the present data would indicate 42 mg of total glycerides per 100 ml of cells.

Hirsch recently showed that complex mixtures of lipids can be separated quantitatively by sorption and elution from silic acid columns(6). We have modified his method so that small quantities of tri-, di-, and monoglycerides can be extracted from 1 ml blood samples, and a direct chemical determination of glyceride glycerol is then performed(7).

Ponder's experience with the cytochemistry of erythrocytes of diverse size and shape led him to express the composition in terms of number of various molecules per cell(8). Taking the volume of a dog erythrocyte as 67 μ^3 and that of the human as 90 μ^3, canine erythrocytes have 0.37×10^8 and human erythrocytes have 0.54×10^8 molecules of glycerides plus NEFA per cell. This is about one-sixth the number of hemoglobin molecules. On the basis of surface areas of 127 μ^2 and 168 μ^2, the glyceride and NEFA fatty acid residues would occur about once in each 175 square Angstroms of cell surface. A surface distribution of this intensity can be compared with the 520 $Å^2$ assigned to one molecule of tetradecylsulfate for production of spheres; "lysis occurs when there is only one molecule per 44 $Å^2$ of cell surface as a minimum"(9).

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Failure to Demonstrate Anti-Hormonal Antibodies in Rats after Maximal Response to Daily Administration of Growth Hormone.* (26023)

J. D. EMERSON AND G. M. EMERSON (Introduced by S. B. Barker)
Department of Physiology, University of Alabama Medical Center, Birmingham

Nearly pure growth hormone (GH), but not pure GH, produced antibodies in rabbit and guinea pig without adjuvant(1). Other reports cited in(2) are open to question because of lack of rigor of proof of purity of GH employed. Pure GH with Freund's adjuvant produces GH-inactivating antibodies(2). Freund's adjuvant can apparently cause autoantibody formation(3), and therefore may have hapten action. Adult Norway rats treated with highly purified GH or GH-containing extracts eventually cease growing at a given dosage per unit weight irrespective of how great this dosage(4). This phenomenon (plateauing) does not result from diabetes (5), ageing of the rat(6), altered steroid production(7), or from altered cardiac output or metabolic rate(8). In the present investigation, serum from plateaued GH-treated giant rats was assayed for anti-GH activity to determine if GH antibodies, or antihormones, are involved in plateauing.

Materials and methods. Eight female Long-Evans rats, plateaued after 248 daily s.c. injections of a pH 10.5 extract of bovine anterior pituitaries(5), at a dose representing 400 mg anterior lobe tissue/kg body weight/day (equivalent to 12.8 mg GH/kg/day), served as source of "treated serum." These rats had attained a mean body weight of 633 g (2.4 times that of their controls). During the first 40 days of treatment, they gained an

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TABLE I. Effect of Serum from Plateaued Rats on Growth Response to GH-Containing Extract.

No. rats	Daily dose	10-day wt gain (g)
5	25 mg* + 1.75 ml treated serum	31.2 ± 3.3†
5	25 " + 1.75 " control "	34.4 ± 3.5
10	25 " + 1.75 " saline	30.0 ± 1.3
8	25 " undiluted	35.7 ± 2.5
10	10 " "	13.7 ± 2.2
17	None	1.9 ± .8

* Dosage is expressed as wet wt of anterior lobe tissue represented in the daily dose; 25 mg of extract is equivalent to 0.8 mg pure GH by 10-day normal rat wt gain assay.

† Stand. error of mean.

average of 113.5 g ± std. error of 4.0 g over their controls, compared to 4.0 ± 4.5 g over controls the last 40 days. These rats were exsanguinated under ether 12 hr after last injection and their serum incubated with aliquots of above pituitary extract in ratio of 1.75 ml serum (8.5% calculated serum of average donor) to 25 mg of extract (10% of daily dose per donor rat during last 40 days of treatment). After 30 min incubation at 10°C, pH 9.0 (no turbidity, possibly due to pH), the mixture was divided into injection vials and stored at –30°C until 30 min prior to injection. Mixtures of same volume of serum from control rats and extract, and of normal saline and extract were prepared and handled identically. These 3 mixtures and 2 dose levels of undiluted extract were assayed for growth potency by daily s.c. injections for 10 days in 5 groups of normal 7-mo-old Long-Evans female rats.

Results. Growth response to the GH extract was not altered significantly by "treated serum" (Table I). The much smaller response of the 10 mg group compared to any of the 25 mg groups indicates that appreciable attenuation would have been detectable. Arthus type reaction with edema, inflammation and some necrosis at site of injection occurred in all 5 rats receiving both extract and treated serum and in no rat in other groups, indicating antibodies to some of the proteins of the extract.

Discussion. The procedures employed do not detect GH-inactivating antibodies in serum of rats plateaued on high dosage of a GH-containing extract. It is conceivable that donor rats were producing GH-inactivating antibodies but at a rate such that the daily GH dosage cleared the antibody from serum for 12 or more hours. It is also possible that plateauing may be due to fixed GH-inactivating antibodies.

Summary. Serum from female rats plateaued while on treatment with a GH-containing extract (after reaching 2.4 times normal adult female size) was ineffective in attenuating GH response of previously uninjected assay rats.

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Spleen Adenosine Deaminase and Guanase Activities after Whole-Body X-Irradiation of Rats. (26024)

HERBERT J. EICHEL

Department of Biological Chemistry, Hahnemann Medical College, Philadelphia, Pa.

It has been shown(1) that the activities per mg of nitrogen (specific activity) of 2 purine-metabolizing enzymes of liver homogenate, adenosine deaminase and inosine phosphorylase, were unchanged from 16 hours to 10 days after rats were given 600 r of whole-