

Prokaryotic Expression and Purification of the Macrophage Migration Inhibitory Factor (MIF) from *Leishmania major*

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Macrophage Migration Inhibitory Factor (MIF) was the first cytokine to be identified from the human T cells and participates in innate and adaptive immune response. MIF has been considered an important factor in the control of parasites infections, presenting a beneficial or a detrimental role, depending on the pathogen. MIF deficient mice are susceptible to the infection by *L. major* and *T. cruzi*. Interestingly, homologues of mammalian MIF which act as a modulator factor of the immune response from infected host have been isolated from parasites species. *Leishmania* is the causative agent of leishmaniasis, a parasitic disease with an estimated 12 million cases worldwide and with 1.5-2 million new cases reported each year. MIF homologue was identified in the soluble extract of vesicles of *L. amazonensis*, however the function of this protein in the trypanosomatid had still not understood. The LmMIF coding sequence region was amplified from *L. major* genomic DNA by PCR and cloned into the pET28a vector. The construct was used for protein expression in *E. coli* BL21(DE3)pLysS, and the recombinant LmMIF containing a His₆-tag was expressed in the soluble form and subsequently purified from the cell lysate by affinity chromatography using a Ni-NTA resin. The secondary and tertiary structure of the rLmMIF was evaluated by circular dichroism and intrinsic tryptophan fluorescence spectroscopy respectively, suggesting that the purified protein is in the native state.

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