

# Orientation of chick embryos in static magnetic fields

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Saali, K. & Juutilainen, J. 1988: Orientation of chick embryos in static magnetic fields. — *Ann. Zool. Fennici* 25:187–189.

After incubation of eggs from 70 to 75 hours in a local geomagnetic field, the orientation of chick embryos differed significantly from the random distribution. When another batch of eggs was incubated in the presence of an artificial field that turned the horizontal component of the resultant field by 90°, there was a corresponding change in the orientation of the embryos. The head-tail axis of the embryos had a tendency to be oriented perpendicularly to the magnetic field. The results suggest that static magnetic fields affect the orientation of chick embryos during their first stages of development.

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## 1. Introduction

Low frequency oscillating magnetic fields have been reported as disturbing the development of chick embryos (Delgado et al. 1982, Ubeda et al. 1983, Juutilainen et al. 1986, Juutilainen 1986, Juutilainen & Saali 1986). The dependence of this effect on the parameters of the field suggests that the mechanism is based on direct magnetic interaction rather than induced electric currents (Juutilainen et al. 1986, Juutilainen & Saali 1986). This kind of effect would necessitate some mechanism of coupling between the embryos and the magnetic field.

In order to study the possible magnetic properties of chick embryos, experiments were carried out to determine whether the embryos orient themselves in the geomagnetic field.

## 2. Material and methods

Fresh fertilized eggs of the breed Mäkelä 16 (a variety of White Leghorn) were obtained from a farm specialized in the production of chickens and fertilized eggs (Ollilan Siitoskanala Ky, Hirvilahti, Kuopio). The eggs were incubated at 37–38°C and 55% relative humidity, in a room with temperature and humidity control and forced air circulation.

The eggs were incubated in a vertical position, with the air space end down in the standard cardboard holder (30×30 cm) used for the transportation of 30 eggs. This orientation of the eggs was chosen after preliminary experiments with other orientations. When the eggs lie with their longitudinal axis in a

horizontal position, the orientation of the embryo is perpendicular to the long axis of the egg because of the determination of the polarity of the blastoderm in the uterus of the hen (Eyal-Giladi 1984). Thus, a horizontal positioning of the eggs is not suitable for this kind of experiment, although some changes in the predetermined orientation suggestive of a magnetic field effect were seen in the preliminary experiments. When the air space end was uppermost, the irregular shape of the air space affected the orientation of the embryos.

Before starting the incubation, each egg was turned several times in order to ensure free floating of the yolk and to prevent the embryo from becoming attached to the egg's shell.

After 70 to 75 hours of incubation, the sharp end of the shell was gently broken and the orientation of the embryo was determined with a transparent compass. The direction of the embryo's head-tail axis was marked on the shell of the egg with a pencil. The compass was then placed on the egg, and, looking through the compass, the angle between the compass needle and the head-tail axis was read off in degrees. Only living embryos floating on the top of the egg were accepted. The local geomagnetic field in the room was 44  $\mu$ T, and its inclination was about 60°.

As the results of the first experiment in the natural magnetic field suggested that the embryos were not randomly oriented, another experiment with an artificial static magnetic field was carried out in order to determine whether the orientation of the embryos in relation to the geographical directions is due to the geomagnetic field or other forces. The artificial static field was generated by two square ten-turn coils (40×40×1 cm) at a distance of 24 cm from each other. This Helmholtz coil system was placed so that the angle between the north-south axis of the geomagnetic field and the axis of the coil system was about 130°. The electric current to the coils was adjusted so that the needle of the compass placed at the centre of the system turned –90° in relation to the horizontal component of the geomagnetic

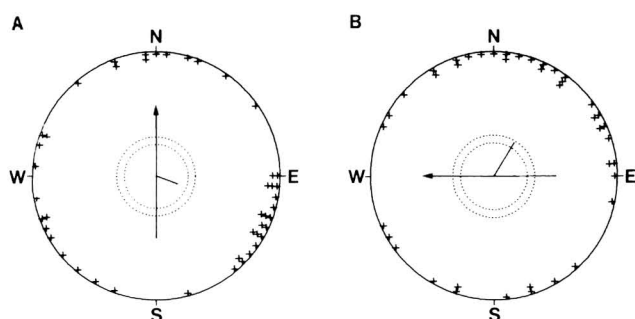


Fig. 1. Orientation of chick embryos in static magnetic field. The direction of the horizontal component of the magnetic field is illustrated by an arrow. The direction of the vector at the centre gives the mean orientation angle of the embryos, and its length gives the significance according to the Rayleigh test. The inner and outer dotted circles represent the vector length required for the 5% and 1% significance levels, respectively. The symbols N, E, S and W refer to geographical directions and not to the poles of the resultant magnetic field. — A. Embryos ( $n=45$ ) in the geomagnetic field. — B. Embryos ( $n=41$ ) in the combination of the geomagnetic field and an artificial static field.

Table 1. Distribution of the orientation angle of the embryos.  $0^\circ$  corresponds to alignment with the horizontal component of the total field with the head towards the north pole of the field. The difference from random (even) distribution was tested with the chi-square test.

Orientation angle (in degrees)	Geomagnetic field	Combined geomagnetic and artificial field
0–89	6	9
90–179	21	19
180–269	9	7
270–359	9	6
	$\chi^2=11.8$ $P<0.01$	$\chi^2=10.4$ $P<0.025$

field. The calculated magnetic flux density of the artificial field at the centre of the coil system was  $34 \mu\text{T}$  (Saali et al. 1986). The determination of the orientation of the embryos was carried out in the same way as in the experiment with the natural field. The artificial field was kept on during this determination.

The chi-square test was used to examine the possible difference of the orientation from the random distribution, and the Rayleigh test (Batschelet 1981, Mardia 1972) was used to test the data for orientation around a certain mean angle (unimodal distribution). The Watson's  $U^2$  test (Batschelet 1981, Mardia 1972) was used to test whether the presence of the artificial field significantly changes the orientation of the embryos.

### 3. Results

In the geomagnetic field experiment, there was a cluster in the orientation angle between  $90^\circ$  and  $150^\circ$  (Fig. 1A). The mean angle of orientation was  $110^\circ$ . The orientation of the embryos differed significantly ( $P<0.01$ ) from the random distribution (Table 1). However, the Rayleigh test did not support the hypothesis of clustering about a certain mean angle,

which suggests that the distribution is not unimodal. Visual impression of the distribution (Fig. 1A) suggest that the distribution may be bimodal, with another, weaker cluster between  $210^\circ$  and  $300^\circ$ .

In the second experiment, the direction of the horizontal component of the resultant static field was turned  $-90^\circ$  from the north-south axis of the geomagnetic field. The orientation of chick embryos seemed to follow this change (Fig. 1B). The mean angle of orientation was  $31^\circ$ . Thus, a change of  $-90^\circ$  in the magnetic field produced a change of  $-79^\circ$  in the mean orientation of the embryos. The change in the orientation was significant at  $P<0.005$  (Watson's  $U^2$  test). Again, the distribution of the orientation differed significantly ( $P<0.025$ ) from the random distribution (Table 1). In contrast to the geomagnetic field experiment, the Rayleigh test suggests a unimodal distribution ( $P<0.05$ ).

### 4. Discussion

Chick embryos seem to have a tendency to be oriented approximately perpendicularly to the geomagnetic field after their first three days of development. The results of the experiment with the artificial field suggested that the embryos followed the horizontal component of the resultant magnetic field rather than other forces (e.g. the coriolis force) related to geographical north-south direction.

The orientation effect is quite weak: all embryos are not clearly oriented in the same direction. This, together with the fact that embryos oriented in unusual directions were morphologically normal, suggests that the magnetic orientation does not necessarily have a physiological meaning for embryos.

It is known that some birds and other organisms are able to orient themselves in the geomagnetic field.

In magnetotactic bacteria, this ability is based on biologically precipitated magnetite ( $\text{FeOFe}_2\text{O}_3$ ) (Kirschvink & Gould 1981). The mechanism of the birds' magnetic sense is not known. Magnetite, as well as other mechanisms, have been proposed (Kirschvink & Gould 1981, Yorke 1981). It is not known whether 0 to 3 days old chick embryos contain magnetite.

For developing embryos, one possible mechanism of orientation could be based on the observations of Jaffe & Stern (1979) that direct electric currents flow through the primitive streak of chick embryos with return currents outside the embryos. The current loops thus formed generate weak static magnetic fields. The currents flow in several directions, but the current patterns measured by Jaffe & Stern (1979) suggest that there is some asymmetry, and the embryo may thus have some net magnetic moment. As the strongest currents (up to  $0.25 \mu\text{A}/\text{mm}^2$ ) seem to flow along the primitive streak, it is possible that the embryo at the primitive streak stage behaves as a magnetic dipole with the north-south axis perpendicular to the embryo's head-tail axis. An external magnetic field could possibly affect the orientation of this dipole during the formation of the primitive streak. Magnetic effects on orientation are less probable at later stages of development because of the large size of the embryo and the complex pattern of currents

flowing in several directions. If it is assumed that a current of  $0.01\text{--}0.1 \mu\text{A}$  flows in a loop with a surface area of  $0.1\text{--}0.5 \text{ mm}^2$ , it can be calculated that the magnetic moment  $\mu$  of the loop is  $10^{-14}\text{--}10^{-15} \text{ Am}^2$ . This is larger than the magnetic moments of about  $10^{-16} \text{ Am}^2$  measured for magnetotactic bacteria (Rosenblatt et al. 1982). Generally, magnetic orientation is possible if the magnetic interaction energy  $-\vec{\mu} \cdot \vec{B}$  ( $\vec{B}$  is magnetic flux density) exceeds the thermal energy  $kT$  ( $k$  is Boltzman's constant and  $T$  is temperature). For a magnetic moment of  $10^{-15} \text{ Am}^2$ , the interaction energy in the geomagnetic field of about  $40 \mu\text{T}$  is about one-order of magnitude higher than  $kT$  at a temperature of  $310 \text{ K}$ .

The results strongly suggest that chick embryos are able to orient themselves in static magnetic fields and thus have properties resembling those of magnetic dipoles. Whatever is the biophysical basis of this interaction, the same mechanism could be one possible explanation to the teratogenic effects of low-frequency alternating magnetic fields (Delgado et al. 1982, Ubada et al. 1983, Juutilainen et al. 1986, Juutilainen 1986, Juutilainen & Saali 1986).

*Acknowledgements.* This work was supported by the Finnish Research Council for Natural Sciences and the Imatran Voima Foundation.

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