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THE EFFECTS OF SENSORY DENERVATION ON THE RESPONSES OF THE RABBIT EYE TO PROSTAGLANDIN E_1, BRADYKININ AND SUBSTANCE P

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1 Six to eight days after diathermic destruction of the fifth cranial nerve in the rabbit, the ocular hypertensive and miotic responses to intracameral administration of capsaicin, bradykinin, and prostaglandin E_1 were greatly reduced or completely abolished. The response to substance P was not abolished.

2 A response could still be obtained to chemical irritants 36 h after coagulation of the nerve and it is deduced that manifestation of the response is dependent upon functional sensory nerve terminals, and is independent of central connections.

3 It is suggested that prostaglandin E_1 and bradykinin act directly upon the sensory nerve endings and that propagation of the response is augmented by axon reflex.

4 In view of the ability of substance P to induce miosis in the denervated eyes, it is presumed that its actions are not mediated via sensory nerves.

5 It is considered possible that the mediator(s) released from sensory nerve endings after chemical irritation or antidromic stimulation may act in the same way as substance P with regard to the miotic effect.

6 Synthetic substance P will only produce ocular hypertension in doses which induce a maximal miotic response. This may either be a question of access or a partial resemblance to the endogenous mediator.

Introduction

The anterior segment of the rabbit eye responds to mechanical or chemical irritation by miosis, vasodilatation and a breakdown of the blood-aqueous barrier. This allows the intraocular pressure to rise and protein to enter the aqueous humour.

The responses to different stimuli, while outwardly similar do not seem to be mediated by the same mechanism, although analysis of these responses has produced a distinctive pattern. The response to mechanical irritation of the rabbit eye, e.g. by scratching the iris, is mediated almost entirely by prostaglandins, whereas that to chemical irritation, e.g. by topical or intracameral application of formaldehyde, appears to act mainly via a nerve pathway (Cole & Unger, 1973; Eakins, 1977; Butler, Unger & Hammond, 1979). This nerve pathway is resistant to conventional autonomic blocking drugs and is now believed to be identical to that with afferent sensory function.

The importance of peripheral sensory nervous involvement in the ocular injury response was first realised by Bruce (1910, 1913) who observed that the effects of chemical irritation of the conjunctiva with nitrogen mustard were reduced after retrobulbar anaesthesia and abolished after postganglionic division and degeneration of the ophthalmic branch of the fifth cranial nerve. Bruce's concept of the sensory axon reflex was compatible with observations of antidromic sensory vasodilatation in the skin (Bayliss, 1901; 1902; Langley, 1923) and was expounded by Lewis (1927) in his interpretation of the triple response. Perkins (1957) re-examined the ocular response to antidromic stimulation of the fifth nerve originally described by Claude Bernard and showed its similarity to that produced by injury. Jampol, Axelrod & Tessler (1976) found that the effects of topical nitrogen mustard on the rabbit eye were reduced after prior infection with herpes simplex and inferred that an axon reflex was necessary to mediate the response. Using the technique of Jancso (1960) of desensitization by capsaicin, Arvier, Chahl & Ladd (1977) showed that in the rat skin, noxious substances including formaldehyde, bradykinin and prostaglandin act directly and exclusively on sensory nerve endings. From these findings they suggested that a

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humoral substance was released which either liberated histamine and 5-hydroxytryptamine from mast cells or acted directly on the blood vessels to cause vasodilatation.

In spite of many attempts to elucidate the mechanism of antidromic sensory transmission, the identity of the putative mediator remains speculative. Histamine-like substances (Ungar, 1935), histamine itself (Kwiatkowski, 1943; Ibrahim, Stella & Ṭalaat, 1951), adenosine 5'-triphosphate (ATP, Holton, 1959), kinins (Chapman, Ramos, Goodell & Wolff, 1961) and prostaglandins (Perkins, 1975) have all been proposed as candidates, but are now generally discounted. Dale (1935) had suggested that in view of the bipolar structure of the sensory neurone the transmitter at the central end of primary afferents was unlikely to differ from that at the periphery. It now seems likely that substance P is the central transmitter (Lembeck, 1953; Amin, Crawford & Gaddum, 1954; Hökfelt, Kellerth, Nilsson & Pernow, 1975; Otsuka & Konishi, 1976) and so it is possible that it may also be a peripheral mediator, evidence for which has been found in the skin (Hökfelt *et al.*, 1975) and in the teeth (Olgart, Gazelius, Brodin & Nilsson, 1977).

Irritant substances that act via sensory nerves should, in theory, produce no response in desensitized or denervated tissue, whereas drugs that act in a similar fashion to the mediator should act independently of functional nerve terminals.

Our experiments were designed to investigate the effects of sensory denervation on the response of the rabbit eye to intracameral infusion of bradykinin and prostaglandin, both of which are known to reproduce an injury-type response with miosis and disruption of the blood-aqueous barrier (Cole & Unger, 1974; Eakins, 1977), and also the effects of intracameral administration of substance P to normal and denervated eyes.

Methods

Sensory denervations

Adult Dutch male rabbits, 2.0 to 2.5 kg in weight, were anaesthetized deeply with sodium pentobarbitone (Sagatal; May & Baker) 45 mg/kg intravenously. Their heads were fixed in a stereotaxic clamp and positioned so that lambda was 1.5 mm below bregma. The fifth cranial nerve was destroyed by diathermic coagulation in the following way.

The skin on the top of the skull was shaved and cut along the midline, and the intersection of the sutures at bregma was located as the primary reference point. The x and y coordinates for the animal's weight were taken from a predetermined chart and a 3 mm diameter hole trephined at their intersection on the left side of the skull. An area of skin on the abdomen was shaved and the positive plate electrode secured in close contact with electrode jelly. A unipolar diathermy needle was inserted vertically down through the brain at the correct coordinates until the tip was felt to penetrate the dura at the base of the skull. Three or four lesions were then placed in and around the fifth nerve, upon which a strong miosis usually developed. This procedure essentially destroys the Gasserian ganglion and some of the epigasserian nerve tract.

If there was loss of corneal sensitivity on awakening, the lashes were trimmed, chloramphenicol ointment was applied to the eye, and the lids were sutured. Each animal was given a single intramuscular injection of a mixture of benethamine penicillin (190 mg), procaine penicillin (100 mg) and benzyl penicillin sodium (120 mg) in a 0.8 ml suspension (Triplopen; Glaxo) and examined daily to ensure that the lids had not parted. If the corneal surface is exposed for any length of time a neuroparalytic keratitis invariably develops.

After 6 to 8 days, degeneration of sensory fibres was considered to be advanced, a presumption which was tested by histological (electron microscopic) examination of the eyes of three operated animals at the end of experiments. Before use the pupillary response to light was examined and those denervated eyes which differed in rapidity or magnitude of response from the normal, or which had developed corneal opacities, were not used. Only 3 out of 40 rabbits had an abnormal light reflex and in these the third cranial nerve was found on necroscopy to have been damaged during coagulation.

Infusion of drugs

Those animals with a denervated eye of normal appearance were anaesthetized with sodium pentobarbitone (30 mg/kg) and a femoral artery and vein were cannulated for blood pressure measurement and intravenous injection respectively. The anterior chamber of each eye was transfixed with needles as described by Nagasubramanian (1974) and each eye was perfused with physiological saline (9.0 g NaCl per litre) at a constant rate of 25 µl per min using the circuit devised by Hammond (1977) (Figure 1) which enables the intraocular pressure to be monitored continuously while drugs are being infused. The intraocular pressure and blood pressure were recorded with Bell & Howell strain gauge pressure transducers. Pupil size was measured with fine dividers.

In eight experiments drugs were infused into the posterior chamber. In these the inflow was via a 25-gauge needle introduced into the posterior chamber through the sclera. The exact position of the needle could be observed through the pupil. The

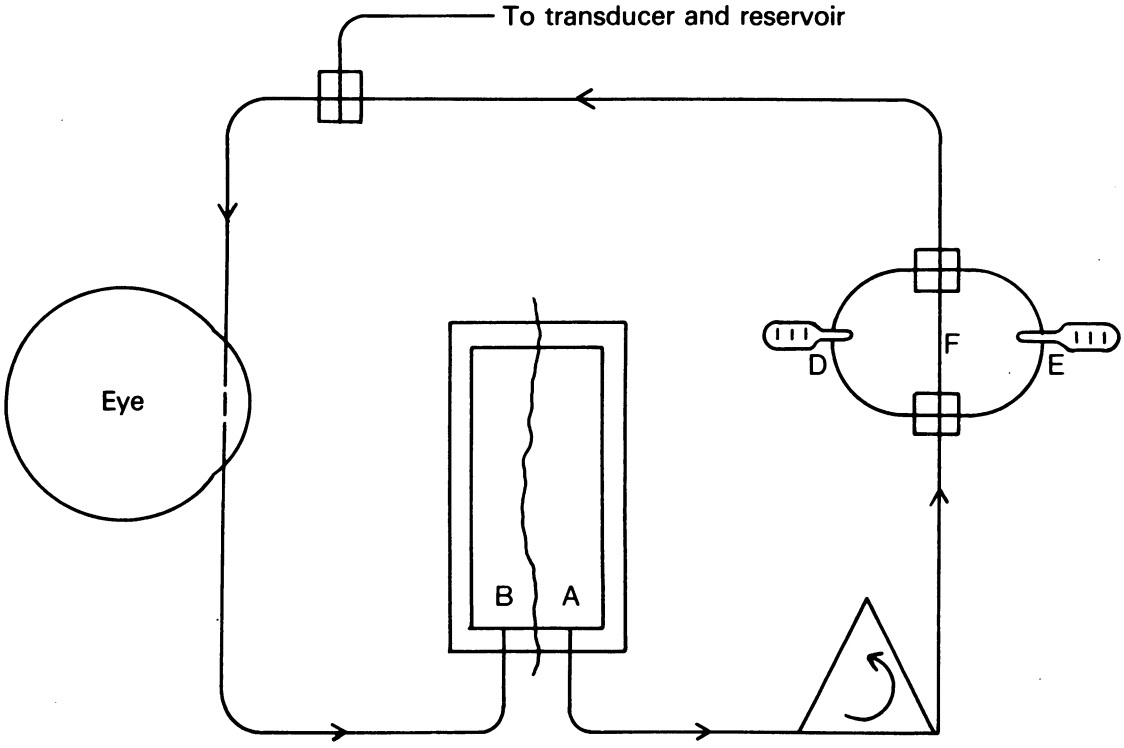

Figure 1 Apparatus for perfusion of rabbit eye. Perfusate is pumped from reservoir A through the eye and back to B; 25 µl volumes of drugs are contained in loops D and E, and are released by moving their clip to F. Two circuits are required to perfuse both eyes simultaneously.

drugs used were capsaicin (8-methyl-*N*-vanillyl-6-nonenamide; Sigma); bradykinin triacetate trihydrate (Koch-Light Laboratories Ltd.); prostaglandin E_1 (Upjohn Company); and substance P (Sigma).

Capsaicin is believed to act specifically upon sensory nerve endings (Jansco, 1960; Jansco, Jansco-Gabor & Szolcsanyi, 1967) and we therefore used it to give an initial indication as to whether complete or effective sensory denervation had been achieved. Drugs were contained in 25 µl loops by clips as illustrated in Figure 1.

At the end of each experiment the skull was opened, the brain removed and the base of the cranium examined. The fifth nerve runs in a channel in the bone beneath the dura at the point of coagulation and in cases where complete destruction of the nerve and immediately surrounding tissues was not visible, or where the third nerve also appeared damaged, results were not included in this study.

Results

Six to eight days after diathermic coagulation of the left fifth cranial nerve, the initial intraocular pressures were significantly lower in the left (denervated) eyes (mean ± s.e. mean, R = 18.7 ± 0.7; L = 13.3 ± 1.5 mmHg; $n = 10$; $P < 0.01$ Student's t test on group values) though there was no difference in the pupil sizes (R = 6.7 ± 1.5; L = 6.9 ± 1.5 mm, $n = 22$) and the extent and rapidity of the light reflex was unaffected.

Histological examinations

No gross evidence of axonal degeneration was visible by light microscopy after selective staining and visual appraisal discerned no apparent difference between normal and denervated iris. However, electron microscopy showed extensive axonal swelling, Schwann cell shrinkage, and loss of microtubules in the larger myelinated bundles in the denervated tissues.

Capsaicin

The hypertensive and miotic responses to intracameral infusion of a high dose of capsaicin (25 µg) were either greatly reduced or completely abolished by effective sensory denervation. (Peak change in pupil diameter (mm) R = −3.8 ± 0.5; L = −0.4 ± 0.1;

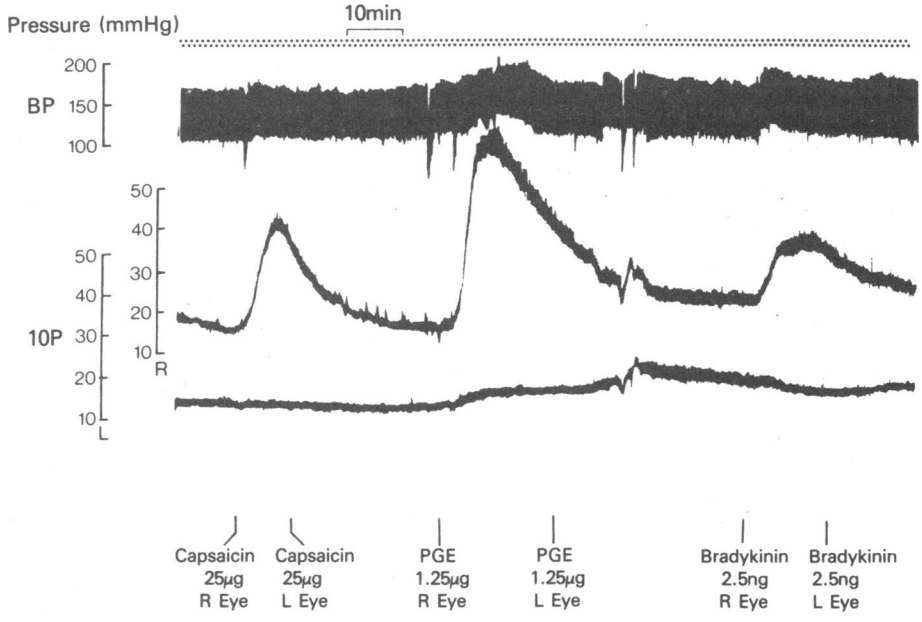

Figure 2 Trace showing a series of intraocular pressure (IOP) responses to capsaicin, prostaglandin E_1 (PGE_1) and bradykinin, 6 days after intracranial coagulation of the left fifth nerve.

$n = 9$; $P < 0.001$; peak change in intraocular pressure (mmHg) R = 19.9 ± 4.8; L = 3.5 ± 4.0; $n = 8$; $P < 0.05$). The normal eye usually recovered sufficiently to enable one or two more drugs to be infused, but the intraocular pressure rarely fell to its initial value again, nor did the pupil size recover fully and after two or three drugs had been infused, the eye became congested and unresponsive.

Bradykinin and prostaglandin E_1

The responses to both bradykinin (2.5 ng to 1.25 μg) and prostaglandin E_1 (6.25 ng to 2.5 μg) were also greatly reduced or completely abolished after destruction and degeneration of the fifth nerve (Figures 2, 3 and 4).

In order to establish that the reduced responses in the denervated eyes were not due to severance of central connections with efferent motor fibres or to oedema and vascular stasis, four experiments were performed at 24 and 36 h after coagulation of the fifth nerve. In these animals the eye on the operated side appeared slightly inflamed, and, although the cornea was clear, some aqueous flare was observed, presumably due to protein remaining from the initial disruption of the barrier. At both 24 and 36 h a somewhat reduced hypertensive and miotic response was still obtainable in the denervated eye to capsaicin, bradykinin and prostaglandin E_1 even though on necroscopy, destruction of the fifth nerve was found to be complete.

Substance P

Intracameral infusion of substance P (250 pg to 2.5 μg) produced an extreme and prolonged miosis not only in normal eyes but also in denervated eyes which were relatively unresponsive to PGE_1 (500 ng) or to

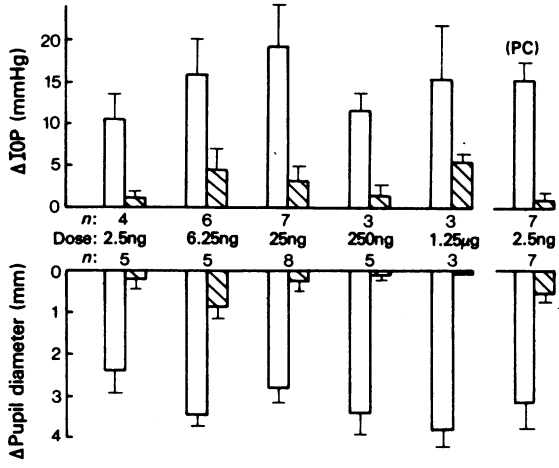

Figure 3 Peak intraocular pressure (IOP) and miotic responses, regardless of time, to intracameral infusion of bradykinin. Open columns represent normal eyes; hatched columns represent denervated eyes. (PC) indicates infusion into the posterior instead of the anterior chamber.

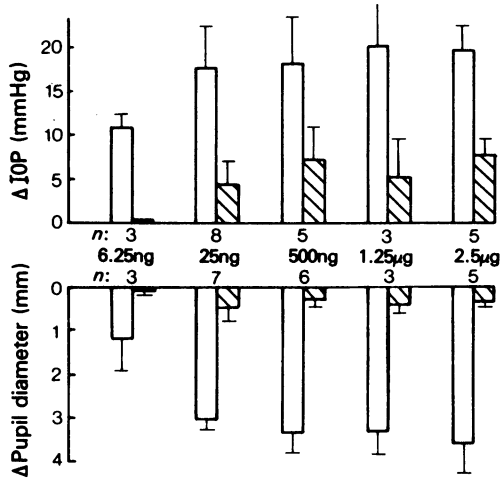

Figure 4 Peak intraocular pressure (IOP) and miotic responses, regardless of time, to intracameral infusion of prostaglandin E_1 (PGE_1). Open columns represent normal eyes; hatched columns represent denervated eyes.

bradykinin (25 ng). After administration of 2.5 ng substance P, the pupil was sometimes not visible, such was the intensity of the response. However only at doses which produced a very severe miosis was there any appreciable rise in the intraocular pressure, even in the normal eye (Figure 5), and this was often attributable to the development of iris bombé and the occlusion of the chamber angle. In 6 out of 20 experiments substance P induced slow and continuous rises in intraocular pressure, the magnitude and gradient of which were similar in normal and denervated eyes (Figure 6). There was often a transient fall in systemic blood pressure after infusion of substance P. This may be attributable to the drug entering the circulation with the outflow of aqueous humour.

Posterior chamber infusions

In a further series of experiments drugs were introduced into the posterior chamber to place them in closer proximity to the ciliary processes which are known to be the site of breakdown of the blood-aqueous barrier (Cole, 1974). When compared with the effects of anterior chamber infusion, bradykinin produced a greater pressure response and slightly reduced pupil constriction in the normal eye, and still no response in the denervated eye (Figure 3). Substance P produced a larger pressure response in both normal and denervated eyes, but the magnitude of this still did not compare with that of the responses evoked by PGE_1 or bradykinin and the miosis was only marginally reduced (Figure 5).

Discussion

With regard to bradykinin and PGE_1 these results are consistent with those findings reported by Arvier *et al.* (1977) in the rat skin. The absence of a response in the denervated eye indicates that the miotic and hypertensive actions of bradykinin and prostaglandin E_1 are largely if not entirely dependent upon an intact

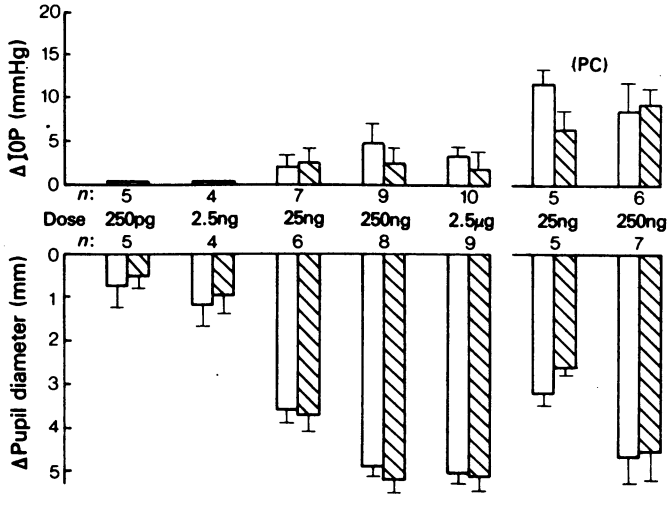

Figure 5 Peak intraocular pressure (IOP) and miotic responses, regardless of time, to intracameral infusion of substance P. Open columns represent normal eyes; hatched columns represent denervated eyes. (PC) indicates infusion into the posterior instead of the anterior chamber.

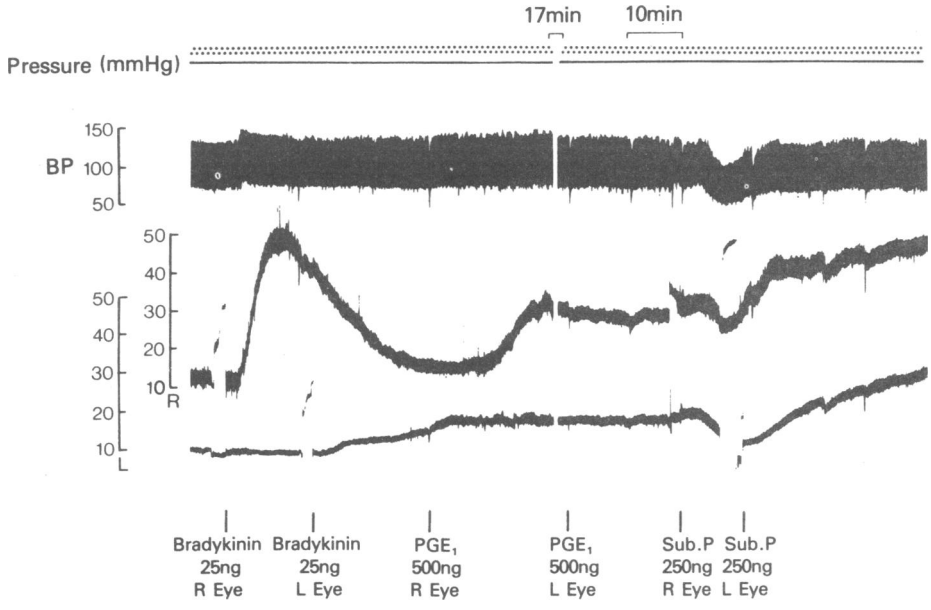

Figure 6 Trace showing a series of intraocular pressure (IOP) responses to bradykinin, prostaglandin E_1 (PGE_1) and substance P 6 days after intracranial coagulation of the left fifth nerve. Discontinuity of the intraocular pressures is due to changing the side loops in the perfusion circuit.

peripheral sensory nervous system. In view of the pain producing properties of these drugs it is to be expected that they would have actions on sensory nerves, but these findings confirm that the fibres have the potential ability to influence the state of the blood-aqueous barrier. The responses obtainable to these drugs 24 and 36 h after section of the fifth nerve indicate that these actions are independent of central connections. It is reasonable to assume that they persist until degeneration of the peripheral fibres has occurred, as observed in the conjunctiva by Bruce (1913) and in the skin by Lewis (1927).

It is unlikely that all the sensory nerves peripheral to the Gasserian ganglion will have degenerated, for in the rabbit the ganglion is a long diffuse structure (Perkins, 1957; Ruskell, 1964) and probably some cell bodies will lie forward of the lesions. However, the lesions were placed as far forward as possible and the greater proportion is likely to have been destroyed or damaged. Surviving neurones may account for the residual response in some animals.

It was considered that the apparently inert denervated eye might be incapable of responding to irritant substances owing to persistent congestion of the vasculature after the initial disruption of the blood-aqueous barrier during fifth nerve coagulation. If the circulation was impaired by stromal oedema, a low perfusion pressure in the capacitance vessels would consequently inhibit the potential increase in the rate of filtration which accounts for the immediate hypertensive response to trauma. This could also account for the lower initial pressures in the denervated eyes and the absence of miosis might in this case be due to iridic ischaemia.

However, there are arguments against this explanation. The pressure in the denervated eye reflects changes in blood pressure, although admittedly not to the extent of that of the normal eye. Vascular congestion is likely to be localized in the ciliary processes which are the main site of barrier disruption (Cole, 1974), and therefore should not affect injury-induced miosis, which is probably mediated by the same transmitter that causes the blood-aqueous barrier to break down. The pupillary response to substance P also excludes the possibility that the sphincter muscle has become inert. Moreover, one would expect more congestion 24 and 36 h after coagulation, but a convincing response was still obtainable to bradykinin and prostaglandin E_1. Also fluorescein angiography has shown that the uveal vascular filling pattern and times are not abnormal in the denervated eye (Butler & Hammond, unpublished observations). It is therefore inferred that the reduced response in the denervated eye can best be explained by the disappearance of the putative mediator from the sensory nerve terminals during degeneration. Release of this substance during degeneration might also explain the inflamed appearance and aqueous flare 24 and 36 h after coagulation.

These observations are intriguing in that the pro-

portion of other such ocular responses, e.g. that to scratching or laser irradiation of the iris (Ambache, Kavanagh & Whiting, 1965; Cole & Unger, 1973; Unger & Bass, 1977) which has hitherto been designated as prostaglandin-mediated would seem necessarily to include this nervous component.

Localization of receptors for prostaglandin E_1 may confine much of its action to the sensory nerve endings where stimulation, with subsequent release of the hypothetical humoral mediator and manifestation of the characteristic response might not be affected by local anaesthetics. The latter would, however, block axonal conduction and so prevent spread of the stimulus. Thus in the interpretation by Unger, Cole & Bass (1977) of the response to laser irradiation of the iris, the 'neurogenic component' blocked by local anaesthetics may be that part of the response which is propagated by axon reflex, whereas the residual response may be that part due to the direct focal action of prostaglandins on the nerve terminals.

It was considered that should substance P act in the same way as the putative mediator, then the presence or absence of functional sensory nerves should have no bearing on the nature or magnitude of the response as they would on that to PGE_1 or bradykinin, which seem to require their presence. In as much as the identical pupil responses in normal and denervated eyes could be explained on this basis, the minimal pressure rise which accompanied submaximal miosis has been the subject of much speculation, as the two features were previously thought to be inseparable components of the injury response. However this provoked the following argument.

The bulk flow of aqueous being from the posterior chamber, drugs infused into the anterior chamber may not gain access to the ciliary vasculature (Figure 7). If this is the case then neither should capsaicin, PGE_1 or bradykinin reach the posterior chamber. If they do not, then one might ask how it is that they cause a breakdown of the barrier in the normal eye? The most likely explanation would be that stimulation of sensory nerve endings by these noxious drugs activates an axon reflex which can produce an immediate response in a vascular bed remote from the primary site of action.

The failure of substance P to induce a similar hypertensive response could therefore be explained in this way as by virtue of its miotic effect on the denervated eye its action does not seem to involve functional sensory nerve endings. Thus to test this hypothesis it was infused into the posterior chamber to place it closer to the ciliary vessels. This did produce a rise in the intraocular pressure in both normal and denervated eyes but it was very much less than the rise produced by equimolar concentrations of bradykinin in the normal eye. Therefore either synthetic substance P is only partially representative of the endogenous mediator and does not act on all the relevant receptors, or it may not reach the resistance vessels responsible for controlling the vascular component of the response. The vessels of the ciliary processes have virtually no contractile elements (Taniguchi, 1962), depending for their flow rates upon the state of the major circle. Also the ciliary processes are sparsely innervated with sensory fibres whereas there is a dense network of sensory nerves terminating around the root of the iris (Ruskell, personal communication). These probably bring the endogenous mediator into close apposition to the major circle which may itself be relatively inaccessible to exogenous substance P.

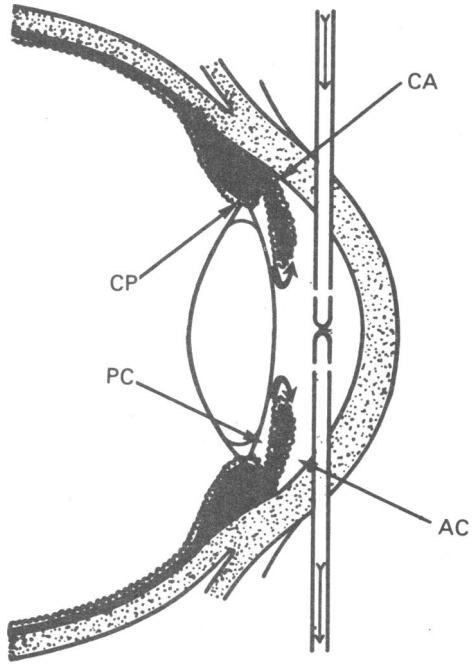

Figure 7 Diagram showing the relationship of the perfusion needle transfixing the anterior chamber (AC) to the ciliary processes (CP) and the flow of aqueous humour from the posterior chamber (PC) to the anterior chamber angle (CA).

In conclusion, the ocular response to PGE_1 and to bradykinin seems to be mediated largely if not mainly via sensory nerves. The response to substance P does not depend upon their functional integrity and it may be that this polypeptide bears some resemblance to the endogenous antidromic mediator, particularly with regard to its actions on the iris sphincter muscle.

References

AMBACHE, N., KAVANAGH, L. & WHITING, J. (1965). Effect of mechanical stimulation on rabbits' eyes: release of active substance in anterior chamber perfusates. *J. Physiol.*, **176**, 378–408.

AMIN, A.H., CRAWFORD, T.B.B. & GADDUM, J.H. (1954). The distribution of substance P and 5-hydroxytryptamine in the central nervous system of the dog. *J. Physiol.*, **126**, 596–618.

ARVIER, P.T., CHAHL, L.A. & LADD, R.J. (1977). Modification by capsaicin and compound 48/80 of dye leakage induced by irritants in the rat. *Br. J. Pharmac.*, **59**, 61–68.

BAYLISS, W. M. (1901). On the origin from the spinal cord of the vasodilator fibres of the hind limb, and the nature of these fibres. *J. Physiol.*, **26**, 173–209.

BAYLISS, W.M. (1902). Further researches on antidromic nerve impulses. *J. Physiol.*, **28**, 276–299.

BRUCE, A.N. (1910). Uber die Geziehung der sensiblen Nervendigungen zum Entzundungsvorgang. *Naunyn Schmiedebergs Arch. exp. Path. Pharmac.*, **63**, 424–433.

BRUCE, A.N. (1913). Vasodilator axon reflexes. *Q. J. exp. Physiol.*, **6**, 339–354.

BUTLER, J. M., UNGER, W.G. & HAMMOND, B.R. (1979). Sensory mediation of the ocular response to formaldehyde. *Exp. Eye Res.*, **28**, 577–589.

CHAPMAN, L.F., RAMOS, A.O., GOODELL, H. & WOLFF, H.G. (1961). Neurohumoral features of afferent fibres in man. *Arch. Neurol. (Chic.)*, **4**, 617–650.

COLE, D.F. (1974). The site of breakdown of the blood-aqueous barrier under the influence of vasodilator drugs. *Exp. Eye Res.*, **19**, 591–607.

COLE, D.F. & UNGER, W.G. (1973). Prostaglandins as mediators for the responses of the eye to trauma. *Exp. Eye Res.*, **17**, 357–368.

COLE, D.F. & UNGER, W.G. (1974). Action of bradykinin on intraocular pressure and pupillary diameter. *Ophthal. Res.*, **6**, 308–314.

DALE, H.H. (1935). Pharmacology and nerve endings. *Proc. R. Soc.* (Biol), **28**, 319–322.

EAKINS, K.E. (1977). Prostaglandin and non-prostaglandin mediated breakdown of the blood-aqueous barrier. *Exp. Eye Res.*, **25** (Suppl.), 483–498.

HAMMOND, B.R. (1977). Perfusion of the rabbit anterior chamber. *Exp. Eye Res.*, **24**, 533–534.

HÖKFELT, T., KELLERTH, J.O., NILSSON, G. & PERNOW, B. (1975). Experimental immunohistochemical studies on the localisation and distribution of substance P in cat primary sensory neurones. *Brain Res.*, **100**, 235–252.

HOLTON, P. (1959). The liberation of adenosine triphosphate on antidromic stimulation of sensory nerves. *J. Physiol.*, **145**, 494–504.

IBRAHIM, F.D., STELLA, G. & TALAAT, M. (1951). The mechanism of antidromic vasodilatation. *Q. J. exp. Physiol.*, **36**, 189–197.

JAMPOL, L.M., AXELROD, A. & TESSLER, H. (1976). Pathways of the eye's response to topical nitrogen mustard. *Invest. Ophthal.*, **15**, 486–489.

JANCSO, N. (1960). Role of the nerve terminals in the mechanism of inflammatory reactions. *Bull. Millard Fillmore Hosp., New York*, **7**, 53–77.

JANCSO, N. JANCSO-GABOR, A. & SZOLCSANYI, J. (1967). Direct evidence for neurogenic inflammation and its prevention by denervation and by pretreatment with capsaicin. *Br. J. Pharmac. Chemother.*, **31**, 138–151.

KWIATKOWSKI, H. (1943). Histamine in nervous tissue. *J. Physiol.*, **102**, 32–41.

LANGLEY, J.N. (1923). Antidromic action. Part II: Stimulation of the peripheral nerves of the cat's hind foot. *J. Physiol.*, **58**, 49–69.

LEMBECK, F. (1953). Zur Frage der zentralen Übertragung afferenter Impulse III. *Naunyn-Schmiedebergs Arch. exp. Path. Pharmac.*, **219**, 197–213.

LEWIS, T. (1927). *The Blood Vessels of the Human Skin and their Responses.* London: Shaw and Sons.

NAGASUBRAMANIAN, S. (1974). The effect of vasopressin on the facility of aqueous humour outflow in the rabbit. *Ophthal. Res.*, **6**, 301–307.

OLGART, L. GAZELIUS, B., BRODIN, E. & NILSSON, G. (1977). Release of substance P-like immunoreactivity from the dental pulp. *Acta physiol. scand.*, **101**, 510–512.

OTSUKA, M. & KONISHI, S. (1976). Substance P and excitatory transmitter of primary sensory neurones. *Cold Spring Harbor Symp. Quant. Biol.*, **40**, 135–143.

PERKINS, E.S. (1957). Influence of the fifth cranial nerve on the intraocular pressure of the rabbit eye. *Br. J. Ophthal.*, **41**, 257–300.

PERKINS, E.S. (1975). Prostaglandins and the eye. *Adv. Ophthal.*, **29**, 2–21.

RUSKELL, G.L. (1964). In *The Rabbit in Eye Research.* ed. Prince, J.H., pp. 568–572. Springfield: Charles C. Thomas.

TANIGUCHI, Y. (1962). Fine structure of blood vessels in the ciliary bodies. *Jap. J. Ophthal.*, **6**, 93–103.

UNGAR, G. (1935). Sur les rapports des appareils peripheriques vaso-dilatateurs avec les terminasions nerveuses sensitives, d'apres la conception de la transmission humorale histaminique. *Ann. Anat. path.*, **12**, 586–596.

UNGER, W.G. & BASS, M.S. (1977). Prostaglandin and nerve-mediated response of the rabbit eye to argon laser irradiation of the iris. *Ophthalmologica Basel*, **175**, 153–158.

UNGER, W.G., COLE, D.F. & BASS, M.S. (1977). Prostaglandin and neurogenically mediated ocular responses to laser irradiation of the rabbit iris. *Exp. Eye Res.*, **25**, 209–220.

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