

[Research]

Impacts of extremely low-frequency electromagnetic fields (50-Hz) on growth performance and survival rate of common carp, *Cyprinus carpio* fingerlings

Mohammadi-Zadeh Khoshroo M¹, Shamsaie Mehrjan M^{1*}, Samiee F², Soltani M³, Hosseini Shekarabi S.P¹

1. Department of Fisheries Science, Science and Research Branch, Islamic Azad University, Tehran, Iran

2. Department of Biomedical Engineering, Science and Research branch, Islamic Azad University, Tehran, Iran

3. Department of Aquatic Animal Health, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran

* Corresponding author's E-mail: m.shamsaie@srbiau.ac.ir

(Received: May 30, 2017 Accepted: Oct. 24, 2017)

ABSTRACT

All organisms are probably exposed to different types of electromagnetic fields (EMFs). This study is an attempt to evaluate the effect of extremely low-frequency EMFs (50 Hz) on some growth parameters and survival rate of common carp fingerling. The fry (n = 120, averaged 16.76 ± 0.88 g in initial weight) was exposed to ELF-EMF at four intensities of 0.1, 0.5, 1 and 2 mT only once for 2 h and then reared for 60 days. The obtained results revealed that all growth indices were significantly ($p < 0.05$) improved by increasing in the EMFs intensity. 2 mT treatment indicated the highest final weight (43.95 ± 0.27 g), weight gain (6.83 ± 0.10 g), weight gain rate (20.94 ± 0.13 %), daily growth rate (0.38 ± 0.00 g day⁻¹), specific growth rate (1.26 ± 0.01 % day⁻¹) and the lowest food conversion ratio ($p < 0.05$). Survival rate had significantly ($p < 0.05$) increased in all exposed treatments compared to the control group. In conclusion, ELF-EMF can enhance the growth indices and survival rate of common carp as one of the most commercially - important cultured fish species over the world.

Key words: Electromagnetic fields, *Cyprinus carpio*, Growth, Survival.

INTRODUCTION

Electromagnetic fields (EMFs) have been widely present in ecosystems as well as aquatic ecosystems. Although, understanding the biological effects of EMFs on different organisms, in particular aquatic animals, have been conducted since the eighteenth century (Berg *et al.* 1999; Cuppen *et al.* 2007). There is a lack of information about the effects of EMFs on aquatic ecosystems (Krylov *et al.* 2014; Nofouzi *et al.* 2015; Khoshroo *et al.* 2017). Electromagnetic radiation or EMFs can be identified according to their physical

parameters such as the type (electricity, magnetic and/or electromagnetic), frequency and intensity (Redlarski *et al.* 2015; Yadollahpour *et al.* 2014). It seems that emitted waves from EMFs are safe and harmless to the health of living organisms (Khurana 2008; Kundi *et al.* 2004). In fact, biological effects of EMFs on aquatic ecosystems are gradually being identified and reported depending on species, populations and ecosystems (Gill *et al.* 2012). Some studies showed the remedial effects of EMFs, known as immunostimulators, on a wide range of

diseases such as musculoskeletal diseases, cancer, neurological disorders, and treating wounds (Mevisen *et al.* 1998; Simko & Mattson 2004; Cuppen *et al.* 2007; Elmusharaf *et al.* 2007a,b; Yadollahpour *et al.* 2014). Some investigators showed that EMFs could affect the growth and survival rate of the exposed organisms. For instance, Peira *et al.* (1992) showed that continuous exposure to EMF at an intensity of 36.1 mT increased the weight of chicken embryo. It was also observed that exposure to EMF (less than 100 Hz) at 0.5 mT intensity for 10 weeks increased the body weight of rats (Gerardi *et al.* 2008). Broiler chick eggs exposed to ELF-EMF (50 Hz) from 0 to 75 min prior to incubation revealed that the EMF had no effects on the weight of the chicks at the time of hatching, feeding intake and FCR (Shafey *et al.* 2011). The present experiment has been designed to evaluate the effects of low frequency EMF (50 Hz) at different field intensities of 0.1, 0.5, 1 and 2 mT on some growth performance and survival rate of common carp fingerlings.

MATERIALS AND METHODS

Fish and experimental design

Healthy common carp fingerling (n = 120, averaged 16.76 ± 0.88 g in body weight and 10.34 ± 0.52 cm in total length) were randomly purchased from a local fish farm (Rasht, Iran). Fish were transferred to Science and Research Branch Laboratory (Tehran, Iran) and maintained for two weeks for adaptation prior to experiment. Fish were then divided in 15 plastic tanks (60 × 50 × 50 cm) with light/dark cycles (12:12 h) under the same experimental conditions (temperature 22 ± 4°C, pH 7.5 ± 0.5, DO = 8.8 ± 0.3 mg.L⁻¹, NO₂ < 0.2 mg.L⁻¹, NO₃ < 4 mg.L⁻¹, and NH₄ < 0.5 mg.L⁻¹) for 60 days. The fish were fed twice a day with commercial balanced carp feed in the form of pellets at the rate of 5% of body weight during acclimation and experimentation. The commercial feed

(Beyza Feed Mill, Shiraz, Iran) contained less than 10% moisture, 36% protein, 10% fat, 5% fibers and 3500 Kcal.Kg⁻¹ digestible energy.

For each treatment, 24 fish were exposed to ELF-EMF only once at different intensity (i.e. 0.1, 0.5, 1 and 2 mT) in a plastic container. The exposed fish were reared for 60 days. A control group without any exposure to EMF was also included.

Electromagnetic field generation system

A cylindrical coil, (80 × 20 cm), made of 490 turn of 2.5 mm diameter enameled copper wire was the generator of electromagnetic field. The system was armed with some additional systems containing digital multimeter (model DT4252, HIOKI, Japan), digital current intensity control system, signal generator (model GHG-8020H, GWINSTEK, Malaysia), digital aquarium temperature controller (model AQ-403, SUNSUN, China), power supply (model PS-605D, DAZHENG, China), and an oscilloscope (model GDS-1072-U, GWINSTEK, Malaysia) (Fig. 1).

The experimental fish were located inward of the coil using a conveyor platform until all samples received the same electromagnetic field intensity. Values in the coil center were calculated based on following equation (Samiee & Samiee 2017):

$$B = \frac{\mu^0 \times N \times I}{L}$$

Where, B is magnetic flux density measured in Tesla (T), N is diameter of enameled copper wire (mm), I is current intensity applied to the coil (ampere), L is length of coil (cm) and

$$\mu^0 = 0.256 \times 10.$$

To create one-way current and conducting different flux densities, we employed a power supply for originating DC (direct or static field) and a signal generator for creating AC (alternating) current flows.

A digital multimeter (volt-meter and ampere-meter) was used to detect the voltage and ampere values. Furthermore, an oscilloscope was used to display voltage versus time and waveform. Magnetic field was parallel to long

axis of fish body in the coil. Digital temperature controller was located inside the coil in order to regulate temperature at 22°C. Moreover, aquarium plastic pipes with cold water

surrounded the coil to prevent its over - heating. The frequency was always stable (50 Hz) during exposure, but the intensity of EMF exposure was changeable.

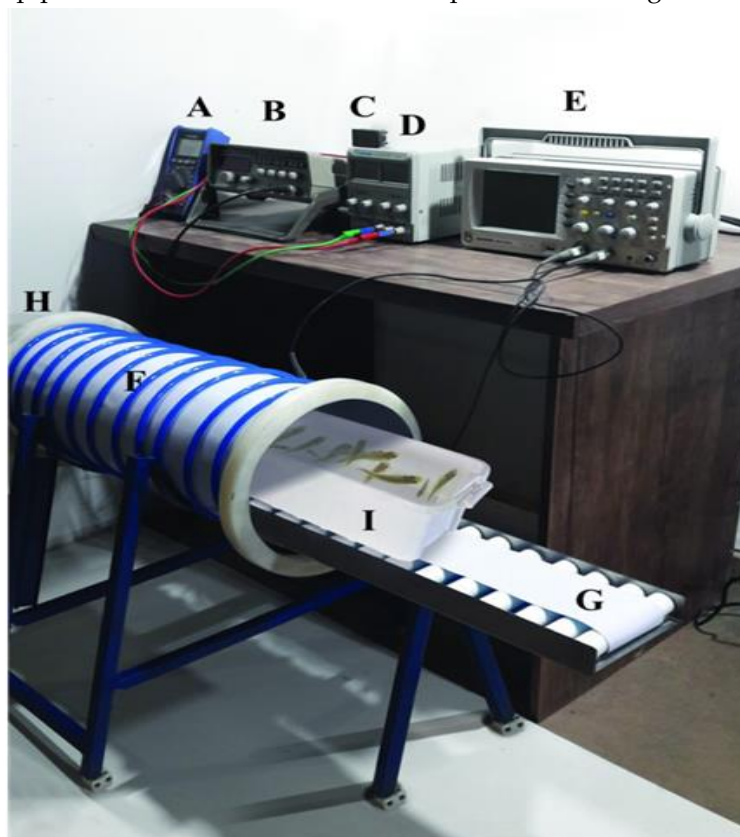


Fig. 1. Experimental setup for low-frequency electromagnetic field exposure to common carp. (A) Digital multimeter, (B) Digital current intensity control system, signal generator, (C) Digital temperature controller screen, (D) Power supply, (E) an oscilloscope, (F) Electromagnetic field generator or coil, (G) elevated platform for placing fish inside the coil, (H) digital temperature controller and air conditioning system and (I) fish container.

Determination of fish growth performance

At the end of experiment, fish from each tank were anaesthetized with clove extract at 100 mg.L⁻¹ (Soltani & Mirzargar 2013), then the length and weight of the fish were measured using a biometric ruler (up to the nearest 1 mm) and also a digital scale (up to the nearest 0.001 g), respectively. The growth factors of fingerling in terms of weight gain (WG), length gain (LG), body weight gain (BWG), daily growth rate (DGR), specific growth rate (SGR), food conversion ratio (FCR), condition factor (CF), and survival rate (SR) were calculated as follows (Luz et al. 2008):

$$WG (g) = W_1 - W_0;$$

$$LG (cm) = L_1 - L_0;$$

$$BWG (%) = W_1 - W_0 / W_0 \times 100;$$

$$DGR (g.day^{-1}) = W_1 - W_0 / T;$$

$$SGR (\% day^{-1}) = [(ln W_1 - ln W_0) / T] \times 100;$$

$$FCR = \text{dry feed (g)} / \text{weight gain (g)};$$

$$CF (\%) = W_1 / L_1^3 \times 100;$$

$$SR = \text{final fish number} / \text{initial fish number} \times 100.$$

Where W_0 refers to the mean initial weight (g), W_1 is the mean final weight (g), L_0 is the mean initial length (cm), L_1 is the mean final length

(cm), and T represents the number of feeding period (days).

Statistical analysis

Prior to analysis, raw data were tested for normality of distribution and homogeneity of variance with Kolmogorov-Smirnov test and Levene's test, respectively.

The group means were compared by One-Way ANOVA followed by Duncan's post-hoc test with 95% confidence level. All statistical calculations were carried out using SPSS version 19.

RESULTS

Fish exposed to 2 mT ELF-EMFs showed (Table 1) the highest final weight (43.95 ± 0.27 g), WG (6.83 ± 0.10 g), RWG ($20.94 \pm 0.13\%$), DGR (0.38 ± 0.00 g), SGR ($1.26 \pm 0.01\%$) and the lowest FCR (0.38 ± 0.00). Maximum values of final length and length gain (LG) were measured in both 2 and 1 mT treatments with no significant differences ($p > 0.05$). However, 1 mT treatment showed a significant ($p < 0.05$) decrease in level of CF ($0.67 \pm 0.00\%$) in comparison with control. All exposed fish with no significant difference ($p > 0.05$) had higher SR than non-exposed fish (Fig. 2).

Table 1. Growth performance of common carp fingerling after 60 days exposure to ELF-EMF at different intensities.

Parameters	Exposure intensity (mT)				
	Control	0.1	0.5	1	2
Final weight (g)	31.63 ± 0.36^d	34.02 ± 0.50^c	35.19 ± 0.45^c	38.83 ± 0.06^b	43.95 ± 0.27^a
Final length (cm)	14.32 ± 0.16^c	14.76 ± 0.21^c	15.87 ± 0.20^b	17.92 ± 0.03^a	18.15 ± 0.11^a
WG (g)	3.1 ± 0.035^d	4.47 ± 0.065^c	4.42 ± 0.056^c	6.15 ± 0.01^b	7.61 ± 0.04^a
LG (cm)	0.41 ± 0.00^d	1.14 ± 0.02^c	1.20 ± 0.02^b	2.15 ± 0.00^a	2.16 ± 0.01^a
RWG (%)	10.86 ± 0.12^e	15.13 ± 0.22^c	14.36 ± 0.18^d	18.81 ± 0.03^b	20.94 ± 0.13^a
DGR (g day ⁻¹)	0.21 ± 0.00^d	0.298 ± 0.00^c	0.294 ± 0.00^c	0.41 ± 0.00^b	0.50 ± 0.00^a
SGR (% day ⁻¹)	0.68 ± 0.01^e	0.93 ± 0.01^c	0.89 ± 0.01^d	1.14 ± 0.00^b	1.26 ± 0.01^a
FCR	0.67 ± 0.01^a	0.52 ± 0.01^c	0.57 ± 0.01^b	0.42 ± 0.00^d	0.38 ± 0.00^e
CF (%)	1.07 ± 0.01^a	1.05 ± 0.02^a	0.88 ± 0.01^b	0.67 ± 0.00^d	0.73 ± 0.00^c

Data represent mean \pm SD of three replicates. Different superscript letters within the same row indicate significant ($P < 0.05$) differences of the means: a>b>c>d>e.

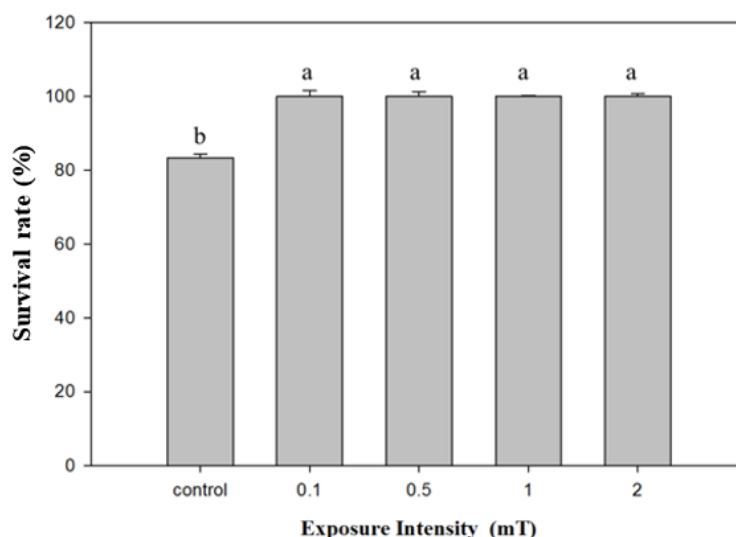


Fig. 2. Survival rate of common carp fingerling after 60 days acute exposure at different intensities of ELF-EMF. Different superscript letters indicate significant ($P < 0.05$) differences of the means.

DISCUSSION

Many studies showed the different bio-reactions of EMFs in neural, neuromuscular activities, cell membrane function, glands, gene expression, and some regulator enzymes to growth performance of exposed organism (Wertheimer & Leeper 1979; Saunders *et al.* 1991; Blank 1993; Kula & Drozd 1996; Koyama & Nakahara 2003; Aaron *et al.* 2004; Büyüksulu *et al.* 2006). Moreover, the growth inhibitory effect of EMF on the growth pattern of single-celled microorganisms was described (Mittenzwey *et al.* 1996; Fojt *et al.* 2004; Falone *et al.* 2007; Yadollahpour *et al.* 2014). There are still a few researches concerning the effects of ELF-EMFs on aquatic organisms like the fish. Hence, the negative and/or positive effects of ELF-EMFs on the health and the growth process of aquatic organisms is unclear (Chebotareva *et al.* 2009; Khoshroo *et al.* 2017).

In the present study, the best results of growth parameters were obtained in the exposed fish, especially in 2 mT treatment. These results are in agreement with the results of Cuppen *et al.* (2007), who revealed the positive effects of LF-EMF (200-5000 Hz) at intensities between 0.15 and 50 μ T on growth performance of fantail goldfish (*Carassius auratus*). As expected, the results were also in agreement with Nofouzi *et al.* (2015), who reported that the growth factors of rainbow trout fry were enhanced following

exposure to ELF-EMFs (15 Hz) at 5 and 50 μ T. Elmusharaf *et al.* (2007b) found the positive effects of EMF on broiler chicks growth performance. Consequently, the growth of zebrafish embryo was inhibited when they exposed to sinusoidal EMF (60 Hz) at an intensity of 1 mT (Cameron *et al.* 1985).

Metabolism of organisms might be affected by EMFs (Gerardi *et al.* 2008; Lai *et al.* 2015). Similarly, Hashish *et al.* (2008) showed that exposure of rats to EMFs with frequency of 50 Hz and intensity of 1.4 mT for 30 days reduced the body weight compared to non-exposed rats. Krylov & Chebotareva (2006) reported that exposure of roach, *Rutilus rutilus* to LF-EMF (500 Hz) with an intensity of 150 μ T reduced the size and weight of the fingerlings due to decline in the growth rate (Chebotareva *et al.* 2009). In addition, Elbetieha *et al.* (2002) found that long-term exposure of male and female rats to sinusoidal ELF-EMF (50 Hz) at an intensity of 25 μ T for 90 days had no significant changes on the body weight. However, Elmusharaf *et al.* (2007 a,b) claimed that EMFs had antagonistic effect and could be considered as a pathogenic agent in broiler chicks. Metabolic changes of organisms are the main reason for different body weights. Fish metabolism seems to be changed by EMF leading to improved growth performance (Nofouzi *et al.* 2015). At the present study, the values of body weight and

length significantly ($p < 0.05$) increased in the exposed treatments, especially in 1 and 2 mT intensity compared to the control group. The increase in the growth indices of common carp might indicate the positive effect of ELF-EMF on carp fingerlings.

Severini *et al.* (2010) reported that exposure of African clawed frog, *Xenopus laevis* to weak EMF (50 Hz) at intensity of 50.76 to 60.69 A.m⁻¹ for 60 days, showed reduction in the growth rate and increase in the metamorphosis period time. However, Zecca *et al.* (1998) showed that daily exposure to ELF-EMF (50 Hz) at intensity of 5 and 100 μ T for 5 days in a week caused no changes on the growth rate of rats. Margonato *et al.* (1995) showed that 22 h daily-exposure to the magnetic field (50 Hz) at intensity of 5 μ T for 32 weeks caused no changes on the growth rate of rats. Zhang *et al.* (1993) found no negative effects of ELF-EMF (60 Hz) on growth performance of chick embryo.

Results of Shafey *et al.* (2011) indicated that chick eggs exposed to EMF (50 Hz) at 75 min before incubation showed new born chicks with high feed intake and low FCR. Exposure of infected broiler to ELF-EMF (200-5000 Hz) at intensities between 0.15 and 50 μ T improved the FCR compared to the control group possibly due to production of cytokines and increased in the immune responses (Cuppen *et al.* 2007). In this study, increasing in the ELF-EMF intensity led to significant reduction of CF and FCR values. Exposure of common frog, *Rana temporaria* to ELF-EMF (50 Hz) at an intensity of 325 μ T, increased the mortality of the frog (Grefner *et al.* 1998). However, Nofouzi *et al.* (2015) found the highest survival rate of rainbow trout exposed to ELF-EMF (15 Hz) at intensities 0.1 to 50 μ T when they were challenged with *Yersinia ruckeri*. Likewise, Cuppen *et al.* (2007) showed that exposure of commercial goldfish to LF-EMF (200-5000 Hz) at intensities between 0.15 and 50 μ T might cause significant differences in the fish survival rate. Keirs *et al.* (2005) showed that the exposure to EMF for 6 years reduced the mortality up to 47.6% among laying hens. The SR results of this study showed that all exposed treatments to

ELF-EMF had higher survival rates in comparison with the control ones. This might be explained by positive effects of these fields on growth function. The best growth functions among different fish species might be explained by increase in the capacity of gastrointestinal tract function (Heidarieh *et al.* 2012; Nofouzi *et al.* 2015). However, the effect of ELF-EMFs on growth performance as well as the behavior of organisms are highly dependent on the environmental conditions, type of EMF fields (constant or alternate), frequency, range and duration of exposure, whereas if the fish expose to EMF for several generations, the effects of these fields will be more effective (Brewer 1979; Krylov *et al.* 2014). In conclusion, acute exposure (2 h) to extremely low frequency electromagnetic fields at higher than 0.1 mT intensities, especially 1 and 2 mT might positively affect the growth performance and the survival rate of common carp fingerlings after 60 days. Obviously, more researches are needed to determine the effects of EMF on aquatic organisms.

ACKNOWLEDGEMENT

A special thank goes to Mr. Fathaliyan and Mr. Tootonchi-Mashhor of Razi Central Laboratory of Islamic Azad University for their kind support and assistance. We also would like to express our highly appreciation to Mr. Fardin Komai, the administrative director of Guilan University, who grammatically edited the last version of this paper.

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اثرات میدان‌های الکترومغناطیس با فرکانس بسیار پایین (۵۰ هرتز) بر شاخص‌های رشد و بقای بچه ماهیان انگشت قد کپور معمولی (*Cyprinus carpio*)

محمدی‌زاده خوشرو م^۱، شمسایی مهرجان م^{۱*}، سمیعی ف^۲، سلطانی م^۳، حسینی شکرابی س.پ^۱

۱- گروه شیلات، واحد علوم و تحقیقات، دانشگاه آزاد اسلامی، تهران، ایران

۲- گروه مهندسی پزشکی، واحد علوم و تحقیقات، دانشگاه آزاد اسلامی، تهران، ایران

۳- گروه بهداشت و بیماری‌های آبزیان، دانشکده دامپزشکی، دانشگاه تهران، تهران، ایران

(تاریخ دریافت: ۹۶/۰۳/۰۹ تاریخ پذیرش: ۹۶/۰۸/۰۲)

چکیده

تمامی موجودات ممکن است با انواع مختلفی از میدان‌های الکترومغناطیسی (EMFs) مواجه شوند. اطلاعات ناچیزی در باره اثرات زیست‌شناختی این میدان‌ها بر آبزیان در دسترس است. در این تحقیق، اثرات میدان‌های الکترومغناطیس با فرکانس بسیار پایین (۵۰ هرتز) بر عملکرد رشد و نرخ بقای بچه ماهیان انگشت قد ماهی کپور معمولی مطالعه شد. تعداد ۱۲۰ عدد بچه ماهی با وزن اولیه 0.188 ± 0.016 گرم در معرض میدان الکترومغناطیسی با فرکانس بسیار پایین (ELF-EMF) با شدت‌های ۰/۱، ۰/۵، ۱ و ۲ میلی‌تسلا (mT) فقط یک بار به مدت ۲ ساعت قرار گرفته و سپس به مدت ۶۰ روز پرورش داده شدند. نتایج این مطالعه نشان داد تمامی شاخص‌های رشد به طور معنی‌داری ($p < 0.05$) با افزایش شدت میدان‌های الکترومغناطیسی بهبود یافت، به طوری که تیمار ۲ mT بیشترین وزن نهایی (0.27 ± 0.043 گرم)، افزایش وزن (0.10 ± 0.068 گرم)، درصد افزایش وزن (0.13 ± 0.020)، نرخ رشد روزانه (0.00 ± 0.038 گرم در روز)، نرخ رشد ویژه (0.01 ± 0.0126 ٪ در روز) و کمترین ضریب تبدیل غذایی (0.00 ± 0.038) را نشان داد. نرخ بقا به طور معنی‌داری ($p < 0.05$) در تمامی تیمارهای مواجهه یافته در مقایسه با گروه شاهد افزایش یافت. در نتیجه، میدان‌های الکترومغناطیسی با فرکانس بسیار پایین می‌توانند شاخص‌های رشد و بقای بچه ماهیان کپور معمولی را به عنوان یکی از مهمترین گونه‌های پرورشی در سراسر دنیا افزایش و بهبود بخشند.

*مؤلف مسئول