ELICITATION AND CHARACTERIZATION OF MONOCLONAL ANTI-IDIOTYPIC ANTIBODIES REACTIVE WITH THE LIGAND BINDING SITES OF MONOCLONAL KININ ANTIBODIES

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Abstract

In an attempt to generate monoclonal antibodies reactive with B2 bradykinin receptors, monoclonal anti-idiotypic antibodies (mAB2s) were elicited against monoclonal kinin antibodies (mAB1s) previously shown to have a binding specificity similar to a B2 bradykinin receptor (Biochem Pharmacol 40:245-251, 1990). Two separate fusions were performed using spleen cells from mice immunized with one of two mAB1s. Five clones were identified from each fusion using an ELISA which tested for bradykinin inhibition of hybridoma tissue culture supernatant binding to a plate coated with mAB1 Fab. Cross-reactivities of the mAB2s with mAB1s were evaluated by examining the ability of four mAB1s with similar kinin binding specificities to inhibit mAB2 binding to a plate coated with mAB1 Fab. The ten mAB2s cross-reacted to varying degrees with the mAB1s. Five of the mAB2s reacted equally well with all four mAB1s. In addition, the abilities of these mAB2s to inhibit the binding of biotinylated-kallidin to the four mAB1s was tested. In each case the mAB2s inhibited biotinylated-kallidin binding. We therefore conclude that the interaction of the mAB2s with the mAB1s occurs with an idiotope in, or in some way related to, the ligand binding sites of the mAB1s. These results are consistent with the mAB2s mimicking the epitope of bradykinin that is recognized by these kinin antibodies. Therefore, these mAB2s may be

mAB2 β type "internal image" anti-idiotypic antibodies.

In order to confirm the "internal image" nature of the mAB2s, experiments were performed to demonstrate their biological activities on B2 bradykinin receptors in PC12 cells. The assay tested the abilities of the mAB2s to mimic, or to inhibit, bradykinin induced increases in cytosolic calcium. The mAB2s did not exhibit agonist or antagonist activities in this system. However, because there have been no reports of any antibodies reactive with BK receptors that either mimic or inhibit bradykinin's effects on PC12 cells, it is possible that this bioassay may not be suitable for the detection of such antibodies.

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