Contribution of heavy chain genes on the binding property of a recombinant human IgG rheumatoid factor

呂思潔

Lee YC;Leu SJ;Hsieh MS;Huang IJ and Yang YY

摘要

Abstract

Previous reports have demonstrated that the heavy chain genes of monoclonal antibodies could determine its binding activity to the corresponding antigen when paired with any light chain and vice versa. Several IgG rheumatoid factors have been isolated in our laboratory using phage display technology. To further determine the influence of the heavy chain on antibody activity to human Fc portion, an antibody library was constructed in the present study by shuffling various heavy chain genes into a rheumatoid factor GG3 clone. After electroporation, the size of the antibody library was estimated to contain 2~90 匀 10 individual clones. DNA restriction analysis revealed that more than 70% of randomly selected clones contained both heavy and light chain inserts. A protein band with a molecular weight of 50 kd was identified using western blotting technique, suggesting that both heavy and light chain fragments were expressed and folded into a Fab heterodimer under non-reducing condition. When examined using an enzyme-linked immunosorbent assay, all the 10 Fab expressing clones showed weaker binding activity to human Fc fragment. Taken together, our results clearly showed that the replacement of heavy chain in the GG3 rheumatoid factor Fab antibody would reduce its Fc-binding activity