecular mass of 12.5kDa.

Reference

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3. Budarf, M.; McDonald, T.; Sellinger, B.; Kozak, C.; Graham, C.; Wistow, G. : Localization of the human gene for macrophage migration inhibitory factor (MIF) to chromosome 22q11.2. *Genomics* 39: 235-236, 1997.
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6. Flaster H, Bernhagen J, Calandra T, Bucala R (June 2007). "The macrophage migration inhibitory factor-glucocorticoid dyad: regulation of inflammation and immunity.". *Mol Endocrinol.* 21 (6): 1267-80.
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