A Critical Period for the Turning of Hens' Eggs

by D. A. T. NEW[1]

From the Department of Anatomy and Embryology, University College London

INTRODUCTION

THE artificial incubation of hens' eggs involves four factors—temperature, humidity, air supply, and at intervals a rotation or 'turning' of the eggs. This last factor is perhaps the most curious and unexpected of the four, yet there is no doubt that it is necessary for development as shown by good hatchability, and in the natural state it is carried out by the sitting hen. Eycleshymer (1906), Chattock (1925), and Olsen (1930) have all concluded from observations on the hen's nest that the hen frequently rotates the eggs during the incubation period; Olsen considers it occurs as often as 96 times in 24 hours.

Various abnormalities have been recorded in eggs incubated without turning. Dareste (1891) stated that absence of turning causes the allantois to adhere to the yolk sac; Eycleshymer (1906) confirmed this, and added that during the first week of incubation, absence of turning may also cause the embryo to adhere to the shell membranes. Randles & Romanoff (1950) have observed further effects: in unturned eggs the formation of the albumen sac is delayed and the physical properties of the amniotic and allantoic fluids differ from normal.

Although the relation between frequency of turning and hatchability has been intensively studied (see Landauer, 1951, pp. 54–55 for references), little appears to be known as to the mechanism by which insufficient turning induces abnormalities. A useful line of approach to this question would be to find the relative importance of turning eggs at different stages of the incubation period. If it were known that at a particular stage of development the embryo was exceptionally sensitive to the absence of turning, it might be possible to deduce what the function of turning was from a consideration of the condition of the egg contents at that time; or at least the range of possible explanations might be greatly narrowed. Such few indications as have been obtained previously suggest that turning is more important in early than in late incubation. For example, Byerly & Olsen (1936) conclude that turning in the third week probably has little effect on hatchability, and Card (1926) observed that eggs turned during the first 6 days hatch nearly as well as those turned throughout incubation, but he gives no figures.

[1] *Author's address*: Department of Anatomy and Embryology, University College London, Gower Street, W.C. 1, U.K.

[J. Embryol. exp. Morph. Vol. 5, Part 3, pp. 293-299, September 1957]

Consideration of the arrangement of the egg contents suggests that a particularly critical time for turning the eggs might be the latter part of the first week of incubation. At this time a large area of chorion lies close to the shell membranes and the layer of albumen between the two has been greatly reduced by a loss of fluid from the albumen to the yolk. Abnormal adhesions between the chorion and shell membranes, therefore, seem a possibility at this stage unless the shell and its membranes are periodically moved relative to the egg contents, i.e. unless the eggs are turned. The present work consists of evidence in support of this explanation. It will be described in two sections, the first indicating that the most important time for turning the eggs is between the 4th and 7th days of incubation, and the second suggesting that turning during this period exerts its marked effect on hatchability by preventing harmful adhesions arising between the chorion and shell membranes.

METHOD AND RESULTS

(*a*) *Hatchability of eggs turned only between the 4th and 7th day*

Table 1 summarizes the results of four experiments designed to test the effect on hatchability of turning the eggs only between the 4th and 7th day (inclusive). The eggs were incubated at 39° C. (heated from above), at about 60 per cent. relative humidity, and turned on the days indicated. Turning was carried out once every 12 hours and the eggs rotated by hand approximately 120° about the long axis; they were rotated alternately right and left.

TABLE 1

Hatchability of eggs after various turning treatments during incubation

Experiment No.	*Turned 4th–7th day*		*Turned 8th–11th day*		*Turned throughout*		*Unturned*	
	Failed	*Hatched*	*Failed*	*Hatched*	*Failed*	*Hatched*	*Failed*	*Hatched*
1 . .	2	6	5	3	2	6	4	4
2 . .	2	7	7	2	5	4	8	1
3 . .	6	3	9	0	2	7	6	3
4 . .	4	5	8	1	2	7	8	1
TOTALS .	14	21	29	6	11	24	26	9

In each experiment the eggs were divided into four equal groups. The first group was turned between the 4th and 7th day, the second between the 8th and 11th day, the third throughout incubation up to the 18th day, and the fourth was not turned at all. The eggs of the four groups were interspersed to avoid differences arising from microclimates within the incubator. The purpose of including a group turned only between the 8th and 11th day was to find whether any improvement in hatchability of the 4–7 day group was restricted to eggs turned specifically during this period, or whether it resulted from turning them

during any 3-day period at about the middle of incubation. Turning of the 'turned-throughout' group was stopped at the 18th day to conform with the normal practice in poultry rearing.

The results of the four experiments added together show clearly that turning the eggs between the 4th and 7th day is similar in its effects on hatchability to turning them throughout the whole incubation period, and turning the eggs only between the 8th and 11th day gives similar results to leaving them completely unturned. Calculation of χ squared values for these two pairs of results also fails to detect significant differences. But χ squared calculated for a comparison of the results of turning 4th to 7th day with those of turning 8th–11th gives a P value of less than 0·001. It seems unquestionable, therefore, that turning the eggs between the 4th and 7th day of incubation gives an improvement in hatchability similar to that obtained by turning the eggs throughout incubation, and the results further suggest that this improvement is limited to eggs which are turned specifically at this stage.

In adding the results of separate experiments together the question of homogeneity arises. The numbers involved in each experiment are here too small to test for homogeneity by the χ squared method. However, if for each treatment the two results are selected which show the greatest difference, it can be shown that the probability of their belonging to different populations is insignificant; e.g. the extreme cases resulting from turning between the 4th and 7th day were 2 failed, 7 hatched; and 6 failed, 3 hatched. The probability of these belonging to the same population is 0·0767.

TABLE 2

Distribution of mortality during incubation

	Infertile	*Died 1st week*	*Died 2nd week*	*Died 3rd week*	*Hatched*
Turned 4th–7th day .	4	1	2	7	21
Turned 8th–11th day .	7	7	7	8	6
Turned throughout . .	4	5	0	2	24
Unturned . . .	6	3	2	15	9

The eggs that did not hatch were opened on the 25th day and the time of death estimated from the extent of embryonic development. Table 2 shows the totals from the four experiments. The division between 'infertile' and 'died 1st week' can only be considered as roughly established since in those cases where the blastoderm died very young it is not always possible to distinguish the egg contents from the infertile condition, particularly after 3 weeks of incubation. The table shows that by far the heaviest mortality of unturned eggs occurs during the third week and it is this mortality which is most affected by turning throughout incubation or from the 4th to 7th day. The results also suggest that turning from the 8th to 11th day reduces the third-week mortality but correspondingly in-

creases mortality in the second week; however, the figures are not large enough to constitute a convincing proof of this.

In those eggs which had died after the 18th day of incubation, malpositions of the chick were fairly common. Of the 26 dead chicks in this group, 6 were found to have the head in the small end of the egg, 4 to have the beak away from the air sac, 3 to have the head under the left wing, and 1 to have the head between the thighs.

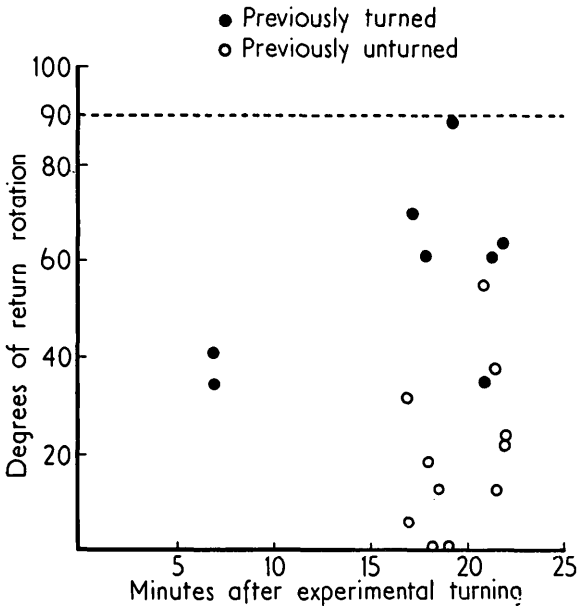
(*b*) *Adhesion between the chorion and shell membranes*

Statements that insufficient turning leads to adhesions between the chorion (or blastoderm) and shell membranes have frequently been made (e.g. Eycleshymer, 1906) but it is not quite clear what evidence they are based on. The chorion is very close to the shell membranes whether the eggs are turned or unturned, and in the second week of incubation the chorio-allantois adheres to the shell membranes as a normal feature of development. Furthermore, a small proportion of unturned eggs develop successfully and produce normal chicks at hatching. It would need, therefore, fairly carefully controlled observations to be certain that abnormal adhesions were occurring.

With this in mind the following experiment was devised. Two groups of fertile eggs were incubated for 7 days. The first group (11 eggs) was left unturned during this period; the second group (8 eggs) was turned every 12 hours between the 4th and 7th day (inclusive). On the 7th day all the eggs were candled and the position of the embryonic membranes marked by drawing a pencil line on the shell over some prominent blood-vessel (usually the sinus terminalis). Each egg was then rotated through 90° and left in this position for a few minutes. On candling for the second time the embryo was seen to have begun to move back towards its former position at the top of the egg, and the embryonic membranes had correspondingly moved relative to the pencil mark on the shell. By measuring with calipers the distance moved by the membranes and also the circumference of the egg, sufficient data were provided for calculating the angle through which the egg contents had turned. The results are plotted in the accompanying graph (Text-fig. 1), which shows the angle through which the egg contents have rotated towards their former position at a given time after the egg was turned through 90°. It can clearly be seen that in the eggs that were unturned during the period of incubation, movement of the embryonic membranes relative to the shell is very much slower than in eggs that were turned between the 4th and 7th days; in fact two of the 'unturned' group have not moved at all.

The eggs were returned to the incubator and examined again 3 hours later. In all those of the 'turned' group the contents had moved through more than 80°, whilst in over half those of the 'unturned' group the contents had rotated less than 60°. The only explanation that seems reasonable is that turning the eggs between the 4th and 7th day of incubation lowers the resistence to relative

movement of the chorion and shell membrane; or in other words, absence of turning encourages adherence between the two.



TEXT-FIG. 1. Each egg was turned through 90°. The subsequent internal rotation by which the egg contents returned towards their former position is plotted (in degrees) against time after the whole egg was turned. Of the two groups of eggs shown, one had been systematically turned during incubation, the other had not.

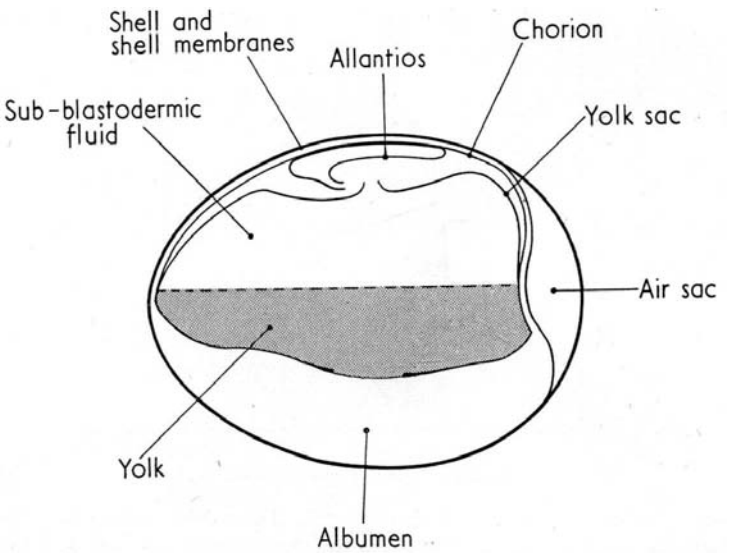
After these observations had been made, incubation of the eggs was continued without any further turning. Of the unturned group (actually turned once through 90° during the experiment on the 7th day) 2 out of 11 eventually hatched. Of the group turned between the 4th and 7th day 6 out of 8 hatched. This lends further support to conclusions reached in the previous section.

DISCUSSION

It is clear that the turning of hens' eggs is particularly important during the latter half of the first week of incubation. If the eggs are turned at this time good hatchability is obtained even if they are left unturned during the remainder of the incubation period. If they are not turned at all during incubation, however, hatchability is low. It has been shown that absence of turning results in an abnormal degree of adhesion between the chorion and shell membranes during the first week of incubation; the probable inference is, therefore, that this abnormal adhesion is closely connected with the causes of the high mortality found in unturned eggs.

Yet there remains the curious fact that, during the second week of incubation,

firm adhesion between the chorion (chorio-allantois) and the shell membranes occurs as a normal feature of development. This normal adhesion is evidently primarily related to the respiratory needs of the embryo. It is important that respiratory surfaces should be applied as closely as possible to the shell; in fact it has been shown (Danchakoff, 1917) that the allantoic capillaries actually move to a position outside the chorionic ectoderm.



TEXT-FIG. 2. Distribution of some of the egg contents at the 6th day of incubation. Based on a sagittal section of an egg fixed by boiling. (For the sake of clarity the distance between the chorion and shell membrane has been exaggerated.)

It seems, therefore, that turning is not a device to prevent adherence altogether, but to prevent it occurring too soon. Why is this so important? The answer may be in the state of development of the chorion and yolk sac during the first and second weeks. Although these two membranes have practically surrounded the yolk by the 4th day, they continue to expand rapidly until the end of the 6th day (Grodzinski, 1934) owing to the increase of the yolk-sac contents as a result of sub-blastodermic fluid formation. Probably, therefore, any adhesion of the chorion to the shell membrane before the 6th day would hinder its normal development, but after this time would not be so serious.

Part of the mechanism involved in premature adhesion is likely to be the rapid water loss from the albumen which is occurring at this time. Until the end of the first week the albumen is constantly losing water (*a*) by evaporation from the shell surface, and (*b*) via the blastoderm to form the sub-blastodermic fluid (New, 1956). Since the albumen layer between much of the chorion and shell membranes has become extremely thin by the end of the first week of incubation and the area of chorion close to the shell membrane has become very large (Text-fig. 2), desiccation of this albumen layer would seem greatly to increase the

chances of adhesion. Turning the egg at this stage might well prevent such desiccation by supplying a fresh layer of watery albumen over the dry regions of the membranes.

As mentioned previously a variety of abnormalities have been observed in unturned eggs. The present work suggests, however, that many of these abnormalities are likely to be secondary effects of an initial interference with development occurring between the 4th and 7th day.

SUMMARY

1. Turning hens' eggs between the 4th and 7th day of incubation gives a hatchability similar to that of eggs turned throughout incubation.

2. Turning the eggs between the 8th and 11th day gives a hatchability similar to that of unturned eggs.

3. New evidence is presented that absence of turning leads to abnormal adhesion between the chorion and shell membranes.

4. The mechanism by which turning effects development is discussed.

ACKNOWLEDGEMENTS

I should like to thank Mr. M. Abercrombie for much useful advice and helpful criticism. I am also indebted to Dr. D. E. Barton for examining the statistics and to the Agricultural Research Council for financial support.

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(Manuscript received 31 : x : 56)