Determination of the Fate of Phosphorus in the Laying Hen by Means of Radiophosphorus (P^{32})[1]

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During the past few years radiophosphorus (P^{32}) has been used in a number of studies with hens (*1–6*). In these studies the P^{32} was either injected as disodium hydrogen phosphate or fed as phosphoric acid. While these results are of value in studying the fate of phosphorus, the methods of administering the phosphorus are not ordinarily used in poultry feeding. Since bone meal (consisting mainly of calcium phosphate) is a common source of phosphorus in poultry rations, an experiment in which this ingredient was replaced by calcium phosphate containing P^{32} was carried out.

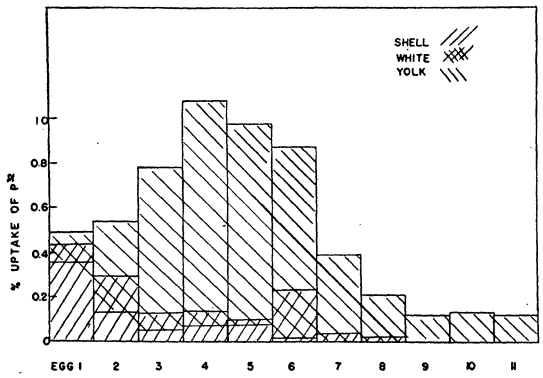

FIG. 1. Percentage of phosphate, fed as a single dose of radioactive $Ca_3(PO_4)_2$, appearing in successive eggs.

In a typical experiment the bone meal (1%) in a laying mash was replaced by 700 mg of $Ca_3(PO_4)_2$ with a P^{32} activity of approximately 10^5 disintegrations/min. In the first trial a hen was given a single feeding of P^{32}. The eggs laid subsequently were hard boiled and separated into shell, white, and yolk. After wet ashing, the phosphorus was converted to magnesium pyrophosphate and its activity determined. After allowing for self-absorption of the sample and decay of the P^{32}, the percentage of phosphate appearing in the different parts of the egg could be determined. About 5% of the phosphorus fed as calcium phosphate appears in the egg, over 80% of this being in the yolk. The P^{32} appearing in successive eggs for this hen is shown in Fig. 1

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If, now, a hen is fed the same dose of active $Ca_3(PO_4)_2$ each day, the amount of labeled phosphorus appearing in each egg should eventually reach an equilibrium value equal to the total measured above, provided the hen is laying at a fairly uniform rate (see Fig. 2).

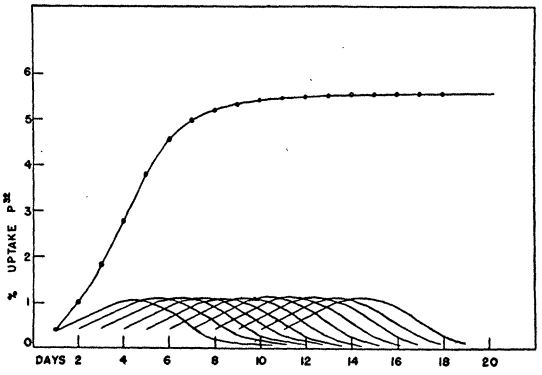

FIG. 2. Graph showing approach to equilibrium in the multiple-feeding experiment, as a summation of the phosphorus distributions for a number of successive feeds.

In order to test this theory, another hen was fed a diet containing 700 mg of calcium phosphate, having the same activity each day at the time of feeding. The results are illustrated in Fig. 3. The hen was fed P^{32} for a period of 25 successive days. These results fit in very well with the idea outlined above.

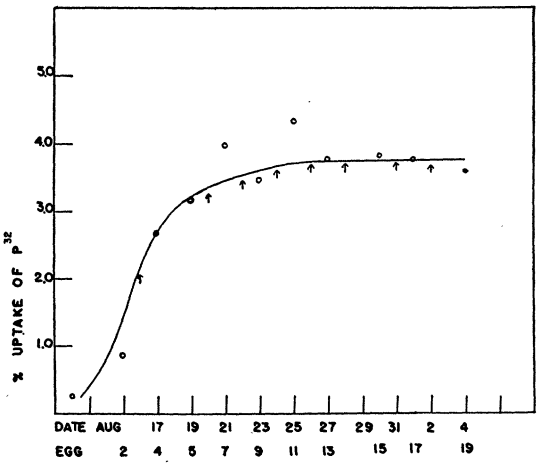

FIG. 3. Percentage of daily feed of phosphate appearing in successive eggs in sustained feeding experiment. (Arrow indicates egg laid but not analyzed.)

It is of interest to record that the phosphorus fed to the hen on a given date was still appearing in the eggs a month later. At this time, the phosphorus must have been coming from the muscles and the bones. This possibility

was confirmed by an analysis of the left tibia of the bird, which 40 days after feeding, showed about 7% of the phosphorus fed.

References

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Some Observations on the Larval Growth Rate and Viability of Two Tumor Strains of *Drosophila melanogaster*

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In a recent paper (*3*) it was pointed out that spontaneous tumors occurring in several genetic tumor strains of *Drosophila melanogaster* showed marked decrease in incidence if the cultures were maintained at a temperature of 30° C. While at lower temperatures responses in terms of incidence varied considerably in the several strains studied, the decrease in incidence at the high end of the range was uniform for all.

The nature of this decrease in incidence is of considerable interest, since the temperature change, like any environmental change, must produce its effect through the physiology of the individual. In their publications dealing with viability and survival of strains of *D. pseudoobscura* (*1*), Dobzhansky and Spassky point out that the survival values of strains homozygous for various chromosomes including wild types may vary considerably with temperature change, even approaching a lethal or semilethal condition at temperatures which might not normally be considered extreme. This being the case, one may wonder if the decrease in tumor incidence noted above is the direct result of such a semilethal or other similar effect related to the chromosomal complement of the tumor strains, which manifests itself at 30° C. On the other hand, it could result from a factor such as an altered growth rate at higher temperature, which does not affect the viability of the culture to any great degree, but may affect the expression of certain genetic characters (*2*).

The paucity of studies dealing with temperature effects on larval growth and survival in tumor strains has made it necessary to probe a few fundamental points before undertaking any detailed investigation of growth rate or survival in relation to the tumor problem.

First, we should have some idea of how the range of 20°–30° C affects the viability of the tumor strains. Second, it would seem important to know if there is any significant variation of growth rate in tumor strains which results from altered temperatures within this range. Finally, if there is variation, does each strain have its own level of reaction, or do they all respond in about the same way? Some preliminary answers to these questions may be found in the data presented in this report. These data are admittedly far from quantitative, since they are derived from experiments set up only as precursors to more thorough studies. They are published now largely as a supplement to the paper mentioned above (*3*).

Two of the tumor strains used in the previous investigation were selected as the basis for this study. These were of the genetic make-up bw tu (a strain showing high incidence of tumors at room temperature as well as low constant temperature, but showing sharp decline at 30° C), and st sr e^s ro ca, tu-36a (a strain low in incidence at 20° C and at 30° C, but showing increased incidence between these extremes). A Burlington, Vermont, wild strain was selected as a control. All three strains were fairly closely inbred, but could not be considered isogenic.

Cultures of these stocks were put in half-pint bottles which were placed in incubators set at 20° ± .5° C, 25° ± .5° C, and 30° ± .5° C. From these cultures eggs were collected on yeast-seeded molasses agar blocks, mounted on the inner surface of the bottle cap (*4*). Eggs were collected over 4-hr periods and, after collection, were transferred to 4″ Petri dishes containing molasses agar well inoculated with yeast. Since the number of eggs placed in any one dish was limited to 150, the number of dishes representing each collecting "run" varied, depending on the number of eggs collected during the period. Five hundred eggs were considered a minimum for study of each strain at each value. In some cases several collecting "runs" were necessary to gather this minimum number; in other cases all eggs were gathered in a single 4-hr period. After transfer of the eggs to the Petri dishes, they were replaced in the incubator, allowed to hatch, and the larvae allowed to develop. Fresh yeast was added from time to time to insure optimum food conditions in all cultures.

Since larval growth only was of interest, the time of pupation, marking the end of the larval period, was used as the basis for comparison of growth rate. The end point arbitrarily selected was the time when the number of pupae equaled half the number of eggs started. In timing the "runs," the 4-hr collection period was not included, the start of the "run" being set as the time of transfer of eggs into the Petri dishes.

Table 1 sets forth the data for the three strains at the three temperature values. Since the cultures were not under constant observation, but were examined at intervals during the day, the end point of 50% pupation is not exact, and the allowance for error in terms of elapsed time between the final reading and the one previous to it is stated. In a few cases this interval was overnight, and so is quite large. However, in these cases it will be noted that this large interval does not