IMMUNOREACTIVITY OF THE *ESCHERICHIA COLI*-SYNTHESIZED POLYPEPTIDE DERIVED FROM A SHORT SEGMENT OF THE HIV-1 *ENV* GENE

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The most widely used approach for identification of individuals infected with the AIDS virus is detection of specific viral antibodies by means of immunoenzymatic methods employing native viral antigens (*1*). When commercial tests have been utilized worldwide by health services, it became clear that their performance may not always fulfill the desirable demands (*2*). This concerns mainly the lowered specificity for the intrinsinc presence of contaminating (cross reacting material) in the purified virus (*3*).

For the production of a non-fused recombinant polypeptide we inserted the short DNA fragment covering the N-terminal part of HIV-1 gp41 into pKK233-2 expression vector (*4*) under the control of the *trc* promotor. As a result of chemically induced depression in transfected *E. coli*, the synthesis of recombinant peptide with molecular weight of 21 kD occured. The synthesized protein was readily detected by anti-HIV 1 positive sera in immunoblot (western) assay. A more detailed evaluation of the immunoreactivity of this genetically engineered protein was performed three panels of sera. In the first group 50 sera were included which were withdrawn from HIV-1-infected individuals. The screening and confirmatory assays scored for this group in concordance positively. In the second group were included 19 sera which were classified as false positive according to discrepant results, i.e. positive in EIA test but negative in immunoblot assay. All 26 sera of the last group originated from healthy blood donors and were devoided from specific anti-HIV-1 antibodies.

Each of the sera classified as positive, with the observed reactivity against p24, gp41, and gp120 antigens on nitrocellulose strips prepared from the whole virus, reacted intensively with recombinant protein. Interestingly, the distinct reaction was also observed with three sera which failed to react with gp41 in a Western blot assay of increased sensitivity (Du Pont). From the sera reacting false positively in screening competitive EIA none gave positive signal in our assay with recombinant antigen. Similarly, no reactions which could consequently be judged as nonspecific were recorded on analysis of sera from healthy subjects.

Our results witness the superiority of the application of recombinant antigen in serological diagnostic tests. Furthermore, the relatively short nucleotide stretch downstream the 5′-end of gp41 gene could code for a peptide comprising the crucial epitopes relevant for serodiagnosis of HIV-1 infection.

References

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