Effect of human growth hormone treatment on adipose tissue in children

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Brook, C. G. D. (1973). *Archives of Disease in Childhood*, 48, 725. **Effect of human growth hormone treatment on adipose tissue in children.** Body fat and the size and number of adipose cells were determined in 22 children with growth hormone deficiency before and after 6 months of replacement therapy with human growth hormone (HGH). A marked reduction in body fat and adipose cell size was found, and the total number of adipose cells increased at the normal rate during treatment. HGH does not appear to induce catch-up growth in the number of adipose cells.

Growth hormone has long been known to cause loss of body fat (Li, Simpson, and Evans, 1949) and to mobilize free fatty acids from adipose tissue in rats (Raben and Hollenberg, 1959). In man this loss of fat may readily be detected by the use of skinfold calipers (Tanner and Whitehouse, 1967) and there is some indication that the magnitude of the loss of body fat may be of value in predicting the acceleration of height velocity produced by treatment with exogenous human growth hormone (HGH), at least in children with isolated deficiency of growth hormone (Tanner *et al.*, 1971). The present paper examines the effects of HGH at a cellular level by measuring the size of adipose cells in children undergoing replacement therapy with HGH, and by estimating the effect on the total number of adipose cells in order to determine whether catch-up growth in cell number is induced.

Materials and methods

Twenty-two children (16 boys and 6 girls) aged 6·7 to 22·0 years were studied. All were participants in the national clinical trial of HGH being carried out by the Medical Research Council subcommittee on Human Pituitary Hormones, some results of which have been published (Tanner *et al.*, 1971). By the design of this trial patients undergo (1) a pretreatment year when baseline measurements are established, followed successively by (2) a treatment year, (3) a 'coasting' year (without treatment and now abandoned in cases with a clear-cut growth response), and (4) further treatment years as indicated. 11 of the 22 patients had treatment with HGH for the first time and 11 were at the start of a

re-entry year, having just completed a coasting year. Details of the patients are shown in the Table; the diagnostic categories and nomenclature follow those of the previous report. Patients with panhypopituitarism, either idiopathic or occurring after surgery for craniopharyngioma, received appropriate replacement therapy with cortisone and/or thyroxine.

For the present study measurements of weight, skinfold thickness, and skeletal maturity were made, and an adipose tissue biopsy was performed first at the start of treatment and then after 6 months of twice-weekly intramuscular injections of HGH in a dose of 20 IU/week.

All patients were weighed on beam scales; skinfold thickness was measured on the left side at 4 sites (biceps, triceps, subscapular, and suprailiac) using a Harpenden skinfold caliper in the standard manner (Edwards *et al.*, 1955). Total body fat was calculated from the skinfold measurements, using different regression equations for children over (Durnin and Rahaman, 1967) and under (Brook, 1971) 12 years of age. Skeletal age was assessed using x-rays of the left hand and wrist taken in the standard manner (Tanner, Whitehouse, and Healy, 1962). A sample of adipose tissue was obtained by aspiration from subcutaneous tissue (usually buttock), and adipose cell lipid content, a measure of adipose cell size, was estimated by measuring the lipid content of the sample by gas-liquid chromatography and by counting in a Coulter counter (model B) the number of cells/wet weight of adipose tissue after fixation in osmium tetroxide (Hirsch and Gallian, 1968). The total number of adipose cells in the body was calculated from the mean weight of lipid/cell and the estimate of total body fat.

Results

Body fat, adipose cell size, and the number of adipose cells are shown in the Table with the changes in these quantities over 6 months of treatment with

TABLE 1
Effect of treatment with human growth hormone on body fat, adipose

Case no.	Sex	Chronological age (yr)	Bone age (yr)	Diagnosis*	First (1) or re-entry (2) treatment	Body fat (% of wt)
1	M	6.7	4.0	HS	1	16.5
2	M	7.0	2.2	HS	1	24.9
3	M	7.9	7.8	HS	1	15.5
4	F	10.8	8.3	HS	1	29.5
5	M	11.7	8.7	HS	1	19.2
6	M	12.2	9.7	HS	1	19.7
7	M	16.8	13.7	HS	1	25.5
Mean ($\pm$ 1SD)						
8	M	8.9	6.6	HS	2	29.5
9	M	9.2	7.2	HS	2	32.9
10	M	9.3	7.1	HS	2	16.8
11	M	13.7	11.9	HS	2	15.5
12	F	14.2	9.6	HS	2	29.1
13	M	16.7	14.1	HS	2	31.6
Mean ($\pm$ 1SD)						
14	M	10.5	5.8	CR	1	33.0
15	F	14.7	10.5	CR	1	27.2
16	M	16.3	14.6	CR	1	30.5
17	M	16.7	13.4	CR	1	24.3
Mean ($\pm$ 1SD)						
18	F	15.6	10.9	CR	2	34.7
19	M	17.3	10.8	CR	2	31.7
20	F	19.7	13.4	CR	2	37.5
21	F	22.0	13.4	CR	2	35.6
22	M	16.6	15.0	PAN	2	29.8
Mean ($\pm$ 1SD)						

Combined means ($\pm$ 1SD)

t values against zero

*HS, hypsomatotropic; CR, craniopharyngioma; PAN, idiopathic panhypopituitarism.

HGH. Because increment in the number of adipose cells with age is to be expected (Brook, 1972a), half the annual increment to be expected at the child's bone age has been subtracted to derive the figures in the last column. Thus, allowance has been made for the varying degrees of growth delay manifested by these children and evidenced by their retarded bone ages.

Division of the patients into four groups (hypsomatotropic and craniopharyngioma, first time and later treated) showed no consistent differences between the diagnostic groups; nor was there an overall difference between the first-time and later-treated patients. Accordingly, the data have been pooled to obtain the figures in the final row of the Table.

There was a highly significant reduction

($P < 0.001$) in both body fat and in adipose cell size during treatment. The total number of adipose cells showed an increase over 6 months ($P < 0.001$) and this was still present ($P < 0.02$), though much reduced, when allowance was made for the normal increase which would be expected. Total body fat at the start of treatment was correlated with the pretreatment height velocity calculated over the whole of the preceding year ($r = -0.51$, $P < 0.01$), as was adipose cell size ($r = -0.42$, $P < 0.05$). There was no correlation, however, between the reduction in body fat or in adipose cell size and the acceleration in height velocity produced by treatment. Thus, the magnitude of the effect of HGH on the adipose organ was not of predictive value in assessing the growth response to treatment. The reduction of body fat was maximal in those who

cell size and number in 22 children with growth hormone deficiency

Change in body fat (%)	Adipose cell size (μg lipid)	Change in cell size	Cell no. $\times 10^9$	Change in cell no.	
				Absolute	Allowing for $\frac{1}{2}$ yearly increment at bone age
-3.1	0.24	-0.02	8.54	+0.19	-0.81
-5.1	0.44	-0.12	9.27	+2.73	+1.73
-2.4	0.45	-0.18	5.55	+2.87	+1.87
-5.3	0.42	-0.10	13.63	+1.73	+0.73
-3.0	0.35	-0.06	9.92	+5.63	+4.63
-3.3	0.62	-0.11	6.87	+0.98	-0.02
-1.7	0.58	-0.17	16.18	+1.35	+0.35
-3.41		-0.11		+2.21	+1.21
(± 1.33)		(± 0.06)		(± 1.78)	(± 1.78)
-2.2	0.69	-0.16	11.30	+2.16	+1.16
-7.6	0.49	-0.13	15.97	+0.35	-0.65
+2.3	0.43	-0.13	7.05	+2.11	+1.11
+1.1	0.31	-0.02	13.12	+3.29	+2.29
-5.2	0.59	-0.15	12.52	+3.00	+2.00
-4.0	0.53	-0.10	28.82	+4.58	+4.08
-2.60		-0.12		+2.58	+1.67
(± 3.78)		(± 0.05)		(± 1.42)	(± 1.57)
-5.5	0.50	-0.15	16.06	+3.40	+2.40
-0.8	0.44	-0.06	19.10	+3.83	+2.83
-3.9	0.63	-0.16	20.26	+3.89	+3.59
-2.8	0.39	-0.04	23.62	+2.29	+1.19
-3.25		-0.10		+3.35	+2.50
(± 1.97)		(± 0.06)		(± 0.74)	(± 1.00)
-10.6	0.87	-0.29	14.32	+0.12	-0.88
-9.6	0.51	-0.16	27.16	-2.97	-3.97
-3.0	0.79	-0.11	26.33	+2.56	+1.56
-2.8	0.71	-0.07	21.32	+0.99	-0.01
-6.7	0.49	-0.15	22.00	-0.78	-1.03
-6.54		-0.16		+0.02	-0.87
(± 3.62)		(± 0.08)		(± 2.06)	(± 2.02)
-3.87		-0.12		+2.01	+1.10
(± 3.04)		(± 0.06)		(± 1.92)	(± 1.95)
5.97		9.33		4.90	2.64

were fattest at the start of treatment ($r = 0.66$, $P < 0.001$).

Discussion

The reduction in adipose cell size and body fat shown in this study is the expected consequence of the lipolytic action of HGH (perhaps of somatomedin) (see Tanner, 1972). There is, in addition, some indication that there has been an increase in the total number of adipose cells during treatment which exceeds the increase normally expected during 6 months. This increased velocity of growth is in accordance with the work of Cheek (1968), showing that HGH tended to correct the deficit in muscle cell number found in hypopituitary children. It is not necessarily at variance with the hypothesis that for an organ, and certainly for the

adipose organ, there is a finite period during which the basic complement of cells is determined (Brook, 1972b). Subsequent events could well affect the rate at which the final number of cells was reached, even though they might not alter it.

The increase in velocity of cell division may, however, be explained by the greater than normal increase in bone age caused by HGH and estimated in hyposomatotropic children as 1.18 years/year (Tanner *et al.*, 1971). The methods of estimating both bone age and adipose cell number are too imprecise to apply such a correction over 6 months to the present data, but if a figure for advance in bone age of around 9 months instead of 6 months in this early treatment phase was taken, then the increase in the number of fat cells would agree precisely. The same could probably be said about

the muscle cell catch-up, which may also be appropriate to the general catch-up in bone maturity and adipose cell number.

The present data do not, therefore, support the concept of a true catch-up to normal cell numbers, unlike the effect on height. Hyposomatotrophic children have an absolute deficit in the total number of adipose cells, and HGH, given at a relatively late stage well after the period of determination for the adipose organ has passed, cannot correct the deficit since it does not allow cell multiplication to proceed at a faster rate than normal. Thus, the final number of adipose cells is likely to be the low number which was determined during the period of organogenesis when growth hormone was absent.

The role of HGH in early postnatal growth has not been clearly established. Brasel *et al.* (1965) reported that more than a third of children with hypopituitarism failed to grow normally in the first year of life, though an impressive body of evidence has also been presented in favour of the hypothesis that HGH plays only a minor role at this time (Seckel, 1960). The consensus of opinion is that HGH is required in the first year of life and the evidence from adipose cell growth is firmly in favour of this.

That the well known fatness of hyposomatotrophic patients might be the result of the lack of the lipolytic activity of HGH seems reasonable. The finding in the present study that total body fat is correlated with pretreatment growth velocity, which may be taken as a measure of the severity of the HGH lack, favours this. However, Tanner *et al.* (1971) found that the reduction in skinfold thickness on treatment was correlated with the acceleration in height velocity ($r = 0.41$) in hyposomatotrophic children. The present data on body composition give a lower correlation coefficient; neither the reduction in total body fat nor the diminution in adipose cell size was of significant value in predicting the magnitude of acceleration in height velocity due to treatment.

The adipose organ, it seems, responds to the lipolytic action of HGH by changes in adipose cell size, and HGH in deficient subjects allows growth in cell number at a rate probably consonant with the slightly increased rate of bone age advancement. However, there is no good evidence, at least in the adipose organ, that HGH can correct the deficit of

cell number which was the result of its lack during the determinative period of organogenesis. Growth in cell number is therefore likely only to proceed as far as the low level previously determined.

I am grateful to the Wellcome Trust, the Medical Research Council, and the Nuffield Foundation for financial support; to the MRC subcommittee on Human Pituitary Hormones, under whose clinical trial these patients were receiving their HGH, for permission to publish these studies; and to Professor J. M. Tanner and Dr. M. Zachmann for criticism.

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