

Production of polyclonal anti- $\beta$ -adrenergic receptor antibody  
and it's effects on receptor ligand binding

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The analysis of membrane receptors for hormones and neurotransmitters has progressed considerably by pharmacological and biochemical means and more recently by the use of specific anti-receptor antibodies. A 14-mer peptide (from Phe102 to Leu115 of  $\beta$ 2-adrenergic receptor) was synthesized and this peptide was coupled to carrier protein Keyhole Limpet Hemocyanin(KLH) by glutaraldehyde method. A 0.5mg of KLH-coupled peptide was emulsified with equal volume of complete Freund's adjuvant and injected via popliteal lymph node to each of the three Newzealnd White rabbits. Booster injections were repeated at 4 weeks interval for three times with incomplete freund's adjuvants. One week after the final injection, serum was prepared from ear artery. Nonspecific immunoglobulins were removed by passing the serum through KLH-Sepharose 6B affinity matrix and further by incubation with bovine lung aceton powder. The titer of the antibody for synthetic peptide which was determined by enzyme linked immunosorbent assay(ELISA) was about 1/1,000. The antibody produced in this study revealed 67kDa protein band in the western blot of partially purified guinea pig lung  $\beta$ -adrenergic receptor preparation. The antibody inhibited  $\beta$ -adrenergic antagonist [3H]Dihydroalprenolol binding to soluble  $\beta$ -adrenergic receptor by 25% while control sera did not show any inhibitory effects. The result of this study suggests that the peptide sequence selected in this study may play some important roles in adrenergic receptor-ligand interaction.