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P0742 Evaluation of the diagnostic value of a recombinant protein mimicking envelope glycoprotein gp105 of HIV2 in the detection of anti-HIV2 antibodies in linear immunoblot format

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Background: Detection of antibodies against envelope glycoprotein (env) gp105 is an essential requirement by WHO for diagnosis of human immunodeficiency virus type2 (HIV2). Immunoblotting (Western blot or Line Immuno Assay (LIA)) is recommended as a confirmatory test. Applying of HIV2 gp105 onto 0,45nm membrane in form of 22 amino acid (aa) long synthetic peptide was complicated due to its small size. Immobilization of a biotinylated form of the peptide through streptavidin molecule has overcome the size problem, but decreased the specificity of the LIA test. The attempt to present the peptide sequence in a form of a tagged recombinant protein was taken.

Materials/methods: Region 306-327 aa of HIV2 envelop protein has been amplified by PCR and cloned as a recombinant protein in fusion with GST+6His tag. A second recombinant protein with the doubled target 22aa-sequence was also constructed. The obtained proteins have been immobilized on a nitrocellulose membrane and tested against 52 HIV-2 positive and 132 HIV-negative samples in LIA format in comparison with the synthetic form of the peptide.

Results: Sensitivity of the LIA test with gp 105 in the form of biotinylated synthetic peptide was 79.5%, specificity was 87.3%; non-specific reactions of serum components with streptavidin occurred. Recombinant protein with one repeat of the 22aa sequence in composition appeared to be inefficient in anti-HIV2 detection. The recombinant carrying the dimer version of the 22aa-peptide sequence has increased the specificity of LIA test up to 98.4%, and the sensitivity remained the same.

Conclusions: Redouble of the target 22aa sequence of HIV2gp105 in the composition of a recombinant protein with GST+6His tag has allowed direct sorption on the nitrocellulose membrane, and has significantly increased specificity of anti-HIV Ab detection in LIA format due to correct presentation of the immunodominant epitope.