

Temporally Incoherent Magnetic Fields Mitigate the Response of Biological Systems to Temporally Coherent Magnetic Fields

T.A. Litovitz, D. Krause, C.J. Montrose, and J.M. Mullins

Vitreous State Laboratory (T.A.L., C.J.M.) and Department of Biology (D.K., J.M.M.), The Catholic University of America, Washington, District of Columbia

We have previously demonstrated that a weak, extremely-low-frequency magnetic field must be coherent for some minimum length of time (≈ 10 s) in order to affect the specific activity of ornithine decarboxylase (ODC) in L929 mouse cells. In this study we explore whether or not the superposition of an incoherent (noise) magnetic field can block the bioeffect of a coherent 60 Hz magnetic field, since the sum of the two fields is incoherent. An experimental test of this idea was conducted using as a biological marker the twofold enhancement of ODC activity found in L929 murine cells after exposure to a 60 Hz, $10 \mu\text{T}_{\text{rms}}$ magnetic field. We superimposed an incoherent magnetic noise field, containing frequencies from 30 to 90 Hz, whose rms amplitude was comparable to that of the 60 Hz field. Under these conditions the ODC activity observed after exposure was equal to control levels. It is concluded that the superposition of incoherent magnetic fields can block the enhancement of ODC activity by a coherent magnetic field if the strength of the incoherent field is equal to or greater than that of the coherent field. When the superimposed, incoherent noise field was reduced in strength, the enhancement of ODC activity by the coherent field increased. Full ODC enhancement was obtained when the rms value of the applied EM noise was less than one-tenth that of the coherent field. These results are discussed in relation to the question of cellular detection of weak EM fields in the presence of endogenous thermal noise fields. ©1994 Wiley-Liss, Inc.

Key words: noise, coherence, incoherence, signal to noise, ornithine decarboxylase

INTRODUCTION

The association of biological effects at the cellular level with exposure to weak extremely-low-frequency (ELF) electromagnetic (EM) fields has remained a controversial subject despite over a decade of such reports [e.g., Blank et al., 1992; Goodman et al., 1992; Litovitz et al., 1991; Martin, 1988; Phillips et al., 1992]. Theoretical arguments based on signal-to-noise considerations are in large measure responsible for the skepticism [Weaver and Astumian, 1990; Adair, 1991]. The

Received for review June 4, 1993; revision received April 11, 1994.

Address reprint requests to T.A. Litovitz, Vitreous State Laboratory, Catholic University of America, Washington, DC 20064.

controversy arises because the data indicate that cells, existing in an electrically noisy environment, respond to external EM fields that are some 100–1000 times weaker than local noise fields resulting from the thermally driven motion of ions in the vicinity of cells. This has led many physical scientists to conclude that it is impossible for weak EM fields to affect cell function. However, the data indicate that they do. How can the externally impressed fields possibly influence cell behavior when cells have evolved in such a way as to function normally in the presence of the much larger local noise fields?

To address this question, Litovitz et al. [1991] explored the role of coherence in the EM stimulation of bioeffects. They demonstrated, using magnetic field-induced ornithine decarboxylase (ODC) activity in L929 murine cells, that in order for this bioeffect to occur the field had to be coherent for times longer than about 5–10 s. It thus appears that the answer to the signal-to-noise dilemma is somehow related to the temporal coherence of externally applied fields and to the intrinsic incoherence of the local, thermal EM noise fields. To explore further the role of coherence, we have hypothesized that by adding a sufficiently large incoherent (noise) magnetic field to a coherent one (such as that from a 60 Hz power line) the total stimulus would appear incoherent, and thus the bioelectromagnetic effect normally caused by the coherent field would not occur. To test this idea, we have conducted experiments using as a biological marker the induced ODC activity in L929 murine fibroblasts. We first measured the enhancement of ODC activity caused by a coherent 60 Hz field. We then superimposed an incoherent magnetic noise field on the 60 Hz field and measured the enhancement of the ODC activity when both fields were simultaneously applied. The effect of the 60 Hz field was measured in the presence of noise fields of different rms amplitudes.

MATERIALS AND METHODS

Cell Cultures

Cultures of the murine L929 cell line (NCTC clone 929; American Type Culture Collection, Rockville, MD) were maintained in active growth using Eagle's minimum essential medium supplemented with 5% donor calf serum and 10 mM HEPES buffer (all obtained from Sigma Chemical Co., St. Louis, MO). Cell stocks were kept at 37 °C in a humidified atmosphere of 95% air, 5% CO₂. Cultures for magnetic field exposure trials were initiated approximately 20 h prior to use. Each 75 cm² flask was given an inoculum of 1.2×10^7 cells, in 15 ml of growth medium, obtained from an actively growing stock culture that was 70–80% confluent at the time of harvesting. These conditions resulted in midlogarithmic phase growth by the onset of the experiment and cultures that were approximately 70% confluent. To avoid large increases in ODC activity due to serum stimulation [Heby et al., 1975; reviewed by Jänne et al., 1978], the culture medium was not replaced prior to exposure.

ELF Exposure Systems

All exposures were carried out in two Helmholtz coil-based, ELF exposure systems. A block diagram of the exposure system is shown in Figure 1. Both systems used E-field shielded Helmholtz coils with average radius of 4.125 inches,

placed within water-jacketed, cell culture incubators. The coils were mounted vertically on a square, MU-metal covered, wooden base. The base was fitted snugly with a MU-metal cover, which enclosed the coils to provide magnetic field shielding. Samples were placed on a 2.75-inch-high sample table mounted on the base at the center of the Helmholtz coils.

Each exposure was carried out with three culture flasks stacked one atop the other and placed on the sample table such that the magnetic field was parallel to the long axis of the flasks. With this arrangement, the growth surfaces of the flasks were positioned within a cylindrical region, of 2.5 inches radius, that was concentric with the axis of the Helmholtz coils and that extended axially to ± 2.4 inches from the center. We refer to this cylinder as the *cell exposure region*.

The magnetic field distribution within the Helmholtz coil was measured using a 0.5-inch-diameter, 0.7-inch-long, 400-turn pickup coil. The pickup coil was positioned over the sample table with its axis parallel to the direction of the magnetic field, and measurements were taken at 0.25 inch intervals along two lines, one parallel with the coil axis and the other perpendicular to it. When the MU-metal cover was not used, the magnetic field variation within the cell exposure region was less than 8% in the radial direction and less than 14% in the axial direction. When the MU-metal cover was used, the magnetic field variation in the radial direction was also less than 10% and decreased to less than 4% in the axial direction. In addition, the field increased by approximately 8% relative to the corresponding field without the MU-metal cover.

The magnetic field distribution was also determined by numerical integration of the Biot-Savart law as applied to a circular current loop. The result indicates that, within the cell exposure region and along the paths over which the measurements were made, the field variation in the radial direction is expected to be less than 11% and less than 6% in the axial direction. Furthermore, the magnetic field over the entire cell exposure region is within $\pm 14\%$ of the field at the center of the coils.

These results show that the magnetic field distribution within our Helmholtz coil without the MU-metal box displays some deviation from ideal behavior, particularly in the axial direction. It is also apparent that the MU-metal enclosure improves the field distribution in the cell exposure region. However, it should be noted that this observation is based only on measurements taken along two perpen-

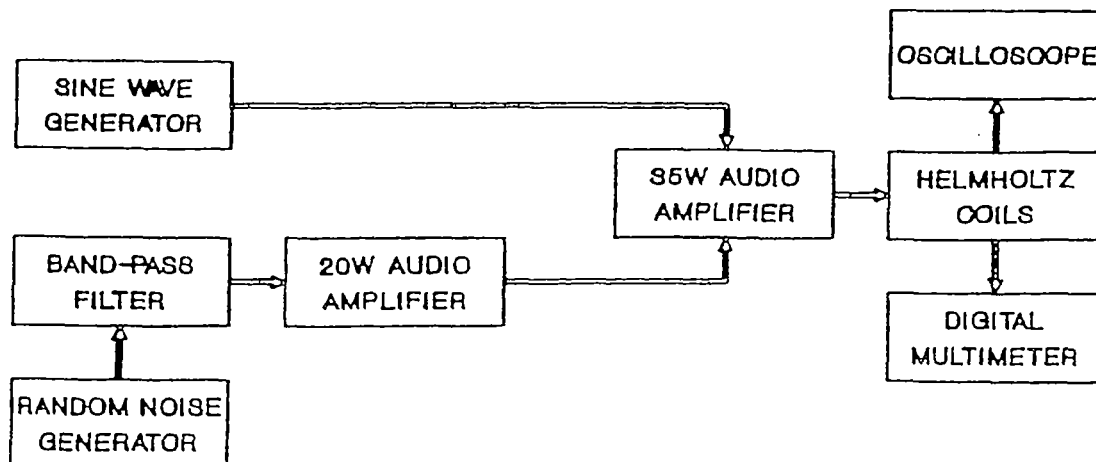


Fig. 1. Block diagram illustrating the components of the exposure system.

dicular paths. If improvement on the field distributions holds over the entire cell exposure region, it is reasonable to assume, on the basis of the above measurements and calculations, that with this exposure arrangement the samples are exposed to fields varying by not more than $\pm 15\%$ of the field at the center of the coil.

The Helmholtz coils were driven either with a pure sinusoid or with a sinusoid plus band-limited noise. Sine wave exposure was carried out at 60 Hz using a signal generator (TENMA model 72-380; MCM Electronics, Centerville, OH) connected to the auxiliary input of a 35 W audio amplifier (Realistic model MPA-45; Tandy Corporation, Fort Worth, TX). To add noise for the second exposure condition, a 60 Hz sinusoid and a noise signal were mixed by connecting them, respectively, to the left and right channels of the auxiliary input of the 35 W amplifier. The noise signal was produced using a General Radio model 1390-B Random Noise Generator followed by a Krohn-Hite (Avon, MA) model 335 Multifunction Variable Filter and a 20 W Realistic (model MPA-25; Tandy Corporation) audio amplifier. The pass band of the filter was set between 30 and 90 Hz. In both cases the Helmholtz coils with the series resistor were connected to the $8\ \Omega$ speaker output of the 35 W amplifier.

The $10\ \mu\text{T}_{\text{rms}}$, 60 Hz exposure field was set by measuring the voltage drop across a $3\ \Omega$ power resistor connected in series with the $3.8\ \Omega$ coil. For this purpose the magnetic field generated by the coil was calibrated as a function of the voltage drop across the series resistor. Measurements were made with a 60 Hz sinusoidal field with the magnetic probe placed in the center of the Helmholtz coil. An IDR-109 60 Hz Magnetic Field Dosimeter (Integrity Design and Research, Buffalo, NY) and a Keithley (Taunton, MA) 197 Digital Multimeter were used for the calibration measurements. The 60 Hz magnetic field was set using this calibration. The noise signal was set by adjusting the rms voltage drop across the $3\ \Omega$ resistor to be equivalent to that of the 60 Hz signal. The rms value of the noise was measured by sampling the noise signal at 5 kHz. The rms value was computed as the square root of the sum of the squares of the sampled points divided by the number of samples.

Magnetic Field Exposure

For each experiment, nine $75\ \text{cm}^2$ culture flasks were used. Three flasks, serving as negative controls (or shams), received no exposure to applied fields. Three flasks, serving as positive controls, were exposed to the 60 Hz, $10\ \mu\text{T}$ stimulating field. The remaining three flasks were exposed to a combination of the 60 Hz stimulating field and a noise field or to a noise field alone. All exposures were for 4 h, an interval previously demonstrated to result in maximal ODC enhancement in response to the stimulating field [Litovitz et al., 1991]. These experiments were conducted over a 6 month period and were done in essentially random order with respect to the amplitude of the noise field employed in a given experiment.

Since cells were exposed to EM fields within a MU-metal enclosure, we examined the possibility that resistance heating from the Helmholtz coils might affect results by raising the temperature of exposed samples. For the highest amplitude field used in these experiments ($100\ \mu\text{T}$ ELF noise), the current in the coils was 68 mA. Given a coil resistance of $3.8\ \Omega$, the expected I²R loss would be 17 mW, a power dissipation that should produce a minimal change in temperature over the 4 h duration of our experiments, even with the MU-metal enclosures in place. To be certain that this was the case, a series of measurements was made using ther-

Mitigation Effect of Incoherent Magnetic Fields

mistor probes positioned within the liquid medium of cell culture flasks. Using either the combined 60 Hz, 10 μ T and ELF noise 10 μ T fields or the 100 μ T ELF noise field alone, we found that even after 16 h of exposure the temperature of the medium in the exposed flasks was, within the 0.1 $^{\circ}$ C resolution of our digital thermometer, the same as that in unexposed flasks. Given these facts, resistance heating was concluded not to be a significant factor.

ODC Assay

At the end of a 4 h experiment, the cells in each flask were washed immediately with two changes of ice-cold, phosphate-buffered saline (PBS). Cells were then gently scraped from the growth surface of the flask in a third change of cold PBS, and the resultant cell suspension was pelleted at 350g for 5 min. After aspiration of the PBS supernatant, cell pellets were stored frozen at -75° C. The ODC assay was performed by the method of Seely and Pegg [1983], which measures ODC specific activity by the release of 14 CO₂ from 14 C-labeled, l-ornithine. As in the Seely and Pegg protocol, 2.75×10^5 cpm of labeled ornithine (specific activity 57.2 μ Ci/ μ M; DuPont NEN Research Products, Boston, MA) was added to an assay solution containing unlabeled ornithine at a concentration of 0.4 mM. Minor modifications to the assay procedure included the addition of 50 μ M pyridoxal 5'-phosphate, 50 μ g/ml leupeptin, and 0.2% Nonidet P-40 (all reagents from Sigma Chemical Co.) to the cell lysis buffer. 14 CO₂ generated by ODC activity was absorbed with 150 μ l of 1.0 N NaOH. Addition of 400 μ l of 20% TCA terminated enzymatic reactions at the completion of the assay. Background counts were determined from preparations in which ODC activity had been eliminated by acid denaturation. Units of ODC activity were expressed as pmoles 14 CO₂ generated/30 min/mg protein.

The cells collected for each exposure condition of an experiment were pooled and then divided into three equal aliquots, which were assayed to obtain a mean value of ODC activity for each exposure condition in an experiment. Each set of three flasks thus constituted a single sample, which was assayed in triplicate. This comparison of replicate samples allowed us to exclude from our results values that clearly resulted from errors in handling of samples or reagents. Furthermore, it provided an averaging of any small differences in ODC activity that might have resulted among a group of flasks due small variations in culture conditions or to the positioning of flasks within the Helmholtz coils. The data from an entire experiment were excluded from consideration if the standard deviation for the triplicate assay of any of the three exposure conditions exceeded $\pm 25\%$ of the respective mean; of 41 separate experiments conducted, five were rejected by this criterion.

ODC Activity as an Effective Marker for EM Field-Induced Response

For this report, as for previously reported work [Litovitz et al., 1991], the specific activity of ODC was selected as a marker for determining magnetic field-induced response. We have found ODC activity to serve reliably in this role, provided that the day-to-day variation in ODC activity displayed by cell cultures is taken into account. Variation in ODC activity between matched cultures, inoculated on the same day, is relatively small. When we compared ODC activities within groups of three or six identical cultures, standard deviations were found to range from 2% to 24% of the mean value. However, ODC activities may vary up to sixfold (range of approximately 5–30 units) among L929 cultures established on different days. Such

variation probably stems from small differences in cell culture conditions and has been observed by others (Dr. Christopher Cain, J.L. Pettis Memorial Veterans Administration Medical Center, Loma Linda, CA, personal communication). To minimize variations in ODC activity, we stockpiled acceptable lots of culture medium and serum to prevent possible variation due to lot differences (although similar responses to magnetic fields have been obtained with four different serum lots), we established a schedule for establishing and maintaining stock and experimental cultures that was maintained consistently each week, and we carefully determined the cell inocula for both stock and experimental cultures. Despite these protocols, day-to-day variation in ODC activity still occurred.

To allow comparisons of results among experiments carried out on different days, we have employed an ODC activity ratio, obtained by dividing the ODC activity of an EM field-exposed culture by that of its matched control. An ODC activity ratio of 2.1 thus indicates that the activity of a field-exposed culture was 2.1 times that of its control. We have found the ODC activity ratio to provide a consistent measure of field-induced effects, regardless of the baseline ODC activity of control L929 cells on a given day. Figure 2 illustrates this fact.

Data compiled from 73 experiments in which L929 cultures were exposed for 4 h to 60 Hz, 10 μ T fields are shown. Experimental results were separated into five groups based on the magnitude of the ODC activities determined for each control culture. Figure 2 demonstrates that, regardless of the ODC activity of the cells at

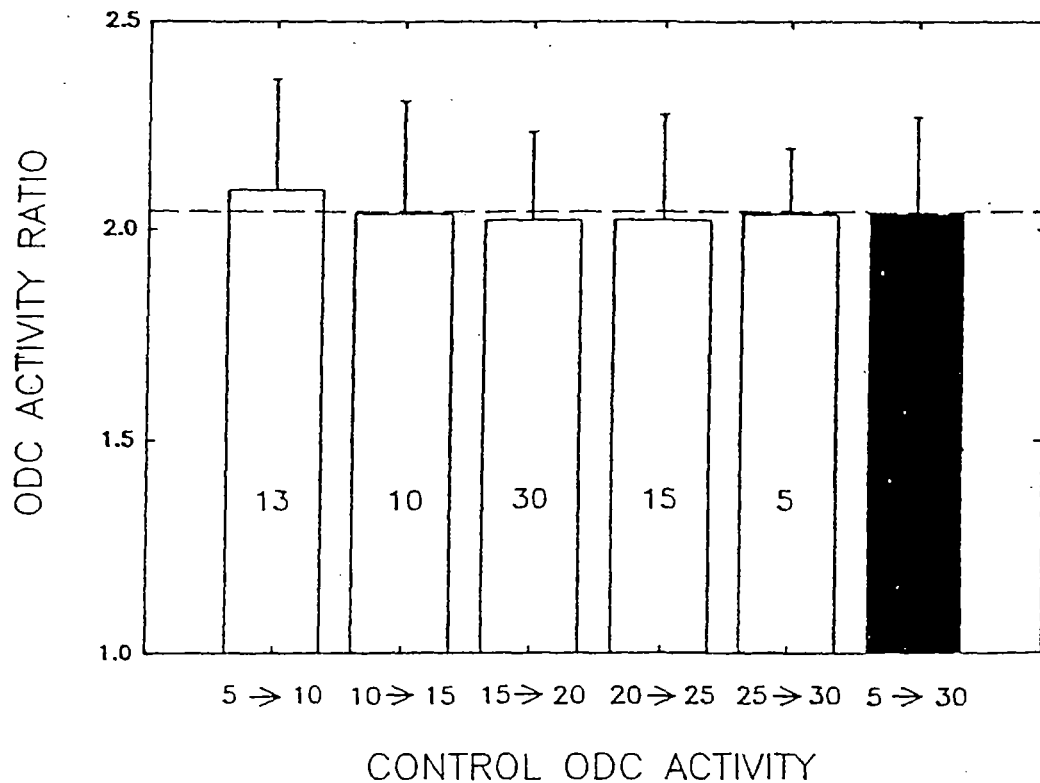


Fig. 2. Illustration of the consistency of the ODC activity ratio as an indicator of response of L929 cells to a 60 Hz, 10 μ T field. Results are grouped according to the ODC activity levels (pmoles 14 C $_2$ generated/30 min/mg protein) of the corresponding control cultures. The number of experiments corresponding to a given range of control ODC activity is given within the corresponding bar; the solid bar represents the mean \pm SD for all 73 experiments.

Mitigation Effect of Incoherent Magnetic Fields

the onset of exposure, all the cultures yielded an approximate doubling of ODC activity in response to the 60 Hz field. The EM field-induced response was independent of the baseline ODC activity of the cultures.

RESULTS

Separate Application of 60 Hz and Noise Fields

We initially compared the ODC response of L929 cells exposed to 60 Hz sinusoidal fields with that obtained for L929 cells exposed to noise fields alone. Consistent with previous results [Litovitz et al., 1991], 4 h exposure of L929 cells to the 10 μ T, 60 Hz stimulating field consistently produced an approximate doubling of ODC activity (ODC activity ratio = 2.1 ± 0.2 , $n = 36$). By contrast, 4 h application of random noise fields with rms amplitudes as high as 100 μ T had no effect on ODC activity (see Table 1).

Simultaneous Application of Noise Plus 60 Hz Fields

When a 10 μ T rms noise field was superimposed on the 10 μ T rms, 60 Hz field, yielding a noise-to-signal ratio (N/S) of 1, the typical, 60 Hz-induced doubling of ODC activity was eliminated. Cells exposed to these combined fields displayed ODC activities that were statistically indistinguishable from those of the negative controls (see Fig. 3). Thus the incoherent magnetic field inhibited the effect of the coherent magnetic field.

We conducted a series of exposures in which the 60 Hz field was maintained at 10 μ T, but the amplitude of the added noise field was held at rms values ranging from 0 to 10 μ T. The results of this series of experiments are plotted in Figure 3. The ODC activity ratio, [ODC], is plotted as a function of the ratio of the rms noise amplitude, N, to that of the rms amplitude of the 60 Hz signal, S, (N/S). It can be seen that, as the noise amplitude (and thus N/S) increased, the 60 Hz field-induced ODC activity decreased. The data from this exposure series were fitted to a curve calculated from the function:

$$[\text{ODC}] = 1 + \frac{1.06}{1 + 76 \left(\frac{N}{S}\right)^2}$$

This function suggests that the inhibitory effect of the noise field is a function of the square of the noise-to-signal ratio, N/S. When N/S was equal to 1, the inhibitory effect was essentially complete.

TABLE 1. ODC Activity Ratios Resulting From Exposure to 60 Hz and Noise Fields

	Sinusoidal (60 Hz)	Noise (30-90 Hz)	
	10 μ T	10 μ T	100 μ T
ODC activity ratio (exposed/control)	2.1 ± 0.2 ($n = 36$)	1.0 ± 0.1 ($n = 6$)	1.1 ± 0.1 ($n = 6$)

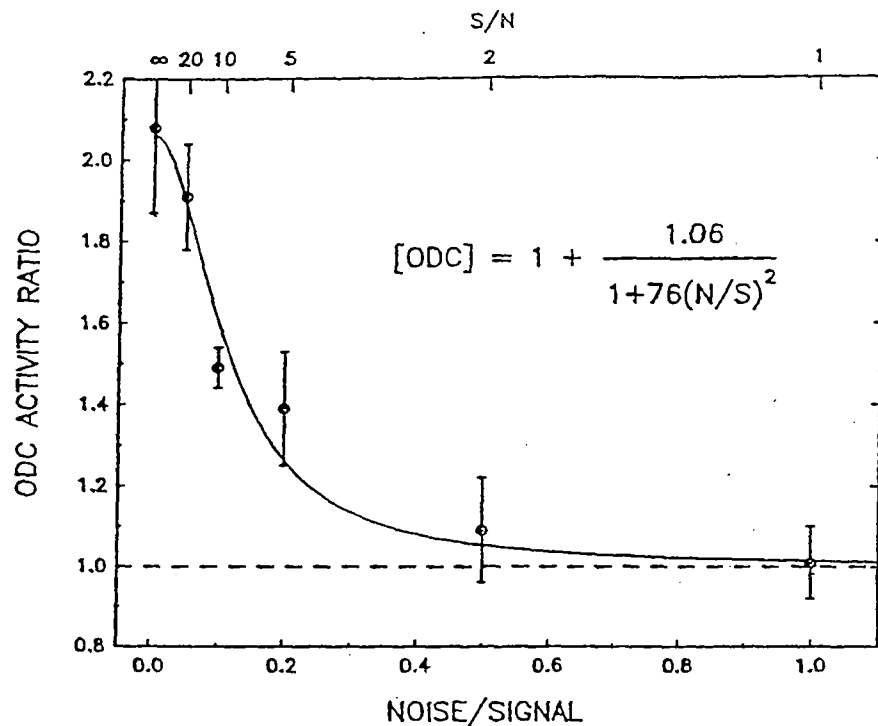


Fig. 3. Effects of 4 h exposure of L929 cultures to a 60 Hz, 10 μ T rms field combined with magnetic noise fields ranging in rms amplitude from 0 to 10 μ T. The ODC activity ratio, [ODC], is plotted against the noise-to-signal ratio (N/S). A scale for the corresponding signal-to-noise ratio (S/N) is provided on the upper x axis. Actual numbers of separate experiments used to calculate results for each noise level are 0.5 μ T (8), 1.0 μ T (6), 2.0 μ T (8), 5.0 μ T (8), 10.0 μ T (6). Since a 60 Hz-exposed condition was carried out for each experiment, the 0 μ T noise value was calculated from the results of all 36 experiments.

Complete inhibition of ODC activity enhancement was realized under conditions of equal rms amplitudes (10 μ T) for the noise and 60 Hz fields, yielding an rms amplitude of 20 μ T for the resultant, applied field. Since it was possible that the observed inhibition of ODC activity resulted simply from the increased rms amplitude of the resultant field, we performed additional exposures to examine this possibility. Two sets of cultures were exposed for 4 h to a 60 Hz field of 20 μ T rms. Enhancement, rather than diminishment, of ODC activity was clearly observed after these exposures (mean ODC activity ratio of 2.5 for the two runs). Furthermore, we obtained a twofold enhancement of ODC activity when 60 Hz fields with rms amplitudes of 100 μ T were used (ODC activity ratio 2.0 ± 0.5 ; $n = 4$). The inhibitory effect produced by addition of the ELF noise to the 60 Hz signal, therefore, appears not to be the result of some sort of "amplitude window."

Since the bandwidth of the noise field (30–90 Hz) encompassed the 60 Hz frequency of the stimulating field, we conducted an additional group of experiments to see whether this frequency relationship was important to our results. L929 cells were exposed for 4 h to the combined noise and 60 Hz fields, as before, but a noise field bandwidth of 90–150 Hz was employed. Noise of this bandwidth also effectively inhibited enhancement of ODC activity by the 60 Hz field, yielding an ODC activity ratio of 1.1 ± 0.1 ($n = 6$).

DISCUSSION

Several aspects of the results reported in this paper are deserving of comment and suggest the formulation of hypotheses to explain them. The basic result is that exposing cultured murine L929 cells to a relatively weak ($10 \mu\text{T}$) 60 Hz sinusoidal field for a period of 4 h effected roughly a doubling of the ODC activity in these cells. (Given the inherent, day-to-day variation in the ODC activity of cultured cells, the ODC response is seen to be a surprisingly good marker for assessing effects of field exposure.) The biological significance of this field-induced increase in ODC activity is uncertain. However, ODC activity is essential for DNA replication and cell proliferation [Heby and Persson, 1990], and the response we observed approximates the threefold [Goto et al., 1991] and fivefold [Cheng et al., 1992] increases in ODC activity observed for osteosarcoma cells specifically stimulated with parathyroid hormone. Given these facts, and the recent demonstration that enhanced expression of ODC may play a critical role in cell transformation [Auvinen et al., 1992; Hibshoosh et al., 1991], it is reasonable to assume that the field-induced response is physiologically relevant.

When a magnetic noise field was superimposed on the sinusoidal 60 Hz signal, the augmentation of ODC activity was reduced. When the rms amplitude of the noise field was significantly smaller than the amplitude of the sinusoidal field, the reduction was small; however, as the strength of the noise field was increased, the enhancement of ODC activity was correspondingly diminished, until finally, when the noise was comparable to the sinusoidal field, the effect was essentially totally suppressed. Neither the specific bandwidth of the applied noise field nor the increase in rms amplitude produced by addition of the noise field to the 60 Hz field accounted for the observed inhibition in ODC enhancement.

In one respect these results are not surprising: Any device, even one of biological origin, that is used to detect a temporally coherent EM field signal can be rendered nonfunctional by the presence of noise. Indeed, previous work [Litovitz et al., 1991] has demonstrated that, unless the impressed sinusoidal signal exhibits temporal coherence for time intervals on the order of 5–10 s, no increase in ODC activity is produced. Because the applied noise fields are temporally incoherent for time scales longer than about 0.02 s, they fail to satisfy this temporal coherence criterion and are incapable of stimulating an ODC response (as was shown). Moreover, the superposition of such an incoherent field on a coherent one (such as a 60 Hz sinusoidal field) leads to a total field that is incoherent, the degree of the incoherence being dependent on the relative amplitudes of the noise and coherent components. At first glance, the situation appears to be rather straightforward.

However, this mitigation of the magnetic field-induced bioeffect by the simultaneous application of a weak noise field (rms amplitude comparable to that of the sinusoid) is puzzling when considered in a slightly different light. Cells exist in an environment that is naturally abundant with electromagnetic noise. The random thermal motion of ions in the vicinity of cells leads to the presence of fluctuating fields that are roughly 1000 times larger than the externally imposed sinusoidal field and noise fields—roughly 0.1 mV/cm for the rms endogenous thermal noise field compared to approximately $0.1 \mu\text{V/cm}$ for the amplitude of the induced electric field component of our externally imposed 60 Hz, $10 \mu\text{T}$ magnetic fields [Adair, 1991; Weaver and Astumian, 1990]. However, the cellular effect of the sinusoidal field

is blocked by the weak external noise field, but it is undisturbed by the much larger thermal noise field. Cells have apparently evolved in such a way that they treat this ever present, random electromagnetic background stimulation as inconsequential.

Although the mechanism of electromagnetic field-cell interactions is not known, reasonable hypotheses can be constructed that account for thermal fields being ignored by cells. The data presented here imply that cells distinguish between applied noise and thermal noise. Some fundamental difference in the properties of these two noise fields must provide the basis for such differentiation. Although both are (by definition) temporally incoherent, we propose that they do exhibit a distinct difference in their spatial behavior: At a given instant, the value (magnitude and direction) of the thermal noise field at any point is uncorrelated with its value at other locations more than a few nanometers or so distant. This follows because the Debye screening length (roughly the range over which an ion is not shielded from other ions) in the extracellular fluid is only about 1 nm. Thus thermally driven localized charge density fluctuations (and, consequently, endogenous thermal noise fields) are spatially incoherent over distances greater than a few nanometers. By contrast, exogenously impressed fields (either temporally coherent or incoherent) always exhibit spatial coherence, since their wavelengths are much larger than cellular dimensions. We hypothesize that it is the spatial incoherence of the thermal noise field that allows cells to ignore it. Conversely, since externally imposed electromagnetic noise is spatially coherent, cells are unable to discriminate against it, so it is capable of confusing the biological EM field-detection mechanism.

If spatial coherence is the crucial field characteristic that enables cells to respond to exogenous EM fields, then one can further infer that the biological targets of electromagnetic fields are spatially extended or distributed. Two possibilities that should be examined are 1) that there is a collection of receptors on each cell that must be coincidentally excited to produce the observed biochemical responses and 2) that a rather large number of cells must be simultaneously stimulated; in either case, some cooperativity among the entities, involving intra- or intercellular signaling, respectively, must be operative.

We have suggested [Litovitz et al., 1994] a possible explanation of the first of these two possibilities. Cellular detection of EM fields is assumed to result from the impressed fields modifying the binding of ligands at their receptor proteins, resulting in a change in cell metabolism. The requirement that many receptors be simultaneously activated—cooperativity—prevents random activation of individual receptors from triggering an erroneous cellular response. It would also demand that electromagnetic field activation occur only if the stimulating field is essentially the same at a number of receptor sites over the cell membrane; this leads to the requirement of spatial coherence in the EM field if a cellular response is to be evoked. In this model, thermal noise fields, which are spatially incoherent, are incapable of causing any modification in cellular behavior.

An alternative explanation that also relies on the spatial coherence of the field is the supposition that communication and cooperation among cells via gap junctions are required for an alteration of cell functioning [Weaver and Astumian, 1992]. Such a multicellular response may account for EMF-induced effects in other systems, but are an unlikely explanation for our results since L929 cells have been shown not to form gap junction couplings [Matthews and Neale, 1989].

It should be noted that cell culture systems provide an electrical environment that is less complex than that encountered by cells in an intact organism. Absent

Mitigation Effect of Incoherent Magnetic Fields

from the culture environment are fields associated with the electrical activity of neurons, smooth and skeletal muscle cells, and myocardial cells. Our results do not address the possible effects of such endogenous, biological fields on the functions of cells in an organism.

ACKNOWLEDGMENTS

The authors acknowledge the Walter Reed Army Institute of Research for support of this work under Contract DAMD17-86-C-6260. The expert technical assistance of Mr. Abiy Desta and Mr. Plameh Doinov is appreciated.

REFERENCES

- Adair RK (1991): Constraints on biological effects of weak extremely-low-frequency electromagnetic fields. *Physiol Rev* 43:1039–1048.
- Auvinen M, Paasinen A, Anderson LC, Holttä E (1992): Ornithine decarboxylase activity is critical for cell transformation. *Nature* 360:355–358.
- Blank M, Soo L, Lin H, Henderson AS, Goodman R (1992): Changes in transcription in HL60 cells following exposure to alternating currents from electric fields. *Bioelectrochem Bioenerg* 28:301–309.
- Cheng S-L, Fausto A, Jänne OA, Avioli LV (1992): Regulation of ornithine decarboxylase by parathyroid hormone in osteoblastic cell systems. *Calcif Tissue Int* 51:370–375.
- Goodman R, Bumann J, Wei L-X, Henderson AS (1992): Exposure of human cells to electromagnetic fields: Effect of time and field strength on transcript levels. *Elec Magnetobiol* 11:19–28.
- Goto H, Matsui-Yuasa I, Otani S, Morisawa S, Nishizawa Y, Morii H (1991): Effects of parathyroid hormone on ornithine decarboxylase activity in human osteosarcoma cells. *Arch Biochem Biophys* 286:316–322.
- Heby O, Persson OL (1990): Molecular genetics of polyamine synthesis in eukaryotic cells. *Trends Biochem Sci* 15:153–158.
- Heby O, Marton LJ, Zardi L, Russell DH, Baserga R (1975): Changes in polyamine metabolism in WI38 cells stimulated to proliferate. *Exp Cell Res* 90:8–14.
- Hibshoosh H, Johnson M, Weinstein IB (1991): Overexpression of ornithine decarboxylase (ODC) on growth control and oncogene-induced cell transformation. *Oncogene* 6:739–743.
- Jänne J, Pösö H, Raina A (1978): Polyamines in rapid growth and cancer. *Biochim Biophys Acta* 473:241–293.
- Litovitz TA, Krause D, Mullins JM (1991): Effect of coherence time of the applied magnetic field on ornithine decarboxylase activity. *Biochem Biophys Res Commun* 178:862–865.
- Litovitz TA, Montrose CJ, Doinov P, Brown KM, Barber M (1994): Superimposing spatially coherent electromagnetic noise inhibits field-induced abnormalities in developing chick embryos. *Bioelectromagnetics* 15:105–113.
- Martin AH (1988): Magnetic fields and time dependent effects on development. *Bioelectromagnetics* 9:393–396.
- Matthews N, Neale ML (1989): Relationship between tumor cell morphology, gap junctions and susceptibility to cytolysis, by tumor necrosis factor. *Br J Cancer* 59:189–193.
- Mullins JM, Krause D, Litovitz TA (1993): Simultaneous application of a spatially coherent noise field blocks the response of cell cultures to a 60 Hz electromagnetic field. In Blank M (ed): "Electricity and Magnetism in Biology and Medicine." San Francisco: San Francisco Press, pp 345–346.
- Phillips J, Haggren W, Thomas WJ, Ishida Jones T, Adey WR (1992): Magnetic field-induced changes in specific gene transcription. *Biochim Biophys Acta* 1132:140–144.
- Seely JE, Pegg AE (1983): Ornithine decarboxylase (mouse kidney). *Methods Enzymol* 94:158–161.
- Weaver JC, Astumian RD (1990): The response of living cells to very weak electric fields—The thermal noise limit. *Science* 247:459–462.
- Weaver JC, Astumian RD (1992): Estimates for ELF effects: Noise-based thresholds and the number of experimental conditions required for experimental conditions required for empirical searches. *Bioelectromagnetics Suppl* 1:119–138.