Br. J. Pharmac. (1983), **80**, 7–9

Anaphylactic contraction (Schultz-Dale reaction) of the bovine bronchial artery *in vitro*

D.I. Alexander, P. Eyre & V.P. Gordon

Department of Biomedical Sciences, Ontario Veterinary College, University of Guelph, Guelph, Ontario N1G 2W1, Canada

Bronchial arteries from sensitized calves contracted on antigenic challenge in the absence and presence of selected anti-allergic drugs and autonomic agents *in vitro*. Anaphylactic reactivity was inhibited by tripelennamine, isoprenaline and aminophylline, and potentiated by indomethacin and dopamine. Disodium cromoglycate, compound FPL 55712 and ketanserin failed to inhibit anaphylactic contraction.

Introduction Considerable interest has been focused on the Schultz-Dale model of Type-I hypersensitivity which occurs in a diversity of animal tissues (Chand & Eyre, 1978). It is a local anaphylactic response initiated when mast cell-bound reaginic antibodies are cross-linked by sensitizing antigen, leading to release of various chemical mediators (Chand & Eyre, 1978).

The reactivity of the bronchial artery to autonomic and autacoid agents and Schultz-Dale reaction in bovine pulmonary vessels were previously reported (Eyre, 1970; 1971b; Arowolo & Eyre, 1979; 1980).

There have been several suggestions that the bronchial artery may participate in obstructive pulmonary disease (Deletonia de la Mata & Aviado, 1961; McLaughlin, Tyler & Canada, 1961; Aviado, 1965; Holm, 1972; Arowolo & Eyre, 1979; 1980), i.e. local inflammation might occlude the vessel and reduce the blood supply (oxygen and nutrients) to the airways and parenchyma.

This paper describes bronchial arterial constriction *in vitro* and its modification by drugs.

Methods Male Jersey calves (1–3 months old) were sensitized to horse serum as previously described (Eyre, Lewis & Wells, 1973). Calves were anaesthetized and killed with sodium pentobarbitone (100 mg kg^{-1}).

Lungs, with heart and aorta intact, were removed and the extrapulmonary bronchial artery was removed as previously described (Arowolo & Eyre, 1979; 1980), trimmed and cut spirally into a single strip (Furchgott & Bhadrakom, 1953; Eyre, 1971b). This was bisected longitudinally into twin strips, each of which was suspended in 10 or 20 ml Krebs-Henseleit (1932) solution at 37°C and bubbled with 95% O_2:5%CO_2 at a tension of 2.0 g and attached to an isotonic lever and myograph-transducer unit. Movements were recorded on a 4-channel physiograph (Model DMP-4A or 4B, Narco Biosystems Inc., Houston, Texas). Control tissues (unsensitized) were obtained from a local abattoir and similarly treated.

Tissues were equilibrated for 60 min; the solution being changed every 20 min. Five or six point cumulative dose-response curves to histamine were established in both strips of each pair (Van Rossum, 1963). After washing, dose-response curves to histamine were again re-established (i) in one strip in the presence of the chosen antagonist to be tested on the Schultz-Dale reaction, and (ii) in the second strip in the absence of antagonist. After 10 min, both strips were challenged with horse serum in cumulative doses of 0.1, 0.2 and 0.3 ml. Antigen-induced contractions were expressed as a percentage of the histamine maximum for each tissue. Means and standard errors were calculated and significance was estimated between control ($n = 30$) and treated tissues ($n = 5$ for each drug) using Student's t test. A value of $P < 0.05$ was considered significant. Drugs employed were: aminophylline (Cyanamid, Montreal, Quebec), dopamine HCl (Nutritional Biochemicals Corp., Cleveland, Ohio), disodium cromoglycate and FPL 55712 (sodium 7 - [3(4 - acetyl - 3 - hydroxy - 2 - propylphenoxy) - 2 - hydroxy propoxy] - 4 - oxo - 8 - propyl - 4H - 1 - benzopyran - 2 - carboxylate, Fisons, Loughborough), indomethacin (Merck, Sharpe and Dohme, Pointe Claire, Quebec), ketanserin (R 41468) (Janssen Pharmaceuticals, New Brunswick, New Jersey), isoprenaline (isoproterenol) HCl (Sigma Chemical Co., St. Louis, Missouri), tripelennamine HCl (Ciba-Geigy, Dorval, Quebec).

Results A response to antigen occurred in all sensitized tissues between 2.5 and 10 min after antigen exposure. Antagonists did not alter this time of onset. The mean response to antigen in untreated vessels was 36% of the tissue histamine maximum. Control tissues did not respond to antigen. Treatment with tripelennamine 10^{-6} M reduced the Schultz-Dale reaction to 4.48% histamine maximum. Indomethacin 10^{-6} M significantly potentiated the anaphylactic reaction to 67.1% histamine maximum.

Incubation with disodium cromoglycate (100 μg ml^{-1}), ketanserin (10^{-6} M), FPL 55712 (10^{-6} M), dopamine (10^{-9} M) or isoprenaline (10^{-9} M) had no significant effect on the anaphylactic response. However, dopamine, 10^{-8} M, produced significant enhancement and isoprenaline, 10^{-8} M, caused significant inhibition, as did aminophylline (10^{-9} M).

Discussion Histamine may play a role in the allergic response of this tissue, as tripelennamine (10^{-6} M) significantly suppressed it. Previous studies (Arowolo & Eyre, 1980) reported the reactivity of the bronchial artery to histamine, and the immunological release of histamine from calf lung (Eyre, 1971c: Eyre *et al.*, 1973). In addition Eyre & Deline (1971) described 5-hydroxytryptamine (5-

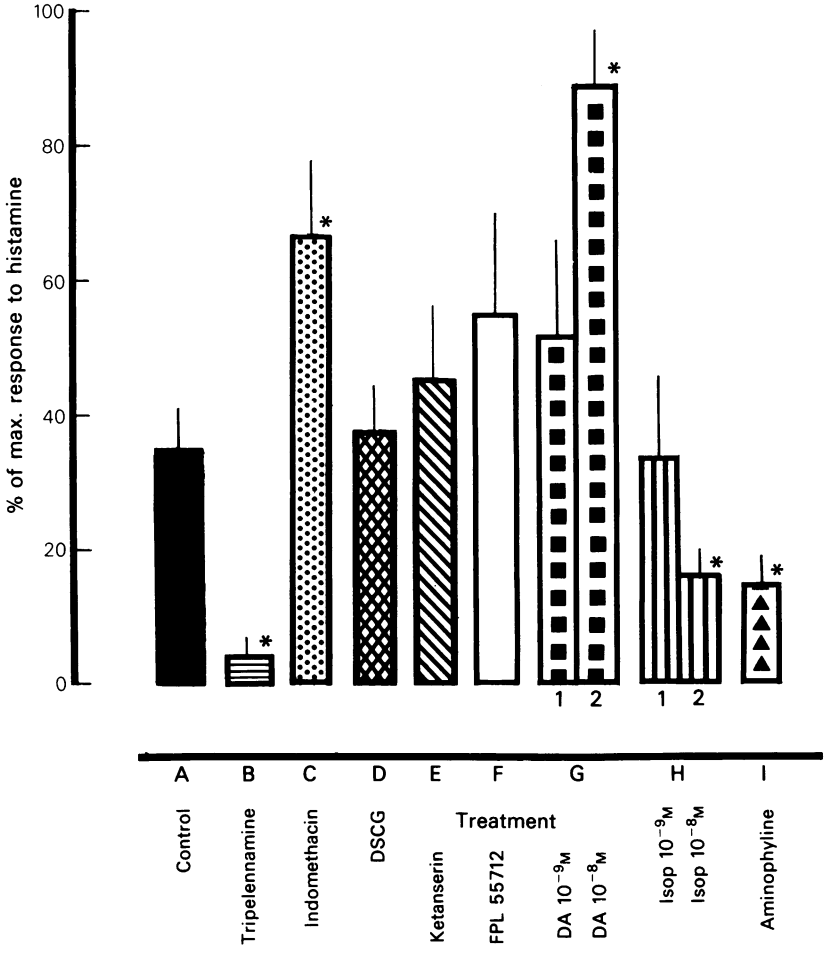

Figure 1 Anaphylactic reactions of bovine isolated bronchial artery to horse serum in Krebs-Henseleit solution at 37°C, expressed as a percentage(%) of the maximum tissue response to histamine. (A) nontreated control; pre-treated with (B) tripelennamine 10^{-6} M; (C) indomethacin 10^{-6} M; (D) disodiumcromoglycate 100 μg ml^{-1}; (E) ketanserin (R 41468) 10^{-8} M; (F) FPL 55712 10^{-6} M; (G1) 10^{-9} M dopamine; (G2) 10^{-8} M dopamine; (H1) 10^{-9} M isoprenaline; (H2) 10^{-8} M isoprenaline; (I) aminophylline 10^{-9} M. All values are expressed as mean with s.e. mean shown by vertical lines; significance (*) was determined by Student's *t* test at $P < 0.05$.

HT) release from bovine lung during anaphylaxis. Although the reactivity of the bronchial artery to 5-HT has been described (Arowolo & Eyre, 1980) no evidence was obtained in this study to suggest that 5-HT is important in bronchial vascular anaphylaxis, as ketanserin failed to inhibit the response.

The potentiation of anaphylaxis by indomethacin may result from blockade of an inhibitory prostaglandin. Alternatively, indomethacin, by inhibiting prostaglandin synthesis, may augment the lipoxygenase pathway, resulting in increased leukotriene production. If so, it is surprising that compound FPL 55712, a selective leukotriene (SRS-A) antagonist, failed to suppress the anaphylactic reaction. Further studies should clarify the role of prostanoids in the vascular Schultz-Dale response. The failure of disodium-cromoglycate to inhibit the allergic reaction confirmed an earlier finding by Eyre *et al.*, (1973) in calves *in vivo*.

Eyre (1971a) reported that dopamine was released by antigen from bovine lung. Burka, Eyre & Holroyde (1976) showed that dopamine enhanced histamine and SRS-A release from calf lung: a positive feedback mediated by dopamine receptors. Potentiation of the Schultz-Dale response by dopamine may operate through this mechanism, and may thus contribute to pulmonary disease in ruminants (Burka *et al.*, 1976). Inhibition by aminophylline and isoprenaline confirmed earlier studies (Castillo, 1948: Lichtenstein & Margolis, 1968).

The Schultz-Dale reaction certainly cannot be said to represent all that may be happening in the bronchial artery *in vivo*. Nevertheless, it is interesting that this blood vessel, taken from a sensitized animal, constricts when exposed to specific antigen. If the bronchial artery contracted *in situ* during hypersensitivity states, some degree of hypoxia of the pulmonary tissues would seem inevitable. Persistent occlusion of the blood vessel could thus contribute to clinical and pathological changes and future investigations should include consideration of the pathophysiology of the bronchial arterial circulation in the aetiology of pulmonary disease.

Ketanserin (R 41468) was a gift from Janssen Pharmaceuticals (New Brunswick, New Jersey, U.S.A.) and FPL was a gift from Fisons, Loughborough, U.K., for which the authors are grateful.

References

ALEXANDER, D.I. (1982). *Autonomic pharmacology of the bovine bronchial artery* in vitro. MSc Thesis, University of Guelph.

AROWOLO, R.A.O. & EYRE, P. (1979). Pharmacological comparison of the isolated bronchial artery and pulmonary vein: possible functional relationships. *Res. Comm. Chem. Path. Pharmac.*, **26**, 227–241.

AROWOLO, R.A.O. & EYRE, P. (1980). Preliminary pharmacological characterization of the bovine bronchial artery strip: a new preparation. *Br. J. Pharmac.*, **68**, 283–288.

AVIADO, D.M. (1965). *The Lung Circulation* 1st ed., Vol. 1 & 2. New York; Pergamon Press.

BURKA, J.F., EYRE, P. & HOLROYDE, M.C. (1976). Dopaminergic (co-mediator) modulation of release of histamine and SRS-A in the calf. *Br. J. Pharmac.*, **58**, 445P.

CASTILLO, J.C. (1948). The tracheal chain II – the anaphylactic guinea pig trachea and its response to antihistamine and bronchodilator drugs. *J. Pharmac. exp. Ther.*, **94**, 412–415.

CHAND, N. & EYRE, P. (1978). The Schultz-Dale reaction: a review. *Agents and Actions*, **8**, 421–430.

DELETONIA, S.M.L., DE LA MATA, R.C. & AVIADO, D.M. (1961). Local and reflex effects of bronchial arterial injection of drugs. *J. Pharmac. exp. Ther.*, **133**, 295–303.

EYRE, P. (1970). The Schultz-Dale reaction in bovine pulmonary smooth muscles. *Br. J. Pharmac.*, **40**, 166P

EYRE, P. (1971a). Release of dopamine from bovine lung by specific antigen and by compound 48/80. *Br. J. Pharmac.*, **42**, 423–427.

EYRE, P. (1971b). Pharmacology of bovine pulmonary vein anaphylaxis *in vitro*. *Br. J. Pharmac.*, **43**, 302–311.

EYRE, P. (1971c). Histamine release from calf lung *in vitro* by specific antigen and by compound 48/80. *Archs int. Pharmacodyn. Ther.*, **192**, 347–352.

EYRE, P. & DELINE, T.R. (1972). Release of 5-Hydroxytryptamine from Sensitized Calf Lung by Antigen and by Compound 48/80. *Archs int. Pharmacodyn. Thér.*, **199**, 245–252.

EYRE, P., LEWIS, A.J. & WELLS, P.W. (1973). Acute anaphylaxis in the calf. *J. Pharmac.*, **47**, 504–515.

FURCHGOTT, R.F. & BHADRAKOM, S. (1953). Reactions of strips of rabbit aorta to epinephrine, isopropyl arterenol, sodium nitrite and other drugs. *J. Pharmac. exp. Ther.*, **108**, 129–143.

HOLM, W.W. (1972). The respiratory distress syndrome: bronchial arteries and catecholamines. *Pediatrics*, **50**, 973–974.

KREBS, A.J. & HENSELEIT, K. (1932). Untersuchungen Uber Die Hainstaffbildung in Tiekorper. *Hoppe-Seylers Z. Physiol. Chem.*, **210**, 33–36.

LICHTENSTEIN, L. & MARGOLIS, S. (1968). Histamine release in vitro: inhibition by catecholamines and methylxanthines. *Science*, **161**, 902–903.

McLAUGHLIN, R.F., TYLER, W.S. & CANADA, R.O. (1961). A study of the subgross pulmonary anatomy in various animals. *Am. J. Anat.*, **108(2)**, 149–165.

VAN ROSSUM, J.M. (1963). Cumulative dose-response curves II. Techniques for the making of dose response curves in isolated organ baths and the elevation of drug parameters. *Archs int. Pharmacodyn. Thér.*, **143**, 299–330.

(Received April 29, 1983.)