

Biological effects of low-frequency pulsed magnetic fields on the embryonic central nervous system development. A histological and histochemical study

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Summary. Numerous experiments have yielded contradictory results on the harmful action of magnetic fields on embryonic development. Pulsed magnetic fields appear to be able to delay normal development of embryos. In the present study, fertilized *Gallus domesticus* eggs were exposed during incubation to pulsed magnetic fields (harmonic signals of 10 μ T for 1 second with silences of 0.5 seconds) of 50 or 100 Hz frequency.

Embryos extracted at 45 h of exposure to fields of 50 Hz or 100 Hz frequency had significantly ($p < 0.05$) fewer somite pairs compared with controls of the same age. At 15 days of incubation, only embryos exposed to a 10 μ T-50 Hz field had a significantly ($p < 0.05$) higher somatic weight. At 21 days of incubation, a significantly lower somatic weight ($p < 0.01$) and development stage ($p < 0.05$) was found in embryos exposed to a 10 μ T-100 Hz field than in controls, while a lower development stage ($p < 0.05$) alone was observed in those exposed to a 10 μ T-50 Hz field. In addition, animals showed higher expression of the neural marker NSE (neural specific enolase) after 21 days of development as determined by immunohistochemistry, with very low expression of glycosaminoglycans identified by alcian blue staining.

These results suggest that pulsed magnetic fields may be able to hinder normal embryonic development *in vivo* and to alter normal neural function, at least at the intensities and frequencies analyzed in the present study.

Key words: Pulsed magnetic fields, Histochemistry, Immunohistochemistry, Central nervous system development

Introduction

Over the past 25 years, a substantial body of literature has been devoted to the biological effects of magnetic fields. Two types of experiments have largely been carried out, those in which the animals are exposed to constant magnetic fields and those in which pulsed magnetic fields are used. While some researchers reported their innocuousness or therapeutic utility, most concluded that they are responsible for a series of disorders that can often lead to an increase in neoplastic processes. The constant use and abuse of electrical energy has increased environmental magnetic contamination to levels far beyond those naturally experienced by humans. Although the importance of the potentially harmful effects of magnetic fields has been underplayed at an official level, concerns have been mounting and an increasing number of experiments have been performed around this issue. However, no definitive conclusion can be drawn from the contradictory results of epidemiological and experimental research published to date.

The therapeutic utility of low-frequency magnetic fields is virtually restricted to their action in accelerating fracture consolidation (Ottani et al., 1991; Fredericks et al., 2000; Trock, 2000). However, most authors have reported that magnetic fields of different intensity and frequency can induce carcinogenesis (Loscher et al., 1993; Rannu et al., 1993; Mevissen et al., 1996, 1998;

Zhao et al., 1999; Saito et al., 2010), although some have refuted their carcinogenic action (Harris et al., 1998; Mc. Cann et al., 2000).

The biological effects of magnetic fields on embryonic development have been experimentally studied. In 1982, Delgado et al. exposed fertilized *Gallus domesticus* eggs to low-frequency magnetic fields during the first 48 h of incubation at intensities of 1.2 - 12 μ T. They found inhibited development of cerebral vesicles, anterior intestine, vessels, and somites, and they reported a significant reduction of glycosaminoglycans in the brain of the animals that might be the cause of this abnormal development (Ubeda et al., 1983). These authors conclude that chick embryos are sensitive to electromagnetic fields of extremely low frequency and intensity, and hypothesize that alterations in extracellular glycosaminoglycans could be a causal factor in the observed malformations (Ubeda et al., 1983). Other authors have also reported that magnetic fields of different intensity and frequency produce some enzymatic, neurochemical and even developmental disorders in chicks (Grandolfo et al., 1991; Moses and Martin, 1993; Rajendra et al., 2004; Saito et al., 2010).

Similar experiments in gestating rats and birds have also shown significantly increased disorders in newborns, including low body weight (Sienkiewicz et al., 1994; Svendenstal et al., 1995; Graham et al., 2000), immature cerebellum (Espinar et al., 1997), and the altered production of enzymes involved in regulating growth of cells and calcium balance, among others (Holmberg, 1995).

Effects on embryonic development have also been reported after other types of magnetic field exposure, interposing silent periods (pulsed magnetic fields) and using different types of low-frequency wave (sinusoidal, square, rectangular, etc.). In 1986, Juutilainen and Saali reported developmental disorders in chick embryos exposed to pulsed magnetic fields with sinusoidal and rectangular waves at 100 Hz frequency. On the other hand, Coulton and Barrer (1991) found no developmental disorder after exposing embryos from day 4 of incubation to 1 or 2 μ T pulsed magnetic fields in 5 ms series every 15 h for 100 h. Nevertheless, in 1994, Ubeda et al. concluded from chick embryo experiments that pulsed magnetic fields can produce an increase in embryonic development anomalies.

In addition, teratologic disorders have also been produced in mice and rats by magnetic fields. Thus, Frölen et al. (1993) reported a reduction in the body weight and length of mouse fetuses exposed to pulsed magnetic fields on day 7 of gestation. Likewise, Huuskonen et al. (1993) observed a significant rise in minor skeletal anomalies in fetuses of rats exposed to low-frequency pulsed (15 μ T) and non-pulsed magnetic fields.

According to the above review, the most of the authors found that pulsed or non-pulsed magnetic fields can produce developmental anomalies that are detectable in the embryo and newborn. The present study was

designed to explore the effects on different phases of pulsed magnetic fields of the same intensity but different frequencies on chick embryonic development and with a silent time.

Materials and methods

Animals and groups of study

Fertilized eggs of *Gallus domesticus* (Leghorn HR7 variety) were incubated at $37.8^{\circ}\pm 0.4^{\circ}\text{C}$ and relative humidity of 49-60% in a model 65 Masalles incubator equipped with forced ventilation and automatic voltage (1 V/h).

The chick embryo was chosen as a model because of its self-sufficient development system, ruling out maternal interference (such as in mammalian models) and allowing a more specific analysis of any direct teratogenic action of the magnetic fields under study. The chick embryo model is also easy to monitor and control throughout the development period.

Eggs in plastic trays were introduced into an incubator sited between two Helmholtz coils placed in parallel and 70 cm apart (each coil has a 70 cm radius of action). This design ensured that all eggs were exposed to a homogeneous magnetic field, confirmed by constant testing of the homogeneity of the field throughout the experiment.

A pulsed magnetic field of 50 or 100 Hz at an intensity of 10 μ T was generated by using a signal generator (Good-Will, model GFG-8015G) and cyclic timer (Omron, model H3de-F) to send pulsed current to the coils. The presence of the magnetic field (1 sec exposures) in the embryo cells was followed by an absence of the field (silences of 0.5 sec) imitating more closely the exposure to magnetic fields experienced in daily life.

The experiment used 440 fertilized eggs randomly assigned to one of the following groups:

Experiment 1. Group A: 123 eggs exposed to magnetic fields of 10 μ T and 50 Hz frequency. Group A-CTR: 98 eggs used as controls. These eggs were incubated in an incubator that was identical to Group A but without any magnetic field exposition.

Experiment 2. Group B: 121 eggs exposed to magnetic fields of 10 μ T intensity and 100 Hz frequency. Group B-CTR: 98 eggs used as controls. These eggs were incubated in an incubator that was identical to Group B but without any magnetic field exposition

Morphological parameters

In both experiments, a first extraction was made at 45 h of 20 treated and 24 control eggs, to study the somitic development; at 15 days, 20 eggs were extracted from each group to study the somatic weight and development stage (third-toe length). In both experiments eggs were randomly selected.

From day 15, half of the treated eggs in groups A

and B were withdrawn from the magnetic fields and placed in identical but unexposed incubators, whereas the other half remained exposed until hatching.

A final sampling was conducted at 21 days of incubation. In each group (A and B), 20 newly-hatched chicks were extracted from eggs that had remained exposed to the magnetic fields for 21 days, 20 from those exposed for only the first 15 days of incubation, and 20 from those that were never exposed.

Embryos extracted at 45 h were fixed with 1% osmium tetroxide solution and observed with a stereomicroscope for somite count and stage determination. The stage of development was determined by using the Hamburger-Hamilton scale. In embryos extracted at 15 and 21 days, the somatic weight was measured and expressed in grams per embryo, and the development stage was determined by third toe length (expressed in millimeters) (Hamburger and Hamilton, 1992).

Immunohistochemistry and histochemistry

In order to determine the protein expression of Human Neuron-Specific Enolase (NSE) in the brains of control and magnetic field-exposed chicken, standard immunohistochemical procedures were carried out on formaldehyde-fixed, paraffin-embedded tissue sections. This antibody was used due to the cross-reactivity previously found in chicken (Sánchez-Montesinos et al., 1996). Briefly, paraffin was removed from the tissue sections using xylene, and endogenous peroxidase was quenched in 3% H₂O₂. Then, we used 0.01 M citrate buffer (PH 6.0) at 98 °C for 5 min for antigen retrieval. Incubation with the primary anti-human NSE antibody was performed for 12 h at 4°C using 1:400 dilutions. Then, secondary biotin-conjugated anti-mouse antibody was used at 1:500 dilution, and a horseradish peroxidase-conjugated streptavidin solution was applied for 40 min. Color was developed with a commercial DAB kit (Vector Laboratories, Burlingame, CA) and samples were then counterstained in Mayer's haematoxylin and mounted on coverslips for optical evaluation.

Finally, the NSE protein expression levels were quantified in four different areas of the optic lobe of the

brain (optic lobe cortex, optic tectum, tectum-spinal area and marginal area of the ventricle), using the map of the chick brain previously described by Tienhoven and Juhasz (1962). NSE quantification was carried out in all groups (A, A-CTR, B and B-CTR) corresponding to exposed (10 μ T and 50 and 100 Hz) and non-exposed animals (controls) at day 21 and animals exposed for 15 days and allowed to hatch without exposition until day 21. For the protein expression quantification, image analysis software (NIS-Elements AR3.0 program) was used at the automatic mode, and the mean pixel intensity -expressed as NIS-Elements intensity units (I.U.)- was quantified at each region of interest.

Histochemistry for detection of glycosaminoglycans in the optic lobe of the brain analyzed here was carried out using alcian blue staining at pH 2.5. Counterstaining was performed with haematoxylin. Glycosaminoglycans were analyzed in exposed and non-exposed animals at day 21 and animals exposed for 15 days and allowed to hatch without exposition until day 21.

Statistical analysis

To compare the different variables analyzed in this work between control groups and chicks subjected to magnetic fields (number of somites, body weight, development stage and NSE protein expression), the non-parametric U test of Mann-Whitney was used. All comparisons were done double-tailed and a p value below 0.05 was considered as statistically significant. These tests were carried out using the SPSS 15.0 program.

To determine the effect-sizes of the different variables considered in this work (mainly, the frequency of the magnetic field and the time of administration) we used a two-way analysis of variance.

Results

Morphological analysis of embryos exposed to magnetic fields

In the embryos extracted at 45 h, the number of somite pairs and the development stage were significantly lower ($p < 0.05$) in the exposed embryos

Table 1. Determination of the number of somites and stage of each embryo after 45 hours of incubation upon pulsed magnetic field (harmonic signals of 10 μ T for 1 second with silences of 0.5 seconds of 50 or 100 Hz frequency).

	Number of somites per embryo (mean \pm SD)		Development stage of each embryo (mean \pm SD)	
Group A-CTR (controls)	16.60 \pm 2.55	$p < 0.05$	11.80 \pm 0.91	$p < 0.05$
Group A (10 μ T-50 Hz)	13.21 \pm 3.53		10.64 \pm 1.27	
Group B-CTR (controls)	18.64 \pm 3.32	$p < 0.05$	12.57 \pm 1.15	$p < 0.05$
Group B (10 μ T-100 Hz)	15.64 \pm 3.67		11.50 \pm 1.34	

Stage was determined from the number of somites according to Hamburger and Hamilton, 1992. P values correspond to the statistical comparison of each experimental group (A and B) versus its control group (A-CTR and B-CTR) using the Mann-Whitney U test.

(Groups A and B) than in the respective controls from both groups (Groups A-CTR and B-CTR) (Fig. 1, Table 1).

After 15 days of incubation (Table 2), exposed embryos from both Groups A and B showed higher somatic weight and development stage as compared to

their controls (Groups A-CTR and B-CTR), although significance was only reached for the comparison of Group A (10 μ T-50 Hz field) versus Group A-CTR (controls).

At 21 days (hatching period), chicks exposed to magnetic fields throughout development (Table 2) (10

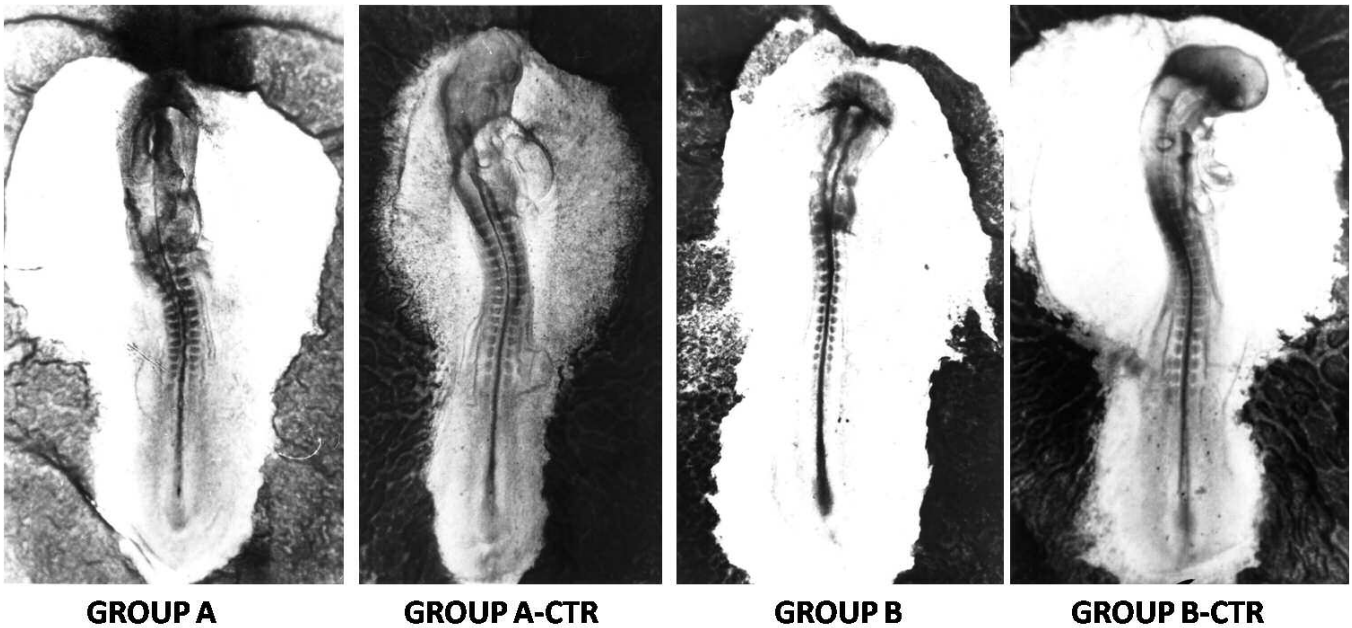


Fig. 1. Development stage of embryos exposed to pulsed magnetic fields (harmonic signals of 10 μ T for 1 second with silences of 0.5 seconds of 50 or 100 Hz frequency) (Groups A and B) and their controls (Groups A-CTR and B-CTR) after 45 h of incubation.

Table 2. Somatic weight (expressed in grams) and development stage after 15 and 21 days of incubation and after 21 days of incubation with exposition to pulsed magnetic fields just for the first 15 days.

	Somatic weight per embryo (mean \pm SD) (g)		Development stage of each embryo (mean \pm SD)	
15 days of incubation				
Group A-CTR (controls)	13.00 \pm 1.12	p<0.05	10.75 \pm 0.57	n.s.
Group A (10 μ T-50 Hz)	13.82 \pm 1.04		11.01 \pm 0.74	
Group B-CTR (controls)	14.35 \pm 2.85	n.s.	9.92 \pm 0.75	n.s.
Group B (10 μ T-100 Hz)	15.59 \pm 2.10		10.06 \pm 0.77	
21 days of incubation				
Group A-CTR (controls)	43.55 \pm 4.99	n.s.	17.93 \pm 0.85	p<0.05
Group A (10 μ T-50 Hz)	42.48 \pm 3.84		17.27 \pm 1.12	
Group B-CTR (controls)	42.34 \pm 4.74	p<0.01	10.15 \pm 0.92	p<0.05
Group B (10 μ T-100 Hz)	37.90 \pm 3.93		9.53 \pm 0.93	
21 days of incubation with exposition to magnetic fields for the first 15 days				
Group A-CTR (controls)	43.55 \pm 4.99	n.s.	17.93 \pm 0.85	n.s.
Group A (10 μ T-50 Hz)	43.03 \pm 3.69		17.54 \pm 2.19	
Group B-CTR (controls)	42.13 \pm 4.81	n.s.	10.09 \pm 0.98	n.s.
Group B (10 μ T-100 Hz)	39.93 \pm 4.16		9.91 \pm 0.64	

The stage was determined by third toe length, and expressed as a numeric stage of the Hamburger-Hamilton scale (Hamburger and Hamilton, 1992). P values correspond to the comparison of each experimental group (A and B) versus its control group (A-CTR and B-CTR) using the Mann-Whitney U test. n.s.: not significant.

Pulsed magnetic fields and brain development

μ T-50 Hz field, Group A) showed a non-significantly lower somatic weight as compared to their controls (Group A-CTR) and a significantly lower development stage ($p<0.05$); those exposed throughout development to a higher frequency (10 μ T and 100 Hz, Group B) showed a significantly lower somatic weight and development stage compared with their controls ($p<0.01$ and $p<0.05$, respectively). Embryos incubated for 21 days but exposed to magnetic fields only for the first 15 days (Table 2) had a slightly but non-significantly lower somatic weight and stage versus controls of the same age.

NSE protein expression analysis of embryos exposed to magnetic fields for 15 and 21 days

In the different brain areas analyzed, the NSE immunoreactivity was significantly higher in the optic lobe of the chicks exposed to different types of magnetic fields in comparison to the unexposed controls ($p<0.001$ for all comparisons except for chicks exposed during only the first 15 days of incubation to 10 μ T, 50 Hz fields, which showed non-significant differences in optic lobe cortex) (Tables 3 and 4). Illustrative images of the immunohistochemical analysis of control and magnetic

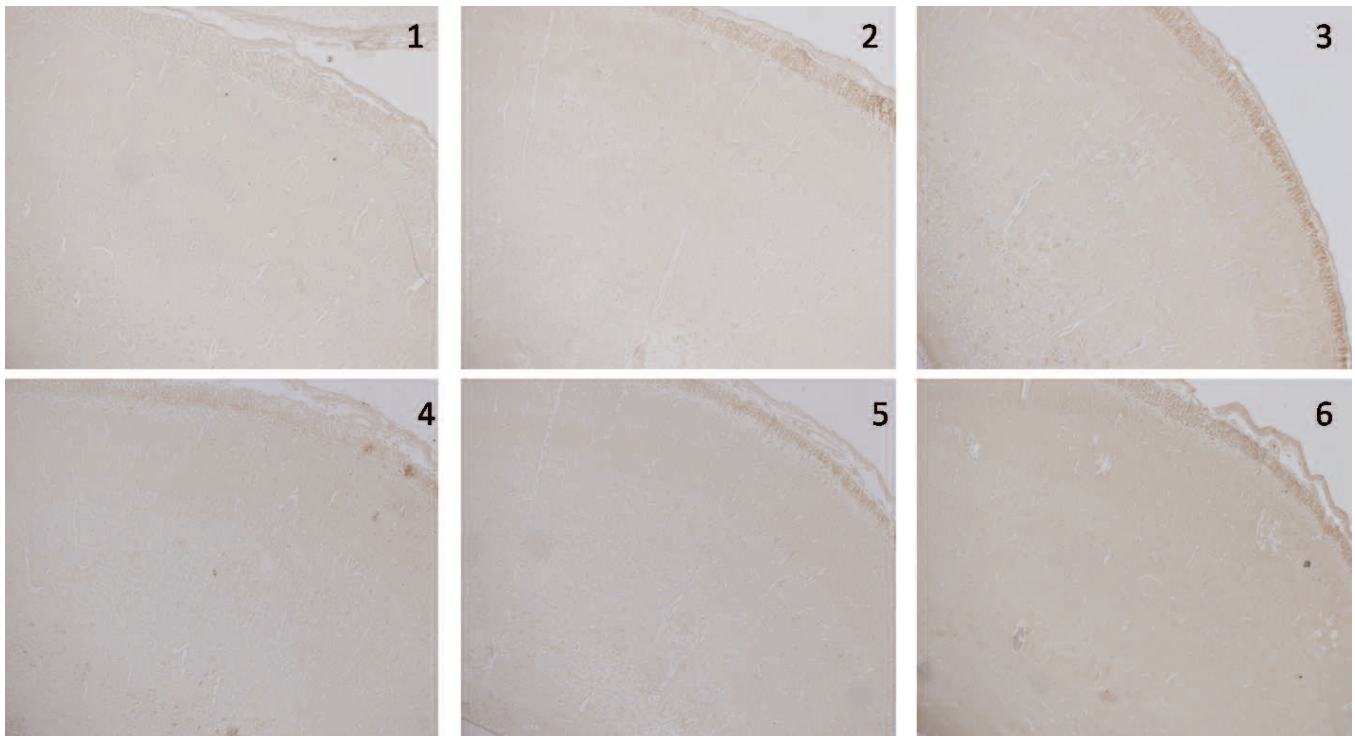


Fig. 2. Neural specific enolase (NSE) protein expression analysis of the optic lobe of the brain of hatching chicks whose embryos were exposed to pulsed magnetic fields (harmonic signals of 10 μ T for 1 Second with silences of 0.5 seconds of 50 or 100 Hz frequency) for 15 and 21 days, as determined by immunohistochemistry. 1. Group A-CTR; 2. Group exposed to 10 μ T-50 Hz only the first 15 days of incubation; 3. Group A (10 μ T-50 Hz); 4. Group B-CTR; 5. Group exposed to 10 μ T-100 Hz only the first 15 days of incubation; 6. Group B (10 μ T-100 Hz).

Table 3. Quantification of neural specific enolase (NSE) in different areas of the optic lobe of chicks exposed to 10 μ T-50 Hz pulsed magnetic fields for 21 days (until hatching).

	Optic lobe cortex		Optic tectum		Tectum-spinal area		Marginal area of the ventricle	
Group A-CTR (controls)	193.00 \pm 9.51	$p<0.001$	198.72 \pm 4.90	$p<0.001$	199.71 \pm 5.08	$p<0.001$	198.19 \pm 6.69	$p<0.001$
Group A (10 μ T-50 Hz)	195.19 \pm 9.37		199.87 \pm 6.18		200.93 \pm 5.86		199.46 \pm 7.71	
Group A-CTR (controls)	193.00 \pm 9.51	n.s.	198.72 \pm 4.90	$p<0.001$	199.71 \pm 5.08	$p<0.001$	198.19 \pm 6.69	$p<0.001$
Group exposed to 10 μ T-50 Hz only the first 15 days of incubation	193.11 \pm 9.20		199.76 \pm 4.86		201.31 \pm 4.84		199.46 \pm 7.71	

Values are expressed as correspond to mean pixel intensity expressed as NIS-Elements intensity units (I.U.).

Table 4. Quantification of neural specific enolase (NSE) in different areas of the optic lobe of chicks exposed to 10 μ T-100 Hz pulsed magnetic fields for 21 days (until hatching).

	Optic lobe cortex		Optic tectum		Tectum-spinal area		Marginal area of the ventricle	
Group B-CTR (controls)	185.39±16.32	p<0.001	186.53±14.51	p<0.001	187.32±14.45	p<0.001	182.86±16.42	p<0.001
Group B (10 μ T-100 Hz)	187.27±10.97		189.77±9.19		191.68±9.97		189.76±10.36	
Group B-CTR (controls)	185.39±16.32	p<0.001	186.53±14.51	p<0.001	187.32±14.45	p<0.001	182.86±16.42	p<0.001
Group exposed to 10 μ T-100 Hz only the first 15 days of incubation	186.26±7.57		187.20±5.12		188.35±4.42		184.24±5.67	

Values correspond to mean pixel intensity expressed as NIS-Elements intensity units (I.U.).

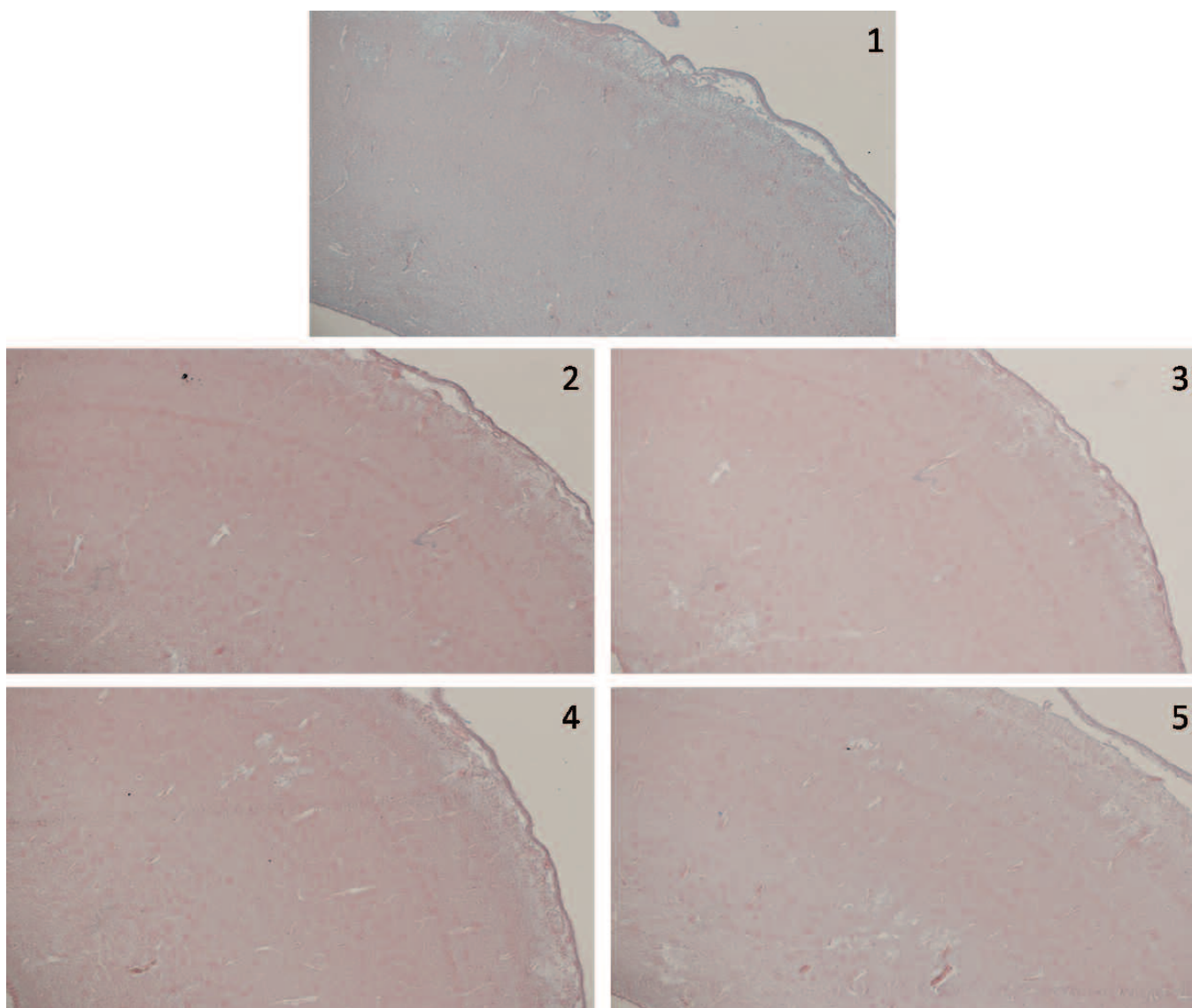


Fig. 3. Analysis of glycosaminoglycans in the optic lobe of the brain of hatching chicks whose embryos were exposed to pulsed magnetic fields (harmonic signals of 10 mT for 1 second with silences of 0.5 seconds of 50 or 100 Hz frequency) for 15 and 21 days (alcian blue staining). 1. Control Group; 2. Group A (exposed to 10 μ T-50 Hz); 3. Group exposed to 10 μ T-50 Hz only the first 15 days of incubation; 4. Group B (exposed to 10 μ T-100 Hz); 5. Group exposed to 10 μ T-100 Hz only the first 15 days of incubation.

field-exposed examined brain areas are shown in Fig. 2. In addition, the two-way analysis of variance revealed that the frequency of the magnetic field used (0, 50 or 100 Hz) was statistically associated to the level of NSE expression in all brain areas analyzed here ($p < 0.001$ for all areas). However, the influence of the time of exposure was statistically significant for the optic lobe cortex ($p < 0.05$) but not for the other areas studied.

Analysis of glycosaminoglycans in embryos exposed to magnetic fields for 15 and 21 days

Glycosaminoglycans in the optic lobe of controls and animals exposed to different types of pulsed magnetic fields for 15 and 21 days showed that the expression of these mucopolysaccharides was reduced in samples corresponding to animals exposed to magnetic fields in comparison to the unexposed controls (Fig. 3).

Discussion

Low-frequency magnetic fields can induce significantly more intense electric currents compared with endogenous currents resulting from physiological chemical reactions, thus inducing an electrical potential that is superimposed on the resting membrane potential of the cell. Any biological effects will depend on the frequency, intensity, and direction of the magnetic field, since these can determine the speed of the chemical reactions (Harkins and Grissom, 1994).

Thus, magnetic fields may produce disruption of the electrochemical balance of cells and therefore of its function (Panagopoulos et al., 2002). The response of a biological entity to magnetic field exposure will depend upon the intrinsic properties of the entity, the characteristics of the field, and the setting in which the phenomenon occurs (Lin, 1994). The main action of magnetic fields is thought to be exerted on the cell membrane, especially the ion channels (Liburdy, 1992), with calcium ion channels being the most affected.

Low-frequency magnetic fields (50–60 Hz) appear to affect numerous biochemical processes (Tenforde, 1991), including protein synthesis, DNA, RNA, hormone production changes, and modifications in cell growth and differentiation.

In the present study, as in the numerous studies of magnetic field exposure of chick embryos by our group using pulsed or non-pulsed fields of similar frequency and intensity (Roda-Moreno et al., 2003; Roda-Murillo et al., 2005) no major embryonic malformations were observed. This contrasts with reported observations in chick and rat studies of agenesis of telencephalic vesicles, severe somatic disorders, and digestive tube anomalies, among others (Delgado et al., 1982; Grandolfo et al., 1991) at 8–10 days of development under exposure to electromagnetic fields.

However, changes in body weight and development stage were observed in the present study, as found by other researchers (Sienkiewicz et al., 1994; Graham et

al., 2000). Rats exposed to electromagnetic fields during the first few days of gestation also gave birth to lower-weight newborns (Sandrey et al., 2002).

The embryos in the present study that were exposed to magnetic fields of the same intensity and different frequencies showed somitic development disorders after only 45 h of incubation, with fewer somite pairs and a lower development stage compared with controls of the same age. Interestingly, these results are in agreement with previous reports from Ubeda et al. (1983) who found that magnetic fields were able to induce several disorders in chicks after 48 h of exposure, including a thickness and disorganization of the neural tubes and a slight retardation of somite development. However, the somatic weight and development stage were higher than controls at 15 days of incubation, although the differences were only significant ($p < 0.05$) in the weight of embryos exposed to 50 MHz fields. In embryos exposed throughout development (21 days), the weight and development stage were significantly lower than controls at hatching, except for those exposed to 50 Hz fields, in which the weight reduction was not statistically significant. Therefore, the longest exposure (21 days) produced the greatest harmful effects in the present study, contradicting some reports that the effect of magnetic fields is more aggressive during initial development stages. The contradictory results of studies on the action of magnetic fields on living beings may be due to the large number of variables involved and differences in the type of research carried out. At hatching (21 days), somatic weight and development stage were not affected in embryos solely exposed to magnetic fields for the first 15 days in comparison to control groups.

In addition, these animals showed higher expression of the neural development marker NSE than the control brains, although the differences were not significant for the optic lobe cortex of the brain when 10 μ T-50 Hz were applied for 15 days. NSE is a superior biomarker for coma and brain damage prognostication. The enolases catalyze the interconversion of 2-phosphoglycerate to phosphoenolpyruvate in the glycolytic pathway, and NSE is the major form found in mature neurons and in cells of neuronal origin. NSE may be envisioned as a marker of neuronal damage because it is a protein-based enzyme found primarily within neurons, and its plasmatic levels rise following traumatic brain injury and correlate with outcome in severe head injury (Dauberschmidt et al., 1983; Skogseid et al., 1992; Yamazaki et al., 1995). For that reason, NSE can be used as a marker of cerebral damage in acute and chronic brain pathology (Barone et al., 1993). In agreement with these previous works, we have demonstrated that NSE expression was increased in the brain of the animals subjected to magnetic fields during the first weeks of development. These results suggest that a significant functional alteration could exist in these brains as a consequence of the magnetic field exposure, although it could also be possible that this

alteration is the result of a developmental inhibition caused by this exposure. In addition, in this work we have been able to demonstrate that an increase of NSE expression occurs *in situ* in the optic lobe of the exposed chicks using immunohistochemical methods, whereas most of the works which previously focused on the relationship between NSE increase and brain damage were carried out using NSE plasmatic levels (Dauberschmidt et al., 1983; Yamazaki et al., 1992; Skogseid et al., 1992). Interestingly, our analysis of variance suggested that although both the frequency and the time of exposure were associated to the expression of NSE in the embryos, most of the variance could be explained by the frequency of the magnetic field used (0, 50 or 100 Hz). This implies that the role of the frequency could be more relevant than that of the time of exposure.

In addition, we have demonstrated that the brain tissues of the chicks exposed to magnetic fields showed a clear decrease of the expression of glycosaminoglycans staining. It is known that acid glycosaminoglycans are important components of the basal membranes and the extracellular matrix of most of the tissues (Ubeda et al., 1983), and play an important role in the regulation of cellular proliferation (Glimelius and Pintar, 1981), and organogenesis. Therefore, the alteration of the synthesis of glycosaminoglycans that we observed in the animals exposed to magnetic fields could be correlated to the developmental and functional alterations found by morphological and immunohistochemical analysis. All these results are in agreement with previous reports suggesting that the expression of glycosaminoglycans is dramatically reduced in brains subjected to magnetic fields (Ubeda et al., 1983).

Although research findings to date do not definitively demonstrate that magnetic fields pose a public health problem, the possibility of negative effects on health suggests that avoidance of this type of energy may be advisable when feasible (Trosko, 2000). Although the results of our work cannot be directly extrapolated to humans, our results suggest that the adverse effects of these types of magnetic fields should be taken into account, especially during pregnancy. New works should be carried out to determine the pernicious effects of these fields from a clinical standpoint.

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