Weight Increase of Body and Lymphatic Tissues in Dwarf Mice Treated with Human Chorionic Somatomammotropin (HCS) (36724)

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It has been demonstrated by several investigators that HCS shows slight but definite growth-promoting activity in the hypophysectomized rat, both in the tibia test (1–3) and the body weight gain assay (4). The activity was estimated to be at best 13% of that of HGH (2). On the contrary, Josimovich (5) did not find any somatotropic activity of HCS, but only a potentiation of the effects of HGH. In view of the possible clinical application of HCS and the marked similarity in the primary structure (6) and immunological behavior (1, 2, 4) of the two hormones, the low somatotropic activity of HCS is rather discouraging. The differences in the chemical structure could certainly suffice to explain the lower somatotropic activity of HCS. However, it is of considerable interest to establish the importance of the animal model used in the assay of growth-promoting potency, in view of the fact that the hypophysectomized rat is the only animal that has been used until now. The dwarf mouse is an excellent alternative (7). In this communication we wish to report that HCS clearly stimulates body-weight gain as well as the thymus and spleen in the dwarf mouse. It will be seen that the effect of HCS on these organs is comparable to that produced by HGH.

Materials and Methods. Human chorionic somatomammotropin was prepared as previously described (3). HCS was found to be homogeneous by gel chromatography and terminal amino acid analysis. It was shown to be biologically active in the pigeon and in the hypophysectomized rat. Human growth hormone was a gift from Dr. C. H. Li.

Thirty-day old dwarf mice of both sexes (Snell-Bagg strain, genetic symbol dw) were used. The animals were descendants of a colony originally obtained from the Jackson Laboratory, Bar Harbor, ME. Rats (Sprague-Dawley strain) raised in our colony, were hypophysectomized by the transauricular method of Falconi and Rossi (8) at 30 days of age and used 14 days after surgery.

Groups of 8 dwarf mice were injected intraperitoneally daily for 22 days, with 80 μg of HCS or HGH in 0.25 ml of saline. The total body weight of each experimental group was checked every 2 days using a Mettler P 2000N balance (precision $\pm$ 0.1 g). Controls were injected with 0.25 ml of saline. At the end of the experiment, the animals were sacrificed and the tibia width determined according to Greenspan *et al.* (9). The organs were removed, washed in saline, gently blotted with filter paper and the wet weight determined on an analytical balance.

Rats were injected daily for four days with 200 or 800 μg of HCS or HGH, dissolved in 0.5 ml of saline. At the time of sacrifice, the organs were removed and weighed. The variance analysis was performed using a computer program 101 type (Olivetti, Ivrea, Italy). The F values were taken from Snedecor and Cochran (10).

Results. As shown in Fig. 1, dwarf mice treated with HCS grew at a faster rate in comparison with animals treated with saline alone, over a period of 22 days. HGH exhibited a more potent activity; the animals treated with the same amount of this hormone attained a body weight approximately 1.5 times greater than that of mice treated with HCS. The results of the tibia test carried out on these animals at the time of sacrifice are shown in Table I. They confirm that HCS is active in dwarf mice, although to a lesser degree than HGH.

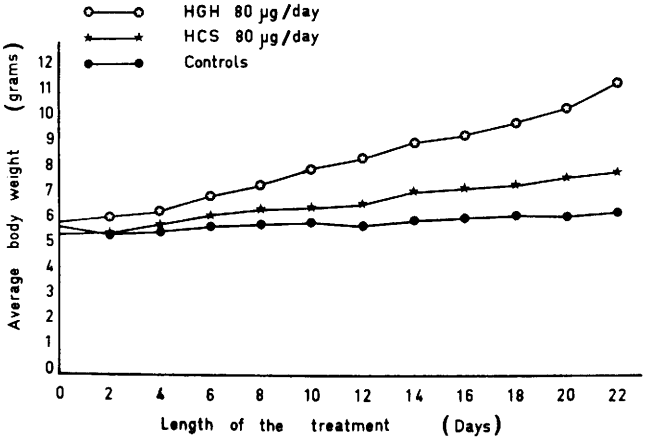

FIG. 1. Effect of HCS and HGH on the growth of the dwarf mouse.

TABLE I. Effect of HCS and HGH on the Epiphyseal Tibia Width of the Dwarf Mouse After 22 Days of Treatment.

Hormone	Total dose (μg)	Tibia width ($\mu \pm$ SE)
Saline (control)	—	144 ± 20
HCS	1760	213 ± 6[a]
HGH	1760	285 ± 21[a]

[a] The variance analysis indicates a highly significant difference in comparison with the control group ($0 \leq F \leq 0.05$).

HCS increased the weight of the spleen and thymus (Table II). HGH showed the same effects as previously demonstrated by Pierpaoli *et al.* (11). No effect was detected in the liver, kidneys or adrenals. The activity of the two hormones on the spleen and thymus is comparable, in contrast with the lower potency shown by HCS in the body-weight gain and tibia test.

The effect on the spleen was also observed in hypophysectomized rats after only 4 days of treatment (Table III).

Discussion. Florini *et al.*, (4) observed that their preparation of HCS was capable of producing an increase in the weight of the spleen in hypophysectomized rats after 14 days of treatment, with daily doses ranging from 0.250 to 16.0 mg. The same effect was also detected in normal mice. Our results, using a lower dose and in a shorter period of time, show that HCS possesses a definite activity in the spleen. This is more clearly evidenced in the experiments using dwarf mice in which both spleen and thymus showed a weight increase, but not the other organs examined. Dwarf mice appear to be more sensitive to HCS than hypophysectomized rats (the average body weight gain in the dwarf mice was 1.5 g over 5.5 g initial weight), while in the rat (4) the maximum increase was of 13.6 g over 184 g initial

TABLE II. Effect of HCS and HGH on Organ Weight in the Dwarf Mouse After 22 Days of Treatment.

Hormone	Total dose (μg)	Organs (% of body wt ± SE)				
		Spleen	Thymus	Liver	Kidneys	Adrenals
Saline (control)	—	0.15 ± 0.00	0.14 ± 0.01	5.00 ± 0.20	1.36 ± 0.15	0.022 ± 0.001
HCS	1760	0.23 ± 0.02[a]	0.26 ± 0.02[a]	4.80 ± 0.14	1.31 ± 0.04	0.027 ± 0.004
HGH	1760	0.39 ± 0.02[a]	0.26 ± 0.02[a]	5.58 ± 0.30	1.37 ± 0.03	0.024 ± 0.001

[a] The variance analysis indicates a highly significant difference in comparison with the control group ($0 \leq F \leq 0.025$).

TABLE III. Effect of HCS and HGH on Weight of Thymus and Spleen in the Hypophysectomized Rat.[a]

Hormone	Total dose in 4 days (μg)	Organ wt (mg/100 mg body wt ± SE)	
		Thymus	Spleen
Saline (control)	—	238 ± 17	220 ± 25
HCS	200	204 ± 12	355 ± 29[b]
	800	272 ± 0	513 ± 48[b]
HGH	200	191 ± 20	580 ± 79[b]
	800	299 ± 0[b]	395 ± 42[b]

[a] 8 animals/group.

[b] The variance analysis indicates a highly significant difference in comparison with the control group ($0 \leq F \leq 0.05$).

weight. These latter results were obtained using a total dose of 670 mg, in comparison with 1.76 mg of our HCS used in the present experiments.

Pierpaoli *et al.*, (11) demonstrated that the dwarf mouse presents hypoplasia of the lymphatic organs and an impaired immunological response; treatment with growth hormone was shown to correct these deficiencies. We have not established whether HCS can restore the immunological response in addition to affecting the weight of thymus and spleen. Studies on these problems are in progress.

Summary. The effect of human chorionic somatomammotropin (HCS) on the body weight gain in the dwarf mouse has been investigated. It was found that the body weight gain was lower than that observed by the same dose of HGH. On the other hand, the effect of HCS in stimulating the hyperplasia of the thymus and spleen was comparable to that of HGH.

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