A STUDY OF THE MECHANISM OF COLLAGEN-INDUCED INTRAVASCULAR PLATELET AGGREGATION IN THE RAT

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In vitro, collagen will aggregate platelets and indomethacin will completely inhibit this aggregation. Since indomethacin prevents the synthesis of the potent aggregating agents, prostaglandin H_2 and thromboxane A_2, it is suggested that collagen acts by inducing the synthesis of these substances. The Technicon Autocounter is an automated system for counting platelets and has been modified to provide an in vivo method for the study of intravascular platelet aggregation without the interference of an anticoagulant (Smith & Freuler, 1973).

Male rats were anaesthetised with pentobarbitone (60 mg/kg IP). The trachea and jugular vein were cannulated. A double cannula, which enables only the withdrawn blood to be citrated, was inserted into the carotid artery and the blood was then pumped through the Counter to give a continuous platelet count on pre-calibrated chart paper.

Adenosine diphosphate (ADP) and collagen both cause platelet aggregation in vivo and produce dose-dependent falls in the circulating platelet count. Indomethacin was found partially to inhibit collagen-induced intravascular aggregation. When collagen, 40 ug/kg, was used the maximum inhibition was 52.2 ± 1.6% at a dose of 6 mg/kg indomethacin and 32.5 ± 2.3% at 4 mg/kg. At a dose of 80 ug/kg collagen, the maximum inhibition was only 33.7 ± 1.8% with indomethacin (4 mg/kg) whilst at 6 mg/kg it was 38.4 ± 1.4%. Indomethacin (6 mg/kg) did not inhibit ADP (25 ug/kg/min for 2 minutes) -induced aggregation. Adenosine (0.25 mg/kg/min for 10 minutes) inhibited ADP-induced but not collagen-induced aggregation. Methysergide (20 ug/kg) had no effect on collagen-induced aggregation but inhibited 5-HT-induced aggregation.

This study has shown that indomethacin inhibits low dose collagen-induced aggregation more than it inhibits high dose collagen-induced aggregation, and, that only partial inhibition occurs. This suggests that collagen exerts its pro-aggregatory effect by more than one pathway. It may be that in vivo indomethacin operates in a reversible manner and is bound to albumin (Ali & McDonald, 1980). However, Huzoor-Akbar and Ardlie (1978) and Best et al (1980) have produced evidence that in vitro low dose collagen and thrombin operate via a thromboxane dependent pathway whilst at higher doses a thromboxane **independent** pathway also operates. Kinlough-Rathbone et al (1977) suggests a third pathway exists in thrombin-induced aggregation. In this study, evidence for the involvement of ADP and 5-HT in collagen-induced aggregation has not been obtained. Using the calcium ionophore, A23,187, Kinlough-Rathbone et al (1975) have found that calcium ion has a direct effect on platelet aggregation independent of the platelet release reaction and prostaglandin synthesis. It may be that collagen, in vivo, exerts part of its effect through a third pathway which involves calcium ion and on infusion, trisodium citrate (0.5 mg/kg/min for 15 minutes) was found to inhibit collagen-induced aggregation.

Further work is in progress to show the involvement of calcium in collagen-induced intravascular platelet aggregation.

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