ELISA is still the most sensitive diagnostic protocol for infectious toxoplasmosis. Comparison with PCR diagnosis.

D. P. Mohapatra and S. Singh
Department of Laboratory Medicine, All India Institute of Medical Sciences, New Delhi 110 029, INDIA

Toxoplasmosis is an emerging infectious disease caused by the intracellular protozoan parasite Toxoplasma gondii. The disease has got a varying epidemiology from ethnic region to region. Moreover it is also regarded as one of the fatal opportunistic infections in immunocompromised hosts. Another important feature of this is that, this parasite exhibit a phenomenal antigenic variation from ethnic region to region.

Thus the diagnostic value of commercially available ELISA based kits are limited to the country or region of production depending upon the immunodominancy of the antigens. With the development of PCR based diagnostic protocols it has been mostly stated that it can diagnose the infection with greater sensitivity. Aiming at this we have analysed the diagnostic efficacy of PCR for the diagnosis of toxoplasmosis and compared the same with the ELISA based kits being developed in our laboratory using purified immunodominant antigens of the parasite. A total of 500 serum samples were analysed and from the analysis of data it has been found that PCR is highly insensitive in comparison to developed ELISA kits for the accurate diagnosis of toxoplasmosis. This proves that ELISA is still the best diagnostic protocol for the diagnosis of toxoplasmosis.

Human-specific Cell Lysis by Intermedilysin, an Exotoxin of Streptococcus intermedius.

H. Nagamune1,2, O. Ohnishi1, K. Hatton1, K. Okhura1, T. Goto1, H. Tsuge1, K. Hirota1, N. Katunuma1, Y. Miyake1, T. Maeda1, H. Kourai1

Introduction: Intermedilysin (ILY) is a human-specific cytolsin secreted from Streptococcus intermedius and is a related toxin to the cholesterol-binding cytolsins (CBCs), but ILY shows different characteristics from CBC such as a low affinity to cholesterol (CHL). In the present study, we performed the molecular modeling of ILY and CBCs to compare the membrane binding domain (domain 4) including the 11 mer region which is necessary for the membrane binding activity.

Methods: Molecular modeling of toxins was performed using InsightII-Discover with Homology module based on the crystallostructure of a CBC, perfringolysin O (PFO), then the structure was minimized. Evaluation of the models was carried out using PROCHECK and X-PLOR. Structure of 11 mer region of toxins was analyzed using MOPAC.

Results and Discussion: ILY was shown to possess the stereocomplementary surface within the molecule and seemed to form a ring cluster of 45 molecules or so by molecular stacking manner in the cell membrane. The 11 mer region will be faced to the inner side of the ring. Because of the electron resonance among the three Trp residues within the 11 mer region of CBC, a strong dipole moment is formed and the moment allows CBC to bind to CHL smoothly. On the other hand, because of the substitution of Trp to Pro93 in the region of ILY, the moment is weakened and its direction is changed. These changes seems to cause the low affinity of ILY against CHL. We currently proceed the construction of various modules based on the domain 4 structure which was revealed in the present study.

Serum Soluble Heat-Shock Protein 60 Is Elevated in Subjects with Atherosclerosis in a General Population.

H. Perschinka, G. Schett, M. Mayr, S. Kiechl, Q. Xu, G. Wick
Rennweg 10, A-6020 Innsbruck, Austria

The work from our laboratory has proven that increased anti-HSP60 antibodies are associated with atherosclerosis and that HSP60 reactive T cells are present in atherosclerotic lesions. Recent studies from others demonstrated that heat shock protein 60 (HSP60) acts as extracellular agonists of cytokines directly activating endothelial cells and macrophages. To explore the possibility that HSP60 exists in circulation where it can exert its functions, we performed an epidemiological study in a free-living population in Northern Italy. A total of 826 subjects aged 45-74 years was recruited and subjected to determination of serum soluble HSP60 (sHSP60), anti-E. coli LPS, anti-HSP60, anti-C. pneumoniae, and anti-H. pylor antibodies. Carotid atherosclerotic lesions were sonographically assessed, and other risk factors, including age, sex, body mass index, LDL, HDL, apoB, apoA, Lp(a), leukocyte number, chronic infections, hypertension, smoking, alcohol consumption and diabetes mellitus, were also evaluated. Our data show that sHSP60 were significantly elevated in subjects with carotid atherosclerosis. Multiple logistic regression analysis documented these associations to be independent of age, sex and other risk factors. Thus, our data provide the first evidence of a strong correlation between sHSP60 and atherosclerosis, suggesting that sHSP60 may play important roles in activating vascular cells and immune system in the development of atherosclerosis.

This work was supported by grant P-12213-MED from the Austrian Science Fund. H.P. is a recipient of the PhD fellowship from the Austrian Academy of Sciences.

© 2000 Biochemical Society