In some clinical and pharmacodynamic conditions, a decrease of kininogen level in plasma can be observed.

The general and circulatory picture during direct release of kinins by trypsin and kallikrein in man has been studied and compared with the clinical pharmacology of some hypotensive peptides such as synthetic bradykinin, kallidin, eledoisin, and physalaemin.[5, 6] The effects of some antifibrinolytic compounds such as Trasylol and ε-aminocaproic acid will be shown.

Some therapeutical trials with synthetic and natural peptides (plasma kinins) will be discussed.

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39. Contribution to the Sedative Action of Substance P. P. STERN (*Dept. of Pharmacology, Medical Faculty, Univ. of Sarajevo, Yugoslavia*).

We have already indicated that substance P (SP) acts as a sedative. In these experiments fighting male mice (fighting induced by keeping mice isolated from each other for 21 days) became tranquil 15 to 30 minutes after the application of SP. This effect has been examined with SP of purity 6 U, 75 U, and 300 U/mg. Groups of mice received from these fractions 5,000 U/kg injected in 0·2 cc/g, i.p. Substance P destroyed with chymotrypsin had no effect on the control group. Tranquil mice became belligerent again if given 25 mg demethylimpiramin i.p. We take this as additional evidence for the central sedative action of substance P. Since in this test impure (6 U) and relatively pure (300 U) SP had the same effect, we believe that the test could be applied for example during the purification of SP. Substance P is a polypeptide that not only contracts the ileum of the guinea pig but has a sedative action as well. We have already shown that the other central effects of pure SP do not parallel the effect on intestine; for example, purified SP (270 U/mg) no longer has antistrychnine-like action. On the other hand, Krivoy has showed that very pure SP (10,000 U/mg) still retains neurotropic action.

40. The Search for Peptides with Specific Antibradykinin Activity. JOHN MORROW STEWART and D. W. WOOLLEY (*The Rockefeller Institute, New York, N.Y., U.S.A.*).

Structure–activity relationships in the bradykinin molecule were studied with the aid of over forty new analogs of bradykinin. These analogs (octa-, nona-, and decapeptides) were synthesized and tested on smooth muscles for bradykinin potency and for their ability to act as antagonists of bradykinin. The compounds were made by slight modifications of the Merrifield method of solid-phase peptide synthesis, and were obtained analytically pure. Single replacements of one amino acid residue by some other were ineffective for formation of antimetabolites, as were changes in the optical configuration of the amino acids. Greatest antibradykinin activity was found among analogs in which both phenylalanines had been replaced by O-methyl tyrosine. These analogs showed antibradykinin activity on rat uterus at low concentrations, while at much higher concentrations bradykinin-like action was observed. The antibradykinin activity fluctuated widely from animal to animal. Various structural alterations, especially in the serine position and in the carboxyl end, were explored in an attempt to obliterate the bradykinin-like action with retention of the antibradykinin effect. No compound has yet been found which showed high potency as an antagonist but had no bradykinin-like activity.

41. An Apparatus to Simplify the Bioassay of Vasoactive Substances. E. STÜRMER and H. WOHLFART (*Biological and Medical Research Division, Sandoz Ltd., Basle, Switzerland*).

An automatic apparatus has been constructed for assaying vasoactive substances by comparing their effect on the blood pressure with that of standard substances. Standard or test solutions are automatically injected by the intravenous route with the aid of two infusion pumps, operated intermittently. Dosage and sequence of the injections follow a prearranged program. The blood pressure changes in response to the injections are recorded by means of a strain gauge and transformed to impulses. The reaction maxima are printed out and the results are evaluated by computer.

This procedure saves manpower and time and makes for greater accuracy.

42. Purification and Some Enzymatic Properties of Bradykinin-Releasing and -Destroying Enzymes in Snake Venoms. TOMOJI SUZUKI, SADAAKI IWANAGA and TADASHI SATO (*Institute for Protein Research, Osaka Univ., Osaka, Japan*).

During purification of the bradykinin-releasing enzyme of the venom of *Agkistrodon halys blom-*

hoffii by DEAE-cellulose chromatography, a proteinase with bradykinin-destroying activity was separated from the bradykinin-releasing enzyme and was found to be identical with proteinase b. Proteinase b, one of the three proteinases with hermorrhagic activity in the venom, hydrolyzed the glycyl-phenylalanyl-R linkage of the bradykinin. When the partially purified bradykinin-releasing enzyme, which had no detectable proteolytic activity, was incubated with physicochemically pure bradykininogen, the bradykinin liberated was destroyed very slowly. The bradykinin-destroying enzyme which was found in the partially purified bradykinin-releasing enzyme preparation was different from proteinase b and hydrolyzed the phenylalanyl–seryl–R linkage of the bradykinin.

The arginine ester hydrolase in the venom of *A. halys blomhoffii* was separated into three entities: bradykinin-releasing, clotting, and capillary permeability-increasing enzymes. The content of arginine ester hydrolytic activity of the purified bradykinin-releasing enzyme was unexpectedly low and was less than 5% of the total activity of the venom. The enzymatic properties of the purified bradykinin-releasing enzyme were similar to those of pancreatic kallikrein. Trasylol inhibited only the activity of the bradykinin-releasing enzyme. The enzymatic mechanisms of liberation of bradykinin from bradykininogen will also be presented.

43. A Possible Role for the Plasma Kinins in Pancreatitis and Shock. ALAN P. THAL (*Robert S. Marx Laboratories of the Dept. of Surgery, Wayne State Univ. School of Medicine, Detroit, Mich., U.S.A.*).

The initial symptoms in human acute hemorrhagic pancreatitis are intense pain, accumulation of edema fluid in and around the pancreas, and accumulation of peritoneal fluid. The local features of edema and pain appear to result from local kinin release. The hypotension which follows appears to be largely the result of fluid loss, but in addition toxic substances emanating from the pancreas may be important. The present study will deal with the measurement of plasma kinins, their precursors, and kininases in acute experimental pancreatitis. Evidence for a proteolytic process will also be supported by measurements of plasma TAME esterase activity in both the experimental and clinical situation.

In other forms of shock as well, evidence will be presented in the experimental animal that endotoxin shock is associated with initiation of the proteolytic process. It will also be shown that endotoxin encourages intracellular and intravascular proteolysis with resultant liberation of plasma kinins. A possible role for these agents in shock and pancreatitis will be discussed. (Supported by Grant AM 06385-02 from the National Institute of Health, the Michigan Heart Association, and the Receiving Hospital Research Corporation.)

44. Kininogenases, Kininases, and Their Inhibitors. I. TRAUTSCHOLD, H. FRITZ and E. WERLE (*Klinisch-Chemisches Institut an der Chirurgischen Klinik der Universität, Munich, Germany*).

Some properties of a pure pancreatic kallikrein preparation will be presented, together with its amino acid composition and kinetic data of the proteolytic and esterolytic action of the enzyme.

Kallikreinogen was prepared from pig serum by a new method, and the possibilities of its activation were studied. New aspects of the already known kininogenase inhibitors such as the trypsin–kallikrein inhibitor from bovine tissues or the submandibular inhibitor from dogs will be discussed. Pancreas from mammals contains a specific trypsin inhibitor which is different from the well-known beef pancreatic trypsin inhibitor of Kunitz. This new inhibitor was isolated and its properties studied. The characteristics of the serum kallikrein inhibitor were also investigated. There is only little evidence for a physiological role of the naturally occurring kininogenase inhibitors in the control of the liberation of kinin.

Under physiological conditions the kinin action is controlled by the kininase system, which is much more active in nearly all organs than in blood. The enzymes that take part in the kinin destruction may be differentiated by selective inhibitors.

45. Effects of Bradykinin on the Specialized Ventricular Conducting System of Dog Heart. R. L. VICK, W. KRIVOY and D. C. KROEGER (*Depts. of Physiology and Pharmacology, Baylor Univ. College of Medicine, and Dept. of Pharmacology, Univ. of Texas Dental Branch, Houston, Texas, U.S.A.*).

Bradykinin (BK), 0·1 to 1·0 μg/ml, has been shown to decrease the action potential duration of the Purkinje, or specialized ventricular conducting system (SCS), of dog heart, with relatively little effect on the action potentials of myocardial cells (*Fed. Proc.* **24**, 137, 1965). By means of KCl-filled micropipettes and direct-current recording techniques, additional effects of the same concentrations of BK have been recorded. In SCS tissue undergoing repetitive stimulation, the maximal diastolic potential and the membrane potential prior to excitation are reduced, and the average conduction velocity is decreased. In spontaneously beating SCS tissue, BK decreases the frequency and stabilizes the membrane potential at a level between maximal diastolic potential and threshold. All these effects are more