Short communications

Effects of calcium on dextran anaphylactoid oedema in the rat

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Low doses of calcium inhibited the local anaphylactoid response of rat paw and skin to dextran.

Voorhees, Baker & Pulaski (1951) reported that a single intraperitoneal injection of dextran into rats produced an anaphylactoid reaction, which involved a characteristic subcutaneous oedema and hyperaemia of the face, tongue, genitals and paws. Later experiments by Harris, Luscombe & Poyser (1967) showed that dextran fractions of molecular weight above 10,000 and below 200,000 were the most active in producing the response, which is said to be induced by the release of biogenic amines from mast cells, as pretreatment with mast cell dischargers such as compound 48/80 inhibit it (Parratt & West, 1957). The principal amine released is 5-hydroxytryptamine (5-HT) since anti5-HT compounds such as BOL 148 greatly inhibit the reaction, whereas antihistamines such as mepyramine only slightly reduce it (Parratt & West, 1957).

Mongar & Schild (1958) reported that the release of biogenic amines during anaphylactic shock in guinea-pig tissues is dependent on the presence of calcium ions. It was therefore of interest to determine whether or not the dextran anaphylactoid reaction in the rat paw or rat skin is affected by calcium.

Methods.—The animals used were female Wistar rats (Tuck strain) weighing 160/180 g. They were allowed food (Dixons 41B) and water *ad libitum*.

Foot experiments

Rats were injected with solutions of drugs into the plantar region of the right hind paw. The left foot received saline alone. Two and a half hours after the feet had been injected, the animals were killed by a blow to the head. The hind feet were removed at the tibio-tarsal joint and weighed. The local anaphylactoid response was assessed by determining the percentage increase in the weight of the right hind foot over that of the left foot.

Skin experiments

Rats were anaesthetized with pentobarbitone sodium (45 mg/kg) and Evans blue (30 mg/kg) was injected into a cannulated jugular vein. The animals then received six intradermal injections into the dorsal skin, which had been shaved 24 h previously. The volume injected was 0.1 ml. Thirty minutes after the intradermal injections, the animals were killed by a blow to the head and the skin of the back was removed. The anaphylactoid reactions were then assessed by measuring the amount of Evans blue in each wheal after extraction according to the spectrophotometric method of Minoru Horada, Mitsuo Takeuchi, Takashi Fukao & Ken Katagiri (1971).

Results.—When 0·05 ml dextran (molecular weight 67,000) was injected into the plantar region of the foot, a dose-response relationship of the oedema was established between the dose range 10 μg and 25 mg (Fig. 1). When calcium was included in the dextran solution the response was reduced; 0·05 ml of 5 mM calcium shifted the dextran curve to the right at all dose levels tested, but 0·05 ml of 50 mM only exerted an inhibitory effect on the lower dose levels of dextran (Fig. 1). The dextran response was not affected by 0·05 ml of 0·5 mM calcium; and 0·05 ml of 5 mM and 50 mM calcium did not increase the weight of the rat's foot. Doses in excess of 100 mM produced gross oedema.

Similar results were found in the skin. A dose-response curve to intradermal dextran was obtained using doses of 10 μg up to 1 mg. The response to dextran was inhibited by 0·05 ml of 5 mM calcium, a dose which failed to modify the histamine (50 μg) and 5-HT (1 μg) responses.

Discussion.—The results show that low doses of calcium inhibit anaphylactoid oedema produced by dextran in both skin and paws of rats. Greaves & Mongar (1968) found that, when the concentration of calcium was increased above the physiological range (2–5 mM), the *in vitro*

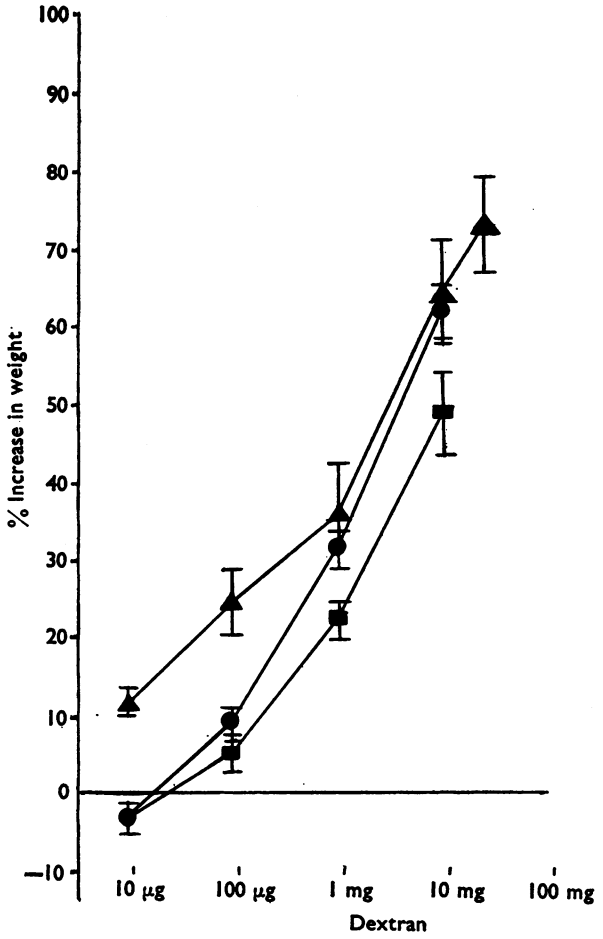

FIG. 1. Effect of calcium on the increased vascular permeability induced by dextran in rat paws. Responses measured as percentage increase in weight (±S.E.M.) of treated foot over weight of contralateral foot. Abscissa, dose of dextran. ▲——▲, Dextran alone ; ■——■, dextran plus 0·05 ml of 5 mM calcium ; ●——●, dextran plus 0·05 ml of 50 mM calcium.

anaphylactic release of histamine from basophils, but not from mast cells was inhibited. Furthermore, Lichtenstein & Osler (1964) reported that high calcium concentrations inhibit anaphylactic release of histamine from human leucocytes.

Recently, Burgen & Spero (1970) showed that raised calcium concentrations inhibited the response of the guinea-pig ileum to carbachol. Inhibition of the dextran response by calcium might therefore involve either a reduced amine release by dextran, or a diminished response of the vascular smooth muscle to the amines released by dextran. The fact that calcium does not reduce the responses to exogenous histamine or 5-HT suggests that inhibition involves reduced output of amines.

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REFERENCES

BURGEN, A. S. V. & SPERO, L. (1970). The effects of calcium and magnesium on the response of intestinal smooth muscle to drugs. *Br. J. Pharmac.*, **40**, 492–500.

GREAVES, M. W. & MONGAR, J. L. (1968). In: *Immunopharmacology*, p. 50. London: Pergamon Press.

HARRIS, J. M., LUSCOMBE, D. K. & POYSER, R. H. (1967). The influence of molecular weight and structure on the vascular permeability responses induced by glucose polymers in rat skin. *Br. J. Pharmac. Chemother.*, **29**, 16–24.

LICHTENSTEIN, L. M. & OSLER, A. G. (1964). Studies on the mechanisms of hypersensitivity phenomena. IX. Histamine release from human leucocytes by ragweed pollen antigen. *J. exp. Med.*, **120**, 507–530.

MINORU HORADA, MITSUO TAKEUCHI, TAKASHI FUKAO & KEN KATAGIRI (1971). A simple method for the quantitative extraction of dye extravasated into the skin. *J. Pharm. Pharmac.*, **23**, 218–219.

MONGAR, J. L. & SCHILD, H. O. (1958). The effect of calcium and pH on the anaphylactic reaction. *J. Physiol., Lond.*, **140**, 272–284.

PARRATT, J. R. & WEST, G. B. (1957). 5-Hydroxytryptamine and the anaphylactoid reaction in the rat. *J. Physiol., Lond.*, **139**, 27–41.

VOORHEES, A. B., BAKER, H. J. & PULASKI, E. J. (1951). Reactions of albino rats to injections of dextran. *Proc. Soc. exp. Biol.*, **76**, 254–256.

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