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Responses of neurons to an amplitude modulated microwave stimulus

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Abstract

In this study we investigated the effects of a pulsed radio frequency signal similar to the signal produced by global system for mobile communication telephones (900 MHz carrier, modulated at 217 Hz) on neurons of the avian brain. We found that such stimulation resulted in changes in the amount of neural activity by more than half of the brain cells. Most (76%) of the responding cells increased their rates of firing by an average 3.5-fold. The other responding cells exhibited a decrease in their rates of spontaneous activity Such responses indicate potential effects on humans using hand-held cellular phones. © 2002 Published by Elsevier Science Ireland Ltd.

Keywords: Cellular telephone; Magnetic field; Health risk; Avian; Central nervous system

The postulated biological effects of electromagnetic fields are highly diverse, ranging from use of natural fields by animals for navigation to thermal cooking that occurs with strong fields such as those produced by microwave ovens [7]. It has been shown that even the weak fluctuations of Earth-strength magnetic fields influence the electrical activity of neurons and pineal cells and the synthesis of melatonin in birds and mammals [1,9,10], including humans [6]. Athermal effects have been the most difficult to explain because the mechanism by which they affect biological tissue is usually unknown. The question arises as to whether there is a particular sensitivity of the neural tissues of the brain to high frequency electromagnetic fields such as is produced by broadcast transmitters.

We tested the effects of electromagnetic radio frequency (RF) signals having a carrier frequency of 900 MHz, unmodulated and pulse modulated at 217 Hz with a duty cycle of 12.5% and a peak power density of 0.1 mW/cm^2. This stimulus was selected because it is similar to that used by the global system for mobile communication (GSM) telephone system. The calculated average SAR of this stimulus for the test subjects was 0.05 W/Kg, based on the equations in Durney and coworkers [8]. The test subjects were 34 adult zebra finches (*Taenopygia guttata*), anesthetized with a mixture of ketamine (0.05 mg/g) and xylazine (0.01 mg/g) injected i.m. into the pectoralis major. The anesthetized bird was mounted in a nonconducting plastic holder. The bird and the holder were placed inside a tuned RF cavity (23.5 cm diameter, 100.5 cm long) made of perforated metal. We used a resonate cavity (length = 3λ) because the resulting electrical field was a standing wave and, therefore, was uniformly distributed within the cavity and was measured accurately at the demodulating stub. The resonant cavity was fitted with two tuned RF stubs (each 16.5 cm [λ/2] from opposite ends): one for emitting the signal and one for monitoring the frequency and power of the signal within the cavity. This arrangement resulted in the two stubs being 2λ from each other causing the signal at the demodulation stub to be synchronized in phase and intensity to the emitted signal. The entire bird was within the cavity and positioned such that the bird's head was at the center of the cavity. This position put the bird's head exactly 1λ from the emitting stub and the demodulating stub. Consequently, the signal the bird's head received was exactly the signal at both of those locations. To record from neurons in the brain of the bird, a small hole (4 mm diameter) was made through the skull. A glass microelectrode (tip diameter 1–2 μm) filled with a conducting solution of physiological saline, to reduce conductivity, was slowly advanced into the

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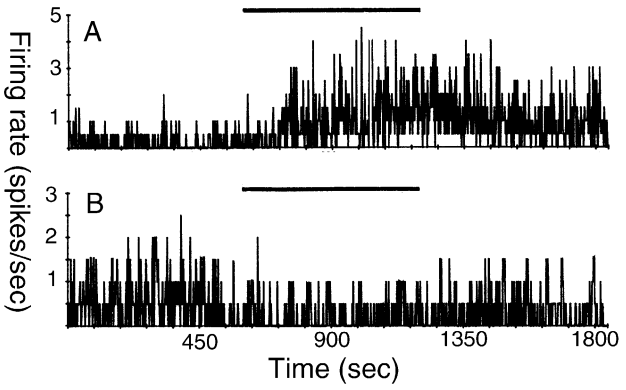

Fig. 1. Examples of neuronal responses in the zebra finch brain to stimulation of a 217 Hz, 12.5% duty cycle square wave modulated 900 MHz carrier signal: (A) simulation and (B) inhibition. The solid bar above each graph indicates the presence of the stimulating RF signal.

brain through this hole until a spontaneously active nerve cell was detected. A silver reference electrode was inserted beneath the skin along the back of the head directly behind the glass microelectrode to complete the circuit. Arranging the electrodes along the long axis of the cavity prevented them from acting as a loop antenna and electrically stimulating the cells. Once a spontaneously active cell was located, it was tested with the stimulus. The protocol for all the testing procedures was a 10 min prestimulus period, a 10 min stimulus period, and a 10 min poststimulus period. The rates of the cell's activity during these three time intervals were compared to detect any effect of the stimulation. A responding cell was one that changed its firing rate during the stimulation by at least 10%.

The microwave stimulus signal was produced by an Amplifier Research amplifier (model 10W1000M7) driven by an HP 8350A sweep oscillator with an HP 83522A RF unit set for 900 MHz. Amplitude modulation of the signal was produced by a free running HP 3314A function generator set for 217 Hz square wave signal with a duty cycle of 12.5%. The output of the amplifier was switched between a matched load and the cable to the waveguide chamber by a SPDT RF switch (HP 8761A). The switch was controlled by a digital signal from the computer program TIDA on an IBM-compatible microcomputer. The frequency and intensity of the emitted signal were monitored using an HP 5342A microwave frequency counter connected to the demodulator stub in the waveguide cavity. All power measurements were of peak power.

We recorded 133 spontaneously active units from 34 anesthetized adult zebra finches. The recording locations were in the cerebrum (Pars occipitalis and Pars parietalis) and Folia of the anterior cerebellum. Ninety-one units (69%) showed some response to the stimulation: 69 (52%) responded with excitation (Fig. 1A) and 22 (17%) responded with inhibition (Fig. 1B). The remaining 42 (31%) cells showed no discernible response. The cells showing excitation responded with increases in their rate of firing to the stimulation (mean rate during stimulation = 3.5 ± 0.30 [SE] times prestimulus rate; Fig. 2). Most of the inhibitory responses were small (mean rate during stimulation = 0.4 ± 0.07 times prestimulus rate; Fig. 2), in part because the cells were firing slowly before the stimulation. There was a significant difference among the firing rates of the three responses and the prestimulus firing rate (Kruskal–Wallis test: $H_c = 216.8$, $P < 0.001$, $v = 5$; see Fig. 3). Based on a non-parametric multiple comparison [13], the firing rates in the three response categories different from one another significantly ($P < 0.05$; Q = 3.817–4.341). There was no significant difference among the firing rates of the nonresponding cells during the prestimulus, stimulus, and post-stimulus periods ($P > 0.05$). All responses we recorded were to power densities of 0.1 mW/cm^2 (SAR = 0.05 W/Kg) and stronger (up to 0.5 mW/cm^2). The mean latency from the initiation of the stimulus to the start of the response was 104 ± 197 s, with the response lasting beyond the end of the stimulus period in half of the responding cells. The mean persistence beyond the end of stimulation was 308 ± 68 s, but there was no correlation ($r = 0.489$, $P > 0.05$) between the latency of the response and how long the cell continued responding beyond the end of the stimulus.

Three cells that responded to the modulated carrier were also tested with an unmodulated signal of the same carrier frequency. The power of the unmodulated signal was tested at two densities: one that equaled the peak power of the modulated stimulus and one that equaled the average power of the modulated stimulus. None of these cells exhibited a response to the unmodulated carrier. In addition to responses to the nominal stimulus, we also tested four cells that did not respond to the 0.1 mW/cm^2 pulsed signal with higher power densities (up to 0.5 mW/cm^2). Three cells did

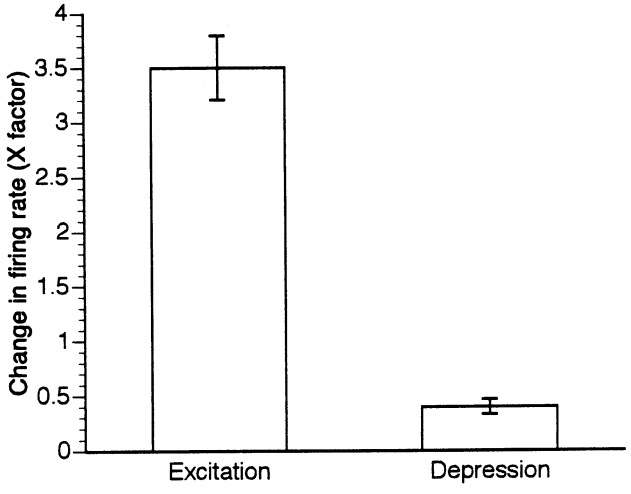

Fig. 2. Mean relative firing rates of cells that responded to the simulated GSM signal and were categorized as excitation or depression. The firing rates are relative to the cells' firing rates during the prestimulus period. The vertical bars indicate 1 standard error.

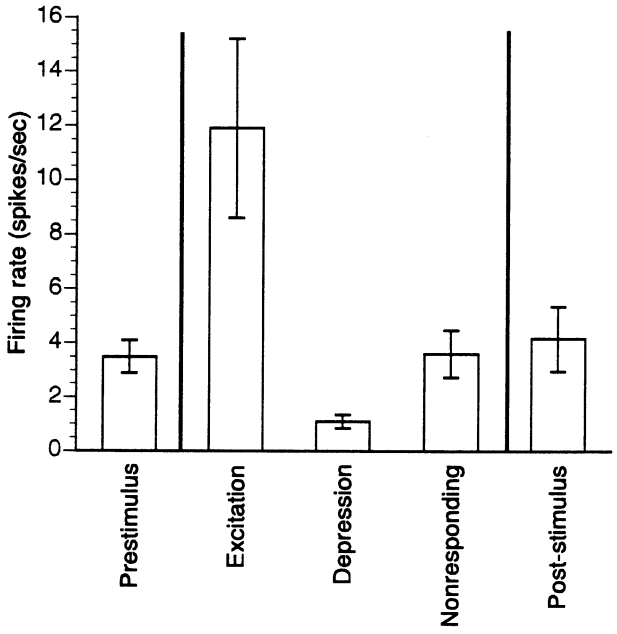

Fig. 3. Firing rates of zebra finch neurons during the prestimulus (10 min), stimulus (10 min), and poststimulus (10 min) periods. The poststimulus values are for the non-responding cells only because the responding cells often continued their response into the poststimulus period (see text for details). The firing rates of the three responses differed significantly (Kruskal–Wallis test: $H_c = 216.8$, $P < 0.001$, $v = 5$). The vertical bars indicate 1 standard error.

not respond to the stronger intensities, but one cell that did not respond to the 0.1 mW/cm^2 stimulus responded to an intensity of 0.3 mW/cm^2 with depression of its rate of activity.

One concern was that the electrodes themselves were acting as an antenna and stimulating the cells electrically. The arrangement of the active and reference electrode centered along the long axis of the waveguide chamber prevented them from serving as a loop antenna. In preliminary experiments we varied the positions of the electrodes to determine whether they could, in fact, act as an antenna. When the electrodes were not aligned longitudinally, the stimulus artifact was detected directly and observed on the oscilloscope display. Whether such a stimulus was strong enough to stimulate the cells is unknown. A second factor that supports the idea that the cells were not stimulated electrically is that not all cells responded to the stimulus, even those in the close neighborhood of a responding cell. This clearly speaks against an artifact.

These high frequency RF fields produced a response in many types of neurons in the avian central nervous system (in both cerebellum and cerebrum) and did not appear to be limited to any specialized receptor. Similar responses (long latency and ongoing higher activity after cessation of the stimulus) also were recorded to a 52 GHz carrier, 16 Hz modulated signal (Semm et al., unpubl. data). Thus, the effect does not appear to be limited to magnetic sensory cells [11], but may occur in any part of the brain. The similar responses to different frequencies point toward a common mechanism of low frequency modulation, perhaps at the cell membrane Such a stimulus might mimic a natural mechanism involved in cell communication, producing responses from many different types of neurons. It is unlikely that the effects we observed are the result of thermal excitation caused by the RF radiation because the power densities we applied were 2–3 orders of magnitude below what is required (10 mW/cm^2) to produce heating of even 0.5 °C [2]. It is also unlikely that localized areas of the brain were heated and thermally stimulated because neurons responded only to the modulated signal and did not respond to unmodulated signals that were the same strength. Consequently, we conclude that the effects we observed are not the result of thermal agitation but at this point we cannot offer an athermal mechanism to account for the observations.

Although individual neurons in the zebra finch brain responded to the pulsed RF stimulus, we do not know whether these responses by the nervous system are manifested in the bird's behavior or its health. Bruderer and coworkers [4,5] reported no behavioral responses of birds to pulsed or continuous RF microwave signals, but their studies involved different frequencies and lower power densities of the stimulus. Thuróczy and coworkers reported neuronal responses of freely moving rats [12] similar to the responses we observed in the zebra finch. During the period of stimulation, sensitive cortical neurons of Long Evans rats showed either an increase or a decrease in the rate of spontaneous activity. The changes in firing rates were less than the changes we observed in the zebra finch: an increase of less than 2× in the rat versus 3.5× in the finch and a decrease to 0.67× in the rat versus 0.4× in the finch. Although the neuronal responses were similar between the rat and the finch, the SAR values of the RF field used with the rat were much greater than that used for the finch. Thuróczy and coworkers also observed behavioral responses by the rat to the GSM signal. In conditioning experiments, the rats' reaction times decreased during stimulation as did their learning rate (as measured by discrimination tasks).

Whether similar neuronal responses occur in other mammals, including humans, requires further investigation. Borbély and coworkers [3] reported that exposure to a RF signal similar to the one we used influenced sleep and sleep electroencephalogram in humans. Their results and the responses we recorded clearly indicate the potential for effects on the human nervous system.

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