down of erythrocytes. It seems probable that the normal lysin is present in the urine of aplastic anemia but is excreted in a bound form. Studies of Ponder[4] and of Ponder and Abels[5] suggest that the action of the ordinary inhibitors of lysis is a physical one on the erythrocyte and not a combination of inhibitor with lysin which renders the lysin inert.

Because of the relatively weak hydrolysis required to free lysin from the bound form in urines from patients with aplastic anemia, it is reasonable to suspect that some loose linkage, such as a glucuronate, is present rather than some more permanent conjugation.

Further studies of the nature of the lysin and of the method of conjugation are in progress.

Summary and Conclusions. 1. The urine of patients with aplastic anemia contains no lysin for erythrocytes when tested directly. 2. Lysin appears after hydrolysis at pH 1 and 100°C for 1 hour. 3. Such hydrolysis does not result in the production of lysin in urines which are non-lytic normally.

10038 P

The Comb of the Baby Chick as a Test for the Male Sex Hormone.

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This work is a continuation of that already reported[1] on the use of comb growth in white leghorn chicks for the assay of male sex hormone activity. The method previously employed involved the daily application of 0.1 cc of sesame oil, containing crystalline androsterone, to the region of the comb; the chicks were started on the 6th day after birth, treatment was continued for 10 days, and the combs were excised and weighed on the 17th day; the total dosage varied from 0 to 500 gamma, which corresponds to 0-50 gamma per daily application. In all of these experiments, there

4 Ponder, E., *Proc. Roy. Soc.*, 1925, **98**, 484.

5 Ponder, E., and Abels, J. C., in press.

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1 Frank, R. T., and Klempner, E., PROC. SOC. EXP. BIOL. AND MED., 1937, **36**, 763.

occurred a progressive increase in comb weight with dosage of androsterone, but the graphs were highly irregular and curved toward the dosage axis as if they might reach plateau values beyond 500 gamma. The chief aim of the present work was to determine what changes in the experimental conditions previously employed must be made in order to effect: (1) An increase in *uniformity* of the comb growth response of any one group of chicks treated simultaneously with the same quantity of hormone, this uniformity being measured by the standard deviation of the mean for the entire group. (2) An increase in *sensitivity* of the reaction, as measured by the mean difference in comb weight per unit weight of androsterone throughout the entire range of dosage employed.

To attain this objective, it has been found desirable to introduce the changes in procedure indicated in Table I.

TABLE I.

Experimental conditions	Previous study	Present study
Age of chick at beginning of application (days)	6	3
Duration of application (days)	10	7
Frequency of application	Daily	Daily
Range of total weight of androsterone per chick (gamma)	0-500	0-50
Volume of sesame oil used daily (cc)	0.1	0.05
Precautions concerning droppings, contact of chicks, segregation of different groups, uniformity of lighting conditions	Maintained throughout	

The use of younger chicks and the reduction of the period of application of the androsterone resulted in a marked increase in *uniformity* of the data; an improvement likewise resulted from the reduction in volume of oil used as vehicle, presumably because of diminished loss by spreading beyond the comb area. The *sensitivity* of the reaction was increased considerably by reducing the dosage range as indicated.

In all, 5 independent series of experiments have been performed, the number of chicks in any one series varying from 70 to 204. With one exception, each series was made up of 6 groups of chicks, each group corresponding to a different dosage value from 0 to 50 gamma inclusive. The final data were plotted as mean comb weight for all the chicks of any one group against the dosage for that group. The graph for any one series approximates a straight line, although the degree of approximation is not as good for the series based on less than 100 chicks as it is for the larger series (Fig. 1). Grossly, the lines all possess the same slopes, but their intercepts on the comb-weight axis are different; similarly, separate plots for the 2 sexes

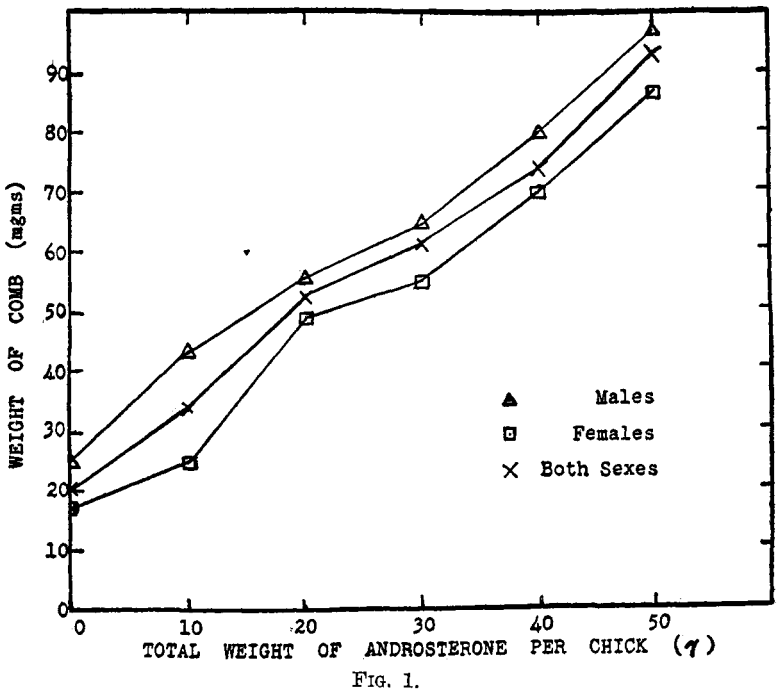

FIG. 1.

Relations between comb weight and androsterone dosage for a single series (S 18) of 150 chicks, uncorrected for control values.

in any one series manifest a higher intercept for males than for females—in conformity with the observations of Dorfman.[2] In other words, the graphs corresponding to the independent series or to different sexes of the same series appear to be distinct but parallel.

Consequently, all these graphs (including those for opposite sexes) tend to coincide if each series of data is corrected by subtracting from the mean comb weight for each group the corresponding value for the untreated chicks (controls) of the same series and sex. The identity of these corrected graphs is sufficiently close to indicate the existence of a characteristic curve for assay purposes; the fact that this curve is practically linear will make its use for these purposes a simple matter. The data for all 5 series of experiments, corrected in this way, have been combined into a single large series of 582 chicks, making adequate allowance for the number of chicks in each group. These combined plots—for separate and mixed sexes—are shown in Fig. 2.

Further efforts to increase the uniformity of the results obtained with small groups of chicks are now in progress. The final report will contain a detailed statistical analysis of the data.

[2] Dorfman, R. I., and Greulich, W. W., *Yale J. Biol. and Med.*, 1937, **10**, 79.

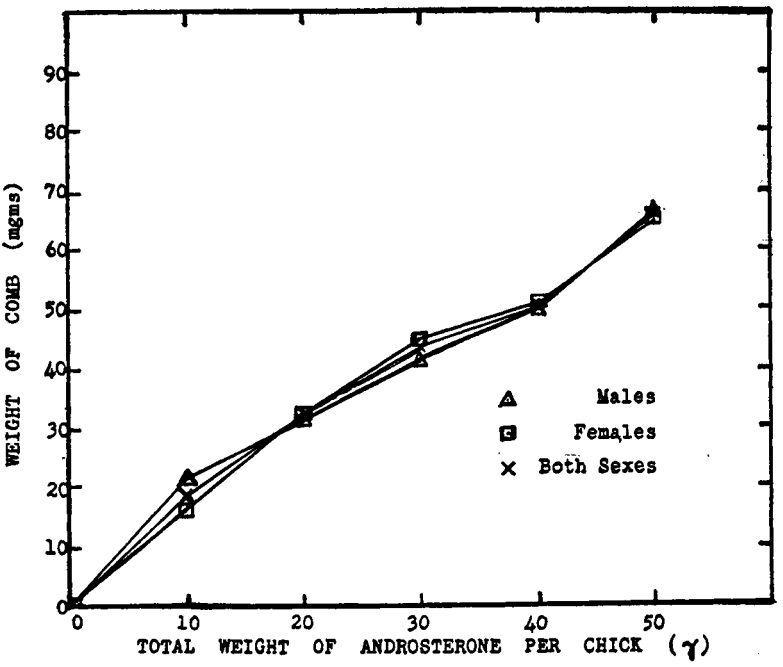

FIG. 2.

Relations between comb weight and androsterone dosage for all 5 series combined, of 582 chicks, corrected for control values.

We desire to express our thanks to Dr. Erwin Schwenk of the Schering Corporation of New Jersey for supplying us with the androsterone used in this investigation.

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Penetration of Radioactive Ions, Their Accumulation by Protoplasm of Living Cells (*Nitella coronata*).

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Exploratory experiments with K*, Na*, Rb* and Br*† reveal hitherto unknown steps of the process of penetration of these ions

* This investigation has been supported by the Board of Research, and the radioactive elements have been provided by the Radiation Laboratory of this University.

† We shall refer to radioactive isotopes of elements as K*, Na*, etc., in contrast to the inactive elements, K, Na, etc.