μg of estrone. When administered simultaneously, neither cortisone acetate nor cortisol acetate had any evident inhibitory effect. DCA, on the other hand, was a definite but weak estrone antagonist (10% as potent as progesterone). Aldosterone was active; its potency appeared to be about 10% progesterone.

1. Roberts, S., Szego, Clara M., *Physiol. Rev.*, 1953, v33, 593.
2. Edgren, R. A., *Ann. N. Y. Acad. Sci.*, 1959, v83, 160.
3. Edgren, R. A., Calhoun, D. W., PROC. SOC. EXP. BIOL. AND MED., 1957, v94, 537.
4. Edgren, R. A., Calhoun, D. W., Elton, R. L., Colton, F. B., *Endocrinology*, 1959, v65, 265.
5. Rubin, Betty, L., Dorfman, Adaline S., Black, Lila, Dorfman, R. I., *ibid.*, 1951, v49, 429.
6. Edgren, R. A., PROC. SOC. EXP. BIOL. AND MED., 1956, v92, 569.
7. Szego, Clara M., Roberts, S., *Am. J. Physiol.*, 1948, v152, 131.
8. Szego, Clara M., *Endocrinology*, 1952, v50, 429.
9. Velardo, J. T., *Ann. N. Y. Acad. Sci.*, 1959, v75, 441.

Received October 13, 1959. P.S.E.B.M., 1960, v103.

Lack of Relationship between Body Weight and Pharmacological Effect Exemplified by Histamine Toxicity in Mice.* (25494)

E. T. ANGELAKOS (Introduced by E. R. Loew)
Dept. of Physiology, Boston University School of Medicine, Boston, Mass.

The pharmacologic effects of many drugs and toxins appear to be proportional to their dose/unit body weight; for some drugs this may not be the case. The following study shows that toxicity of histamine in mice is not directly related to body weight.

Methods. Albino mice of Swiss strain weighing 15 to 25 g were used. Each group tested consisted of mice with a mean weight within $\pm$ 1 g of the desired mean body weight and with no individual mouse deviating by more than 1 g from the mean. Histamine diphosphate was dissolved in appropriate concentrations in 0.9% NaCl so that each animal received the desired dose in 0.2 ml of solution. Within each weight group all animals received the same dose of histamine diphosphate regardless of exact body weight of each animal. All injections were made intraperitoneally and mortality was recorded over a 24 hour period even though all animals that succumbed terminated within 6 hours after injection.

Results. A total of 360 mice were used in 18 groups of 20 mice each. The LD_{50} of histamine injected intraperitoneally was first determined in mice weighing 18 $\pm$ 1 g. Seven dose levels were utilized. Results and their statistical analysis are shown in Table I. Doses of 1.6 to 2.5 mg/g body weight produced submaximal responses. This dose range was then used to reevaluate the LD_{50} in 2 groups of mice, one of 18 $\pm$ 1 g and another of 25 $\pm$ 1 g. Both groups were tested simultaneously at 3 identical dose levels on a mg/g basis. Results are tabulated in Table II. When the doses are taken on a mg/g basis the smaller mice (18 $\pm$ 1 g) seem to have a larger LD_{50} than larger mice (25 $\pm$ 1 g). This difference is statistically significant and could have been interpreted as indicating that smaller animals are more resistant to histamine than larger. However the ratio of the two LD_{50}'s, *i.e.*, 1.29 with confidence limits of 1.15 to 1.44, is very close to the inverse ratio of weights of the 2 groups 25/18 = 1.39 suggesting that the difference between the 2 groups may be entirely due to the assignment of doses on body weight basis. In fact when the doses are expressed as mg/mouse, the results of the 2 groups fit a single dose-response probit regression indicating that they correpond to a single homogeneous population with an LD_{50} of 40 mg/mouse. This is further substantiated from the results of 3 different

* Supported by grant from Parke, Davis and Co., Detroit, Mich.

TABLE I. Histamine Toxicity Mice Weighing 18 ± 1 g* (20 Animals/Dose Group).

	Histamine diphosphate (mg/g body wt)	(mg/mouse)	Mortality (%)
	1.5	28	0
	1.7	31	20
	1.9	34	30
	2.1	38	35
	2.3	41	45
	2.5	45	65
	2.7	49	85
LD_{50}	2.3	41	
95% conf. limits	2.2 to 2.4	40 to 43	

* Within each dose group all animals were inj. with the same dose of histamine diphosphate intraper. Mortality was recorded over 24 hr after inj.

weight groups given the same dose on a mg/g basis (Table III A). As expected, mortality was higher in the larger animals than in smaller mice given the same dose on unit weight basis. When the doses are translated in mg/mouse the same data (Table III A) form a single regression with an LD_{50} of 36 mg/mouse (95% confidence limits 32 to 40). By contrast mortalities are very similar when 3 groups injected with the same dose on a mg/mouse basis are compared (Table III B). By translating the mortalities into probits the variance of the results under A and B (Table III) can be compared. The variance of the results is about 9.5 times larger when doses

TABLE II. Histamine Toxicity in Mice Weighing 18 ± 1 and 25 ± 1 g (20 Animals/Dose Group).

Mouse wt (g)	Histamine diphosphate (mg/g)	(mg/mouse)	Mortality (%)
18 ± 1	1.6	29	20
"	2.0	36	30
"	2.4	43	60
LD_{50}	2.2	40	
95% conf. limits	2.0 to 2.5	36 to 45	
25 ± 1	1.6	40	30
"	2.0	50	80
"	2.4	60	90
LD_{50}	1.7	43	
95% conf. limits	1.5 to 1.9	38 to 48	
Potency ratio 18 *vs* 25 g	1.29	.93	
95% conf. limits	1.15 to 1.44	.85 to 1.02	

Both groups combined on basis of mg/mouse give an LD_{50} (and 95% confidence limits) of 40 (36 to 44).

are expressed on a mg/g basis as compared to the results when doses are given on a mg/mouse basis (Tables III A *vs.* III B).

Discussion. It is apparent from the results and statistical analyses that histamine toxicity in mice is represented more generally by a regression relating dose per animal rather than dose per unit body weight. This seems to be applicable in the range of body weights tested (15 to 25 g).

When doses are expressed in terms of unit body weight (mg/g) histamine appears to be less toxic in smaller than in larger animals. This is clearly an artifact as it is entirely dependent upon the assumption that the pharmacological effect of a given dose is a function of body weight. This assumption can not be justified by the data. The simplest explanation of the available data is that histamine toxicity in mice is an example of a pharmacological action where the dose producing a given effect is not a function of body weight, at least in the weight ranges tested.

TABLE III. Histamine Toxicity in Mice of 3 Different Weight Groups (20 Animals/Group).

Mouse wt (g)	Histamine diphosphate (mg/g)	(mg/mouse)	Mortality (%)	(probits)
		A		
15 ± 1	2.0	30	30	4.47
20 ± 1	2.0	40	60	5.26
25 ± 1	2.0	50	85	6.04
		B		
15 ± 1	2.67	40	50	5.00
20 ± 1	2.0	40	60	5.26
25 ± 1	1.6	40	40	4.75

Probit variance for mortalities of groups under A is 0.616 and that of those under B is 0.065 giving a variance ratio of 9.48.

It may be suggested that other variables inherent in estimation of histamine toxicity in mice may be so large as to mask any contribution produced by differences in body weight. This suggestion is not tenable in this case because correction for body weight actually increases variation to the extent that differences in toxicities between different weight groups become statistically significant. Alternatively it may be held that protective mechanisms (such as rate of inactivation and excretion) may bear the same relationship to body weight as those factors which contribute to toxicity

(such as rate of absorption and total body water) and thus the contribution of body weight is cancelled. In any event, the applicability of the results reported here may not be limited to this particular experimental model since it has been concluded(1) that for several pharmacological agents the lethal dose for small animals is usually a little larger than for large animals when both are expressed on a unit weight basis.

The above considerations are not intended to imply that the same relationship exists with other drugs or species or even with the same drug when a different pharmacological effect is evaluated. However they do emphasize that dependence of the dose on body weight can not be implicitly assumed in the absence of data showing that a relationship actually exists. Such a dependence may be practically zero, as in the present case, or it may be represented by any fraction of unity. In the later case it is best established by covariance analysis, which has been reported(2). It is noteworthy that a similar lack of a relation between body weight and toxicity has been shown to exist in the case of the lethal effects of botulinal toxins on mice(3) and alpha-naphthyl-thiourea in rats(4).

Summary. Data from 360 mice are presented which show that the toxicity of intraperitoneal histamine is independent of body weight. Adjustment for body weight in the range of 15 to 25 g increased the variation so that smaller animals appeared to be less sensitive to histamine than larger mice. Possible mechanisms are discussed.

1. Gaddum, J. H., *Pharmacology,* Oxford Univ. Press, 1953, p508.

2. Deutsch, S., Angelakos, E. T., Loew, E. R., *Endocrinology,* 1956, v58, 33.

3. Lamanna, C., Jensen, W. I., Bross, I. D. J., *Am. J. Hyg.,* 1955, v62, 21.

4. Rall, D. P., North, W. C., PROC. SOC. EXP. BIOL. AND MED., 1953, v83, 825.

Received October 19, 1959. P.S.E.B.M., 1960, v103.

Initiation of Lactation in Rats with Hypothalamic or Cerebral Tissue.*† (25495)

JOSEPH MEITES, P. K. TALWALKER AND C. S. NICOLL
Dept. of Physiology and Pharmacology, Michigan State University, East Lansing

Recently we demonstrated that epinephrine, acetylcholine, serotonin and reserpine can initiate lactation in estrogen-primed virgin rats, and maintain mammary secretion in postpartum rats after litter removal(1,2). Numerous other drugs and a variety of stressful stimuli can also initiate mammary secretion in rats (unpublished). These drugs and stressful stimuli are believed to induce release of prolactin and probably ACTH from the anterior hypophysis, since administration of prolactin alone is ineffective in initiating lactation in intact rats and requires the addition of ACTH or glucocorticoids(3). Large doses of ACTH or glucocorticoids alone may induce mammary secretion in intact rats, presumably by stimulating pituitary prolactin release(4). Harris(5) and others have suggested that the hypothalamus and perhaps other portions of the brain produce neurohormones which reach the anterior pituitary *via* the hypophyseal portal vessels and induce release of its hormones. Thus hypothalamic extracts have been shown to elicit ACTH discharge(6,7) and pineal extracts to evoke aldosterone release(8). It was of interest, therefore, to determine whether hypothalamic or cerebral tissue from rats could induce release of sufficient prolactin and probably ACTH to initiate lactation in rats.

Methods. Brain tissue was taken from postpartum or estrogen-treated mature female rats (Carworth strain). They were killed with

* Published with approval of Mich. Agr. Exp. Sta. as Jour. Art. No. 2516.

† This work was supported in part by NIH grant.