ascitic fluid. Production of rabbit anti murine IgE, rendered specific for the ε-chain has allowed us to develop a radioimmunoassay for the quantitation of murine IgE, and to study the specificity of the hybridoma product. Furthermore, we are at present investigating the morphology and localisation of mast cells from Balb/c mice bearing the hybridoma and fed with cow's milk or β-lactoglobulin.

Structure of phosphoribosylanthranilate isomerase: Indoleglycerolphosphate synthase (PRAI:IGPS) at 5 Å resolution

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PRAI:IGPS from *Escherichia coli* is one of the few simple representatives of multifunctional enzymes. It is a monomer which catalyzes the 2 metabolic steps of tryptophan biosynthesis preceding the tryptophan synthase reaction. Biochemical and genetic studies showed that the N-terminal portion of the polypeptide chain is responsible for IGP-synthase activity while the C-terminal part comprises the active site of PRA-isomerase. - The enzyme crystallizes in space group $P4_1$ with $a=b=105$ Å and $c=67.9$ Å. 3 heavy atom derivatives were found, their respective Patterson maps interpreted and heavy atom parameters refined. A 5 Å electron density map was calculated. In the map one can distinguish separate molecules of size $70\times50\times40$ Å in which the 2 domains can be recognized. Crystal data of the enzyme in the presence of a substrate analogue for both reactions should reveal the locations of the 2 active sites.

In vitro effects of citrate on calcium handling by isolated rat liver mitochondria

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Energized mitochondria respond to small amounts of EDTA or $CaCl_2$ to the suspension medium by a net release resp. uptake of Ca^{2+}, so as to maintain the Ca^{2+} concentration of the extramitochondrial fluid at a steady value (set point). As measured with a pCa electrode, the set point established by mitochondria incubated in a KCl-medium with succinate + rotenone (pH 7.4, 25 °C) was decreased from 1.18 ($\pm$0.05) to 0.75 ($\pm$0.02) ng-ion Ca^{2+}/ml by admixing 1.2 mM citrate to the medium ($n=4$). With increasing amounts of added $CaCl_2$, on the other hand, the capacity of mitochondria to accumulate and retain Ca^{2+}, which is limited by the occurrence of Ca^{2+}-induced uncoupling with collapse of the Ca^{2+} gradient across the inner membrane, was markedly increased by citrate. Strong effects of citrate were also observed with Pi present (1-2 mM). Citrate effects on both the set point and the Ca^{2+}-retention capacity resemble known effects of ATP.

The mechanism of action of cholera toxin: inhibition by blockers of protein synthesis

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Pretreatment of macrophages with cycloheximide blocked the intracellular accumulation of cAMP after exposure to cholera toxin (CT) in a time- and dose-dependent manner which paralleled the inhibition of protein synthesis. The response to isoproterenol, on the other hand, was not affected, nor did the number of receptors for CT decrease. In broken cells, however, adenylate cyclase could be stimulated by the A_1 fragment of CT regardless of pretreatment. - Presumably, the A protomer of CT has to enter the cell in order to be reduced and release the active A_1 fragment. Our experiments showed that the generation of A_1 after incubation of cells with ^{125}I-CT was inhibited in pretreated cells. Furthermore, ^{125}I-CT bound to treated cells was not degraded. - Since both the receptor for CT and the adenylate cyclase were intact, we concluded that the translocation of the A subunit of CT through the cell membrane is inhibited by pretreatment with blockers of protein synthesis and that a cell membrane protein is necessary for the translocation to take place.

Ultrastructural localization of Kunitz trypsin inhibitor in soybeans using gold granules labeled with protein A

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The Kunitz trypsin inhibitor (SBTI) is one of the best characterized inhibitor of serine proteases but its cellular location is unknown and its physiological role in soybeans is obscure. SBTI has now been localized at the ultrastructural level on thin sections of *Glycine max* (Soybean) var. Maple Arrow by the gold method (Horisberger and Rosset, J. Histochem. Cytochem. *25*, 295, 1977) using protein A. Thin sections were incubated with an anti-SBTI immunoglobulin fraction and then exposed to gold granules (12 nm in size) labeled with protein A. SBTI was found localized in the cell wall and in most of the protein bodies but not in the cytoplasm. However, in the embryonic axis, marking was also associated with the cytoplasm. Cotyledons after 4 days germination were also examined. The results were similar. These observations were corroborated by immunofluorescence staining. Numerous controls indicated that the methods were specific.

Spatial structures of snake venom toxins by NMR: Sequential ^{1}H NMR assignments in cardiotoxin $V^{II}2$ from *Naja mossambica mossambica*

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Cardiotoxins from snake venoms produce a variety of toxic effects, e.g. depolarization of membranes, hemolysis and synergistic action with phospholipase A_2. Relatively little is known about the mechanisms of action of cardiotoxins and additional insight might come from a knowledge of the spatial structures. We have started work on the determination of the spatial structure of cardiotoxin $V^{II}2$ from *N. moss. moss.* (obtained from Prof. M. Lazdunski), following a strategy based on the use of 2-dimensional ^{1}H NMR at 500 MHz as was recently described in detail (Wüthrich, Wider, Wagner and Braun, J. molec. Biol., in press). Nearly complete ^{1}H NMR assignments will be presented and the gross features of the conformation of toxin $V^{II}2$ will be discussed.

Replicases appear to be highly conserved enzymes

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DNA polymerases responsible for chromosomal DNA replication of the bacterium *Escherichia coli*, the fungus *Ustilago maydis*, the fly *Drosophila melanogaster*, the chicken and