ABSTRACT

Bacterial Immunoglobulin (Ig)-Receptors and IgY Technology

An Assessment of their Potential and Future Perspectives

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Immunoglobulin (Ig) receptors are widely distributed among species of bacteria as mechanisms of evasion from the host's immune system response. However, once purified or constructed by genetic engineering procedures, they can be widely used for antibody purification immunodiagnostic or therapeutic purposes. This thesis describes for the first time, the binding of some mammalian and avian immunoglobulins with the bacterial Ig receptors: Staphylococcal protein A (SpA), Streptococcal protein G (SpG) and Peptostreptococcal protein L (SpL) and a recombinant construction Protein LA (SpLA). Various techniques, with increased ranges of sensitivity were used to determine the reactivity of bacterial Ig receptors with Ig molecules. Bacterial Ig receptors did not react with a lectin protein preparation extracted from tissue fluids of the mussel *Isognomon alatus*. SpA or SpG were successfully used as antiglobulins for the detection of specific antibodies against human red blood cells (anti-RBC). The anti-rbc detection was performed in an agglutination assay, which used poly-ethylene-glycol as agglutination enhancer. Since chimeric recombinant Ig-receptors have shown a wider spectrum of affinity to immunoglobulins, chimeras were developed, by chemical coupling of these bacterial proteins, with unlabeled and antibody labelled enzymes, generating Ig-binding reagents with higher Ig-reactivity than those commercially available and / or described. A potentially useful and new chimeric conjugate was horseradish peroxidase (HRP) labelled with SpLA, SpL, SpG and HRP labelled rabbit anti-IgY antibody (SpLAG-anti-IgY-HRP). It bound to several avian and mammalian immunoglobulins, overcoming the limitations of commercially available conjugates, which preferentially bind to antibodies from mammals. This conjugate was applied in an indirect enzyme-linked immunosorbent assay (ELISA) for detection of antibodies against HIV glycoprotein-120 peptide raised in chickens and cats. These antibodies were obtained as part of preliminary experiments, which demonstrated that anti-gp-120 antibodies are present in the eggs of chickens immunized with gp-120 peptide. Feeding of the eggs to cats resulted in the production of anti-anti-idiotypic gp-120 antibodies, which were detectable in cat serum. These results suggest that immunized eggs may be useful in the development of anti-idiotypic vaccines with the option for oral administration.

Keywords: Protein; Immunoglobulin; HIV; Coomb's test; ELISA; Vaccine.