explained as secondary phenomena resulting from the hyperphagia.

On the other hand, the development of unquestionable obesity in some of our lesioned rats which did not gain weight more rapidly than the controls suggests that they deposited fat at the expense of other body constituents. Apparently a slowing or cessation of growth can obscure the significance of weight gain as a criterion of obesity. Cessation or diminution of growth together with obesity was observed by Hetherington and Ranson in hypophysectomized HO rats(11) and by Scow(12) in force-fed thyroidectomized rats. In both of these cases a combination of growth hormone deficiency (resulting in negative N balance) and excessive food intake could explain the phenomenon(12). The same explanation might well hold for our HO rats which did not exhibit an excessive weight gain. Support for this concept comes from the observation(13) that the anterior pituitaries of many of the lesioned rats show a striking degranulation of the acidophiles, similar to that found after thyroidectomy. This suggests that the secretion of growth hormone by these cells might be severely disturbed by hypothalamic lesions which also produce obesity.

Summary. 1. Hypothalamic obesity (HO) was produced in rats by means of electrolytic lesions. Glucose intestinal absorption determinations yielded the following glucose coefficients of absorption (GCA): Control, 114.0; HO, 107.4; HO thyroxinized, 173.5; thyroxinized, 191.6; thyroidectomized, 75.2. These results indicate that the intestinal absorption of glucose need not differ from control values in order for obesity to occur. 2. Thyroxine elevated the GCA of HO rats as well as that of unlesioned rats. Unlesioned thyroidectomized and thyroxinized rats served as method controls. 3. Observations on HO rats which did not exhibit excessive weight gain suggest that hypothalamic lesions may interfere with the secretion of growth hormone by the pituitary.

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Comparison of Effects of Various Brands of ACTH on Normal Subjects.* (19894)

HENRY D. KAINE. (Introduced by Paul Starr.)

From the University of Southern California, School of Medicine, Department of Medicine and the Los Angeles County General Hospital.

The purpose of this study was to compare the effects of several types of commercial brands of ACTH in their ability to produce a reduction in circulating eosinophils and to increase the urinary excretion of 17 ketosteroids in human subjects. Additional studies were aimed at depleting the ascorbic acid contents in the adrenal glands of hypophysectomized rats by means of injections of the same lots of ACTH as were used in the human subjects. Commercial preparations of ACTH have their dosages based on the rela-

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TABLE I. Change in Blood Eosinophil Levels Before and After Administration of the Different Brands of ACTH and Normal Saline, as Related to Specific Brands and the Individual Subjects.

Drugs		Subjects J.B.	R.C.	L.S.	L.M.	M.M.	Avg % decrease/brand
	No. of eosinophils						
Brand 1	at 0 and 4 hr →	218– 73	113– 48	138– 44	144– 50	350– 52	70
	% decrease →	75%	57%	68%	65%	85%	
2		116– 58	149– 94	84– 20	166– 78	367– 98	57.8
		50%	37%	76%	53%	73%	
3		147– 95	125– 59	84– 89	95– 33	56– 25	40.4
		35%	53%	+6%	65%	55%	
4		170– 53	111– 33	127– 43	211– 78	144– 45	67.4
		69%	70%	66%	63%	69%	
5		135– 61	175–117	92– 73	200– 94	102– 42	44.2
		55%	33%	21%	53%	59%	
Norm. saline		259–183	272–218	116–100	170–128	232–130	26.4
		29%	20%	14%	25%	44%	
Avg % decrease/subject		56.8%	50%	45%	59.9%	68.2%	

Columns of percentages at far right and at extreme bottom are the avg percentage changes in eosinophils as related to individual brands and subjects respectively. Eosinophils are expressed in total No./cc.

tive potency of their products as determined by the biological assay of Sayers, Sayers and Woodbury(1). This assay depends on the ability of intravenous ACTH to deplete the ascorbic acid content present in the adrenal glands of hypophysectomized rats. Greenspan and coworkers(2) however have noted the lack of correlation of ACTH potency as determined by the Sayers method and other adrenal measurement tests such as the maintenance test. Reinhardt and coworkers(3) have concluded that the ascorbic acid depletion test is but one effect, and as such, is an incomplete measure of the various possible responses to ACTH administration in experimental animals.

Methods. For purposes of this study, young male medical students were selected. The lots of ACTH were prepared by E. R. Squibb and Son, United Laboratories, Ltd., Wilson Laboratories, National Drug Co., and Armour Co. All ACTH preparations were made into solution just prior to use unless originally prepared in a liquid state. At intervals of no less than one week, intramuscular injections of one of the various lots of ACTH or normal saline were given to each subject. At no time were the subjects aware of the type of injection received, and eventually they all were given each lot of ACTH and normal saline. The dosage of ACTH was equivalent to 25 mg and that of normal saline, 1 cc. Eight hours prior to testing oral intake was restricted to either water or orange juice. A 2 hour urine specimen was obtained from 6 a.m. to 8 a.m., just before receiving the injection. At 8 a.m., capillary blood specimens were also obtained. Beginning 1½ hours after the injection of the unknown, another 2½-hour urine specimen was collected between 10:30 a.m. and 12 noon. At this last hour, capillary blood was again drawn. The tests performed with these collections were the 4-hour Thorn Eosinophil Test(4) and the measurement of urinary 17 ketosteroids as determined by the method of Talbot and coworkers(5). Additional studies with hypophysectomized rats were used employing the biological assay of Sayers to evaluate the ability of these lots of ACTH and the Armour standard (La-1 A) to deplete the ascorbic acid content in their adrenal glands.† A dosage of 2 gamma of ACTH 100 g rat weight was used.

Results. An average decrease in blood eosinophils of 57% or greater was obtained with 3 of the 6 products tested in the human subjects (Table I). Brands No. 1, 2, and 4 were associated with decreases of 70%, 67%, and 57% respectively. Brands No. 3 and 5, with average decreases of 40% and 44%,

† Sayers' bioassay data are the results obtained from the United Laboratories, Ltd., Pasadena, Calif., through the courtesy of Mr. Richard Bruner.

TABLE II. Changes in mg of 17 Ketosteroids/Hr Before and After Administration of Different Brands of ACTH and Normal Saline, as Related to Specific Brands and Individuals.

Drugs		Subjects J.B.	R.C.	L.S.	L.M.	M.M.	Avg change/brand
	mg of 17 keto-						
Brand 1	steroids →	.895–1.788	1.300–1.102	.846–1.156	.875–1.338	.463–.690	.361+
	Change in levels→	.893 ↑	.198 ↓	.310 ↑	.463 ↑	.227 ↑	
2		.748– .940	.849– .783	.895–1.305	.714– .937	.513–.692	.188+
		.192 ↑	.066 ↓	.410 ↑	.223 ↑	.179 ↑	
3		.719– .944	.702– .860	1.019– .952	.583– .556	.484–.645	.090+
		.225 ↑	.158 ↑	.067 ↓	.027 ↓	.161 ↑	
4		.749–1.727	.669– .938	.710–1.250	.442– .702	.513–.711	.449+
		.978 ↑	.269 ↑	.540 ↑	.260 ↑	.198 ↑	
5		.700– .929	.895– .769	.746– .778	.648–1.065	.527–.967	.208+
		.229 ↑	.126 ↓	.032 ↑	.417 ↑	.440 ↑	
Norm. saline		.540– .951	.832– .779	.780–1.003	.466– .596	.542–.498	.133+
		.411 ↑	.053 ↓	.223 ↑	.130 ↑	.044 ↓	
	Avg change/subject	.543 ↑	.001 ↑	.244 ↑	.329 ↑	.241 ↑	

Avg change in quantitative amounts is recorded in columns to extreme right and at bottom of the chart, and reflects changes among individual brands and subjects respectively. Direction of arrow indicates either a rise or fall in the change of levels.

had 3 responses which were greater than 50%, but failed to elicit greater decreases in other tests. The average decrease with normal saline was 26% with variations from 14 to 44%. All subjects demonstrated an average decrease in eosinophils of at least 45% or more with ACTH. The responses to normal saline were much less pronounced. Subject L.S., having the least response with ACTH, showed the least change with normal saline whereas subject M.M., showing the best response, almost demonstrated an ACTH-like response with normal saline.

The individual 17 ketosteroid responses as related to the specific lots of ACTH and normal saline are summarized in Table II. All the drugs, including normal saline, were associated with some varying degree of increase in levels in the urine specimens obtained after the injections were made. In one instance, the changes with normal saline exceeded those of ACTH. Considering the total amounts, the greatest differences among the ACTH preparations were within close range of each other. Although the individual subjects tended to have initial levels within a close range, they varied widely in their responses. Subject J.B. demonstrated the greatest increases, but the response to normal saline was also relatively excessive. The 17 ketosteroid levels in subjects L.M. and M.M. showed the greatest differences in responses to ACTH and normal saline. Subject R.C. demonstrated only slight changes with all products.

The results of the comparison of the ascorbic acid depletion tests as caused by the different lots of ACTH and the Armour standard are summarized in Fig. 1. All lots of ACTH possessed a varying degree of potency, but always that below the standard. Those lots associated with the most intense depletion were Brands No. 2, 3, and 5. Brands No. 1 and 4 when tested in humans showed the most marked responses with 17 ketosteroid increases and eosinopenia, but in the animal test they showed the least.

Summary. An analysis of the accumulated results indicates the following: 1. There is a statistically significant difference between ACTH and normal saline as related to induced eosinopenia. Furthermore, differences among brands are apparent with Brands No. 1, 2, and 4 producing the more profound effects. 2. 17-Ketosteroid levels are slightly increased by all the brands and also by normal saline. Although Pincus(6) has been able to produce sharp hourly rises in urinary 17 ketosteroids under situations of induced stress in man, it seems that an intramuscular injection of 25 mg of ACTH is not capable of duplicating this, at least from urine specimens collected at these

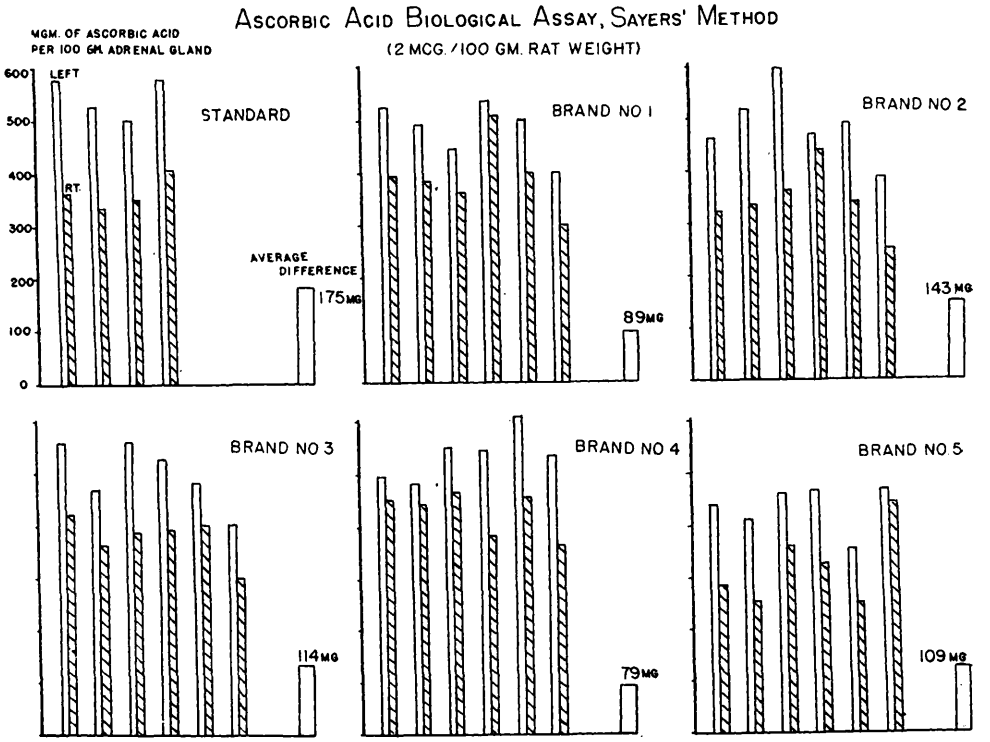

FIG. 1. Different brands of ACTH are compared to the known Armour standard by the use of the Sayers' ascorbic acid biological assay. The plain and striped paired columns indicate mg of ascorbic acid/100 g adrenal gland before and after administration of ACTH. Columns are labeled right and left, indicative of the respective glands, and depleted levels in the right adrenal glands occurred following ACTH. The other column at extreme right in each grouping shows avg differences in ascorbic acid levels of the paired adrenal glands.

time intervals. 3. There is no direct correlation among the three tests performed, but there seems to be an inverse relationship between the biological assay in rats and the tests in humans. 4. Because of this variation between animal and human responses, and the results of other investigative work as referred to above, it seems to be questionable whether the present ascorbic acid depletion animal bioassay is a completely satisfactory method of determining the degree of activity of ACTH in man.

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