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Binding of cholera toxin B subunit to intestinal epithelial cells

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We have prepared ^{125}I-labeled cholera toxin B subunit (^{125}I-labeled CT-B, a specific activity of 98 Ci/mmol) and found that it binds to rat intestinal epithelial cell membranes, rat IEC-6 and human Caco-2 intestinal epithelial cells with high affinity. The binding of labeled protein was completely inhibited by unlabeled thymosin-α1 (TM-α1), interferon-α2 (IFN-α2), and the synthetic peptide LKEKK that corresponds to residues 16-20 in TM-α1 and 131-135 in IFN-α2, but was not inhibited by the synthetic peptide KKEKL with the inverted amino acid sequence. Thus, TM-α1, IFN-α2, and the peptide LKEKK bind with high affinity and specificity to the cholera toxin receptor on rat intestinal epithelial cell membranes, IEC-6, and Caco-2 cells. It was found that CT-B and the peptide: LKEKK at concentrations of 10-1000nM increased in a dose-dependent manner the nitric oxide (NO) production and the soluble guanylate cyclase (sGC) activity in IEC-6 and Caco-2 cells. Taking into account that NO acts as a primary activator of sGC, it can be assumed that the effect of CT-B and the peptide: LKEKK on the target cell is realized in the following way: increase in the iNOS expression → increase in the NO production → increase in the sGC activity → increase in intracellular levels of cGMP

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