diameter. The solution in the micropipette was connected with a Ag/AgCl electrode via an external circuit to a reference Ag/AgCl electrode immersed in the outflow of the bathing solution. A Bell stimulator in conjunction with a Devices isolated stimulator (type 2533) provided positive square wave DC pulses to the micropipette at 10 Hz, 20 ms and a current of 100 μA which were monitored by an oscilloscope in the circuit.

The amount of white body formation was assessed by counting, at a magnification of 350. the total thrombi adhering during the first 4 min after stimulation. A graded response could be produced by varying the duration of stimulation. Stimulation for 1, 2 or 4 s produced (mean ± s.e. mean (*n*)), 2.91 ± 0.54 (12), 3.8 ± 0.33 (17), 5.25 ± 0.49 (8) white bodies respectively. n refers to the number of 1, 2 or 4 s stimulations in six hamsters. The difference between the responses to 1 and 4 s were statistically significant ($P < 0.001$).

The method has so far been used to study the effects of sulphinpyrazone and aspirin which are being currently assessed in man as anti-thrombotic agents (Barnett, 1973). In addition, oxprenolol has been examined since β-adrenoceptor blockers have been reported to inhibit platelet aggregation (Bucker & Stucki, 1969).

Groups of 4-7 male hamsters, 100-120 g were given orally 18 h and 1 h before injury, one of the following treatments: 3, 10, 20 or 65 mg/kg of sulphinpyrazone (0.5 ml/100 g); 1, 10, 30, 60, 100 or 200 mg/kg of sodium aspirin (0.5 ml/100 g); or oxprenolol, 0.1, 1 or 5 mg/kg (0.2 ml/100 g).

Significant reduction in white body formation was found with all three drugs. Sulphinpyrazone produced a significant reduction for all periods of stimulation ($P < 0.001$ and $P < 0.05$) at 20 and 65 mg/kg respectively, a small reduction at 10 mg/kg and no effect at 3 mg/kg. Oxprenolol produced a significant reduction ($P < 0.05$) at 5 mg/kg but not at 1 mg/kg. Aspirin behaved differently in producing a bell-shaped dose response curve. Whereas 10 and 30 mg/kg produced a significant reduction ($P < 0.005$), both a lower dose (1 mg/kg) and higher doses (60 and 100 mg/kg) produced no significant reduction, while 200 mg/kg enhanced white body formation.

These results support the view that sulphinpyrazone is an effective anti-thrombotic agent and suggest a similar action for oxprenolol. However, the effectiveness of aspirin is variable according to dosage.

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The effect of sulphinpyrazone on the thrombocytopenia occurring in the Arthus reaction

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Sulphinpyrazone is a uricosuric agent, which, *in vitro*, inhibits platelet aggregation and release caused by a large number of agents including collagen, ADP, antigen/antibody complexes, viruses and bacteria (Mustard & Packham, 1975). The drug has also shown, in a number of clinical trials, to have anti-thrombotic activity, and to reverse shortened platelet survival (Steele, Weily & Genton, 1973; Steele, Weily, Davies & Genton, 1974). However, some *in vitro* findings suggest that the *in vivo* effect of sulphinpyrazone may not be due to its direct interference with the aggregation of platelets (Mustard & Packham, 1975). In order to investigate further the problem of the mode of action of the drug, an examination of its influence on intravascular immune platelet

aggregation and lysis occurring in the Arthus reaction in the rabbit has been commenced.

Rabbits were immunized using alum precipitated ovalbumin (1.0 mg, s.c.) 6 weeks prior to challenge with aqueous ovalbumin (10 μg or 1 mg) injected s.c. at each of six sites on the back. Thrombocytopenia, measured as a fall in the circulating level of homologous ^{51}Cr-labelled platelets injected (i.v.) 24 h earlier occurred acutely following antigenic challenge. Its magnitude varied with the antigenic dose, so that after 6 mg or 60 μg challenges, the radioactivity in whole blood fell to 24.5 ± 1.9% and 74.3 ± 6.7% of control value, respectively. Similar results were obtained by absolute cell counts but the variation between animals was larger. The thrombocytopenia persisted for 4 h after both doses of antigen. An obligatory role for complement was confirmed since depletion of complement to 10% of control level through the use of anti-complementary factor from Cobra venom was found to result not only in the abolition of the Arthus reaction but in the thrombocytopenia as well.

Sulphinpyrazone (50, and 30 mg/kg), administered as the sodium salt intravenously 1 h before challenge with 60 μg of antigen completely inhibited the thrombocytopenia. Upon challenging with 6 mg of antigen, sulphinpyrazone (50 mg/kg) restored the platelet count significantly ($P < 0.01$) to 44.5 ± 4.9% of control, but a dose of 30 mg/kg was not active. The inhibitory effects persisted for at least 4 h in all cases.

These results confirm that sulphinpyrazone can act relatively quickly to protect platelets against complement-mediated immune lysis in the Arthus reaction, an effect which is possibly analogous to its action in endotoxin shock (Evans & Mustard, 1968) and the Forsmann reaction (Tsai, Taichman, Pulver & Schönbaum, 1973). However, it remains to be established whether such an effect is due to a direct action on platelets or on the endothelium of the vasculature.

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Inhibition of 5-hydroxy-[^{3}H]-tryptamine binding to rat blood platelets by 5-HT antagonists and uptake inhibitors

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Rat blood platelets undergo a shape-change in response to sub-micromolar concentrations of 5-hydroxytryptamine (5-HT), and can also accumulate 5-HT against a high concentration gradient by active transport. We have recently shown that three [^{3}H]-5-HT binding sites exist on intact rat blood platelets (Drummond & Gordon, 1975), and in this present study we investigated the relationships between binding of the two highest affinity sites, production of the 5-HT-induced shape change and the active uptake of 5-HT. The experiments were performed with rat citrated platelet-rich plasma, and all the techniques used have been previously described in detail (Gordon & Drummond, 1974; Drummond & Gordon, 1975).

Table 1 Effects of 5-HT antagonists and uptake inhibitors on the high-affinity binding of [^{3}H]-5-HT to rat blood platelets and upon the platelet shape-change induced by 5-HT

Compound	IC_{50} value (nM) against 5-HT-induced shape-change	[^{3}H]-5-HT binding at 4° C
Pizotifen	1.2	1.3
D-LSD	5.5	1.2
Cyproheptadine	3.0	1.0
Cinanserin	2.8	1.4
Methysergide	5.5	2.0
Xylamidine	16.0	11.0
Chlorpromazine	32.0	24.0
Imipramine	250.0	100.0
Chlorimipramine	400.0	500.0
Lilly 110140 3-(p-trifluoromethylphenoxy)-N-methyl-3-phenylpropylamine	4000.0	700.0
Lilly 103947 3-(p-trifluoromethylphenoxy)-3-phenylpropylamine		