Effects of Magnetic Field Intensities for Various Lengths of Time on Orientation of Fowl Spermatozoa

Du Ngoc Pham[1], Akihisa Shinjo* and Katsunori Sunagawa

Faculty of Agriculture, University of the Ryukyus, 1 Senbaru, Nishihara-cho, Okinawa 903-0213, Japan

ABSTRACT : This study used fowl sperm from three White Leghorn rooster reared at our laboratory. Semen samples were exposed to the magnetic field strengths of from 650 to 5700 Gauss for one, two, or three days to investigate the influence of magnetic field on the orientation of fowl spermatozoa. Fowl spermatozoa were found to orient with their long axis of heads perpendicular to the magnetic field direction. The fowl spermatozoa were initially influenced when magnetic field intensities were from 650 to 5700 Gauss and the highest values (70.67, 72.49 and 71.79%) were found in the 5700 Gauss treatment at one, two, and three days exposure, respectively. Although percentages of the perpendicular oriented fowl spermatozoa increased along with the enhancement of the magnetic field intensity, the degree of orientation was only significantly higher in the treatments having the magnetic field strength from 1500 to 5700 Gauss than that in the control treatment at all exposure time. In addition, the experimental results also showed that the percentages of all orientational types of fowl spermatozoa (perpendicular category including upward perpendicular and downward perpendicular and parallel type consisting of leftward parallel and rightward parallel) in all treatments tended to be stable during exposure time. From the results of this study, it is suggested that (1) the diamagnetic anisotropy of the inside structural components of fowl spermatozoa induce them to orient perpendicular to the magnetic field direction, (2) the degree of orientation increased according to the enhancement of magnetic field strengths, (3) fowl spermatozoa had not an high sensitivity to the magnetic field, and the level of perpendicular orientation of fowl spermatozoa in this study is nearly similar to that of cattle sperm in the study of Suga et al. (2000). ***(Asian-Aust. J. Anim. Sci. 2001. Vol 14, No. 10 : 1367-1373)***

Key Words : Field Intensity, Fowl Spermatozoa, Orientation, Exposure Time

INTRODUCTION

In recent years, there have been numerous studies on the effect of various magnetic field strengths on several biological systems. Studies on bull sperm under the effect of a weak magnetic field of 600 Gauss by Suga and Shinjo (1997), 1000 Gauss by Suga et al. (1995) and 1600 Gauss by Shinjo and Utimura (1992), demonstrated that the motility and viability significantly increased. Also, Formiki et al. (1990) reported that magnetic field strengths from 60 to 100 mT significantly induced increased percentages of activated spermatozoa and prolonged the time of different phases of fish sperm movement (Danube salmon).

The effects of magnetic field were also observed in other objects. Kwee and Raskmark (1995) reported that human cell proliferation increased in a magnetic field of 500 Gauss. However, in the experiment of Brewer (1979), magnetic field caused an reduction of the gestation period as well as the spawn rate of the Lebistes Reticulatus (Guppy).

As for the magnetic orientation, several experiments were conducted on deoxygenated sickle erythrocytes (Brody et al., 1985), on glutaraldehyde fixed erythrocyte (Higashi et al., 1996), on lipid tubule (Rosenblatt et al., 1987), on the polymerization of fibrin fibers (Yamagishi et al., 1990) and on non motile bull sperm (Suga et al., 2000). The results showed that the organic materials, depending on the structure of their inside components, oriented perpendicular or parallel to the magnetic field direction.

The present experiment was carried out to investigate the orientation as well as the susceptibility of fowl spermatozoa under the effect of various magnetic field intensities from 650 to 5700 Gauss for several exposure times.

MATERIALS AND METHODS

Animals and preparation of semen sample

Three one-year-old commercial While Leghorn roosters reared at our laboratory were used in this study. They were placed in single cages and fed *ad libitum* on a commercial breeder diet.

Semen was collected by the method of abdominal massage. Semen of each individual was accumulated to 1 ml, and stored in an incubator at 4°C. These semen samples were checked for sperm density and then mixed in a plastic vial (after treating them to get the same sperm density in each). The final semen sample was preserved again in the incubator at 4°C and used throughout the experiment. One hundred μl semen was added to 4 ml NaCl 0.15 M. Fifty μl semen was then dropped into the chamber of a slide which

* Address reprint request to Akihisa Shinjo. Tel: +81-98-895-8756, Fax: +81-98-895-8756, Email: ashinjo@agr.u-ryukyu.ac.jp

[1] Permanent address: Faculty of Agriculture, Cantho University, Cantho City, Vietnam.

Received April 24, 2001; Accepted June 8, 2001

was made according to the method of Suga et al. (2000); then a cover-glass covered it immediately. This cover-glass was moved to any direction so that the distribution of fowl spermatozoa was at random. Nail enamel was used to seal around the border of the cover-glass to prevent the sample fluid from flowing or evaporating. Then these slides were placed on the wood stages between magnets, with the magnetic field strengths varying from 650 to 5,700 Gauss for one, two or three days at room temperature (approximately 25°C). The method of exposing the semen samples to the magnetic field is shown in figure 1. The control sample was placed in a room unaffected by the artificial magnetic field.

Measuring the magnetic orientation of fowl spermatozoa

The magnetic orientation of fowl spermatozoa was classified using the method described by Suga et al. (2000). However, in this study we only ranked fowl spermatozoa into four categories because of their structural characteristic. The categories were: (U) upward perpendicular orientation, with the long axis of sperm head perpendicular to the magnetic field direction and upward (+45°< long axis < +135°); (D) downward perpendicular orientation, with the long axis of sperm head perpendicular to the magnetic field direction and downward (+225° < long axis < +315°); (L) leftward parallel orientation, with the long axis of sperm head parallel to the magnetic field direction and leftward (+135° < long axis < +225°); (R) rightward parallel orientation, with the long axis of sperm head parallel to the magnetic field direction and rightward (+315° < long axis < +45°).

Statistical analysis

Data on magnetic orientation of fowl spermatozoa of each treatment were collected each four times with a number of about one thousand spermatozoa or more. Then the weighted means were analyzed by using SAS (1990).

The Chi-square test of FREQ procedure for equal population was used to analyze the orientation data, which were classified into categories, such as perpendicular orientation and parallel orientation, upward perpendicular orientation and downward perpendicular orientation, leftward parallel orientation and rightward parallel orientation. Besides, contrasts of CATMOD procedure were used according to the technique described by Shinjo (1994) to compare the difference between treatments non exposed and exposed to the magnetic field strengths, and to examine effects of exposure time.

RESULTS

The orientation of fowl spermatozoa to the magnetic field direction after exposing for one, two or three days is presented in table 1. This table described the difference between two major orientational directions of sperm in treatments, which were perpendicular and parallel orientation of fowl spermatozoa to the magnetic field direction. The perpendicular orientational category consist of upward perpendicular orientation plus downward perpendicular orientation, and the parallel orientation compose of leftward parallel orientation plus rightward parallel orientation.

Table 1 shows that percentages of perpendicular oriented fowl spermatozoa considerable increased from

Table 1. Mean oriented percentages of fowl spermatozoa after exposing to the magnetic field strengths for various times

Magnetic field value (Gauss)	Exposure time (days)											
	1			2			3			Weighted mean		
	N[1]	Perp[2]	Para[3]	N	Perp	Para	N	Perp	Para	N	Perp	Para
Control	1,026	$49.51^{a,A}$	50.49	1,013	$49.85^{a,A}$	50.15	1,019	$50.15^{a,A}$	49.85	1,019	49.85^{a}	50.15
650	1,043	$53.40^{ab,A}$	46.60*	1,034	$54.06^{ab,A}$	45.94**	1,028	$54.38^{ab,A}$	45.62^{**}	1,033	53.91^{ab}	46.09*
1,500	994	$55.94^{b,A}$	44.06**	1,017	$55.56^{bc,A}$	44.44**	1,033	$57.31^{bc,A}$	42.69^{**}	1,027	56.26^{bc}	43.74**
2,700	1,010	$57.43^{b,A}$	42.57**	1,069	$59.03^{cd,A}$	40.97**	1,028	$58.37^{bc,A}$	41.63^{**}	1,040	58.30^{cd}	41.70**
3,600	1,044	$62.55^{c,A}$	37.45**	1,037	$60.75^{de,A}$	39.25**	1,037	$61.62^{c,A}$	38.38^{**}	1,036	61.63^{de}	38.37**
4,700	1,062	$65.35^{c,A}$	34.65**	999	$64.66^{e,A}$	35.34**	1,021	$66.01^{d,A}$	33.99^{**}	1,015	65.34^{e}	34.66**
5,700	1,050	$70.67^{d,A}$	29.33**	1,036	$72.49^{f,A}$	27.51**	1,013	$71.79^{e,A}$	28.21^{**}	1,035	71.64^{f}	28.36**

[1] Weighted means of the number of sperms.

[2] Oriented spermatozoa each with its long axis pependicular to the magnetic field direction.

[3] Oriented spermatozoa each with its long axis parallel to the magnetic field direction.

** Significant differences between perpendicular and parallel oriented percentages (** $p<0.01$).

* Significant differences between perpendicular and parallel oriented percentages (* $p<0.05$).

^{a,b,c,d,e,f} Oriented percentages in the same column with different superscript differ ($p<0.01$-0.05).

^{A} Oriented percentages within a row with like superscript are not different ($p>0.05$).

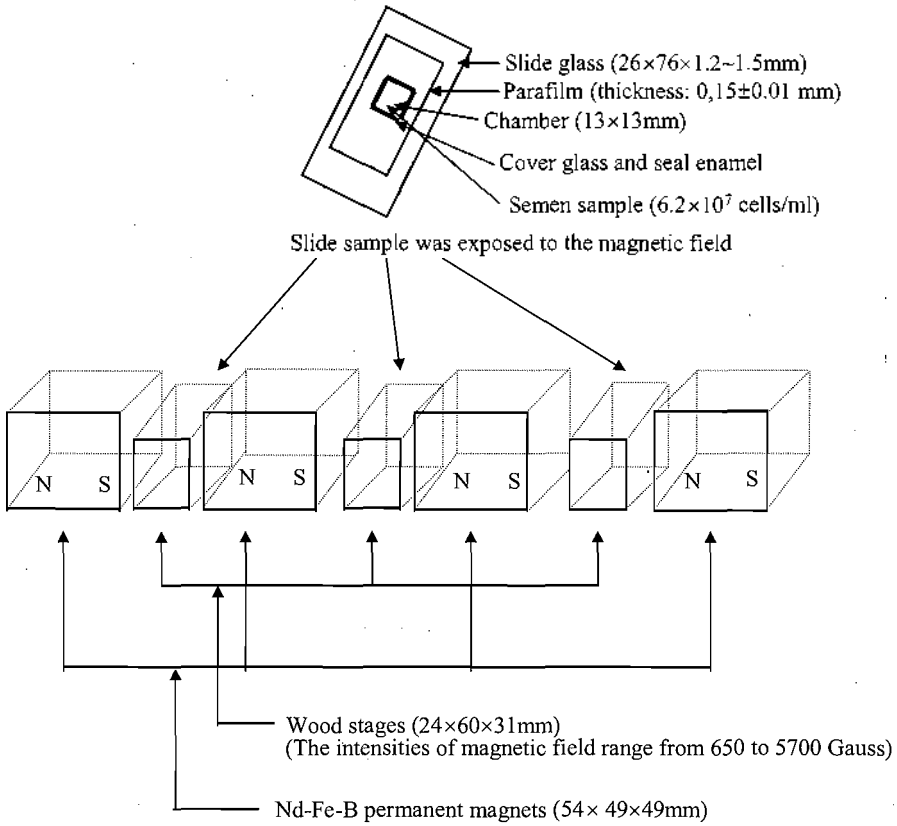

Figure 1. Method of exposure of fowl spermatozoa to the magnetic field

53.40 to 70.67%, 54.06 to 72.49% and 54.38 to 71.79% at one, two and three days in all of the exposed treatments, respectively, according to the enhancement of the magnetic field strengths from 650 to 5,700 Gauss as compared to those in the control treatment (49.51, 49.85 and 49.85% at one, two and three days, respectively), and reached on the highest values of 70.67, 72.49, and 71.79% in the 5,700 Gauss treatment at one, two and three day exposure, respectively. However, the significant difference was only found in the exposed groups when the magnetic field strengths ranged from 1,500 to 5,700 Gauss at one, two and three day exposure as compared to the control treatment. Percentages of perpendicular oriented fowl spermatozoa were similar in the 650, 1,500 and 2,500 Gauss treatments at one and three day exposure whereas, at two days exposure, a difference between the 650 and 2,700 Gauss treatments was found. Similarly, between 3,600 and 4,700 Gauss treatments, there were no difference between one and two days exposure, but at three days exposure a significant difference between two treatments was found.

Table 1 also shows that percentages of the perpendicular oriented fowl spermatozoa in all exposed groups were significantly higher than those of the parallel orientation at one, two and three days exposure, and disclosed a stability over exposure time.

For interpreting further the magnetic orientational characteristics of fowl spermatozoa, and since there was no significantly difference in perpendicular orientational category of exposed groups during exposure time, we added up the data of all exposure times of each treatment to calculate the weighted mean, and then used this value to compare the difference in perpendicular oriented percentages between exposed groups as well as between perpendicular and parallel types. Percentages of orientational types of treatments are presented in the right-side of table 1; it also showed to be similar to those at two day exposure as described previously. In addition, these results also indicated that the degree of magnetic orientation of fowl spermatozoa depended on the magnetic field intensities (figure 3).

A further comparison of the magnetic orientation of fowl spermatozoa was performed and shown in table 2. This table describes the difference between upward and downward perpendicular oriented fowl spermatozoa of treatments non or exposed to the magnetic field. Results showed that percentages of the downward perpendicular oriented fowl spermatozoa in the exposed treatments, in which the magnetic force ranges from 1500 to 5700 Gauss, were increased significantly as compared to the control treatment for all exposure times, except for the 650 Gauss

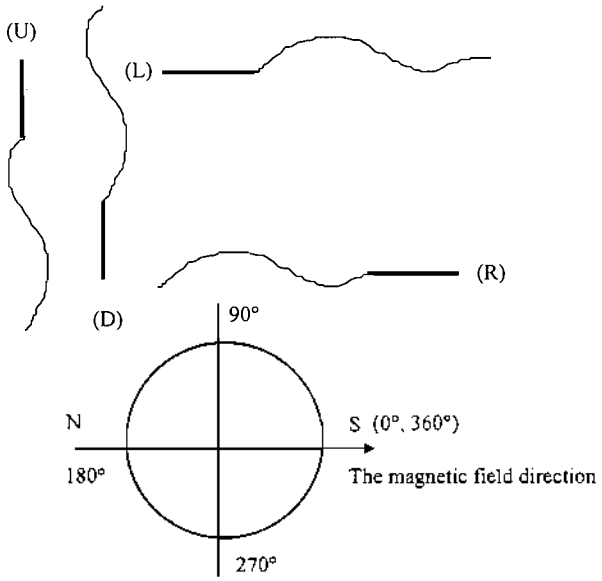

Figure 2. Method of ranking the magnetic orientation of fowl spermatozoa.

(U) Upward, with sperm head perpendicular to the magnetic field direction and upward

(D) Downward, with sperm head perpendicular to the magnetic field direction and downward

(L) Leftward, with sperm head parallel to the magnetic field direction and leftward

(R) Rightward, with sperm head parallel to the magnetic field direction and rightward

N The north pole of a magnet

S The south pole of a magnet

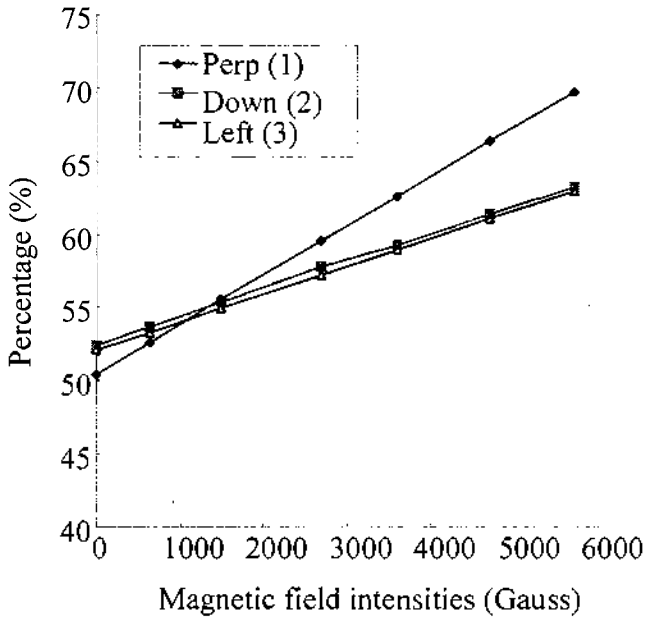

Figure 3. Correlation between the orientation of fowl spermatozoa and the magnetic field intensities

(1) Oriented spermatozoa each with its long axis perpendicular to the magnetic field direction ($Y=50.3904+0.0034X$, $R^2=.9695$, $p<0.01$)

(2) Oriented spermatozoa each with its long axis perpendicular to the magnetic field direction and downward ($Y=52.4289+0.0019X$, $R^2=0.8692$, $p<0.01$)

(3) Oriented spermatozoa each with its long axis parallel to the magnetic field direction and leftward ($Y= 52.0586+0.0019X$, $R^2 = 0.8593$, $p<0.01$)

treatment in which percentages of downward perpendicular oriented fowl spermatozoa were similar to the control treatment (figures 4a, 4b). Although percentages of the downward perpendicular oriented fowl spermatozoa in exposed groups were considerably increased according to the enhancement of the magnetic field intensities (table 2 and figure 3), the significant difference between these treatments was not found except for the 5700 Gauss treatment at two or three day exposure, in which percentages of the downward perpendicular oriented spermatozoa were significantly higher than those of the other exposed treatments at three day exposure, or of the 650, 2,700 and 4,700 Gauss treatments at one and two day exposures.

The weighted mean (right-side of table 2, calculated from data of all exposure time) of treatments also showed that percentage of downward perpendicular oriented spermatozoa with the 5,700 Gauss treatment was significantly higher than those of the control, 650, 1,500 and 2,700 Gauss treatments.

Table 3 presented the magnetic orientations of fowl spermatozoa, which consisted of leftward and rightward parallel orientations. For two and three day exposed treatments, percentages of leftward parallel oriented spermatozoa significantly increased in all the exposed groups (from 56.86 to 63.16% and from 58.28 to 65.73% at two day and three day exposure, respectively) as compared to the control treatment (49.80 and 49.21% at two and three day exposures, respectively). At one day exposure and the weighted mean (right-side of table 3) a significant difference was only found in the treatments that the magnetic force started from 1,500 to 5,700 Gauss. Besides, a difference between exposed groups was not found.

The above results showed that magnetic orientations of fowl spermatozoa (perpendicular, downward and leftward), had an positive correlation with field intensities (figure 3).

DISCUSSION

Frequently, if non motile organic materials are placed inside a magnetic field, they reveal an behavior by orienting either perpendicular or parallel to the magnetic field direction. This is an ordinary physical phenomenon.

Table 2. Comparison of the oriented direction between upward and downward orientation of fowl spermatozoa after exposing to the magnetic field strengths for variuors of times

Magnetic field value (Gauss)	Exposure time (day)											
	1			2			3			Weighted mean		
	N[1]	Downw[2]	Upw[3]	N	Downw	Upw	N	Downw	Upw	N	Downw	Upw
Control	508	$50.00^{b,A}$	50.00	505	$49.90^{c,A}$	50.10	511	$51.47^{c,A}$	48.53	507	50.49^{a}	49.51
650	557	$52.96^{ab,A}$	47.04	559	$54.92^{bc,A}$	45.08**	559	$54.20^{bc,A}$	45.80**	559	54.03^{ab}	45.97
1,500	556	$57.73^{a,A}$	42.27**	565	$58.58^{ab,A}$	41.42**	592	$55.91^{bc,A}$	44.09**	571	57.44^{b}	42.56**
2,700	580	$59.31^{a,A}$	40.69**	631	$56.42^{b,A}$	43.58**	600	$60.00^{b,A}$	40.00**	603	58.54^{b}	41.46**
3,600	653	$58.81^{a,A}$	41.19**	630	$60.16^{ab,A}$	39.84**	639	$58.69^{b,A}$	41.31**	640	59.22^{bc}	40.78**
4,700	694	$60.81^{a,A}$	39.19**	646	$57.89^{b,A}$	42.11**	674	$58.75^{b,A}$	41.25**	671	59.17^{bc}	40.83**
5,700	742	$62.94^{a,A}$	37.06**	751	$63.91^{a,A}$	36.09**	728	$65.38^{a,A}$	34.62**	740	64.05^{c}	35.95**

[1] Weighted means of the number of sperms

[2] Oriented spermatozoa each with its long axis perpendicular to the magnetic direction and downward.

[3] Oriented spermatozoa each with its long axis pependicular to the magnetic direction and upward.

** Significant differences between upward and downward oriented percentages (** $p<0.01$).

* Significant differences between upward and downward oriented percentages (* $p<0.05$).

[a,b,c] Oriented percentages in the same column with different superscript differ ($p<0.01$~0.05).

[A] Oriented percentages within a row with like superscript are not different ($p>0.05$).

Table 3. Comparison of the oriented direction between leftward and rightward orientation of fowl spermatozoa after exposing to the magnetic field strengths for various of times

Magnetic field value (Gauss)	Exposure time (day)											
	1			2			3			Weighted mean		
	N[1]	Leftw[2]	Rightw[3]	N	Leftw	Rightw	N	Leftw	Rightw	N	Leftw	Rightw
Control	518	$50.00^{a,A}$	50.00	508	$49.80^{a,A}$	50.20	508	$49.21^{a,A}$	50.79	511	49.71^{a}	50.29
650	486	$54.12^{ab,A}$	45.88	475	$54.53^{ab,A}$	45.47*	469	$54.58^{ab,A}$	45.42*	476	54.41^{ab}	45.59
1500	438	$56.16^{ac,A}$	43.84**	452	$56.86^{bc,A}$	43.14**	441	$58.28^{b,A}$	41.72**	443	57.11^{bc}	42.89**
2700	430	$58.60^{bc,A}$	41.40**	438	$58.45^{bc,A}$	41.55**	428	$57.01^{b,A}$	42.99**	432	58.10^{bc}	41.90**
3600	391	$57.29^{bc,A}$	42.71**	407	$58.48^{bc,A}$	41.52**	398	$57.79^{b,A}$	42.21**	399	57.89^{bc}	42.11**
4700	368	$59.24^{bc,A}$	40.76**	353	$59.49^{bc,A}$	40.51**	347	$59.65^{bc,A}$	40.35**	356	59.55^{bc}	40.45**
5700	308	$62.34^{c,A}$	37.66**	285	$63.16^{c,A}$	36.84**	288	$65.73^{c,A}$	34.27**	293	63.82^{c}	36.18**

[1] Weighted means of the number of sperms

[2] Oriented spermatozoa each with its long axis parallel to the magnetic direction and leftward.

[3] Oriented spermatozoa each with its long axis parallel to the magnetic direction and rightward.

** Significant differences between leftward and rightward oriented percentages (** $p<0.01$).

* Significant differences between leftward and rightward oriented percentages (* $p<0.05$).

[a,b,c] Oriented percentages in the same column with different superscript differ ($p<0.01$~0.05).

[A] Oriented percentages within a row with like superscript are not different ($p>0.05$).

The degree of orientation depend on the magnetic field intensity. The greater the magnetic field , the higher the degree of the orientation.

The results of the present study showed that fowl spermatozoa (non-motile spermatozoa) in exposed groups oriented with their long axis of heads perpendicular to the magnetic field direction, as shown in table 1 and figures 4c and 4d. In the control treatment, which was placed outside the magnetic field, they were dispersed at random (figure 4a). The degree of orientation of fowl spermatozoa increased according to a gradual increase of magnetic field strength from 650 to 5,700 Gauss (figure 3). The orientation could be explained from the diamagnetic anisotropy of the inside components of fowl spermatozoa, which are DNA and protein molecules. According to Worcester (1978) the diamagnetic anisotropy in nucleic acid is due to aromatic rings of base pairs, many of which are parallel in a DNA molecule because of the persistence length. For protein molecules, the diamagnetic anisotropy was attributed due to both the orientation of aromatic group and peptide bonds. In this study, perpendicular oriented percentages of fowl spermatozoa only significantly increased as compared to the control treatment when the magnetic field strength was from 1,500 to 5,700 Gauss. Moreover, the highest oriented percentage of fowl spermatozoa was 72.49% in the 5,700 Gauss treatment at two day exposure; this value approximated to that of 75.9% of bull spermatozoa at a magnetic field strength of 5,400 Gauss in the study of Suga

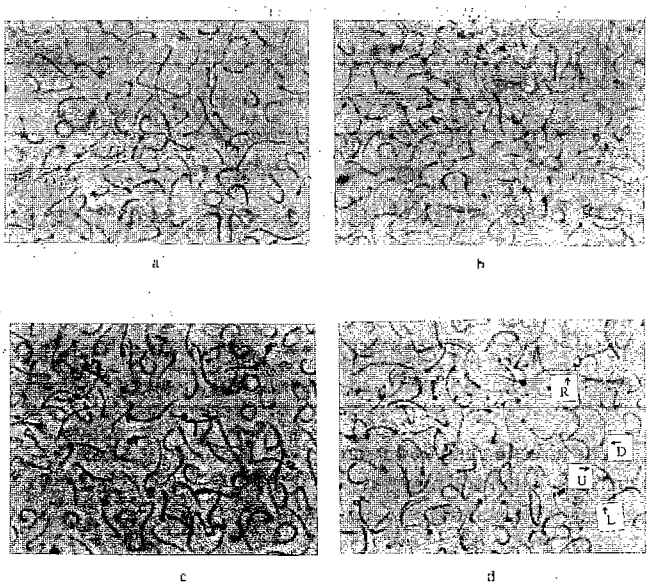

Figure 4. The orientation of fowl spermatoza after exposure to various magnetic field intensities for length of time (original magnefication 400×)
a The control treatment
b The 650 Gauss treatment at two day exposure
c The 3,600 Gauss treatment at two day exposure
d The 5,700 Gauss treatment at two day exposure
U Upward, with sperm head perpendicular to the magnetic field direction and upward
D Downward, with sperm head perpendicular to the magnetic field direction and downward
L Leftward, with sperm head parallel to the magnetic field direction and leftward
R Rightward, with sperm head parallel to the magnetic field direction and rightward

et al. (2000). From this result, it is suggested that fowl spermatozoa had not a high anisotropic magnetic susceptibility. The diamagnetic anisotropy was also found in other organic material, such as bull sperm (Suga et al., 2000 and Ashida et al., 1996), glutaraldehyde-fixed erythrocytes (Higashi et al., 1995), deoxygenated sickle erythrocytes (Brody et al., 1985) and collagen fibrils (Murthy, 1984).

The results in this treatment also showed that the differences between upward perpendicular and downward perpendicular, or between leftward parallel and rightward parallel were found significantly in all exposed groups except the 650 Gauss treatment. This was due to the fact that magnetic anisotropy induced fowl spermatozoa to orient to constant positions in the field direction; these were downward perpendicular orientation (table 2) and leftward parallel orientation (table 3), which nearly had the same direction as downward perpendicular orientation (figure 4).

In the present study, the magnetic orientation of fowl spermatozoa was stable during exposure time. From this result, it is suggested that one day is sufficient time for fowl spermatozoa to orient to the magnetic field. Thus, prolonging the exposed time does not induce them to orient further.

It can be concluded from this study that magnetic field enhanced fowl spermatozoa oriented with their long axis of heads perpendicular to the magnetic field direction because of the magnetic anisotropy of the inside components of sperm (DNA and protein molecule). Degree of orientation increased according to the increase of magnetic field strength. In addition, the results in this treatment showed that fowl spermatozoa had not a high sensitivity to magnetic field intensity, and the degree of perpendicular orientation of fowl spermatozoa is nearly similar to that of cattle sperm in the study of Suga et al. (2000).

REFERENCE

Ashida, N., T. Higashi and T. Takeuchi. 1996. Orientation of bull sperm in static magnetic field. Jap. J. Biomagn. Bioelectromagn. Soc. 9:29-34 (in Japanese).

Brewer, H. B. 1979. Some preliminary studies of the effects of a static magnetic field on the life cycle of the *Lebistes Reticulatus* (Guppy). Biophys. J. 28:305-314.

Brody, A. S., M. P. Socrette, C. A. Gooding, J. Listerud, M. R. Clard, W. C. Mentzer, R. C. Brasch and T. L. James. 1985. Induced alignment of flowing sickle erythro-cytes in a magnetic field. Invest. Radiol. 20:560-566.

Formiki, K., A. Sobocinski and A. Winnicki. 1990. Motility of spermatozoa of Danube salmon (*Hucho Hucho* L.), exposed to magnetic field prior to activation. Pol. Arch. Hydrobiol. 37:439-477.

Higashi, T., A. Yamagishi, T. Takeuchi, M. Date. 1995. Effect of static magnetic flied on erythrocyte rheology. Bioelectrochem. Bioenerg. 36:101-108.

Higashi, T., S. Sagawa, N. Ashida and T. Takeuchi 1996. Orientation of glutaral-dehyde-fixed erythrocytes in strong static magnetic fields. Bioelectromagnetic. 17:335-338.

Kwee, S. and P. Raskmark. 1995. Changes in cell proliferation due to environmental non-ionizing radiation. 1. ELF electromagnetic fields. Bioelectrochem. Bioenerg. 36:109-114.

Murthy, N. S. 1984. Liquid crystallinity in collagen solution and magnetic orientation of collagen fibrils. Biopolymers. 23:1261-1267.

Rosenblatt, C., P. Yager and P. E. Schoen. 1987. Orientation of lipid tubules by a magnetic field. Biophis. J. Biophys. Soc. 52:295-301.

SAS Institute Inc. 1990. SAS/STAT User's Guide: Version 6. 4th edn. SAS Institute Inc., Cary, North Carolina.

Shinjo, A. and M. Utimura. 1992. Effect of magnetic field on the viability of the cattle sperm. Jap. J. Vet. Inform. 29:13-15.

Shinjo, A. 1994. Introduction to statistics by PC SAS (in Japanese). Tokai University Press, Japan.

Suga, D., A. Shinjo, S. Nishikawa and Y. Chiba. 1995. Effect of a weak static magnetic field on the viability, motility, and velocity of cattle sperm. Jap. J. Magn. Soc. 19:872-874.

Suga, D. and A. Shinjo. 1997. Effect of 600 Gauss static

magnetic field on the freezing of cattle sperm. Jap. J. Magn. Soc. 21:1134-1137.

Suga, D., A. Shinjo, E. Kurnianto and T. Nakada. 2000. Magnetic orientation of bull sperm treated by DDT or heparin. Asian-Aus. J. Anim. Sci. 13:10-18.

Worcester, D. L. 1978. Structure origins of diamagnetic anisotropy in proteins. Proc. Natl. Acad. Sci. USA. 75:5475-5477.

Yamagishi, A., T. Takeuchi, T. Higashi and M. Date. 1990. Magnetic field effect on the polymerization of fibrin fibers. Phisica B. 164:222-228.