plasma exudation, as in the guinea-pig, but that addition of PGE_1 or PGE_2 to bradykinin or histamine before i.d. injection produced marked potentiation, in doses of prostaglandin down to a few nanograms. PGE_1 and PGE_2 had similar potentiating potency, as did the analogues 16, 16-dimethyl PGE_2 and (15S)-15-methyl-PGE_2. PGA_1 and PGA_2 showed less potentiating activity. Potentiation was produced by PGD_2 and $PGF_{2\alpha}$ but only at high doses (1 μg/dose). The evidence linking potentiation with vasodilatation is as follows. Firstly, the ranking order of the exudation potentiating activity of the prostaglandins studied, correlated with their vasodilating potency. Secondly, other vasodilators, e.g. adenosine diphosphate (1 μg/dose), produced significant potentiation of histamine responses. Thirdly, PGEs, in spite of their lability, produced prolonged dilatation in skin, and when histamine was administered as a supra-injection (i.d.) 30 min after PGE_2 injection, potentiation was still in parallel with the observed increased blood flow. This was also the case with the more persistent effects of the methylated analogues. Fourthly, addition of vasoconstrictors, e.g. angiotensin II, to histamine/prostaglandin mixtures before intradermal injection reduced local blood flow and reduced exudation potentiation in parallel.

Thus it would appear that in rabbit skin, vasodilatation (perhaps by increasing transmural hydrostatic pressure gradients and vessel wall area), may be responsible for the potentiation of inflammatory exudation produced by prostaglandins.

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The effects of anti-inflammatory steroids on levels of prostaglandin in adipose tissue *in vitro*

J. CHANG*, G.P. LEWIS & PRISCILLA J. PIPER

Department of Pharmacology, Institute of Basic Medical Sciences, Royal College of Surgeons, London WC2A 3PN

Lewis and Piper (1975) have postulated that anti-inflammatory steroids inhibit the prostaglandin mediated vasodilatation accompanying lipolysis in adipose tissue by preventing the release of prostaglandin (PG). This hypothesis has now been tested further using an *in vitro* system.

Female New Zealand white rabbits were killed and the epigastric fat depots removed. The fat tissue was rinsed in Krebs medium and divided into portions (5g), chopped into pieces about 2 mm^3 and washed three times with Krebs medium. The chopped fat was placed in fresh Krebs medium to a final volume of 10 ml. Each portion was pre-incubated for 20 min at room temperature with or without the anti-inflammatory drug before the addition of $ACTH_{1-24}$ (0.1 μg/ml) and then incubated for a further 120 minutes. After incubation, the Krebs medium was separated by filtration; a fresh volume was added to the chopped fat and again filtered. The two filtrates were combined. The chopped fat was placed in ice-cold ethanol (10 ml) and both the fat and supernatant extracted into ethyl acetate. Since only small amounts of PG were released on incubation with $ACTH_{1-24}$, radioimmunoassay (Hennam, Johnson, Newton & Collins, 1974) using an antiserum which cross-reacted 100% with PGE_2 and 44% with PGE_1, was used to measure the PG content.

In order to provide further evidence of identification of the PG, pooled ACTH stimulated extracts were subjected to thin layer chromatography in the AII system. The plate was then divided into 1 cm zones, silica gel scraped off and PGs estimated on rat stomach strip, chick rectum and rat colon. The activity recovered from the plates corresponded to PGE_2.

The release of glycerol, measured by the method of Eggstein (1966), was used to monitor the lipolytic action of ACTH. The increase of

glycerol levels during ACTH stimulation was not affected by the doses of indomethacin and corticosteroids used, indicating that the anti-inflammatory drugs did not reduce lipolysis.

When the chopped fat tissue was incubated in Krebs medium alone a small amount of PGE_2 (measured as ng/5g fat tissue) appeared in the fat sediment (5.4 ± 0.4) and in the supernatant (4.9 ± 0.5).

After stimulation with ACTH there was a statistically significant increase in the PG content of the fat (13.4 ± 0.7) and of the supernatant (17.0 ± 1.3). When incubated in the presence of indomethacin (1 μg/ml), ACTH failed to cause an increase either in the fat (1.5 ± 0.3) or supernatant (2.3 ± 0.5), the PG levels remaining below control values. On the other hand, in the presence of the corticosteroid, betamethasone (10 μg/ml) the content of PGE_2 in the supernatant was lower (5.8 ± 0.8) whilst that in the fat was higher (24.4 ± 1.5) than with ACTH alone. Hydrocortisone (10 μg/ml) produced the same effect.

Thus whereas a non-steroid anti-inflammatory agent, indomethacin, reduced the total amount of PG formed during lipolysis, anti-inflammatory steroids did not reduce the total but increased the tissue/supernatant ratio. These results support the hypothesis that in rabbit adipose tissue corticosteroids inhibit the release of prostaglandins but not their synthesis.

J.C. is an M.R.C. research student.

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Effects of steroid hormones on tissue levels of prostaglandin 15-hydroxydehydrogenase in the rat

G.J. BLACKWELL & R.J. FLOWER

Wellcome Research Laboratories, Langley Court, Beckenham, Kent BR3 3BS

Blackwell, Flower & Vane (1975) showed that prostaglandin 15-hydroxydehydrogenase (PGDH) —the enzyme catalysing the initial step in prostaglandin metabolism—has a short life within the cell, and suggested that the enzyme might be under hormonal control many metabolic processes and there is already some evidence that steroids, after PGDH levels in the pregnant rabbit (Bedwani & Marley, 1975; Sun & Armour, 1974). We have now studied the tissue activity of PGDH in rats in which steroid hormone levels were altered by adrenalectomy, ovariectomy or pregnancy.

PGDH activity was estimated in the high speed supernatants of kidneys (and sometimes in lungs) by measuring the conversion of [^{3}H]-PGE_2 to its 15-keto derivative as previously described (Blackwell, Flower, Parsons & Vane 1975). Metabolism in the particle-free fractions of kidneys from adrenalectomized rats was greatly (140%) increased above control levels (see Table 1); however, after hydrocortisone hemisuccinate (two doses of 5 mg i.p., 8 h apart) PGDH activity was returned to control levels or below. A synthetic glucocorticoid, dexamethasone (two doses of 1 mg i.p., 8 h apart) also reduced the tissue levels of PGDH in control rats by about 40%.

Metabolism in high speed fractions of ovariectomized rat kidneys was again higher (116%) than in the control group. Administration of oestradiol-17β (1 mg each day for 3 days) reduced metabolism to 17% of control levels whilst progesterone (same dose) had only a very slight inhibitory effect ($< 5\%$).

None of the exogenous steroids had any direct effect on enzyme activity *in vitro*.

Metabolism of PGE_2 in the lungs and kidneys of pregnant rats varied; for example in well advanced pregnancy (day 18) metabolism by lung was higher than in the ovariectomized controls (185%). However, in rats during parturition the enzyme levels were extremely low, less than 3% of control levels. Similar effects were seen on kidney PGDH levels.

These results suggest that exogenous or endogenous steroid hormones can greatly modify PGDH levels. In particular the profound