environmental rather than hormonal stimuli.[26] It seems clear from our study that the injection of sex hormones alone are ineffective in augmenting lactogen output by the pituitary or initiating crop milk secretion in pigeons.

It appears remarkable that the pigeon pituitary should be refractory to the administration of sex hormones, particularly estrogens which are such powerful activators of lactogenic hormone secretion in the pituitaries of mammals. Neither do estrogens increase the weight of the pigeon pituitary as in mammals. It would be of interest to determine what effect the sex hormones have on the cytology of the pigeon hypophysis.

Summary. The effects of estrone, diethylstilbestrol, progesterone and testosterone-propionate were determined on the lactogen content and weight of the pituitary, and on the proliferation and weight of the crop glands of common pigeons of both sexes. The results were all negative. It is concluded that the pigeon pituitary, unlike the mammalian, is refractory to the administration of sex hormones and is not stimulated by them to increase the secretion of lactogenic hormone.

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Statistical Evaluation of Growth Curves.

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A problem that frequently confronts research workers in the biological sciences is the comparison of growth curves of groups of experimental animals subjected to different treatments. The customary practice has been that of visual estimation of significant differences from a plot of weight against time (Fig. 1), or the application of the "t" test to the mean weights at a particular time. These conventional methods of evaluation often give an ambiguous or unconvincing analysis of the results.

The purpose of this paper is to demonstrate how time-weight data (growth curves) have been treated more critically in a comparative study of exposure of rats to vapors

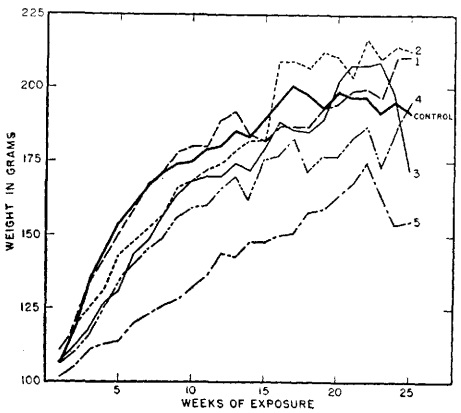

FIG. 1.
Combined growth curves of male and female rats.

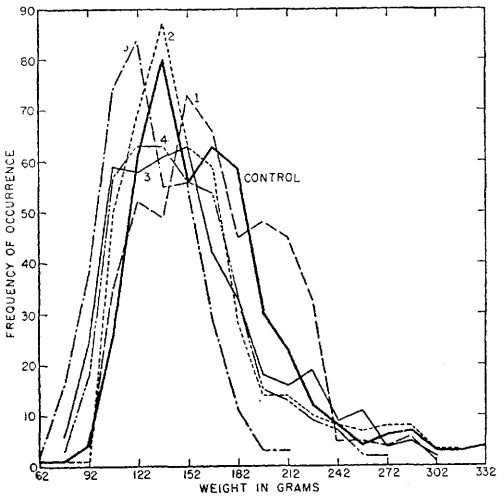

FIG. 2.
Curves showing the frequency of occurrence of individual weights for male and female rats, throughout the period of treatment.

TABLE I.
Summary of Data Showing Differences in Weight Response.

Group	No. of rat wts	Mean wt	Standard dev.	"t" at 12 wks exposure	Probability level for 12 wk "t"	$3\sigma d/d$	Chi square where $\chi^2 = \Sigma \left(\frac{(fo - ft)^2}{ft}\right)$	Probability level for chi square
				Combined Weights of Male and Female Rats.				
1	464	168.4	40.6	0.72	0.50	3.74	19.8	0.14
Control	451	166.1	45.8	—	—	—	—	—
2	444	157.3	47.7	0.44	0.65	1.07	16.8	0.27
3	429	154.0	46.0	0.72	0.50	0.77*	31.1	0.008*
4	395	147.7	36.9	1.03	0.30	0.46*	31.4	0.002*
5	371	127.4	25.4	2.94	0.005*	0.20*	126.6	<0.001*
				Weights of Male Rats.				
1	220	195.2	48.8	0.05	0.95	2.08	7.6	0.66
Control	209	188.0	54.5	—	—	—	—	—
3	227	169.2	54.8	0.54	0.60	0.84*	17.3	0.24
2	234	167.3	53.0	0.64	0.54	0.74*	9.7	0.70
4	180	157.1	44.2	1.03	0.32	0.49*	24.0	0.013*
5	**200**	**133.0**	28.1	2.77	0.014*	0.23*	72.5	<0.001*
				Weights of Female Rats.				
1	244	158.0	31.4	1.74	0.11	0.66†	21.4	0.003†
Control	242	146.6	23.6	—	—	—	—	—
4	215	140.0	27.1	0.12	0.91	1.09	9.8	0.20
3	202	137.1	24.0	0.35	0.73	0.72*	14.8	0.01*
2	210	135.1	20.0	0.82	0.43	0.53*	17.6	0.004*
5	171	119.4	25.6	2.18	0.04*	0.27*	57.6	<0.001*

* Decrease from control group is statistically significant.
† Increase above " " " " "

of a series of 5 analogous halogenated hydrocarbons. Each group consisted of about 30 animals, with males and females in approximately equal numbers; the sexes being kept separately. These groups were exposed on alternate days for 25 weeks, and each animal was weighed weekly. A control group was exposed to air alone. Fig. 1 shows the combined growth curves of the male and female rats as normally plotted, with time the abscissa and weight the ordinate. With the exception of Group 5, it is impossible to designate which group does or does not vary significantly from the control. The "t" test was applied to the weights of the 12th week of exposure as at this time the majority of the original animals were still alive. Again, with the exception of Group 5, nothing of significance was found. As a solution to the problem, the weight data were expressed in the form of frequency distributions and the weight curves were compared as one treats normal curves of other biological data. Each individual weekly weight of each rat alive at the time of weighing was considered a point in the distribution. Separate frequency distributions were prepared for the male and the female rats. These data were combined to show the overall effect of the materials. Fig. 2 illustrates the shape of these curves, where weight is the abscissa and the frequency of occurrence of each weight is the ordinate.

With the data in the form of frequency distributions, statistical comparison of the curves is simple and straight-forward. The method of chi square reveals similar probability levels of significance, as does the ratio of 3 times the standard error of the difference, to the difference between the mean weights. In each case exposed rats were compared to controls. A summary of the chi square and "t" data is presented in Table I. The coefficient of skewness was calculated for the distributions of the control groups, and was found to be 0.48 for the combined male and female group, 0.19 for the male and 0.55 for the female controls. These values justify the use of normal statistics.

Discussion. The use of chi square is more valid for the comparison of weight-response curves than is a test of differences between means. In the chi square test the frequency with which a given weight range occurs in the treated group is directly compared to the frequency with which the same weight range occurs in the control group and there is made no arbitrary selection of a particular period for weight evaluation as is done in the "t" test. It was necessary to combine some weight classes at the extremities of distributions where the rats did not attain the spread in range of weights of the controls and where the theoretical frequencies of the classes were less than 5.

The use of a frequency distribution, in which each individual weekly weight is a point, yields a sufficient amount of valid data to make a good comparison of the weights of comparatively small groups of animals. In Table I the values for the groups of animals are listed in decreasing order of the mean weights. A remarkable relationship may be seen between the rank of the groups by this method of listing and by the magnitude of chi square and the $3\sigma d/d$ ratio.

Summary. By the application of frequency distributions to growth data, familiar statistical comparisons may be applied to determine whether significant differences exist. Previously no valid method was available to compare adequately growth curves in their entirety. The use of the chi square test on the distributions is deemed the method of preference for showing such differences.