

liver, for, unlike the dog, the rat tolerates hepatic artery ligation without antibiotic protection(7,8) and, again unlike the dog, the normal rat's liver does not harbor bacteria(9).

Summary. 1. The average portal venous pressure of rats was 15 cm H₂O in the control period, 9 cm H₂O after bleeding to an arterial pressure of 45 mm Hg, and 14.5 cm H₂O immediately after blood replacement. 2. Direct microscopy disclosed reduction in blood flow and narrowing of sinusoids, central venules and sublobular venules after bleeding from 0.7 to 1.0 cc/100 cm² of body surface. These changes became extreme after a hemorrhage of 2.1-2.4 cc/100 cm². Replacement transfusion after 3 hours of shock restored normal vessel caliber and blood flow, whether recovery followed or not.

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Embryonic Development in Turkey Eggs Laid 60-224 Days Following Removal of Males. (20202)

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Turkey spermatozoa remain viable within the body of the female for a far longer time than those of chickens. Fertile turkey eggs have been obtained and even poults hatched from eggs laid more than 50 days after removal of the male or following a single artificial insemination(1-3). An occasional fertile egg was encountered as long as 70 days after a single insemination(4).

Routine fertility checks of segregated turkey females were made by breaking incubated eggs laid by these hens prior to the time the birds were to be used in later matings. During the course of these fertility checks, eggs were encountered showing what appeared to be a delayed and retarded type of embryonic development. Furthermore, eggs showing these forms continued to be laid beyond the expected time limit recorded previously for the duration of fertility in turkeys. These observations made it desirable to study in greater detail the factor or factors responsible

for eggs showing development of this type.

Materials and Methods. A group of 30 Beltsville Small White turkey hens, 32 to 38 weeks of age were segregated from males, on Jan. 21, 1952. Prior to the time of housing these 30 hens had been with sexually mature males of their own age but none of the females had produced any eggs. Every precaution was taken to assure that no male came in contact with these females after January 21. The first egg was laid by this group of 30 hens on March 18, 1952, 54 days after the removal of the males. All eggs produced thereafter were saved for incubation and were stored in a refrigerated room maintained at a temperature of about 55°F. The relative humidity averaged about 80%. The eggs were placed in an incubator each week and each setting of eggs was incubated for a period of 7-11 days. Upon removal of the eggs from the incubator they were broken and examined macroscopically for evidences of development. The

TABLE I. Incubation Data on Eggs Laid by 30 Beltsville White Turkey Hens at Various Periods Following Removal of Males.

Month eggs laid	No. hens	Time after removal of males, days	Total No. eggs set	No. eggs showing em- bryonic de- velopment	%
Mar.	30	54- 67	39	12	30.8
April	30	68- 97	288	52	18.1
May	29	98-128	435	65	14.9
June	29	129-158	97	20	20.6
July	3	159-189	42	4	9.5
Aug.	3	190-220	27	2	7.4
Sept.	3	221-229	6	1	16.7
Totals and avg			934	156	16.7

germinal discs of representative eggs showing embryonic development were removed, fixed in Bouin's, and sectioned later. On June 4, 1952, 29 of the original 30 hens remained. Twenty-six of these were placed in breeding pens with mature males on that date. The remaining 3 hens were transferred to wire cages where they remained isolated from males. Eggs laid by these three hens were incubated until Sept. 15, 1952 when egg production ceased.

Results. Data are presented in Table I showing the total number of turkey eggs laid each month and the number and percentage of such eggs that showed some degree of embryonic development. The development encountered was of a delayed type first becoming visible before the egg candler after 4 days of incubation in contrast to normal embryonic growth which is visible at 24 hours. Furthermore when it did occur, it continued in most cases for only a limited time. Growth of the extra-embryonic membranes usually stopped after covering an area of the yolk surface equivalent to that occupied by a normal 2-or 3-day embryo. Typical examples of this delayed and retarded type of development as the membranes appear on the yolk are shown in the photographs in Fig. 1 and 2.

In most instances no definite embryo could be detected on gross macroscopic examination. Cross sections of these embryonic areas however often showed groups of centrally located cells attempting, with various degrees of success, to form embryonic folds and organs. A number of embryos were found however where blood islands had formed and in a few instances blood vessels were in evidence.

There were a number of noticeable exceptions however where more or less normal appearing embryos developed, though retarded. These included 2 live embryos from eggs laid 78 and 94 days respectively after the females were confined to the house. In both instances, the embryos after 8 days of incubation had attained a size equivalent to a normal 4-day embryo. The heart of each of these embryos was still beating at the time the eggs were examined. In both instances there was a well-developed vascular system.

A third embryo was encountered which had attained a size equivalent to that of a normal 7-day embryo. The egg in this instance had been incubated for 11 days, and was laid 195 days after the female had last been in contact with a male. This embryo, although not alive at the time of examination, still showed a well-developed system of blood vessels as is indicated in Fig. 3. A cross sectional view of this same embryo is shown in Fig. 4 where more or less normal embryonic folds and organs are in evidence.

The incidence of these retarded forms varied considerably in the eggs laid by different hens. Twenty-eight of the original 30 hens produced one or more eggs showing delayed and retarded type of development during the period covered by this experiment.

Of those hens producing eggs with this type of retarded development, 8 individuals laid eggs having less than 10% of such forms. On the other hand, 7 others produced eggs in which 30% to 60% showed embryonic development. The average for the eggs of all birds during the entire period was 16.7%.

The percentage of eggs showing delayed

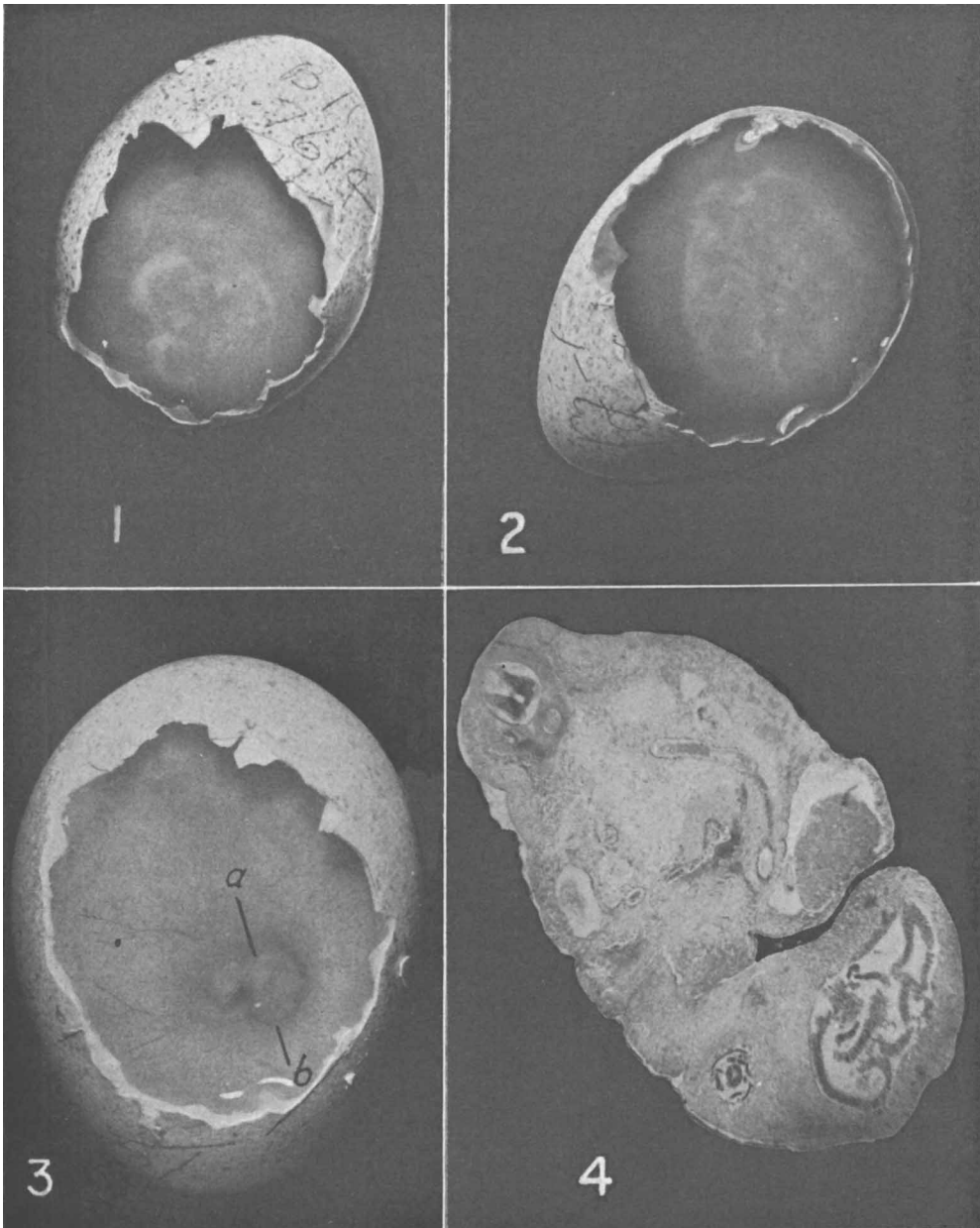


FIG. 1 and 2. Eggs of 2 turkeys showing embryonic development which occurred during a 7-day incubation period; eggs laid more than 200 days after females were isolated from males.

FIG. 3. Embryo in egg laid 195 days after isolation of the female and following an 11-day incubation period. Embryo approximately the size of normal 7-day embryo.

FIG. 4. Cross section of embryo shown in Fig. 3. Section shown is through region indicated by line ab.

embryonic development during July and August was considerably lower than that recorded for the prior months. This apparent decrease in the incidence of eggs showing these delayed forms (Table I) can be

accounted for when the records of the 3 hens used for this particular portion of the experiment are examined. One of these 3 hens never produced an egg throughout the entire season that showed these retarded embryonic

forms. A second hen laid eggs averaging only 5% of these forms while the third hen produced eggs, 13.3% of which showed embryonic development during the entire season.

Discussion. It is evident from the data presented that delayed and retarded embryonic development occurred in some eggs produced by the Beltsville Small White turkeys referred to in this paper. The activating factor is uncertain in this instance since the eggs were produced by turkey hens which had at one time been in contact with sexually mature males. This much is certain however, that some of these developmental forms occurred in eggs laid by hens which had been separated from males for more than 200 days at the time the eggs were laid. This is about 3 times longer than the 70-day figure previously given (4), as the maximum duration of fertility in Broad Breasted Bronze turkeys. This would lead one to suspect, if we assume that sperm initiated this form of development, that the spermatozoa were stored for these prolonged periods in a more favorable environment than that found in the upper end of the turkey oviduct. One likely location would be within the young follicles on the ovary. It has already been shown in the case of the domestic hen that sperm are able, if deposited on the surface of the immature ovary, to enter the young ova and live there until the egg matures and in some instances to initiate development after the ovum leaves the ovary (5). Fertile chicken eggs were reported as long as 5 months after the sperm had been applied to the surface of the immature chicken ovary.

If it is concluded that sperm were not the activating factor in this instance, then it must be assumed that the egg itself, in the absence of the sperm, was activated by some other stimulus and induced to undergo some type of development. If this were true it might be a case of natural but delayed parthenogenesis in a higher order of animal. To our knowledge no case of natural parthenogenesis, to this extent has ever been reported in birds. It is true that several investigators have reported what they considered to be cells, some nucleated, in the blastodisc of newly laid infertile chicken eggs produced by non-mated

and virgin females (6-10). This, however, involved at the most only a relatively few cells.

Until more critical experiments are conducted in which turkey poults of the two sexes are separated at an early age, so that the role of sperm can be ruled out, the activating factor involved must remain in doubt. Such critical experiments are not being conducted at the United States Agricultural Research Center, Beltsville, Md.

Summary. A delayed and retarded type of embryonic development occurred in 16.7% of the Beltsville Small White turkey eggs laid 54-224 days after the females were separated from males. This development in most instances could not be detected macroscopically earlier than the 4th day of incubation. The embryonic membranes usually discontinued their growth after covering an area of the yolk equal to that occupied by a normal 2- to 3-day turkey embryo. In most instances no embryo could be detected macroscopically although serial cross sections showed a definite organization on the part of centrally located cells. Two living embryos were found in eggs laid 78 and 98 days respectively after the hens were separated from the males. These embryos, after 8 days of incubation had attained a size equivalent to a normal 4-day turkey embryo. The embryonic hearts were still beating and there was a well developed vascular system in both cases. A 7-day embryo, with a well-developed vascular system was also found in an egg which had been incubated 11 days. This egg was laid 195 days after the female had been separated from males.

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