pensions of HeLa was constant for an hour but decreased slightly after first hour. Metabolism of two lines of HeLa cells was compared. Respiration and glycolysis of cells grown in media containing homologous human serum was higher than that of cells grown in media containing horse serum. Human HeLa cells had a metabolism like that reported for tumor tissue in that respiration was low ($Q_{O_2} = 6.0$) and aerobic glycolysis moderate ($Q^{O_2}_{L.A} = 12.0$). They differed from tumor tissue in that anerobic glycolysis was low ($Q^{N_2}_{L.A} = 5.5$) and p-phenylenediamine stimulated respiration 338% ($Q_{O_2} = 26.3$). There was no Pasteur Effect and Fermentation Excess was negative.

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Absence of Appreciable Resistance to Growth Inhibiting Action of Cortisone After Prolonged Treatment.* (22825)

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Chronic administration of nitrogen retaining hormones, to the plateaued adult Norway rat, leads to period of rapid growth followed by attainment of a new weight plateau on which there is no further growth although dosage of hormone per animal or per unit weight, is unchanged. This phenomenon of plateauing on chronic treatment with anabolic hormones appears to be identical in protein anabolic steroids(1,2) in anterior pituitary growth hormone(3-5); and in anabolic steroids plus growth hormone(6). Glucocorticoids have been shown to have a striking growth inhibiting action(7,8) due to their protein catabolic action. It would, therefore, be of interest to determine if there is a failure of constant dosage of glucocorticoid to give a constant inhibition of growth; indeed, the data of Tolksdorf *et al.*(8) suggest that this might be the case.

The present investigation was performed to further clarify possible development of resistance to growth inhibiting action of glucocorticoids. Since the time required for growth cessation, on a constant dosage of anterior pituitary growth hormone, is highly sensitive to the dose level(9), a range of dose levels was employed.

Methods. A microcrystalline suspension of cortisone acetate† (Δ^4-Pregnene-17β, 21-diol-3,11,20-trione-21-acetate) in isotonic saline was prepared by trituration with Tween-80 (polyoxyethylene sorbitan monooleate). Concentration of the suspension was adjusted to contain 3.2 mg cortisone/ml and less than 0.2 % Tween-80. After dilution, the suspension was sealed in injection vials and stored at 2 to 4°C until just before injection. Single daily subcutaneous injections were given under strictly aseptic conditions, at systematically varied sites along dorsal surface of the rat. Dosage was adjusted every 2 days

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TABLE I. Weight Gain of Cortisone Treated Young Female Rats, in Successive 10 Day Intervals, before and during Treatment.

Cortisone dose, mg/kg/day	No. of rats	Wt gain (g) 10 days pretreat.	10 day intervals of treatment 1st	2nd	3rd	4th	5th	Mean wt (g) 1st day of treatment
0	15	40.9 ± 2.0*	35.3 ± 1.7	27.0 ± 1.6	19.2 ± 1.3	12.1 ± 0.9	12.3 ± 1.1	98.4
8	8	38.3 ± 2.8	17.0 ± 2.7	17.0 ± 2.3	14.5 ± 2.1	6.7 ± 1.8	5.2 ± 1.4	99.0
16	8	40.6 ± 2.6	5.7 ± 2.2	12.1 ± 2.8	10.8 ± 1.0	6.8 ± 1.7	6.2 ± 1.5	98.5
24	8	41.6 ± 3.1	−2.6 ± 2.8	8.9 ± 1.4	10.6 ± 1.6	3.1 ± 1.6	2.2 ± 2.2	99.6
32	8	41.9 ± 2.4	−4.1 ± 2.0	4.4 ± 2.1	6.1 ± 2.0	8.0 ± 2.1	3.1 ± 1.1	99.9

* Mean ± stand. error of mean in all cases.

to maintain dosage/kg body wt constant. The rats were rewarded with 0.5 ml imitation maple syrup after each injection. Split litter controls were given the same amount of syrup, but were not given injections of the suspending medium since previous work in this laboratory has shown 1 cc/day of 0.2% Tween-80 to be devoid of effect on growth. In experiments reported on anabolic hormones, stanolone (Androstane-17β-ol-3-one) was suspended in saline with Tween-80 in the same manner as cortisone and adjusted to concentration of 10 mg stanolone/ml. Ant. pituitary growth hormone was prepared in this laboratory by modification of the method of Wilhelmi, Fishman and Russell(10). The growth hormone was dissolved in sodium carbonate-saline at pH 9.5; adjusted to growth hormone concentration of 2 mg/ml; divided into injection vials and stored frozen at −30°C until just prior to injection. All experiments were carried out in the room in which rats had been raised from birth. Temperature was maintained at 22 to 24°C and all groups were fed Purina Laboratory Chow *ad lib*. The rats were an all black strain, developed in this laboratory from original Long-Evans stock. Rats on the cortisone experiment were young females with mean age of 38 days, and mean wt of approximately 100 g on the day treatment was initiated. Rats on the anabolic hormone experiments were plateaued young adult females, 8-10 months of age, having a mean wt of 250 g at onset of treatment.

Results. Table I gives growth of young female rats on cortisone treatment during 10 days prior to treatment and during 5 successive 10 day periods of treatment. The decrease in growth rate of control animals during each successive 10 day interval is the characteristic plateauing as adult size is approached. This is probably also the explanation of the successive decrease in growth in the group treated with 8 mg/kg/day. A complete cessation of growth was achieved only at the 24 mg/kg/day and 32 mg/kg/day dose levels, and then only during the first 10 day period. There appears to be no meaningful increase in growth in any group after the second 10 day period on treatment. Furthermore, during the final 10 day period, neither the 24 mg/kg/day, nor the 32 mg/kg/day group achieved a growth equal to 10% of that shown by normal controls when at the same body wt.

In comparison, Table II shows the typical decrease in resumed growth of adult animals given growth promoting hormones over intervals corresponding to those used in the cortisone experiments.

Discussion. We interpret the data presented above to indicate that no appreciable loss in growth inhibiting action of glucocorticoids occurs, at any dose level utilized, within a time interval comparable to that required for cessation of the resumed growth of adult rats treated with growth promoting hormones.

Summary. Cortisone acetate was given daily at 4 dose levels, for 50 days, to young female Long-Evans rats, initially 38 days of age and weighing approximately 100 g each. The upper 2 dose levels were in the range giving nearly complete cessation of growth. In the upper 2 dose levels, there appeared to be a slight reduction in growth inhibiting action after the first 10 days, but all 4 groups still had a marked depression of growth during the 40th to 50th day of treatment. For comparison, anterior pituitary growth hor-

TABLE II. Weight Gain of Plateaued Adult Female Rats Treated with Pituitary Growth Hormone and Stanolone, in Successive 10 Day Intervals before and during Treatment.

Treatment and dose	No. of rats	Wt gain (g) 10 days pretreat.	10 day intervals of treatment 1st	2nd	3rd	4th	5th
Growth hormone, 1 mg/rat/day	8	0.9 ± 2.0*	38.6 ± 2.8	21.8 ± 4.6	−5.8 ± 5.7	−3.0 ± 3.1	−1.6 ± 1.0
Growth hormone, controls	20	0.7 ± 0.7	4.9 ± 1.3	2.8 ± 1.1	−1.2 ± 1.3	2.9 ± 1.1	−2.4 ± 1.0
Stanolone, 5 mg/rat/day	6	1.2 ± 1.6	19.3 ± 2.3	6.3 ± 1.3	6.0 ± 0.5	3.7 ± 1.6	
Stanolone, controls	18	3.1 ± 0.8	2.3 ± 0.7	0.0 ± 1.4	5.1 ± 1.1	2.0 ± 1.2	

* Mean ± stand. error in all cases.

mone and stanolone were given to plateaued adult female rats of the same strain. These hormones, at dose levels capable of giving a major growth response during the first 20 days of treatment, gave little response after the first 20 days.

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Serum Protein-Bound Carbohydrates and Lipids in Experimental Tuberculosis. (22826)

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Guinea pigs inoculated with tubercle bacilli reveal characteristic serum protein changes which resemble to a great degree those in human tuberculosis; serum gamma globulin is elevated and albumin reduced(1,2). These serum protein changes, especially the gamma globulin elevation, have been related to chronic inflammation and hypersensitivity reactions. However, similar changes occur in chronic hepatic disease. It appeared therefore desirable to further characterize the serum protein changes produced by chronic tuberculosis.

A rise of serum mucoproteins (which are alpha globulins)(3) in experimental tuberculosis(4), in human tuberculosis(5) and in experimental brucellosis(6) has been demonstrated. This rise is absent in hepatic injury (7). Since methods have become available recently to demonstrate serum protein-bound lipids(8) and carbohydrates(9) by zone electrophoresis it appeared worthwhile to study their levels in experimental tuberculosis. To accentuate the serum protein changes, animals with long standing tuberculosis were investigated. The levels were compared not only with those of healthy control guinea pigs but also with animals in which treatment with