

[Biochem. Inetr., 16, 647 (1988)]

**A Comparative  $^1\text{H}$  NMR Study of Mouse  $\alpha(1-53)$  and  $\beta(2-53)$  Epidermal Growth Factors.**

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The three-dimensional structure of the mouse epidermal growth factor (EGF) in solution was studied by comparison of the  $^1\text{H}$  NMR spectra of  $\alpha\text{EGF}$  (1-53) and  $\beta\text{EGF}$  (2-53, des-asparaginyll<sup>1</sup> form). Using pH dependence of chemical shifts and a two-dimensional difference spectrum, the effect of the N-terminal deletion was investigated based on the complete assignment of the proton resonances. The affected residues were all found to be located exactly in the triple-stranded,  $\beta$ -sheet core in the N-terminal domain of the EGF molecule. We have demonstrated here the usefulness of the 2D difference spectrum method for detecting structural changes in proteins.

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**Immunohistochemical Localization of Human Epidermal Growth Factor/ $\gamma$ -Urogastrone in Submandibular Glands and in Their Obstructive Lesions.**

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Immunohistochemically detectable human epidermal growth factor (hEGF)/ $\gamma$ -urogastrone ( $\gamma$ -UG) was demonstrated with the use of polyclonal anti-hEGF/ $\gamma$ -UG antiserum in normal human submandibular glands and in their obstructive lesions. hEGF/ $\gamma$ -UG was isolated and purified from human urine and tested for its biological specificity by radioreceptor assay, and prepared anti-hEGF Fab' fraction. Histochemical expression of hEGF/ $\gamma$ -UG was confined to the intercalated and striated duct cells, and staining intensities differed depending on the fixations used. PLP fixed frozen sections showed the highest staining for hEGF/ $\gamma$ -UG in ductal segments.

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**Evidence for the Expression of a Primitive Intestinal-like Alkaline Phosphatase in the Intestinal 407 Cell Line.**

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Intestinal-like alkaline phosphatase (I-ALP) was found to be expressed in the intestinal 407 cell line. This enzyme was identified by use of monoclonal antibodies specific for human placental and intestinal alkaline phosphatases separately. Purification of this isozyme by use of two different monoclonal antibody immunoaffinity chromatographies demonstrates a single protein band on SDS-PAGE indicating that this enzyme is formed as a homodimer. Our data suggest that the I-ALP in the intestinal 407 cell line displays properties intermediate of the intestinal and placental isozymes which may reflect the existence and reexpression of a new primitive isozyme.