During this work the lipogenic activity of adipose tissue from B-complex deficient rats has also been studied. The per cent of total acetate incorporated into lipid per 100 mg tissue in the deficient rats averaged 1.59% (CO_2 formation 1.53%). One week following supplementation of the diet with B-complex vitamins, lipogenic activity of adipose tissue from these rats was found to average 19.08% (CO_2 formation 0.81%). Tuerkischer and Wertheimer(1) have found that addition of dried yeast to the diet of B-complex deficient rats will cause deposition of adipose tissue glycogen.

Summary. 1. The incorporation of C^{14}-acetate into lipid and carbon dioxide by rat adipose tissue (*in vitro*) was investigated. This investigation concerned the effect of the nutritional status upon the lipogenic activity of adipose tissue. 2. Fasting was found to produce an increase in nitrogen concentration and a decrease in lipid concentration and lipogenic activity. 3. In normal rats lipogenic activity of the tissue was found to increase during recovery feeding following fasting. The increased activity was paralleled by increased nitrogen and glucose content of the tissue and by decreased lipid content. 4. Lipogenic activity of adipose tissue of rats could be made relatively constant by depletion of fat stores followed by recovery feeding.

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Effect of Pituitary Growth Hormone on Transplantable Mouse Tumors. (23374)

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In a recent study of the effect of growth hormone on a mammary adenocarcinoma transplantable in DBA/1 mice, Mirand(1) and Mirand and Hoffman(2) reported that tumor volume was significantly increased when the host mouse was given hormone beginning at the time of inoculation. Reid's (3) review indicates that mouse tumor response to growth hormone was not a consistent finding. We examined this problem further with more quantitative experiments on 4 mouse tumors using purified growth hormone which we assayed for its effect on hypopituitary dwarf mice(4,5).

Materials and methods. The effect of growth hormone was studied on 4 transplantable tumors. 1. The DBA/1 mammary adenocarcinoma which originated spontaneously in the DBA/1 mouse, 2. the Marsh Albino (MA) mammary adenocarcinoma which originated spontaneously in the MA mouse. 3. and 4.: Tumors 3 and 4 were fibrosarcomas induced in the DBA/1 and MA mouse strains respectively by desoxycorticosterone acetate in sesame oil(6). These 4 tumors grew in castrated mice indicating their lack of hormonal dependence on the host. All tumors were grown subcutaneously in male mice having an average age of 3 months at the time of inoculation. Cell suspensions of the tumors were made by the method of Reinhard, Goltz and Warner(7).

TABLE I. Assay of a Purified Beef Growth Hormone Preparation (Horner's)* in Adult Hypopituitary Dwarf Mice† by Body Weight.

Dwarf mouse No.	Wt in g: Initial	After 1 wk .4 mg/day	After 2nd wk .6 mg/day	After 3rd wk 1 mg/day	After 4th wk 1.5 mg/day
1—♂	11.3	15.1	20.5	22.5	25.5
2—♂	8.7	14.5	19.5	23.5	26.5
3—♂	11.0	17.5	21.5	25.5	28.5
4—♀	8.5	13.5	17.5	20.5	23.5
Avg	9.9	15.2	19.8	23.0	26.0
Controls, saline inj.					
1—♂	11.0	10.5	11.5	12.0	12.0
2—♂	10.5	11.0	12.5	11.0	12.0
3—♀	8.5	9.0	9.0	9.5	9.0
Avg	10.0	10.2	11.0	11.0	11.0

* Growth hormone preparations contaminated with ACTH showed no growth promoting action after the 4th wk in hypopituitary dwarf mice. Growth hormone preparation contaminated with TSH after the 4th wk gave an avg body wt value of 38 ± 2.5 g in hypopituitary dwarf mice.

† Syracuse Strain.

A purified beef growth hormone preparation (Lot #GE-5-F. W. Horner Ltd., Montreal, Canada) was given immediately after tumor cell inoculation and in increasing doses weekly according to the following schedule. In the first week at the time of tumor inoculation 0.4 mg per day, second week 0.6 mg per day, and third week 1.0 mg per day. On the eighteenth day the animals were weighed and sacrificed. The excised tumors from these animals were weighed by the water displacement method. Potency of hormone was assayed beforehand on hypopituitary dwarf mice that had attained their maximum adult body weight. Other growth hormone preparations (from Armour Co. and Princeton Laboratories) were assayed on hypopituitary dwarf mice, but were not used because a maximum growth response on dwarf mice was not evident. Growth hormone preparations contaminated with ACTH did not show growth promoting action on the hypopituitary dwarf. Growth hormone preparations contaminated with TSH gave a greater effect than the purified growth hormone preparation (Horner's) used in this study. Table I shows the increase of body weight in the hypopituitary dwarfs at 4 successive week intervals with progressively increased hormone dosage. Control dwarf mice were injected with saline, the same volume as given above. Results given in Table I show when growth hormone is given at least a 2-fold increase in body weight of the hypopituitary dwarf mouse occurs in 4 weeks.

Results. Tables II A and B show the results for these tumors and the host mouse body weights when the animals were sacrificed on the eighteenth day after inoculation. They also show that variation of number of cells per inoculum by a factor of 50 had no significant effect on tumor growth, either in the final weight of tumor or in the observed latent period. The reasons for this lack of variation with inoculum size are unknown, but one important factor is probably the constancy of the final tumor mass attained in a given host mouse, as will be described below.

Comparison between the hormone treated animals and the controls, which were saline injected, shows that the host animal underwent approximately a 20% increase in body weight and that there is about a factor of 3 increase in tumor weight in the presence of administered hormone. Also, the tumor latent period is reduced by about 25%, a fact which points to an increased multiplication rate. In order to estimate the shortening of generation time, measurements were made of the tumor volume when it first became palpable. These showed it to be of the order of 2 cmm. This figure permits an estimate of the generation time(8,9) in the DBA/1 fibrosarcoma (Table II A) to be 14 hours in the controls. In the hormone treated animals the tumor cell generation time was 10 hours.

TABLE II A. Effect of Growth Hormone on Average Tumor and Host Weights. DBA/1 fibrosarcoma inoculated subcut. in DBA/1 males.*

	Hormone treated					Controls (saline inj.)				
Cells per inoculum	No. of animals	Body wt (g) Initial	Final	Tumor wt (g)	Latent period (days)	No. of animals	Body wt (g) Initial	Final	Tumor wt (g)	Latent period (days)
596	12	16.5	25.4	.25	11.7	12	17.2	21.3	.06	15.7
2112	12	16.6	25.2	.22	12.2	12	16.5	20.4	.07	15.7
5026	12	16.9	24.8	.24	10.8	12	16.7	19.8	.07	14.4
10200	12	15.5	23.8	.25	11.4	12	16.2	19.9	.07	15.3
25700	12	16.3	24.8	.23	11.1	12	15.3	19.3	.07	14.3

TABLE II B. DBA/1 mammary adenocarcinoma inoculated subcut. in DBA/1 males.*

	Hormone treated					Controls (saline inj.)				
515	12	16.8	26.9	.48	12.9	12	17	22.2	.16	15.5
2010	12	16.7	27.0	.53	10.9	12	17.1	23.2	.28	13.8
4825	12	16.3	25.6	.49	11.4	12	16.5	22.4	.19	14.6
10850	12	16.7	25.5	.64	11.2	12	16.9	21.5	.18	15
26850	12	16.5	25.1	.73	10.5	12	17.3	21.4	.22	13.5

* All tumors and final mouse weights were measured on 18th day after inoculation.

However, the growth period from inoculation to sacrifice was not always fixed at 18 days. Experience showed that it was desirable to terminate growth of tumor at various times beginning as early as 11 days after inoculation depending on growth rate. For example, in the DBA/1 mammary adenocarcinoma, (Table II B), some of the tumors at 18 days had attained a volume of about 1 ccm. It was felt that such large tumors were undesirable for quantitative measurements because of possible limitation of vascular supply.

A variable growth time for the tumors made it necessary to introduce the host body weight as a basis of reference for comparison of the various experimental conditions. Since the mice had a spread of body weight, it was necessary to express the increase in body weight at any time after tumor inoculation as the ratio of final to initial body weight. Thus, in the various strains used, the ratio of final to initial body weight ranged from 1.00 to 1.80 in the hormone treated mice and from 1.00 to 1.5 in the saline treated controls. In the MA fibrosarcoma, which is the first entry in Table III, the average mass of the tumor in control mice was 0.08 g, while the body weight ratio varied from 1.1 to 1.4. These control tumors had growth times ranging from 14 to 22 days. The same tumor in hormone treated males averaged 0.15 g over a range of body weight ratio from 1.3 to 1.6. The growth times were less than those in the controls and varied from 13 to 20 days. The hormone shifted both the tumor and body weights to values greater than those of the controls.

The other 3 tumor entries in Table III show the same pattern as that described for the MA fibrosarcoma. The last 2 entries are for the one tumor, the DBA/1 mammary adenocarcinoma, the experiments being carried out at different times of the year, namely May and August. The tumor masses are greater in the August run than in the May run both in the controls and in the hormone treated animals. While the tumor masses are greater, the approximate constancy with body weight ratio is maintained, and hormone influence is manifest in increased mass of both tumor and host.

The data for the fourth tumor, the DBA/1 fibrosarcoma, are presented graphically in Fig. 1 to illustrate the pattern of response seen in each of the entries of Table III. The abscissae are ratio of final to host body weight and are common to both Fig. 1 a and 1 b.

Discussion. The effectiveness of a purified beef growth hormone preparation is demonstrated in the assay on hypopituitary dwarf mice. Table I shows that average body weight of these mice is slightly more than doubled after 3 to 4 weeks of treatment. Each mouse receiving growth hormone underwent a

TABLE III. Average Tumor Weight as a Function of Ratio of Final to Initial Host Body Weight.

Ratio of final to initial body wt	1.0/ 1.099	1.1/ 1.199	1.2/ 1.299	1.3/ 1.399	1.4/ 1.499	1.5/ 1.599	1.6/ 1.69	1.7/ 1.8
MA fibrosarcoma in MA mice								
Tumor with GH (g)				.12	.17	.17		
No. of animals				4	13	7		
Control tumors (g)		.07	.08	.08				
No. of animals		4	19	1				
MA mammary adenocarcinoma in MA mice								
Tumor with GH (g)		.22	.21	.17	.19	.20	.23	
No. of animals		1	6	16	21	25	3	
Control tumors (g)		.07	.06	.07	.07			
No. of animals		19	40	12	1			
DBA/1 mammary adenocarcinoma in DBA/1 mice (May run)								
Tumor with GH (g)		.22	.24	.22	.22	.25	.26	
No. of animals		9	12	15	17	3	4	
Control tumors (g)	.09	.07	.09	.15				
No. of animals	6	23	28	1				
DBA/1 mammary adenocarcinoma in DBA/1 mice (Aug. run)								
Tumor with GH (g)		.45	.45	.59	.49	.56	.56	.69
No. of animals		1	1	6	11	19	8	13
Control tumors (g)	.25	.19	.18	.22	.23		.20	
No. of animals	1	7	18	22	7		2	

consistent increase in body weight during the 4 weeks' time during which the exogenous hormone supplemented the deficient pituitary hormone.

In the non-deficient mice the hormone effect was not so pronounced although clearly evident. The 60 males of the DBA/1 strain in Table II A, for example, saline injected had an average body weight increase of from 16.4 to 20.1 g in 18 days. The 60 males treated with GH increased from 16.3 g to 24.8 g in the same time interval.

More pronounced, however, is the effect of GH on the transplanted fibrosarcoma (Table II A) whose mass was increased by at least a factor of 3 over the saline injected controls. The results of Tables II A and B parallel the results on tumor fragment transplantation of Smith *et al.*(10) who found that GH increased both the body weight and tumor growth rate of C3H mice bearing a transplantable mammary adenocarcinoma. Evidence for increased growth rate in the DBA/1 fibrosarcoma is exemplified in the reduction of the average latent period from 15.7 days in controls to 11.7 days in the first run of 12 mice in Table II A.

No one of the 4 different tumors failed to

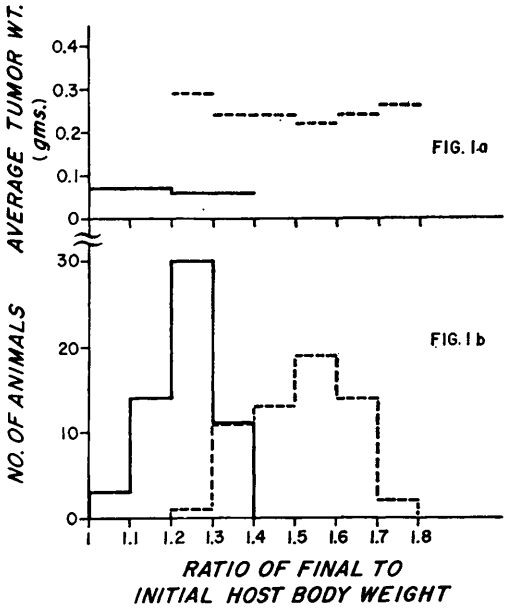

FIG. 1a. Avg tumor weights (ordinates) for corresponding ratio of final to initial host body wt (abscissal). DBA/1 fibrosarcoma in DBA/1 male mice, solid lines tumor wt in controls, saline inj. Dashed lines, tumor wt in mice treated with growth hormone.

FIG. 1b. Histogram of distribution of numbers of animals treated with growth hormone and controls, saline inj., plotted against ratio of initial to final wt of host body wt of tumor bearing animals in Fig. 1a.

show a response in increased growth. Referring, for example, to the fibrosarcoma in the DBA/1 males (Table II A) the average control tumor weighed 0.07 g compared to the average of 0.238 g in the GH treated mice. The mammary adenocarcinoma in DBA/1 males (Table II B) had a control weight averaging 0.21 g as compared with 0.57 g in the GH treated mice. The ratio of GH treated fibrosarcoma weight to its control is 3.4 whereas in the mammary adenocarcinoma the ratio is 2.7. In the MA strain its mammary adenocarcinoma had a higher ratio than did the fibrosarcoma (Table III). No significance is attached to the differences in ratios. Thus, the 4 tumor types studied all have about the same sensitivity to GH in either the DBA/1 or MA male mice.

The constancy of maximum attainable tumor mass in a given host is of special interest. In Tables II A and B the inoculum size varied from about 500 to 26,000 or by a factor of 50, yet the final tumors had the same average mass. This is especially notable in the fibrosarcoma where the 5 separate runs had average tumor masses from 0.22 to 0.25 g and the controls averaged 0.06 to 0.07 g. This is interpreted as meaning that regardless of inoculum size the tumors attained a fixed, characteristic mass and grew no further. By the eighteenth day all tumors had attained this average volume.

As the data of Table II and Fig. 1 show, the host body weight may increase from 10 to 60%, yet the average tumor mass remains at the level of the fixed characteristic value both in control and in GH treated mice. The graph in Fig. 1a summarizes the GH effect in displacement of the line for the average tumor mass. It is displaced upward because tumor size is increased, and to the right because average body weight of the host is also increased by GH. While host weight may increase 10 to 15% tumor average increases from a minimum of 50 to 300% depending on the tumor type and the host.

The complexity of growth hormonal action is indicated by many reports(1). There has been controversy as to whether the anterior pituitary hormones which promote or assist to promote normal (and perhaps cancerous) mammary growth are due to specific mammogenic hormones working in concert with ovarian hormones, or whether the hypophyseal contribution can be wholly attributed to the action of one or more of the 6 generally accepted and characterized anterior-pituitary hormones. The data in this report indicate that mammary tumor cells as well as fibrosarcoma cells can respond to growth hormone and that the effect is not entirely a general metabolic one. Pearson, *et al.*(11,12) reported recently the anabolic and symptomatic effects of human growth hormone in hypophysectomized patients with osseous mammary metastases. Their data suggested that pituitary growth hormone is an important factor for mammary carcinoma growth. From the reports of other investigators, and ours, it would seem reasonable to assume that growth hormone in some way plays a role in carcinogenesis(13-17) and in supporting neoplastic growth.

Summary. 1. Growth hormone was assayed for potency by its effect on body weights of adult hypopituitary dwarf mice. Their body weights increased by more than a factor of 2 during 4 weeks' administration of the hormone. 2. Four different transplantable tumors responded to growth hormone administered to the host mice. The latent period of the tumor is reduced and the mass of the tumor is much greater than that of the controls. 3. Tumor bearing host mice showed a significant body weight increase under action of growth hormone. Enhanced tumor growth due to the hormone did not reduce the host body weight. 4. Maximum average tumor mass attained by the 4 different tumors is independent of the host body weight. While growth hormone increases the tumor as well as the hosts weight, average mass of tumor grown is constant and characteristic of the tumor-host combination. 5. Data suggest that growth hormone plays a supportive role in mammary carcinoma.

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Transplantable Mastocytoma in the Mouse Containing Histamine, Heparin, 5-Hydroxytryptamine.* (23375)

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Mast cells are among the current problem cells of the body. Several vital functions have been attributed to them but the relation of any functions to the mast cells has not been firmly established. They are said to be the major, if not the sole, source of heparin (1); so they may be involved in the clotting mechanism. Their relation to connective tissue and their high content of heparin suggest that they may be involved in the elaboration of the mucopolysaccharides of the collagenous ground substance. They are a rich source of histamine(2) and may play a part in processes of inflammation and hypersensitivity. It has been reported that they contain 5-hydroxytryptamine (serotonin, 5-HT)(3), a naturally occurring amine with pharmacological actions on smooth muscle. The distribution of mast cells in certain locations and in certain tumors apparently follows a definite pattern(4), the significance of which is not clear.

Tumors composed of mast cells are well known in animals; none has been reported in man. The most studied is that occurring occasionally in dogs. It was first described by Bloom(5) and its high heparin content established by Oliver *et al.*(6). Deringer and Dunn(7) have described a mouse mastocytoma. Transplantation of a mastocytoma, apparently carried out simultaneously with that described here, has just been reported by Dunn and Potter(8). A survey of the relevant literature is beyond the scope of this report.

Origin. The tumor was found in a 500-day-old mouse of the LAF_1 strain, that had been irradiated at approximately 7 weeks of age with 475r over the entire body. The left adrenal of the animal was removed 5 weeks following irradiation. This was one of a large

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