1.1

APPLYING THE TECHNIQUES OF MOLECULAR BIOLOGY TO PHYSIOLOGICAL RESEARCH. S. Chien. Univ. of California, San Diego, La Jolla, CA

Most physiological functions are carried out by proteins, which are genetically coded as DNA in cell nuclei. The transcription of DNA to RNA and the translation of RNA to proteins provide the flow of genetic information, and the regulation of these processes lead to the control of gene expression and physiological functions. Molecular cloning techniques have led to the establishment of the amino acid sequence, the prediction of the secondary structure, and the correlation of molecular structure with physiological function of the protein. Comparisons of proteins with structural homologies have generated new insights into their tissue specificity, developmental differentiation, and evolutional changes. Techniques of molecular manipulations such as site-specific mutagenesis and transgenic mice have led to the identification of known functions with specific sites on a given protein molecule. The applications of these molecular biological techniques have also helped to elucidate the molecular basis of pathophysiological functions in disease states.

REFERENCES:

1.

2.

3.

COMPARATIVE INTESTINAL NUTRIENT TRANSPORT

5.1

VERTEBRATE INTESTINE APICAL MEMBRANE MECHANISMS OF ORGANIC SOLUTE TRANSPORT. Bruce R. Stevens, Dept. of Physiology, Coll. of Med., Univ. of Florida, Gainesville, FL USA 32610.

The intestinal epithelium apical membrane serves as the major interface between the environment and blood, controlling the net absorption of organic nutrients. Mechanisms of amino acid and sugar transport include un-regulated simple passive diffusion and regulated transporter (carrier) proteins. Transporter activities have been shown [1] to be regulated locally in enterocyte membranes at several levels: (*a*) ion and substrate kinetic activation and binding order, (*b*) membrane electrical potential modulation, (*c*) interactions among transporter protein subunits, and (*d*) intracellular phosphorylation events associated with enterocyte differentiation, as recently demonstrated in my laboratory. Organic nutrient accumulation occurs in enterocytes by energetic coupling to the electrochemical gradient associated with 2 Na^+ ions [2]. The use of high-energy electron radiation inactivation experiments and kinetic modeling has revealed that the sodium-activated glucose transporter, and also the proline transporter, each behaves *in situ* in the apical membrane as a tetramer of four subunits [3]. Inasmuch as intestinal transporter characteristics are shared among the vertebrate species studied, there may exist a universal vertebrate intestinal apical membrane mechanism of ion-activated organic solute transport.

REFERENCES:

1. B.R. Stevens and E.M. Wright
Kinetics of the intestinal brush border proline (Imino) carrier.
J. Biol. Chem.
262 (1987):6546-6551.

2. G.A. Gerencser and B.R. Stevens
Energetics of sodium-coupled active transport mechanisms in invertebrate epithelia.
Am. J. Physiol.
257 (1989):R461-R472.

3. B.R. Stevens, A. Fernandez, B.Hirayama, E.Wright, E.Kempner
Intestinal brush border membrane Na+/glucose cotransporter functions in situ as a homotetramer.
Proc. Natl. Acad. Sci. USA
87 (1990):1456-1460.

5.2

ARE INVERTEBRATE GUT DIVERTICULA NUTRIENT ABSORPTIVE ORGANS?
G. A. Ahearn, G. A. Gerencser, M. Thamotharan, & R. D. Behnke
Dept. of Zoology, Univ. of Hawaii, Honolulu, Hi 96822 and Dept. of Physiol., Univ. of Florida, Gainesville, Fl 32610.

Physiological mechanisms of gastrointestinal nutrient absorption among invertebrates remain severely underinvestigated. We briefly review some of the biochemical and biophysical forces affecting nutrient transfer across epithelial cells and membranes of vertebrate and invertebrate absorptive organs. Next, we present some of the described processes for sugar and amino acid transport in the tubular portion of the gastrointestinal tracts of three major invertebrate groups: echinoderms, molluscs, and arthropods. We then discuss in detail recent nutrient transport studies using purified epithelial brush border membrane vesicles of two invertebrate gut diverticula, the crustacean hepatopancreas and the starfish pyloric caecum. Lastly, transepithelial nutrient transport studies of crustacean hepatopancreatic epithelial cell monolayers grown in primary culture and mounted in flux chambers are used to demonstrate the applicability of these methods to structurally complex invertebrate organs as well as to characterize the cellular mechanisms responsible for movements of solutes across these cell layers. It is concluded that invertebrate gut diverticula may make a significant contribution to total nutrient absorption. Supported by NSF grant no. DCB89-03614.

REFERENCES:

1. G. A. Ahearn (1988)
Nutrient transport by the invertebrate gut.
IN: Advances in comparative and environmental physiology (R. Gilles, Ed.)
Vol. 2, pp. 91-172.

2. G.A. Gerencser and B.R. Stevens (1989)
Energetics of sodium-coupled active transport mechanisms in invertebrate epithelia. Am. J. Physiol. 257: R461-R472.

3. R.D.Behnke, R.K.Wong, S.M.Huse, S.J.Reshkin, and G.A.Ahearn (1990)
Proline transport by brush border membrane vesicles of lobster antennal glands.
American Journal of Physiology 258: F311-F320.