in pathological responses, such as local reactions to injuries, whereas prostaglandins may well be involved in normal function. For the prospects of their clinical use, this distinction may be important. Because kinins are probably involved in pathological responses, there seems little prospect of using them therapeutically, except perhaps for diagnosis or for increasing local blood flow. On the contrary, the antagonists of kinins may well be useful clinically and there is evidence suggesting that aspirin may act as an analgesic by antagonizing the release of an active substance by kinins. Because the roles of prostaglandins in the body are probably mainly concerned with normal function, the possibility of their clinical use looms larger. Later in this symposium we shall learn of some successful clinical trials.

REFERENCES
Berry P A (1966) PhD Thesis, London; p 91
Berry P A & Collier H O J (1964) *Brit. J. Pharmacol.* 23, 201
Bhoola K D, Collier H O J, Schachter M & Shorley P G (1962) *Brit. J. Pharmacol.* 19, 190
Clarke C A (1969) *Brit. med. J.* i, 256
Collier H O J
(1968) *Endeavour* 27, 14
(1969*a*) *Nature (Lond.)* 223, 35
(1969*b*) *Advanc. Pharmacol.* 7, 333
(1970) In: Proceedings of the International Symposium on Cardiovascular and Neural-Actions of Bradykinin and Related Drugs. Ed. N J Back, M Rocha e Silva & F Sicuteri. New York; p 461
Collier H O J, Dinneen L C, Johnson C A & Schneider C (1968) *Brit. J. Pharmacol.* 32, 295
Collier H O J, Dinneen L C, Perkins A C & Piper P J (1968) *Naunyn-Schmiedebergs Arch. exp. Path. Pharmak.* 259, 159
Collier H O J, James G W L & Schneider C (1966) *Nature (Lond.)* 212, 411
Collier H O J & Lee I R (1963) *Brit. J. Pharmacol.* 21, 155
Collier H O J & Shorley P G (1960) *Brit. J. Pharmacol.* 15, 601
Collier H O J & Sweatman W J F (1968) *Nature (Lond.)* 219, 864
Cook R A (1946) Allergy in Theory and Practice. Philadelphia; p 161
Elliott D F *et al.* (1960) *J. Physiol. (Lond.)* 153, 473
Emele J F & Shanaman J (1963) *Proc. Soc. exp. Biol. (N.Y.)* 114, 680
Feldberg W & Lewis G P (1964) *J. Physiol. (Lond.)* 171, 98
Gjuris V, Heicke B & Westermann E (1964) *Naunyn-Schmiedebergs Arch. exp. Path. Pharmak.* 248, 540
Graeff F G *et al.* (1969) *Brit. J. Pharmacol.* 37, 723
Greeff K & Moog E (1964) *Naunyn-Schmiedebergs Arch. exp. Path. Pharmak.* 248, 204
Helfer H & Jaques R (1968) *Helv. physiol. pharmacol. Acta* 26, 137
Horton E W
(1963) *Nature (Lond.)* 200, 892
(1969) *Physiol. Rev.* 49, 122
James G W L (1969) *J. Pharm. Pharmacol.* 21, 379
Jaques R (1965) *Helv. physiol. pharmacol. Acta* 23, 156
Lecomte J, Troquet J & Dresse A (1961) *Arch. int. Physiol.* 69, 89
Lewis G P & Reit E (1965) *J. Physiol. (Lond.)* 179, 538
Lim R K S, Guzman F, Rodgers D W, Goto K, Braun C, Dickerson C D & Engle R J (1964) *Arch. int. Pharmacodyn.* 152, 25
Lim R K S, Miller D G, Guzman F, Rodgers D W, Rogers R W, Wang S K, Chao P Y & Shih T Y (1967) *Clin. Pharmacol. Ther.* 8, 521
Main I H M (1964) *Brit. J. Pharmacol.* 22, 511
Moog E & Fischer J (1964) *Naunyn-Schmiedebergs Arch. exp. Path. Pharmak.* 249, 384
Northover B J & Subramanian B (1961) *Brit. J. Pharmacol.* 17, 107
Pearson R S B (1963) In: Salicylates: an International Symposium. Ed. A StJ Dixon *et al.* London; p 170
Piper P J & Vane J R (1969) *Nature (Lond.)* 223, 29
Pisano J J (1968) *Fed. Proc.* 27, 58
Schachter M (1969) *Physiol. Rev.* 49, 509
Sweatman W J F & Collier H O J (1968) *Nature (Lond.)* 217, 69
Vargaftig B B (1966) *Experientia* 22, 182

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Role of Kinins and Prostaglandins in Inflammation

Prostaglandins (PGs) and kinins are families of highly active substances naturally present in tissue. We are only just beginning to learn where and when these substances are present in significant amounts and what they can do. Their formation and destruction is not easy to follow and although we know that they must contribute to pathophysiological changes during inflammation, direct proof is meagre. One can nevertheless make an interim assessment of their virtues and vices and tentatively indicate whether medical intervention might be advantageous. Table 1 outlines a general scheme of the factors expected to initiate, increase, or prolong the action of kinins and PGs directly during inflammation. Other mediator substances such as histamine, active components of complement, lymphocyte factors, adenosine derivatives, and possibly 5-hydroxytryptamine cannot be ignored because the release of one substance will often lead to the activation or potentiation of another. The pattern of events is therefore complex and dynamic, and subject to enormous variation depending on the degree and site of inflammation.

Kinin is activated by very mild damage involving increased vascular permeability or activation of the clotting mechanism. Several authors (*see* Rocha e Silva 1964) have shown that quite mild heating (47°C) causes formation of kinin. Damage caused by turpentine was studied by Spector & Willoughby (1963) and others who established that kinins were involved. These studies relied heavily on the use of aprotinin (Trasylol), a pancreatic inhibitor, to suppress the formation of kinin, and thus diminish the changes observed.

It is well established that bradykinin plays a part in anaphylactic shock, and Sicuteri & Periti (1963) detected kinins in blood during severe

Table 1
Development and maintenance of inflammation: roles of kinins and prostaglandins

Triggers
Trauma, heat, chemicals
Micro-organisms
Antigen + antibody
Early reactions and mediators (K = kinins P = PGs)
Vascular changes, e.g. increased permeability, damage to endothelium (K)
Active complement factors (K, P ?)
Leukocytes (K, P ?)
Damaged or moribund tissue releasing mediators and/or lysosomal enzymes (K ? P ?)
Established inflammation
Low pH and oxygen tension
Fibrin deposits
Impaired paracapillary circulation and drainage

human asthma. Cells upon which reaginic-type antibody is fixed are damaged when the antibody reacts with antigen, and a tissue kallikrein is activated. This differs from plasma kallikrein, and complement is not involved. In saline-perfused sensitized tissue, kinin is not found on challenge with antigen because no substrate is present. However, incubation of the effluent perfusate with kininogen shows that kallikrein is activated by the antigen-antibody reaction. Kinins are found in the blood *in vivo* for a short time at the height of shock, and if the antigen is administered at low concentration for an hour, there is a striking fall in plasma kininogen, showing that a very large amount of kinin must have been activated, but quickly destroyed (Lahiri 1962). It has long been known that plasma containing antibody would generate kinin when antigen was added. Jonasson & Becker (1966) showed that the activation of kallikrein from its precursor in plasma required the activation of complement, and that precipitating IgG was the antibody concerned. Kinin can thus be generated either by cell damage or by activation of complement in the circulation or tissue spaces.

Complement activated extravascularly will cause increased extravasation of blood proteins because the third and fifth components of complement are permeability factors. This will bring both kininogen and kallikreinogen to the site, and produce kinin. The experimental œdema produced by carrageenin given extravascularly is partly due to kinin and involves C^1 depletion, thus adding further suggestive evidence of a relationship between these events. But, apart from direct activation, the lytic action of complement will damage cells and must be expected to activate or release chymotrypsin-like enzymes able to form kinin. This mechanism has been demonstrated in polymorphs by Greenbaum & Kim (1967). Direct damage to cells, including the effects of complement activated by nonimmune processes, could also release such enzymes. Thus at some stage or stages in almost any form of allergic or other form of tissue damage, conditions will favour the formation of kinins.

The more severe kinds of cell damage resulting in enzymic damage to the plasma membrane or even disruption of the cell, may well form prostaglandins. These can be extracted from most normal tissues, and can be formed from polyunsaturated aliphatic acids, such as arachidonic acid, by a common lipid oxidase (Bergstrom *et al.* 1968). The amount produced seems to be governed by the availability of substrate, and the type of prostaglandin produced varies according to the presence of different co-factors. Considerable quantities of PGs – mainly of the E type – are found when cells are undergoing autolysis, as during menstruation and the late stages of experimental inflammation (Pickles 1967). The participation of lysosomal enzymes seems obvious, notably the phosphohydrolases, which could split suitable fatty acids from lecithin present in cell membranes, and thus supply substrate for conversion to prostaglandins.

But inactive phospholipases are thought to exist on the cell surface also, and are said to be responsible for the disruption of mast cells during anaphylaxis, so there may be sources of effective enzyme other than lysosomes. Vane has shown that, both *in vivo* and *in vitro*, anaphylactic shock triggers the appearance of prostaglandin E_1 (Piper & Vane 1969). At a late stage in tissue damage, lysosomal enzymes are said to be discharged from overactive phagocytes when phagolysosomes are emptied by exocytosis. We have recently observed correspondence between the time when prostaglandins and lysosomal enzymes appeared in an inflammatory exudate, but this may be associated with cell death as well as phagocytosis.

Acid conditions in the tissue favour both the rate of formation and the persistence of kinin, and also the activity of lysosomal enzymes. Reduced paracapillary circulation, attributable to poor tissue drainage or loss of protein into the tissue spaces, would seem to generate a vicious circle. In fact, changes in pH and oxygenation may not be very serious because there is generally hyperæmia and the plasma proteins will exert a considerable buffering effect. The plasma also contains inhibitors of some enzymes and may well provide substrate for others, thus minimizing tissue damage. Until enzymes having the ability to destroy cells are inactivated or diluted it is obviously not advantageous to permit their free drainage from the inflamed area.

On balance, it seems to me that kinins may be beneficial in moderating and limiting a local inflammatory reaction. As for the prostaglandins, they have differing biological activity at different sites, and the proportion of the different types will determine the net result. Indeed the PG system could well be 'self-limiting', by generating different types of PG as the local concentration of thiols and the oxygen tension alter with variations in the effective circulation. But such speculation does not contribute to our assessment of the good and bad features of PGs. Direct tests in man suggest that PGE_1 and PGE_2 are very potent but rather benign vasodilators, with the F series much less active. Injection of 25 ng PGE_1 in 0·1 ml intradermally causes a little œdema, and a strong flare reaction lasting over thirty minutes. There is a sensation of warmth but no pain and no aftereffects (Crunkhorn & Willis 1969). In conclusion, it seems that in our present state of knowledge, there is no good general case for attempting to abolish the effects of kinins or PGs.

REFERENCES
Bergstrom S *et al.* (1968) *Pharmacol. Rev.* **20**, 1
Crunkhorn P & Willis A L (1969) *Brit. J. Pharmacol.* **36**, 216P
Greenbaum L M & Kim K S (1967) *Brit. J. Pharmacol.* **29**, 238
Jonasson O & Becker E L (1966) *J. exp. Med.* **123**, 509
Lahiri S C (1962) PhD thesis, Edinburgh
Pickles V R (1967) *Biol. Rev.* **42**, 614
Piper P J & Vane J R (1969) *Nature (Lond.)* **223**, 29
Rocha e Silva M (1964) *Ann. N.Y. Acad. Sci.* **116**, 899
Sicuteri F & Periti P (1963) *Folia allergol.* **10**, 523
Spector W G & Willoughby D A (1963) *Bact. Rev.* **27**, 117

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Role of Kinins and Prostaglandins as Mediators of Functional Hyperæmia

The blood supply to many organs is increased when they are active; at rest the blood flow is small and the vessels are constricted, the smaller ones sometimes being closed altogether. When an organ is activated, the vessels are dilated and blood flow is increased.

This fundamental vascular reaction varies according to the special needs of each organ. In skeletal muscle or exocrine glands, activity is intermittent and each burst of activity is accompanied by a strong short-lasting vasodilatation. In endocrine glands or adipose tissue, on the other hand, activity is almost continuous with prolonged periods of increased activity during which there is a moderate and prolonged increase in blood flow.

There have been two principal hypotheses based upon work on skeletal muscle and exocrine glands. First, Claude Bernard in 1858 postulated 'the nervous mechanism'; according to this the chorda tympani, the secretory nerve to the submandibular salivary gland, was reported to contain special vasodilator nerve fibres which, when stimulated, liberated a chemical mediator causing a vasodilatation. This view has been held by many workers, most of whom go on to say that these supposed vasodilator nerves are cholinergic nerves liberating acetylcholine from their endings. As the main nerve bundle in the chorda tympani is indeed cholinergic, it is not surprising that acetylcholine could be shown to be present about the gland. However, as the secretory response to chorda stimulation is abolished by small doses of atropine, whereas the vasodilator response is only slightly reduced (as shown in Fig 1), it seems unlikely that this vascular response is wholly due to liberated acetylcholine. Such a response due to injected acetylcholine is readily abolished by atropine. The acetylcholine liberated from the secretory fibres, however, may well account for that part of the vasodilatation which is atropine-sensitive.

Gaskell, in 1877, believed that the great increase in blood flow following muscle contraction was brought about by vasodilator nerve fibres which were stimulated at the same time as the motor fibres. In 1880, however, he advanced the theory that locally produced metabolites act on the vessels to bring about the vasodilatation. This 'vasodilator metabolite' theory was later championed by Joseph Barcroft (1914). He observed that, although atropine abolished the secretory effect of chorda stimulation, it did not completely inhibit the increased uptake of oxygen due to the metabolic activity which resulted from nerve stimulation. Barcroft interpreted this result as proof that metabolic activity in the gland cells results in the liberation of vasodilator metabolites.

The metabolic factors generally accepted as possible mediators of the vasodilatation, such as O_2 lack, lactic acid and pH changes have now been virtually excluded from playing any major part in the response. There has so far been no adequate demonstration of the appearance of a vasodilator substance during muscle contraction. However, Grant (1938) obtained indirect evidence for the release of vasodilator substances. He observed that, if a muscle is made to contract while its blood flow is occluded, a normal vasodilator response is seen when the flow is re-established.

In 1953 Hilton and I took up this problem of the mechanism of functional vasodilatation in the salivary gland. We found that when the gland is activated, an enzyme called kallikrein is liberated from the secretory cells into the interstitial space where it acts upon an α_2-globulin, forming, as shown in Fig 2, a vasodilator polypeptide kinin (Hilton & Lewis 1955*a*, *b*, 1956). I should like to recall our main points of evidence. First, we were able to repeat the experiment of Grant to show that a vasodilator substance was produced on activation of the gland which was more stable than acetylcholine because it could be held within the gland during occlusion of the blood supply. In the experiment of Fig 3, the time course and extent of the hyperæmia following 15 sec stimulation of the chorda are seen at (*a*); occlusion of both artery and vein for 1 min produced a small reactive hyperæmia at (*b*). However, when the chorda was stimulated during the first 15 sec of the same period of circulatory unrest, the hyperæmia on re-establishing the circulation at (*c*) was as large as that seen after chorda stimulation in the ordinary way. This result is in agreement with the view that acetylcholine is released to act on the gland cells which, when activated, produced in their turn a more stable vasodilator substance.

Secondly, when we perfused the salivary gland with Locke's solution and collected the venous effluent, then a vasodilator substance which was not acetylcholine was present during activation.