Investigation on the Mechanism of Prostaglandins Release

The release of Prostaglandins has been investigated by many investigators who found that PGs are released from the spleen, kidney, stomach, etc, by different stimuli. We investigated the release of PGs by expanding the extracellular space and/or the intravascular volume of anaesthetized rats.

Materials and method. The experimental animals were male Wistar-type rats weighing 500–550 g. They were permitted free access to food and water up to the time of the actual experiments. The animals were anaesthetized with sodium ethylmethylbutyl barbiturate (Mebubarbital Abbott®) 30 mg/kg i.p., then their tracheae were catheterized, and they received 600 U/kg of heparin in 1–3 ml/kg of saline.

a) *Blood-bathed organ technique:* Blood was withdrawn from the right femoral artery (RFA) of an anaesthetized rat weighing 500–550 g, at the rate of 5.0 ml/min[1]. The blood was analyzed for its hormonal content by cascading over the assay organs, arranged in series. There were 3 assay organs, rat colon (RC) chick rectum (CR) and rat stomach strip (RSS), continuously superfused according to procedure described by VANE[2,3] and GADDUM[4]. The blood was collected into a reservoir and returned at the same rate to the animal, via a catheter inserted into the right femoral vein (RFV) by the same roller pump (Figure 1). Arterial blood pressure (ABP) was recorded through a polyethylene catheter (0.03 inch ⌀) inserted into the left femoral artery (LFA) and connected to a strain gauge manometer (Physiograph E and M instrument). The extracellular and/or the intravascular space was expanded by i.v. infusion of 5–6 ml[5] saline and/or 3–5 ml[5] of blood or plasma, respectively, at a rate of 1–2 ml/min, through a catheter inserted into the left femoral vein (LFV).

b) *Cross-circulated rats:* The animals were connected for cross-circulation by polyethylene tubes leading from the right femoral artery (RFA) of the donor to the left femoral vein (LFV) of the recipient, and from the left femoral artery (LFA) of the recipient to the right femoral vein (RFV) of the donor. The rate of the blood flow was controlled by a roller pump at 3.0 ml/min. Arterial blood pressures were recorded through polyethylene catheters (0.03 inch. ⌀) inserted into the LFA of the donor and the RFA of the recipient, by a strain gauge manometer. The extracellular and/or the intravascular space of the donor was expanded by i.v. infusion of 5–6 ml of saline and/or 3–5[5] ml of blood or plasma, respectively, at a rate of 1–2 ml/min, through a catheter inserted into the LFV.

[1] Many animals did not support the rate of 5.0 ml/min, but when the rate was smaller than 4.0 ml/min, the contraction of the rat colon was almost imperceptible. For this reason many experiments took place by withdrawing blood from 2 animals simultaneously at the rate of 2.5 ml/min from each.

[2] J. R. VANE, Br. J. Pharmac. *35*, 209 (1969).

[3] S. H. FERREIRA and J. R. VANE, Nature, Lond. *216*, 868 (1967d).

[4] J. H. GADDUM, Pharmac. Rev. *11*, 241 (1959).

[5] We used the smallest quantities of saline or blood which could produce the release of Prostaglandin-like substances.

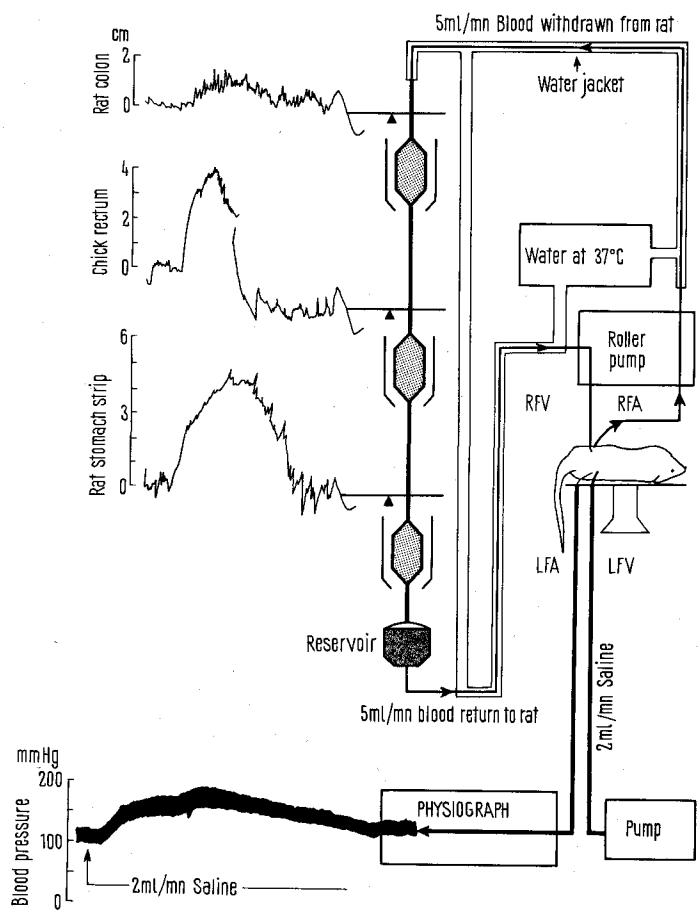

Fig. 1. Blood was withdrawn from the right femoral artery (RFA) of the anaesthetized rat, at the rate of 5 ml/min. The blood was analyzed for its hormonal content by cascading over the 3 assay organs, rat colon (RC), chick rectum (CR) and rat stomach strip (RSS) and was collected into a reservoir and returned at the same rate to the animal, via a catheter inserted into the right femoral vein (RFV) by the same roller pump. Blood pressure was recorded through a catheter inserted into the left femoral artery (LFA) and connected to a strain gauge manometer (physiograph). The extracellular space was expanded by i.v. infusion of 5–6 ml of saline at the rate of 2 ml/min via a catheter inserted into the left femoral vein (LFV).

Results and discussion. The identification of PGs in blood has been limited by the lack of sensitivity in available chemical methods. The blood-bathed organ technique which was used to detect Prostaglandin-like substances (PLS) in these experiments possesses the required sensivity for detection of nanogram quantities of PGs. GADDUM[4] considered the evidence provided by parallel pharmacological assay to be more important in the identification of a substance than that provided by same biochemical procedures.

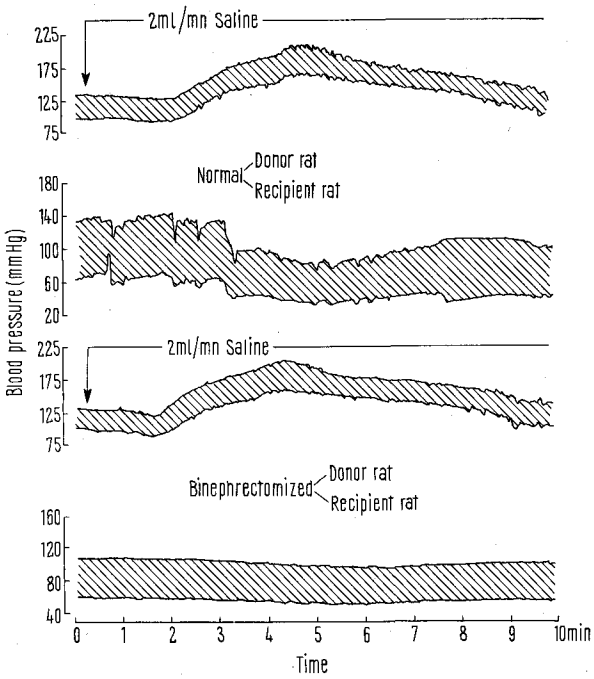

Fig. 2. Cross-circulation experiments. The changes of blood pressure (BP) of normal and binephrectomized rats. Diminution of the BP of the normal recipient rat, while the BP of the donor increased following expansion of the extracellular space. This phenomenon was almost imperceptible on binephrectomized rats.

In all the experiments, expansion of the extracellular and/or of the intravascular space produced the release of PLS which contracted the assay organs (Figure 1). No other known circulating hormones in physiological concentration will produce the same pattern of response as PGs in the 3 specially chosen assay organs[2,3]. The results of changes observed in the animals in cross-circulation show (Figure 2) that the arterial pressure (AP) of the donor was increased during the expansion, while the AP of the recipient diminished. The phenomenon could be repeated during all the experiments.

It is clear that a very active vasodepressor substance was released into the circulation of the donor during the expansion which was capable of diminishing the AP of the recipient. This phenomenon was almost imperceptible when the rats were binephrectomized. This observation suggests a close relationship between the vasodepressor factor and the kidney. Perhaps the kidney is the main organ which produces this vasodepressor factor.

Further investigation by chromatographic methods must take place for identification and separation of PGs which are released through this particular mechanism.

Résumé. Des substances qui semblent être des prostaglandines, détectées par des méthodes de bioassay, se libèrent dans la circulation des rats anesthesiés lors de l'expansion soit de l'espace extracellulaire, soit du volume intravasaculaire. Lors de l'expansion du donneur, en circulation-croisée, il se libère une substance capable de produire une baisse de la tension artérielle du receveur.

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Resistance to Flow Through the Pancreatic Duct by the Isolated Cat Sphincter of Oddi

In cat the principal pancreatic duct and the common bile duct share a common entrance into the duodenum (reviewed by HALLENBECK[1]). BOYDEN[2], in his study on muscle arrangement in the cat choledochoduodenal junction, reported that in the proximal part of the intramural sphincter of Oddi the pancreatic and the bile ducts are separated by a muscle septum, which according to him might selectively influence the bile duct. The ducts are then jointed unto the papillary orifice.

It was thought of interest to investigate the effect of various agents known to affect the sphincter on the resistance to flow through the isolated cat sphincter measured both as perfusion pressure through the common bile duct and as perfusion pressure through the pancreatic duct.

Method. 15 adult cats of both sexes fasted 24 h were used. They were anaesthetized with pentobarbitone sodium (Abbot) and bled. The detailed description of cat sphincter of Oddi by BOYDEN[2] was used as dissection guide. The distal pancreatic duct was dissected free from tissue and cannulated with polyethylene catheter (Clay-Adams PE 10). The catheter was gently moved through the sphincter of Oddi into duodenum. Similarly the distal common bile ducts was cannulated (Clay-Adams PE 50). Then the sphincter of Oddi was dissected free from duodenal tissue. The catheters were withdrawn and the perfusion catheters (PE 90) were ligated just before the entrance of the ducts into the sphincter. The preparation was mounted in an isolated organ bath as shown in Figure 1. Both ducts were perfused with Krebs solution at a constant rate, common bile duct 3 ml/h and pancreatic duct 1–3 ml/h (Perfusor B. Braun Melsungen 71100). Longitudinal isometric tension changes in the sphincter (Grass force displacement transducer FT 03 B) and the perfusion pressures (Statham pressure transducers P 23 AC) were recorded on a Grass Polygraph (7 Pl). The tension was initially adjusted to (0.1–0.4 g). The bath (100 ml) contained Krebs solution gassed with carbogen and maintained at 38°C. A small part of the perfusion catheters just distal to the perfusion pump was replaced by a rubber tubing so that drugs could be given in the perfusion fluid as well as in the solution bathing the sphincter preparation.

[1] G. A. HALLENBECK, in *Handbook of Physiology. Alimentary Canal* (Ed. W. HEIDEL; Williams and Wilkins Co., Baltimore 1967), vol. 2, p. 1007.

[2] E. A. BOYDEN, Surgery *41*, 773 (1957).