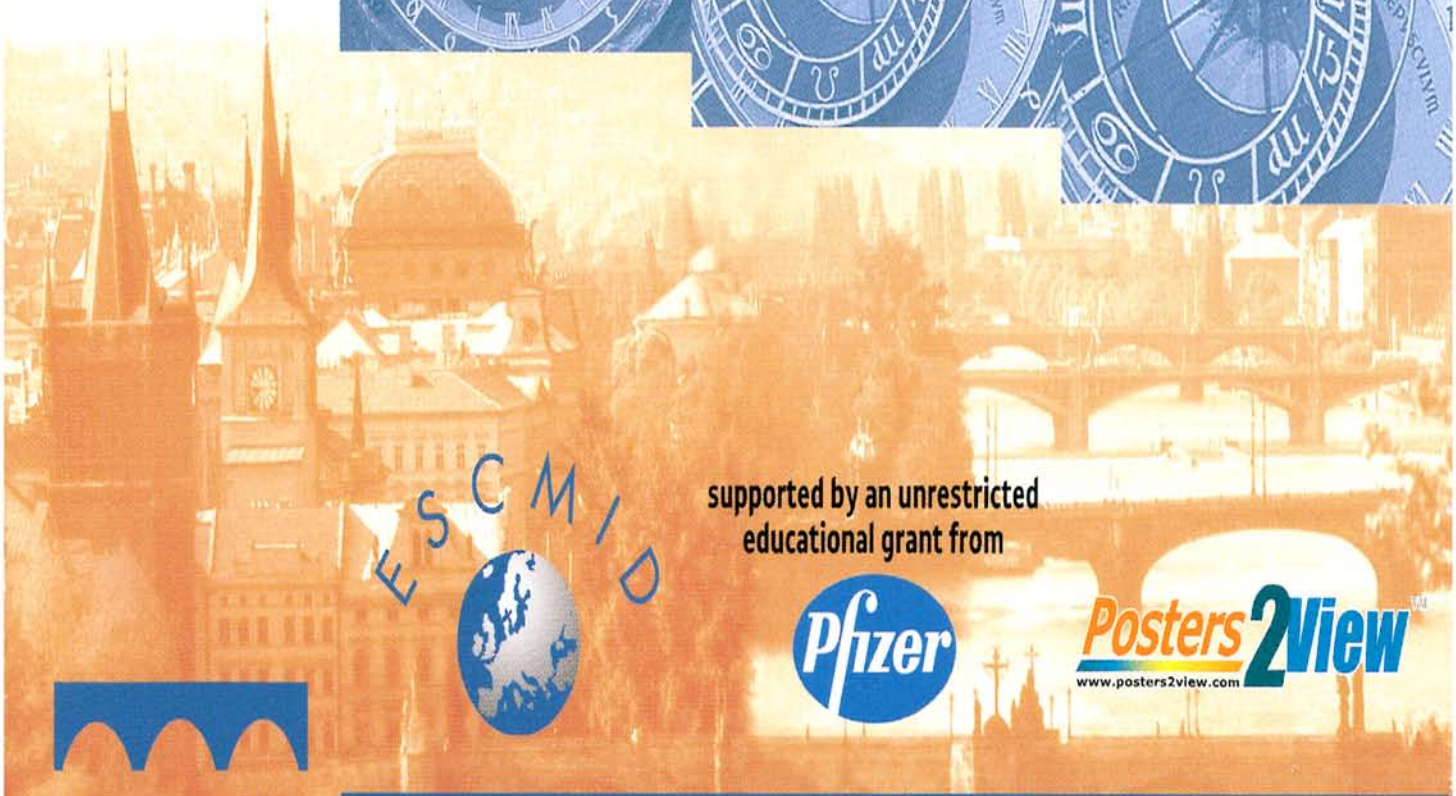
PRAHA 14th ECCMID

14th European Congress of Clinical Microbiology and Infectious Diseases



ESCMID

supported by an unrestricted educational grant from

Pfizer

Posters2View
www.posters2view.com

PRAHA 2004
14th ECCMID

Prague/Czech Republic, May 1-4, 2004

P844

A new enzyme immunoassay for the detection herpes simplex virus type 2 – specific antibodies

A. Obriadina, S. Klimashevskaya, M. Gladisheva, V. Puzyrev, A. Burkov, T. Ulanova
Nizhniy Novgorod, RUS

Objectives: The purpose of this study was to develop enzyme immunoassay (EIA) for the detection of IgG anti-HSV-2 activity using two new recombinant proteins as antigenic targets, and to evaluate these EIA with the aid of statistical methods.

Methods: Fragments of glycoprotein G (gG-2), comprising residues 525 to 578aa of herpes simplex virus type 2 (HSV-2) and glycoprotein D of HSV-2 (gD-2(266-394aa)), were expressed in the *E. coli* as GST fusion proteins to develop an assay for the detection and HSV-2 type-specific antibodies.

Results: A new enzyme immunoassay for the detection of IgG anti-HSV-2 (IgG-EIA) in sera was developed using two new recombinant proteins. The IgG-EIA was evaluated using serum specimens obtained from patients with culture-proven HSV-2 infection (CP) (n = 13) and from normal blood donors (BD) (n = 629). All specimens were additionally tested for IgG anti-HSV-2 activity by two commercially available EIAs. This new IgG-EIA detected anti-HSV2 activity in all specimens from HSV2 infected patients. When BD were tested the overall concordance between these three assays varied between 39 and 63.6%, concordance between positive samples ranged from 18.4 to 46.7%. In the absence of a gold standard the accuracy of these EIAs was assessed by the computer program based on a maximum likelihood approach using a ‘latent class’ model. This analysis estimated the IgG-EIA sensitivity and specificity to be within the range 98–100% and 95–100%, respectively.

Conclusion: The results show that the new two proteins-based enzyme immunoassays may be useful tools for the detection of type-specific HSV-2 antibodies. However, if only one assay is performed, careful interpretation of the results is indicated and for determination of the definitive HSV-2 serostatus, statistical models may be necessary.

P1075

Antigenic property of different sequence variants of the hepatitis B surface antigen wild types and vaccine escape mutants

T. Ulanova, J. Yokosawa, A. Burkov, V. Puzyrev, A. Obriadina
Nizhniy Novgorod, RUS; Atlanta, USA

Objectives: The purpose of this study was to determinate and evaluate of sequence heterogeneity on antigenic properties of hepatitis B surface antigen (HBsAg).

Methods: A number of recombinant HBsAg wild subtypes adw2, adw4, ayw1, ayw2, adr as well as ‘vaccine escape mutants’ adw2 T126S, Q129R, Q129L, T143K, Q145R and ayw1 Q145A were synthesised, purified and tested by enzyme immunoassay with a panel of 43 commercial available monoclonal antibodies specific for different determinants of HBsAg.

Results: Recombinant proteins were tested at the same concentration 1 ng/µL. All recombinant HBsAg proteins were immunoreactive and demonstrated very different level of reactivity. Among wild subtypes mostly immunoreactive with this panel was recombinant HBsAg subtype adw2. This protein has been detected by 90.6% of used MABs. Average S/Co was 43.7. Recombinant HBsAg subtype adw4 was least immunoreactive (55.8% MABs and average S/Co 3.7). Point mutations affected very different on HBsAg antigenic properties. The immunoreactivity of ‘vaccine escape mutants’ strains with MABs panel varied from 88.4 to 34.8%.

Conclusion: These data suggest that HBsAg sequence heterogeneity has a significant effect on the antigenic properties of this antigen. Diagnostic test development requires careful selection of MABs as diagnostic reagents. Recombinant HBsAg point mutants, especially ‘vaccine escape mutants’, may be used efficiently for evaluation of sensitivity commercial and ‘in house’ EIA for HBsAg detection.