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MECHANISMS OF FORSSMAN-INDUCED BRONCHOSPASM AND THEIR INHIBITION

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1 The bronchospasm induced in the guinea-pig by the injection of Forssman antiserum was biphasic in nature in both the sublethal and the lethal reaction.

2 The development of both phases of the bronchospasm in the sublethal reaction was dependent upon the presence of the intact complement system and circulating platelets. In the lethal reaction the phase II bronchospasm did not appear to depend on these factors.

3 The compounds used in this study inhibited phase I bronchospasm of the sublethal reaction in the order, methysergide > indomethacin > aspirin = sulphinpyrazone and phase II in the order, indomethacin > sulphinpyrazone > aspirin. Methysergide was inactive.

4 Aspirin, indomethacin and sodium salicylate all prevented the inhibitory action of sulphinpyrazone in reducing the phase II bronchospasm of the sublethal reaction in the order, indomethacin > sodium salicylate > aspirin, when the drugs were administered prior to sulphinpyrazone.

5 The inhibitory action of aspirin on the sulphinpyrazone effect could be prevented by administering sulphinpyrazone before aspirin. All drug-induced inhibitions of sulphinpyrazone by aspirin, indomethacin and sodium salicylate were dose-dependent.

Introduction

Forssman shock is an acute lethal syndrome provoked in normal guinea-pigs by the intravenous injection of a rabbit antiserum raised against sheep erythrocyte stroma. Within a few minutes after challenge the animals show signs of profound respiratory distress culminating in violent convulsions and death. The lungs of the animals appear oedematous and haemorrhagic with fluid often filling the respiratory passages. Not all of the pathological events responsible for the death of the animal are clear but some contributing factors are understood. The rabbit antiserum contains antibodies which cross-react with antigens situated on or near the vascular endothelium of the lungs (Tanaka & Leduc, 1956) and the activation of complement subsequent to the injection of Forssman antiserum appears to be important for the development of the reaction, since death only occurs in animals with an intact complement system (May & Frank, 1972). In addition there is considerable evidence that platelets play a vital role for the full development of the lethal (Tsai, Taichman, Pulver & Schonbaum, 1973) and sub-lethal syndrome (Butler & White, 1979).

The biphasic bronchospasm associated with the lethal reaction induced by high doses of antiserum has been studied previously (Pelczarska & Roszowski, 1973). These workers suggested a pharmacological mediation of phase I, and that phase II bronchospasm resulted from mechanical factors (e.g. pulmonary oedema and haemorrhage of the small blood vessels in the lungs). In this investigation we have mainly studied the non-lethal reaction induced by low doses of antiserum since this experimental situation appears less complicated with respect to the pathological changes (Baker, Bullock, Butler, White and Williamson, unpublished observations) and open to more certain interpretation.

Methods

Guinea-pigs (Dunkin-Hartley, 300 to 400 g) were obtained from Hacking and Churchill, Huntingdon.

Guinea-pigs genetically deficient in the C_4 component of the complement system were obtained from a breeding colony maintained in our own laboratories.

Bronchospasm was measured in guinea-pigs (300 to 400 g), anaesthetized with sodium pentobarbitone (50 mg/kg i.p.), by the method of Mathe, Strandberg & Fredholm (1972) and recorded via a pressure transducer on a Devices 4 channel recorder. Animals were ventilated with a constant volume respirator at 40 strokes/min with a 5 ml stroke volume. All injections were given through a cannula in the external jugular vein.

The platelet count was determined as described by Butler, Pay, Roberts and White (1979).

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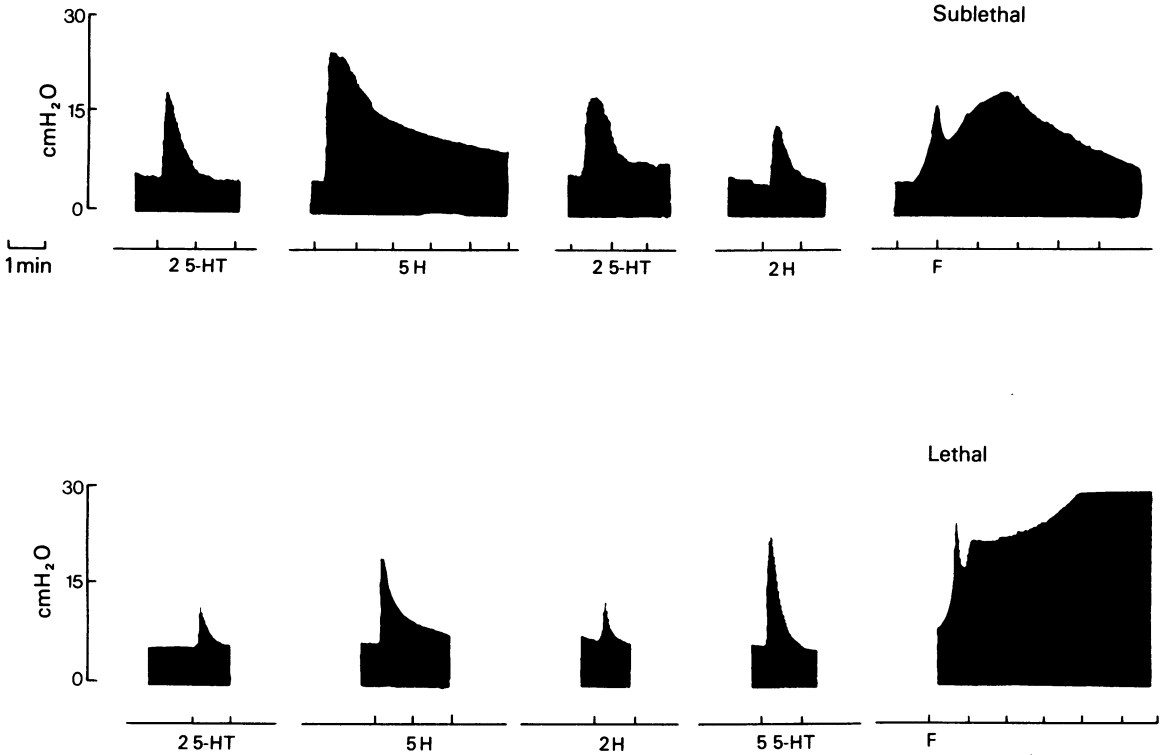

Figure 1 Typical traces of the bronchial pressure response of the guinea-pig to intravenous injections of 5-hydroxytryptamine (5-HT), histamine (H) and anti-Forssman antiserum (F). The autacoid doses are shown in μg and the dose of antiserum used was 0.2 ml (sub-lethal) and 0.5 ml (lethal). Increases in pressure were measured as cmH_2O. The time intervals start at the time of injection of the drug or antiserum.

Forssman antiserum

New Zealand White rabbits were immunized with four simultaneous 1 ml subcutaneous injections of sheep erythrocyte stroma (Cordis Laboratories) suspended in Freund's complete adjuvant (Difco). Two weeks later two identical booster injections were given subcutaneously. After a further 5 weeks the animals were bled and serum was prepared, pooled, divided into 5 ml aliquots and stored frozen at −20°C.

Antiplatelet antiserum

Blood was collected from guinea-pigs by cardiac puncture into 10 ml syringes containing 1 ml 3.8% trisodium citrate and mixed by inversion. The blood was centrifuged at 200 *g* for 15 min at 4°C to obtain platelet-rich plasma (PRP). The PRP was removed, the platelet count determined and then the PRP centrifuged at 400 *g* for 20 min at 4°C to obtain a platelet pellet. The plasma was removed and replaced by an equal volume of physiological saline. The platelets were then resuspended in saline, centrifuged and then resuspended in fresh saline to give a platelet concentration of 10^{10} cells/ml. The platelets were then disrupted by freezing and thawing three times and the material was used to immunise rabbits after suspending 1 ml in 3 ml Freund's complete adjuvant. An immunisation procedure identical to that described above for the preparation of Forssman antiserum was used.

Drugs

The following drugs were used: histamine acid phosphate, 5-hydroxytryptamine (serotonin) creatinine sulphate and salicyclic acid (BDH), aspirin (acetylsalicyclic acid), sulphinpyrazone (Geigy Pharmaceuticals), indomethacin (Merck, Sharpe & Dohme), cobra venom factor (Cordis Laboratories) and neuraminidase (Sigma).

Aspirin (1 g) and sulphinpyrazone (1 g) were dissolved in an equivalent of 1N NaOH, adjusted to pH 7.4 and made up to 10 ml with physiological saline to give a final concentration of 100 mg/ml. Indomethacin (70 mg) was dissolved in 1N NaOH (2.1 ml) adjusted

Table 1 Increase in pulmonary resistance (cm of H_2O) to intravenous histamine, 5-hydroxytryptamine and anti-Forssman antiserum in control and genetically C_4-deficient guinea-pigs

Experiment	*No. of animals*	*Histamaine* 2 μg	*5-HT* 2 μg	*Forssman reaction* Phase I	Phase II
Control animals (0.2 ml antiserum)	31	6.6 ± 0.7	7.5 ± 0.7	6.5 ± 0.8	11.4 ± 0.7
C_4 deficient animals (0.2 ml antiserum)	9	6.2 ± 1.7	12.3 ± 2.3	6.1 ± 1.9	0.33 ± 0.33*
Control animals (0.5 ml antiserum)	6	4.1 ± 1.3	5.9 ± 1.4	5.7 ± 1.3	12.9 ± 0.9

Values are mean ± s.e. mean.
*Denotes a statistically significant difference from the corresponding value in the control group given 0.2 ml antiserum: $P \leq 0.05$.

Table 2 The effects of various drug treatments on the bronchospasm induced by intravenous administration of a sub-lethal dose (0.2 ml) of anti-Forssman antiserum

Compound	*Dose*	n	*% Inhibition* *Hist*	*5-HT*	*Forssman* Phase I	Phase II
Neuraminidase	2 u/kg	5	33*	69*	100*	100*
Antiplatelet antiserum	0.5 ml/kg	5	35*	37*	100*	100*
Cobra venom factor	200 u/kg	5	0	15	100*	100*
	(mg/kg)					
Methysergide (a)	0.3	5	0	100*	100*	0
	1	7	14	100*	100*	22
	3	5	0	100*	100*	0
Mepyramine (a)	0.3	5	100*	49*	31	33*
	1	7	100*	38*	100*	64*
Aspirin (b)	30	5			6	11
	100	17	0	0	79*	33*
Indomethacin (b)	1	5	0	0	0	0
	10	5	0	0	68*	71*
Sulphinpyrazone (b)	10	5	0	0	0	3
	30	17	0	0	0	71*
	50	5	0	0	0	95*
	100	17	0	0	62*	61*
Sodium salicylate (b)	100	5	0	0	0	0

n denotes the number of animals used in each group and the results are expressed as mean percentage inhibition. The values for histamine (Hist) and 5-hydroxytryptamine (5-HT) were based on the response to a 2 μg dose (being approximately the ED_{50}). The standard errors for the means were of the same order as shown in Table 1 for the 0.2 ml antisera dose. The mean percentage inhibition of histamine and 5-HT was assessed by comparison of the autacoid response before and after drug treatment, except with the results following neuraminidase, cobra venom factor and antiplatelet antiserum which were compared with values in control animals (see Table 1). Inhibition of the phases of Forssman bronchospasm were assessed in all cases by comparison with the control results for the sublethal reaction shown in Table 1.
*denotes a statistically significant inhibition: $P \leq 0.05$.
(a) denotes drug administered i.v. 15 min before Forssman antiserum; (b) denotes drug administered i.v. 40 min before Forssman antibody.

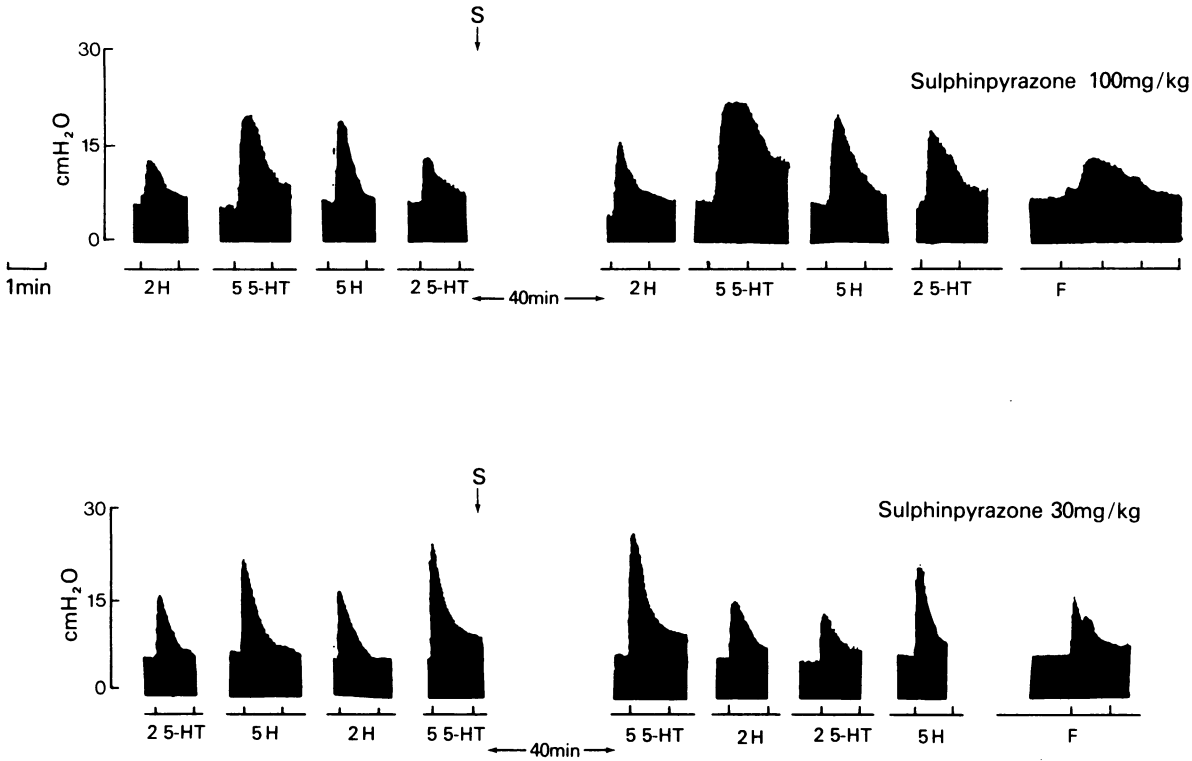

Figure 2 Typical traces of the bronchial pressure response of the guinea-pig to intravenous injections of 5-hydroxytryptamine (5-HT) and histamine (H) before, and 5-HT, histamine and anti-Forssman antiserum (F) after the intravenous administration of sulphinpyrazone (S). The autacoid doses are shown in μg and the dose of antiserum used was 0.2 ml. Increases in pressure were measured as cmH_2, and the time scale starts at the time of injection of either autacoid or antiserum.

to pH 7.4 and made up to 7 ml with saline to give a final concentration of 10 mg/ml.

All other drugs were dissolved in physiological saline.

Results

Changes in pulmonary pressure

In normal guinea-pigs, anaesthetized and artificially ventilated as described, an intravenous injection of 0.2 ml (sublethal) Forssman antibody resulted in a biphasic increase in pulmonary pressure (Figure 1). The initial increase (phase I) occurred within 30 s and was followed by a second increase which was maximal between 1 to 2 min after the injection of antiserum and which returned to control levels by 5 min.

A lethal reaction was induced by increasing the dose of antiserum used to 0.5 ml. A biphasic bronchospasm similar in magnitude to that found in the sublethal reaction was observed but the animals given the higher dose died before the second phase was reversed. The magnitude and variability of the increase in pulmonary pressure in response to Forssman antiserum, histamine and 5-hydroxytryptamine (5-HT) are shown in Table 1.

The demonstration that complement activation starting with the C142 complex is required for the full development of the Forssman reaction was obtained in genetically C_4 deficient guinea-pigs. When 0.2 ml Forssman antiserum was administered intravenously eight of the nine C-deficient animals studied showed a monophasic response, the remaining animal also developed a small phase II bronchospasm.

Inhibition of changes in pulmonary pressure

The influence of various drugs on the control changes in pulmonary pressure after 0.2 ml antiserum is shown in Table 2 and the basic experimental design in Figure 2. In these studies responses to doses of histamine and 5-HT giving submaximal effects were measured at approximately 5 min intervals. The compound under study was administered and the responses to the autacoids were again measured. Then the response to a

sublethal (0.2 ml) dose of Forssman antiserum was measured. Animals served as their own control for a determination of mean percentage inhibition of autacoid responses except where the animals had been treated with neuraminidase, cobra venom factor or antiplatelet antisera for several days before experimentation. In these responses, a comparison was made between the experimentally treated and the 31 untreated control animals (Table 1). Statistical analysis was performed with an unpaired *t* test.

Neuraminidase (2 units/kg) administered 24 h before or antiplatelet serum (0.5 ml/kg) administered 72, 48 and 24 h before the administration of the Forssman antiserum resulted in a reduction of the platelet count from 255 ± 10 to 26 ± 13 and 17 ± 2 × 10^3 platelets per mm^3 whole blood (mean ± s.e. of 5 experiments) respectively. These forms of pretreatment resulted in not only total abolition of both phases of the bronchospasm but also a significant reduction in the responses to histamine and 5-HT.

The administration of cobra venom factor (200 units/kg i.p.) 96, 72, 48 and 24 h before the administration of the sublethal Forssman dose did not reduce the platelet count but did reduce the circulating levels of complement to 10% of control vales. This again resulted in total abolition of both phases of the bronchospasm but did not impair the response of the animal to either histamine or 5-HT.

Of the compounds investigated methysergide, at all doses tested, selectively blocked the response to 5-HT and phase I bronchospasm.

Mepyramine was a less selective inhibitor than methysergide, inhibiting the responses to both histamine and 5-HT. Mepyramine (1 mg/kg) inhibited both phases of the Forssman bronchospasm but at 0.3 mg/kg only partially blocked phase II of the bronchospasm.

Aspirin and indomethacin inhibited both phases of the Forssman bronchospasm but only at 100 and 10 mg/kg respectively. Sulphinpyrazone also inhibited both phases (100 mg/kg) but lower doses (50 and 30 mg/kg) were only found to inhibit phase II (Figure 2). Sodium salicylate did not inhibit either phase I or phase II.

Inhibition of changes in pulmonary pressure during a lethal Forssman challenge

A summary of these limited studies is given in Table 3. The same comments as made in the previous section apply to the statistical comparisons, except that in this study the untreated control group consisted of only 6 animals (Table 1).

Administration of antiplatelet antiserum, as previously described, again resulted in the inhibition of both phases of bronchospasm and also impaired the response to 5-HT. Both neuraminidase and cobra venom factor inhibited phase II bronchospasm after a lethal Forssman challenge without impairing the autacoid response.

Of the drugs investigated in this study, only aspirin (100 mg/kg) inhibited both phases of the bronchospasm while sulphinpyrazone (100 mg/kg) and mepyramine (1 mg/kg) only inhibited phase II. Methysergide was inactive on the bronchospasm of the lethal reaction.

Interactions of aspirin, sodium salicylate and indomethacin with the effect of sulphinpyrazone

A summary of these studies in which aspirin, sodium salicylate and indomethacin were administered before sulphinpyrazone is given in Table 4. The mean percentage inhibition of the responses to the autacoids and Forssman antiserum was determined by comparison with the 31 untreated control animals (Table 1).

Table 3 The effects of various drug treatments on the bronchospasm induced by intravenous administration of a lethal dose (0.5 ml) of anti-Forssman antiserum

Compound	*Dose*	n	*% inhibition*			
			Hist	*5-HT*	*Forssman*	
					Phase I	Phase II
Neuraminidase	2 u/kg	5	0	0	23	71*
Antiplatelet antiserum	0.5 ml/kg	5	0	56*	69*	97*
Cobra venom factor	200 u/kg	6	5	20	0	52*
	mg/kg					
Methysergide (a)	1	3	67	100*	0	28
Mepyramine (a)	1	5	100*	69*	3	98*
Aspirin (b)	100	6	0	0	65*	87*
Sulphinpyrazone (b)	100	5	0	0	0	75*

Details of experimental procedure are as in Table 2

Table 4 Interaction of aspirin, sodium salicylate and indomethacin with the effect of sulphinpyrazone on the Forssman bronchospasm

Drug combination A + B	*Drug doses* (mg/kg A + B)	n	*% inhibition* *Hist*	*5-HT*	*Forssman* Phase I	Phase II
Sodium salicylate (A)	10 + 30	5	0	0	0	41*
and sulphinpyrazone (B)	30 + 30	4	0	0	0	0
	100 + 30	5	0	0	0	0
Indomethacin (A) and	0.3 + 30	5	0	0	0	44*
sulphinpyrazone (B)	1 + 30	5	0	0	0	0
Aspirin (A) and						
sulphinpyrazone (B)	1 + 30	5	0	0	0	45*
	3 + 30	5	5	32	36*	63*
	10 + 30	4	0	0	31	57*
	30 + 30	5	43*	24	71*	26
	100 + 30	4	0	0	0	0
Sulphinpyrazone (A)	30 + 100	5	0	8	0	86*
and aspirin (B)						

Drug A was administered 10 min before drug B and the antiserum given 40 min later except when sulphinpyrazone was given before aspirin. In this experiment sulphinpyrazone was given 30 min before aspirin which was followed 10 min later by the antiserum.
Percentage inhibitions were determined as described in footnote to Table 2.

Sodium salicylate (100 mg/kg) and indomethacin (1 mg/kg) did not inhibit either phase I or phase II of the Forssman bronchospasm (see Table 2). When either of these compounds was administered in these doses 10 min before the administration of sulphinpyrazone (30 mg/kg) then the inhibitory effect of sulphinpyrazone on phase II bronchospasm was not observed. However, the sulphinpyrazone effect was observed when the dose of sodium salicylate was lowered to 10 mg/kg and that of indomethacin to 0.3 mg/kg (Table 4).

The effect of aspirin on the inhibitory action of sulphinpyrazone on phase II bronchospasm was similar to that obtained with sodium salicylate; inhibition of the bronchospasm was obtained on lowering the dose of aspirin to 10 mg/kg.

The results obtained with respect to phase I bronchospasm were more complex. When some doses of aspirin (30 mg/kg or 3 mg/kg) were given before sulphinpyrazone an inhibition of phase I was observed. Since neither dose of aspirin nor the dose of sulphinpyrazone used had any effect individually this would appear to suggest synergy. However, when aspirin at 10 or 100 mg/kg was used there was no inhibition of phase I bronchospasm even though the latter dose of aspirin alone had previously been inhibitory (Table 2).

Aspirin (100 mg/kg) was not found to prevent the inhibitory action of sulphinpyrazone (30 mg/kg) on phase II bronchospasm when administered after instead of before the sulphinpyrazone.

Discussion

After a low dose of anti-Forssman antiserum the phase II bronchospasm was reversible but when the dose of antiserum used was increased to 0.5 ml then the bronchospasm became irreversible and continued until the death of the animal.

The role of complement in the generation of the Forssman reaction was demonstrated previously by May & Frank (1972) and was also confirmed by the present results obtained with the C_4-deficient and cobra venom factor (CVF)-treated animals. Animals which had previously received CVF treatment when given a sublethal dose of antiserum displayed neither a phase I nor phase II bronchospasm, while in C_4-deficient animals no phase II was observed. This would suggest that the alternate pathway through C_3 activation was adequate to account for the initial phase I bronchospasm but that the classical pathway of complement activation was required for the development of the phase II bronchospasm in the sublethal reaction. May & Frank (1972) demonstrated that the reaction following a higher dose of antiserum was not lethal in the absence of an intact complement system. However, even in CVF-treated animals given a normally lethal dose of antiserum, a phase II broncho-

spasm was still observed indicating that part of this bronchospasm was not dependent on the presence of complement in the intravascular or extravascular component.

Both neuraminidase and antiplatelet antiserum given in doses that caused a pronounced thrombocytopenia abolished the bronchospasm caused by a sublethal dose (0.2 ml) of Forssman antibody (Table 2) indicating a dependency of the reaction on the presence of platelets. Moreover, the results of drug-induced inhibition of phase I are consistent with mediation by platelet-derived 5-HT since methysergide brought about complete inhibition (Table 2) and the potency of the other inhibitory drugs was in the order indomethacin > aspirin > sulphinpyrazone, an order which applies to inhibition of collagen-induced secretion of 5-HT by platelets *in vitro* (Zucker & Petersen, 1970) and inhibition of prostaglandin synthesis (Ziel, personal communication; Ziel and Krupp, 1975; Ali, Zamecnik, Cerskus, Stoessl, Barnett & McDonald, 1977). In contrast the results of drug-induced inhibition of phase II sublethal bronchospasm followed a different pattern. Platelets again appeared to be essential but mediation by 5-HT or by prostaglandins appeared to be unimportant in view of the inability of methysergide to inhibit phase II bronchospasm and the fact that sulphinpyrazone was a more potent inhibitor even than aspirin. In this connection it is worth noting that sulphinpyrazone is capable of inhibiting the thrombocytopenia of the sublethal Forssman reaction whereas aspirin is unable to do so (Butler & White, 1979). The observation that sulphinpyrazone has also a unique ability to protect endothelial cells against damage (Harker & Ross, 1978) may provide the link between these two observations.

In the lethal reaction, treatment with neuraminidase or antiplatelet antisera both reduced phase II bronchospasm but only the antiplatelet antiserum reduced phase I. The role of platelets in the lethal reaction has already been clearly demonstrated (Taichman, Creighton, Stephenson & Tsai, 1972; Tsai *et al.*, 1973; Taichman & Tsai, 1975). However, the present results must also be considered in the light of the possibility that the antiplatelet antisera caused hypovolaemic shock and could have reduced the circulating complement levels through this mechanism. These factors might explain the differences between the results obtained with neuraminidase and antiplatelet antisera on the phase I bronchospasm in the lethal (0.5 ml) reaction.

Tsai *et al.*, (1973) and Taichman & Tsai (1975) demonstrated that non-steroidal anti-inflammatory drugs such as aspirin and indomethacin protected against death but did not prevent the platelet sequestration. The present studies showed that they also prevented the bronchospasm and the findings are consistent with the inhibition of the release of platelet mediators. Taichman & Tsai (1975) also demonstrated the presence of polymorphonuclear (PMN) leucocytes in the vasculature. In view of the findings of Henson (1970) that the presence of neutrophils during the reaction of platelets with immune complexes increased the release of vasoactive amines, it is possible that part of the drug action might be mediated via this cellular interaction. The inhibition observed by mepyramine is not easily explained but probably does not reflect a specific anti-histamine action, since Humphrey & Mota (1959) demonstrated that anti-Forssman antiserum causes capillary damage without inducing the mast cell degranulation typically found in the IgE mediated reaction in guinea-pig lung.

The main reasons for this difference in these models is that the Forssman reaction relies on the binding of IgG or IgM to or near the vascular endothelium and not to the mast cells, with the platelet and not the mast cell appearing to be the major cellular mediator in this reaction. The model described in this paper relates more closely to models of pulmonary embolism such as that described by Daly (1974) with barium sulphate-induced embolism and so probably has little relevance to clinical asthma from a mechanistic viewpoint. It may however bear some relevance to arterial thrombosis in which platelet-vessel wall interactions are considered to be prime factors in the induction of arterial thrombosis and this model does illustrate a distinction between aspirin and sulphinpyrazone with respect to the prevention of the thrombocytopenia associated with this reaction by sulphinpyrazone but not by aspirin.

Drug interactions indicating interference between salicyclic acid and aspirin or other non-steroidal anti-inflammatory drugs have been demonstrated previously (Van Arman, Nuss & Risley, 1973; Ezer, Palosi, Hajos & Szporny, 1976). Recently Lefort and Vargaftig (1978) and Vargaftig (1978) have demonstrated inhibition of the effect of aspirin by salicyclic acid consistent with inhibition at the level of the cyclo-oxygenase. This might explain the inhibitory action of sodium salicylate, aspirin and indomethacin upon the effects of sulphinpyrazone. However, since the inhibitory action of the various drugs on the effects of sulphinpyrazone are dependent upon the concentration of sulphinpyrazone present, then the question of competitive binding arises. This could have the effect of speeding the elimination of sulphinpyrazone by inhibiting protein-binding. In contrast this would not happen if sulphinpyrazone was given before aspirin.

References

ALI, M., ZAMECNIK, J., CERSKUS, A.L., STOESSL, A.J., BARNETT, W.H. & McDONALD, J.W.D. (1977). Synthesis of thromboxane B_2 and prostaglandins by bovine gastric mucosal microsomes. *Prostaglandins*, **14**, 819–827.

BUTLER, K.D., PAY, G.F., ROBERTS, J.M. & WHITE, A.M. (1979). The effect of sulphinpyrazone and other drugs on the platelet response during the acute phase of the active Arthus reaction in guinea-pigs. *Thromb. Res.*, **15**, 319–340.

BUTLER, K.D. & WHITE, A.M. (1979). Inhibition of platelet involvement in the sublethal Forssman reaction by sulfinpyrazone. In *Cardiovascular Actions of Sulphinpyrazone: Basic and Clinical Research* ed. McGregor, M., Mustard, J.F., Oliver, M.F. and Sherry, S. pp. 3–17 Miami: Symposia Specialists.

DALY, M.J. (1974). Pulmonary mechanical effects of experimental lung embolism and their modification by bronchodilator drugs in the guinea-pig. *Br. J. Pharmac.*, **51**, 599–601.

EZER, E., PALOSI, E., HAJOS, G. & SZPORNY, L. (1976). Antagonism of the gastrointestinal ulcerogenic effect of some non-steroidal anti-inflammatory agents by sodium salicylate. *J. Pharm. Pharmac.*, **28**, 655–657.

HARKER, L.A. & ROSS, R. (1979). Prevention of homocysteine-induced arteriosclerosis: sulphinpyrazone endothelial protection. In *A New Approach to Reduction of Cardiac Death*. International Symposium VIIIth World Congress of Cardiology, Tokyo, Japan. ed. Abe, T. & Sherry, S. pp. 59–71 Bern, Stuttgart, Vienna: Hans Huber.

HENSON, P.M. (1970). Mechanisms of release of constituents from rabbit platelets by antigen-antibody complexes and complement. II. Interaciton of platelets with neutrophils. *J. Immunol.*, **105**, 490–501.

HUMPHREY, J. H. & MOTA, I. (1959). The mechanism of anaphylaxis: specificity of antigen-induced mast cell damage in anaphylaxis in the guinea pig. *Immunology*, **2**, 31–43.

LEFORT, J. & VARGAFTIG, B. B. (1978). Role of the platelets in aspirin-sensitive bronchoconstriction in the guinea pig; interactions with salicyclic acid. *Br. J. Pharmac.*, **63**, 35–42.

MATHE, A. A., STRANDBERG, K. & FREDHOLM, B. (1972). Antagonism of prostaglandin $F_{2\alpha}$ induced bronchoconstriction and blood pressure changes by polyphloretin phosphate in the guinea pig and cat. *J. Pharm. Pharmac.*, **24**, 378–382.

MAY, J.E., & FRANK, M.M. (1972). Complement-mediated tissue damage: contribution of the classical and alternate complement pathways in the Forssman reaction. *J. Immunol.*, **108**, 1517–1525.

PELCZARSKA, A.B. & ROSZKOWSKI, A.P. (1973). Inhibitors of Forssman guinea-pig 'anaphylaxis'., *J. Pharmac. exp. Ther.*, **185**, 116–126.

TAICHMAN, N.S., CREIGHTON, M., STEPHENSON, A. & TSAI, C.-C. (1972). Ultrastructure of pulmonary vascular lesions produced by Forssman antiserum in guinea pigs. *Immunology*, **22**, 93–102.

TAICHMAN, N.S. & TSAI, C.-C. (1975). Platelets, drugs and intravascular immune reactions. In *Platelets, Drugs and Thrombosis*, Symposium, Hamilton 1972. ed. Hirsh, J., Cade, J.F., Gallus, A.S. & Schönbaum, E. pp. 169–181. Basle: Karger.

TANAKA, N. & LEDUC, E.H. (1956). A study of the cellular distribution of Forssman antigen in various species. *J. Immunol.*, **77**, 198–212.

TSAI, C.-C., TAICHMAN, N.S., PULVER, W.H. & SCHÖNBAUM, E. (1973). Heterophile antibodies and tissue injury. III. A role for platelets in the development of lethal vascular injury during Forssman shock in guinea-pigs. *Am. J. Path.*, **72**, 179–200.

VAN ARMAN, C.G., NUSS, G.W. & RISLEY, E.A. (1973). Interactions of aspirin, indomethacin and other drugs in adjuvant-induced arthritis in the rat. *J. Pharmac. exp. Ther.*, **187**, 400–414 (including Appendix).

VARGAFTIG, B.B. (1978). The inhibition of cyclooxygenase of rabbit platelets by aspirin is prevented by salicylic acid and by phenanthrolines. *Eur. J. Pharmac.*, **50**, 231–241.

ZIEL, R. & KRUPP, P. (1975). The significance of inhibition of prostaglandin synthesis in the selection of non-steroidal anti-inflammatory agents. *Internat. J. clin. Pharmacol.*, **12**, 186–191.

ZUCKER, M.B. & PETERSON, J. (1970). Effect of acetyl-, other non-steroidal anti-inflammatory agents and dipyridamole on human blood platelets. *J. Lab. clin. Med.*, **76**, 66–75.

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