

emetic dose of dihydroergotamine methanesulfonate increases the number of *Macaca mulatta* which vomit following oral administration of staphylococcal enterotoxin. The possibility that increased incidence of vomiting might be due to summation effects of 2 similar pharmacological compounds has been considered, but no adrenolytic effect of enterotoxin corresponding to that of DHE-45 has been found. The other drugs tested have not significantly affected the incidence of emesis in monkeys following feeding of enterotoxin.

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Received February 21, 1958. P.S.E.B.M., 1958, v97.

Food Utilization and Weight Gain in Hypophysectomized Rat as Function of Handling. (23917)

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Previous studies have shown that normal young rats handled or gentled by the experimenter gain significantly more weight and length than unhandled control animals(1-9). Furthermore, these effects of handling appear to be a function of better food utilization rather than increased intake(4,6,7). The literature on this subject has recently been reviewed in detail(10).

This paper reports the effects of handling on weight gain, skeletal growth, food utilization and body composition in the young hypophysectomized rat. It is the first of a series of investigations designed to study the mechanism of various effects of handling in experimental animals.

Methods. Twenty-eight male hypophysectomized rats of Sprague-Dawley strain, obtained from Hormone Assay Laboratories, (Chicago), were used. Hypophysectomy was performed on nineteenth day of age. Animals were weighed and then divided at random into 2 equal groups: control and experimental.

Beginning on twenty-first day of age, each animal of experimental group was handled individually by one of us (L.B.) for 10 minutes every day at the same time for 6 weeks. Handling consisted of holding the animal in one hand, and stroking it with the other from head to tail repeatedly, with animal facing experimenter. Control animals were never touched after their original placement in cages. A weighed amount of food in petri dish was placed in each cage daily and leftover food weighed daily to determine daily food consumption. The diet used was of the following content and proportion: 480 g canned dog food (Pard®), 2 slices of crumbled whole wheat bread, 480 ml skim milk, and 2.5 ml Vi-Daylin®* (a liquid multi-vitamin preparation). These ingredients were thoroughly mixed and proved suitable for these animals (tender-jawed following hypophysectomy), and food loss by spillage from petri dish was

* Kindly supplied by Mr. V. Yeager, Abbott Laboratories, N. Chicago, Ill.

TABLE I.

		Weeks							Significance (<i>p</i>)
		0	1	2	3	4	5	6	
Wt (g)	U	38.6	51.3	63.1	69.0	73.2	81.3	89.2	.05
	H	38.6	51.3	63.6	69.9	74.9	84.1	92.8	
Food consumption (g)	U		163.0	235.8	272.0	290.0	319.0	314.4	.05
	H		153.9	225.6	265.0	283.9	306.4	302.8	
Food utilization									
Food consumption (g)	U		12.9	16.2	22.1	27.8	30.0	31.6	.001
Wt gain (g)	H		12.2	15.2	20.6	25.6	27.2	28.3	
Body composition									
% protein	U							21.1	.6
	H							21.5	
% fat	U							9.1	.5
	H							9.8	
Skeletal growth									
Tibial epiphyseal car- tilage (μ)	U							151.5	.9
	H							150.8	

U = Unhandled rats. H = Handled rats. values are calculated from data at 6 wk.

All figures are mean values.

Significance

minimum. Five % glucose was supplied as drinking water. All animals survived the 6 week experiment and all appeared in good condition throughout the period. The animals were housed individually in wire cages measuring 20 x 17 x 23.5 cm. Cages were arranged in rack so that handled and unhandled animals alternated, and with equal numbers of each group at each level in the rack. The animals were weighed weekly in their cages and weight of cage subtracted so that no handling was involved in the process. The temperature of the animal room was maintained by thermostat at $80 \pm 2^\circ\text{F}$. At end of experiment the animals were sacrificed by gas inhalation and whole carcasses analyzed for protein and fat content. Carcasses were ground in meat grinder and dried to constant weight. Fat was extracted repeatedly with ether, then dried, and weighed. Total protein was obtained by multiplying total nitrogen (macro-Kjeldahl) of dry carcass by 6.25. Skeletal growth was measured by means of the rat tibia test(11), used for assay of pituitary growth hormone.[†] The sellas of the 5 heaviest animals of each group were removed for histological study and completeness of hypophysectomies was confirmed in all animals.

[†] These assays were performed in Research Laboratories of Frank Horner Ltd., Montreal, through the courtesy of Dr. L. Mitchell.

Results. Table I summarizes data on weight gain, food consumption, carcass analysis and tibial epiphyseal measurements of the 2 groups of rats. Weight gain of handled group was significantly ($p = .05$) greater than that of unhandled group. The handled animals consumed significantly ($p = .05$) less food than unhandled ones. When a ratio of grams of food consumed/g weight gained is computed, the difference between the 2 groups of rats becomes much more significant ($p = .001$). Another method of demonstrating this difference between the 2 groups of rats is by analysis of covariance(12), in which weight gains may be adjusted for variation in amounts of food ingested. The computed value F (26.5) is significant beyond the 0.1% level of confidence.

Carcass analysis for percentage of fat and nitrogen showed no significant difference between the 2 groups of animals. The same was true of tibial epiphyseal measurements.

Discussion. The results indicate that handling has a significant enhancing effect on weight gain of young hypophysectomized rat. The magnitude of response was considerably less than that previously reported in the young normal rat(1,3,4,5,6,7,8,9). This is probably due to limitations imposed by lack of pituitary hormones necessary for normal growth and development. It is noteworthy

that increased weight gain in hypophysectomized animals was achieved despite a significantly smaller quantity of food ingested. When weight gains are corrected for amount of food ingested, increases in weight of the handled over unhandled group are greatly magnified. In the normal rat, handling has no influence on amount of food ingested(6). The findings indicate that handling increases efficiency of food utilization (increased intestinal absorption and/or diminished metabolism) and that this effect is not dependent upon presence of pituitary or adrenal hormones.

It is apparent that increased skeletal growth previously reported in the handled normal young rat(6), is abolished by hypophysectomy. This suggests that the effect of handling on skeletal growth requires pituitary hormones, presumably growth hormone and/or thyrotropic hormone.

It is pertinent to consider the contribution of handling to weight gains previously reported in hypophysectomized rats treated with insulin(13) or force-fed(14). Since these studies involved extra handling of experimental animals as compared with control animals, it is conceivable that weight gains observed were due in part to extra handling.

Summary. Handling in young hypophysectomized rats resulted in significantly increased weight gain despite ingestion of significantly less food. No effect on skeletal growth or

percentage of carcass protein and fat was observed. Increased weight gain in the handled hypophysectomized rat is due to greater efficiency of intestinal absorption of food and/or diminished metabolism rather than a direct stimulation of growth or increased food ingestion.

We wish to acknowledge the technical assistance of Y. Arai and A. Smith.

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Received February 24, 1958. P.S.E.B.M., 1958, v97.

Pyridoxal Phosphate (B₆-al-PO₄) Levels in Organs, Leukocytes and Blood of Rats with Developing Vit. B₆ Deficiency.* (23918)

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Recently a method has been described for determination of pyridoxal phosphate (B₆-al-PO₄), the coenzymatically available active form of Vit. B₆, in whole blood and isolated leukocytes(1). Using this technic we estimated the B₆-al-PO₄ content in the only readily accessible prototype tissue, *i.e.*, leuko-

cytes in blood of healthy, nonpregnant controls, mothers at term, and in cord blood of their babies(2). It was found that the pyridoxal phosphate levels of circulating leukocytes were significantly lower in pregnant women than in nonpregnant controls. Since cord blood showed the highest levels, it was concluded that the growing fetus can successfully compete with the mother for this impor-

*This work was aided by grant-in-aid from Nat. Vit. Fn.