Mechanism of action of anti-inflammatory steroids on membrane fluidity and phospholipase activity

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A major structural feature of cell membranes is the lipid bilayer, and one of its important physical properties is its fluidity. The lipid molecules may exist either in a rigid gel-like state where they are closely packed so that little motion is possible. Alternatively they may exist in the liquid-crystalline state where molecules within the bilayer are separated so that considerable motion of the fatty acid chain occurs. As the temperature of the lipid bilayer is raised, at the transition temperature (Tc), it undergoes a sharp change from gel- to liquid-crystalline state (Chapman, 1975).

Saturated or unsaturated lecithin dispersed as liposomes can be hydrolysed by phospholipase A_2 only near the Tc (Op den Kamp, de Gier & Van Deenen, 1974). The irregularities in the packing of the lipid molecules at Tc may favour ion permeability and insertion of enzymes into the lipids. Phospholipase A_2 activity may therefore be affected not only by drugs which react directly with the enzyme, but by those which alter membrane fluidity. It was possible to distinguish between these two actions by measuring Tc using differential scanning calorimetry (DSC) or measuring the rate of hydrolysis of dipalmitoyl lecithin (DPL) liposomes by phospholipase A_2.

Since mepacrine is an inhibitor of phospholipase A_2 (Vargaftig & Dåo Hai, 1972; Flower & Blackwell, 1976) its interaction with DPL was studied by DSC. Up to 400 μg mepacrine/mg DPL did not affect Tc, but the rate of hydrolysis induced by phospholipase A_2 from pig pancreas or Naja naja venom could be inhibited by concentrations of 0.1–1 mg/mg DPL in a dose-related manner up to 100%. These findings suggest that mepacrine interact directly with the phospholipase.

Compounds known to modify membrane fluidity such as cholesterol (Ladbrooke, Williams & Chapman, 1968) were also tested. As the concentration of cholesterol was raised from 28 to 132 μg/mg DPL, the rate of hydrolysis of DPL by phospholipase A_2 was inhibited in a dose-related manner up to 100%. The same range of concentrations of cholesterol lowered the Tc.

We recently reported that arachidonic acid release from the phospholipids in fat cell ghosts is inhibited in the presence of anti-inflammatory steroids, suggesting that phospholipase A_2 was inhibited either directly or by an action of the steroids on the membrane (Lewis, Piper & Vigo, 1979). When the effect of the steroids was examined on the Tc using DSC of DPL-steroid mixtures concentrations up to 100 μg steroid/mg DPL, (i.e. much higher than used in fat cell ghosts) did not affect Tc. Furthermore, concentrations of dexamethasone, betamethasone or hydrocortisone up to 1:1 w/w (steroid/DPL) were found to be without effect on the rate of hydrolysis of DPL by phospholipase A_2. Therefore, the anti-inflammatory steroids do not appear to act either by altering membrane fluidity or by directly inhibiting the phospholipase.

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