

Failure of Cortisone and Hydrocortisone to Replace Pteroylglutamic Acid and Citrovorum Factor in Microbial Growth. (20820)

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An unsuspected mechanism by which steroid hormones might influence unicellular metabolism was suggested in the reports of Gaines and her associates indicating that dehydroisoandrosterone(1) and cortisone(2) replace pteroylglutamic acid (PGA) and citrovorum factor (CF) in supporting the growth of certain lactic acid-producing bacteria. These reports conceivably offered a method of investigating steroid hormone action and, in the light of the observed effects of adrenocortical hormones in pernicious anemia(3) and in sprue(4), were of particular interest with respect to hematopoiesis. As a collateral aspect of studies on PGA metabolism in this laboratory, an attempt was made to confirm and to explore further by microbial technics the relationship between cortisone and PGA and CF.

Methods. The methods described by Gaines *et al.*(2) were duplicated as far as possible. The PGA replacing activity of cortisone and of hydrocortisone was assayed with *S. faecalis* 9790, employing the basal medium of Rabinowitz and Snell(5), and with *L. casei* ATCC 7469, employing the medium of Flynn(6). *Le. citrovorum* ATCC 8081, used in the assay of CF activity, was grown in the medium of Sauberlich and Baumann(7), substituting acid-hydrolyzed casein for part of the amino acid mixture. All organisms were grown at 37°C, and growth was measured titrimetrically in all instances after 72 hours' incubation, and turbidimetrically in some instances after 20 hours' incubation. Crystalline cortisone acetate, cortisone, and hydrocortisone†

were each tested in duplicate at least once with each organism. Alcoholic solutions of steroid were evaporated under vacuum in the assay tubes, pre-autoclaved growth media were added, and the tubes were then shaken for 30 minutes prior to inoculation. Serial concentrations of steroid from 0.1 µg/ml to 500 µg/ml were tested, although undissolved crystals of steroid remained visible in the media at concentrations of 50 µg/ml and above.

Results. No growth was observed at any concentration of cortisone acetate, cortisone, or hydrocortisone with any of the organisms tested. Furthermore, cortisone acetate at a concentration of 50 µg/ml did not potentiate the growth response of *S. faecalis*, *L. casei*, or *Le. citrovorum* to a wide range of concentrations of PGA and CF.

Summary. Cortisone and hydrocortisone, added to standard assay media, failed to support or to potentiate the growth of *S. faecalis*, *L. casei*, or *Le. citrovorum*.

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5. Rabinowitz, J. C., and Snell, E. E., *J. Biol. Chem.*, 1947, v169, 631.

6. Flynn, L. M., Williams, V. B., O'Dell, B. L., and Hogan, A. G., *Anal. Chem.*, 1951, v23, 180.

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