the subjects experienced pain at the point of 'partial rupture' of the ligament. In 5 subjects a small amount of anæsthetic was then injected at this point. Following the injection, renewed attempted abduction did not produce any muscular responses (Fig. 1 *B*). It seems likely that the reflex motor effects obtained prior to anæsthetization were due to the activation of pain receptors.

Detailed accounts of these experiments on cats and man will be published elsewhere.

BERTIL STENER

Department of Physiology,
Department of Clinical Neurophysiology,
University of Göteborg,
Göteborg.

July 10

[1] Partridge, E. J., *J. Anat.*, **58**, 346 (1924).
[2] Gardner, E., *Amer. J. Physiol.*, **161**, 133 (1950).
[3] McCouch, I. D., Deering, I. D., and Ling, T. H., *J. Neurophysiol.*, **14**, 191 (1951).
[4] Skoglund, S., *Acta Physiol. Scand.*, **36**, Suppl. 124 (1956).
[5] Cohen, L. A., and Cohen, M. L., *Amer. J. Physiol.*, **184**, 433 (1956).

Peptides with Oxytocic and Pressor Activity obtained from Acidified Rat Serum

INCUBATION at 38° produces an increase in oxytocic activity in the acidified serum of normal rats (Croxatto, H. B., and Croxatto, H., unpublished work). Earlier studies have revealed that pepsitensin-like substances can be obtained from hypertensinogen under similar conditions[1]. This communication describes experiments carried out to determine some of the properties of the substance (or substances) extracted from acidified and incubated rat serum.

Blood was drawn without anticoagulant by aortic puncture from rats anæsthetized with ether or bromethol. It was centrifuged at 0° and the serum was acidified to *p*H 3·8 with hydrochloric acid. The period of incubation at 38° varied from 1 to 30 hr. The proteins were then precipitated with nine volumes of absolute ethanol. The supernatant was evaporated at low temperature and the residue suspended in distilled water for biological assay. Further purification was achieved by removing the lipids with petroleum ether. The aqueous solution remaining was again evaporated and the residue dissolved in glacial acetic acid. Ten volumes of a mixture of petroleum ether and absolute ethanol were then added. The residue was dissolved in distilled water to half the original volume of the serum. The solution was tested on the isolated rat uterus suspended in Tyrode solution containing atropine and dibenamine. Extracts from 1 ml. rat serum showed an oxytocic effect similar to that of 10–50 mU. oxytocin and contracted the isolated rat ileum like 0·5–1 U. of pepsitensin or hypertensin I.

The solubility characteristics and the effect of proteolytic enzymes on the unknown oxytocic substance suggest a polypeptide structure. It is rapidly destroyed by chymotrypsin and carboxypeptidase and partially destroyed by pepsin and trypsin (30–50 per cent). Under similar conditions, trypsin completely inactivates vasopressin and hypertensin I and II. That the substance is different from oxytocin is demonstrated by its lack of milk ejection activity in the lactating rabbit, its resistance to the action of sodium thioglycollate and by its different reaction to carboxypeptidase, pepsin and trypsin.

The peptide differs also from the neurohypophysial hormones in its vasopressor effects: whereas it produced a striking increase in the blood pressure of rats nephrectomized 12–24 hr. previously, no effect or a slight fall was seen in intact anæsthetized rats.

The progressive liberation of this substance is probably due to an enzymatic action at 38–40° at an optimum *p*H of approximately 3·8. Incubation for 24 hr. liberates practically all oxytocic and vasopressor activity from acidified rat serum. Little or no active substance was obtained when serum was heated for 30 min. at 58°C. before acidification and incubation. Incubation for a few hours at 38° at normal *p*H was also ineffective. The enzyme active in acidified serum is not renin, since serum from nephrectomized animals yields higher activity. A gastric origin of the enzyme can also be excluded because similar quantities of active peptide can be obtained from the serum of gastrectomized animals. The pressor and oxytocic activities are rapidly destroyed when purified samples of the peptide are incubated at neutral *p*H in the presence of small volumes of blood serum.

The polypeptide liberated by the simple procedure of acidification and incubation is different from pepsitensin and hypertensin I and II, but judging from its action on the rat uterus and guinea pig ileum it resembles bradykinin[2]. However, bradykinin has a strong vasodepressor effect in several mammalian species and is less active on the hen's rectal cæcum. Further investigations will be necessary to ascertain whether the peptide liberated under the above described conditions is related to the oxytocic substance described by Hawker[3] and to peptides obtained by the action of pepsin on blood serum. The substance is now being purified to obtain information on the possibility that it has a physiological function and plays a part in the pathogenesis of renoprival hypertension.

A full account of these experiments will be published elsewhere.

We wish to thank the Gildemeister Foundation for the financial aid given us in the work here reported. We are indebted to Dr. L. T. Skeggs for his generous supply of natural hypertensin I and synthetic hypertensin II.

H. CROXATTO
T. PEREDA
R. MELLADA

Laboratorio de Fisiologia,
Universidad Catolica de Chile.

[1] Alonso, O., Croxatto, R., and Croxatto, H., *Proc. Soc. Exp. Biol. Med.*, **52**, 61 (1943).
[2] Rocha e Silva, N., 'Polypeptides which stimulate Plain Muscle', edit. by Gaddum, J. H. (E. & S. Livingstone, Ltd., Edinburgh, 1955).
[3] Hawker, R. W., and Robertson, P. A., *Endocrin.*, **63**, 242 (1958).

VIROLOGY

Antiviral Chemotherapeutic Activity of Isatin β-thiosemicarbazone in Mice infected with Rabbit-pox Virus

ISATIN β-thiosemicarbazone has been found to have a very powerful chemotherapeutic effect in mice infected intracerebrally with the *IHD* strain of neurotropic vaccinia virus[1], and will give complete protection against death after infection with doses of virus as high as 100,000 *LD*50[2]. The compound is inactive