Heberden Society

Clinical Meeting, Oxford, July 14, 1972

At a clinical meeting, held at the Oxford Polytechnic and the Nuffield Orthopaedic Centre, the following papers were given.

Effect of Some Adrenal Steroid Hormones on Skin Fibroblast Replication *in vitro*. By W. HARVEY and R. GRAHAME (Guy's Hospital Medical School, London)

We have previously drawn attention to the fact that, in patients suffering from rheumatoid arthritis, the familiar steroid-induced skin-thinning effect which occurs in those given long-term oral corticosteroid medication is absent in those treated with ACTH (Grahame, 1969). This observation is compatible with the earlier observations of Savage, Copeman, Chapman, Wells, and Treadwell (1962) and West (1961) that 'steroid bruising' is rare in ACTH-treated rheumatoid arthritis patients. This study was undertaken to investigate this apparent disparity of effect between ACTH and oral corticosteroids.

It is well established that *in vitro* corticosteroids (including synthetic analogues) inhibit fibroblast growth and replication (Berliner, Bartley, Kenner, and Jee, 1970). It follows that the 'protective' effect of ACTH could be due to a direct effect of ACTH itself, or perhaps to the effect of androgenic steroids released by the adrenal cortex in response to ACTH stimulation, since it has been shown that the skin of hirsute women is thicker and contains more collagen (Shuster, Black, and Bottoms, 1970) and that their urine contains increased amounts of testosterone (Nabarro, personal communication).

Controlled experiments were carried out to determine if such an effect occurred *in vitro*.

Human fibroblasts (Biocult BCL D2) were grown in 5 cm. disposable tissue-culture Petri dishes in an atmosphere of 95 per cent air and 5 per cent CO₂, using Eagles' Minimal Essential Medium reinforced with 10 per cent foetal bovine serum. The effects of varying concentrations of some adrenal androgens both with and without a standard concentration of cortisol were studied in terms of the rate of cell division over a 5-day incubation period.

It was demonstrated that the inhibition of growth caused by 10 µg/ml. cortisol is reversed by physiological concentrations of many of the commercially-available androgenic steroids, including testosterone, dihydroxy-testosterone, androstenedione, and dehydroepiandrosterone sulphate.

No direct effect of ACTH could be demonstrated.

Discussion

DR. J. GLYN (London) Have you used any of the non-virilizing anabolic hormones such as methandienone or nandrolone? It may be that a good case could be made out for giving these routinely to patients receiving long-term corticosteroids. Conversely, have you tried any of the other anti-inflammatory steroids such as triamcinolone, which has a slightly different effect on collagen?

DR. HARVEY We have not tried any of the other anti-inflammatory steroids, but have taken cortisol as the standard. We shall try others in due course. We are just about to start experimenting with other anabolic and non-virilizing steroids and I agree that there does seem to be a case for giving these concurrently with anti-inflammatory steroids.

DR. A. WHITE (Horsham) I find these results extremely interesting, but one thing that disturbs me slightly is that you have to go to such a high level of cortisol in the medium to reduce the growth of the fibroblasts. I should have thought that the level of cortisol *in vivo* was some two orders of magnitude less—something around 10⁻⁷ molar in water content of the fibroblasts of the skin—and this raises the possibility that, while you obviously are antagonizing something you still may not *in vivo* antagonize the effects of cortisol, which is responsible for the thinning of the skin. I say this with some feeling because we have shown in rats that anabolic steroids and testosterone do reverse many of the biochemical changes induced by a number of anti-inflammatory catabolic steroids in skeletal muscle (Bullock, Christian, Peters, and White, 1971), but they still do not change the rate of weight loss in the experimental animal.

DR. HARVEY Although the cortisol level used was higher than the *in vivo* level, I cannot agree that the inhibition we antagonized is unrelated to the thinning of skin. Current experiments, furthermore, suggest that the rates of DNA and collagen synthesis are affected by cortisol at 0.1 µg/ml., allowing direct comparison with the *in vivo* situation.

DR. M. J. O. FRANCIS (Oxford) In the last slide you stated that the levels of collagen were soluble collagen levels. I wonder what proportion of collagen in the medium was soluble?

DR. HARVEY I do not have that information.

References

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Properties of Skin Polymeric Collagen in Patients with Rheumatoid Arthritis and Normal Controls. By M. J. O. FRANCIS, A. G. MOWAT, J. ELLIS and D. C. MACMILLAN (Nuffield Orthopaedic Centre and Radcliffe Infirmary, Oxford)

The clinical impression that the skin of some patients with rheumatoid arthritis is thin and/or transparent has been