

## RECOMBINANT EXPRESSION OF VARIABLE SURFACE GLYCOPROTEINS: LITAT 1.3 AND LITAT 1.5 OF *TRYPANOSOMA BRUCEI GAMBIENSE*

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Human African Trypanosomiasis is caused by the *Trypanosoma brucei gambiense* and *T. b. rhodesiense* parasites. These parasites express variable surface glycoprotein (VSG) coats, used in diagnostic tests, however their acquisition is of high risk to staff handling the parasites. This study aimed at recombinantly expressing selected VSGs, for possible use as alternatives in diagnostics.

Genes encoding VSG LiTats 1.3 and 1.5 were amplified from either genomic DNA or cDNA, cloned into a T-vector and sub-cloned into expression vectors, prior to recombinant expression in *E. coli* BL21 DE3 and purification by Ni-affinity chromatography. Amplification and subsequent cloning yielded the expected 1.4 kb and 1.5 kb for the *LiTat 1.3* and *LiTat 1.5* genes respectively. Recombinant expression in *E. coli* was only successful with the constructs cloned from cDNA. Purification of the 63 kDa cLiTat 1.3<sub>His</sub> protein following solubilising and refolding did not yield pure protein and signs of protein degradation were present. For comparison, expression was carried out in *P. pastoris* with expression only successful with the LiTat 1.3-SUMO construct yielding a 62.7 kDa protein. Purification of LiTat 1.3<sub>SUMO</sub> surpassed that of cLiTat 1.3<sub>His</sub> with no degradation.

With successful expression and purification, the recombinant proteins may potentially be used in diagnostics.

**Keywords:** Protein biochemistry, recombinant expression, Trypanosomiasis, *T. b. gambiense*, VSG.