Microbiological Research 172 (2015) 26–33

Contents lists available at ScienceDirect

Microbiological Research

journal homepage: www.elsevier.com/locate/micres

Bacterial growth rates are influenced by cellular characteristics of individual species when immersed in electromagnetic fields

Lucas W.E. Tessaro[a,b,*], Nirosha J. Murugan[a,b], Michael A. Persinger[a,b]

[a] Behavioural Neuroscience Program, Laurentian University, Sudbury, Ontario, Canada P3E 2C6
[b] Department of Biology, Laurentian University, Sudbury, Ontario, Canada P3E 2C6

ARTICLE INFO

Article history:
Received 23 May 2014
Received in revised form
15 December 2014
Accepted 19 December 2014
Available online 19 January 2015

Keywords:
Electromagnetic fields
Bacteria
E. coli
Rates of growth
Biophysics

ABSTRACT

Previous studies have shown that exposure to extremely low-frequency electromagnetic fields (ELF-EMFs) have negative effects on the rate of growth of bacteria. In the present study, two Gram-positive and two Gram-negative species were exposed to six magnetic field conditions in broth cultures. Three variations of the 'Thomas' pulsed frequency-modulated pattern; a strong-static "puck" magnet upwards of 5000 G in intensity; a pair of these magnets rotating opposite one another at ~30 rpm; and finally a strong dynamic magnetic field generator termed the 'Resonator' with an average intensity of 250 μT were used. Growth rate was discerned by optical density (OD) measurements every hour at 600 nm. ELF-EMF conditions significantly affected the rates of growth of the bacterial cultures, while the two static magnetic field conditions were not statistically significant. Most interestingly, the 'Resonator' dynamic magnetic field increased the rates of growth of three species (*Staphylococcus epidermidis*, *Staphylococcus aureus*, and *Escherichia coli*), while slowing the growth of one (*Serratia marcescens*). We suggest that these effects are due to individual biophysical characteristics of the bacterial species.

© 2015 Elsevier GmbH. All rights reserved.

Introduction

The behavioural effects of electromagnetic fields (EMFs) have been reported since the 1970s (Persinger 1970; Persinger and Pear 1972). Recent developments in the field have shown that exposure to non-ionizing, electromagnetic radiation can induce numerous and quite varied biological effects (Adey 1981; Santini et al. 2009; Cifra et al. 2010). For example, exposure to extremely low frequency (ELF)-EMFs, ranging from 0 to 100 Hz, are capable of activating cellular immune responses (Simkò and Mattson 2004; Akan et al. 2010). ELF-EMFs are known to produce voltage gradients in tissues in the range of $10–10^{-1}$ V/cm. These gradients are essential to various physiological functions at the level of the cell (Adey 1993).

The majority of research regarding non-ionizing radiation has fixated on the probability of analogous effects. Despite this, there have been an increasing number of studies suggesting that exposure to certain patterned ELF-EMFs can inhibit the growth of cancer cells (Hu et al. 2010; Karbowski et al. 2012). Other beneficial effects have been reported in studies where bisected planarian flatworms exposed to various ELF-EMFs have increased the rate of regeneration of the worms (Novikov et al. 2008; Goodman et al. 2009; Tessaro and Persinger 2013).

One field in which the application of EMFs has not been well-defined is the influence of EMF on microorganisms. Goodman et al. (1994) conducted a series of studies on the bacterium *Escherichia coli* which demonstrated that 60-min exposure to weak, frequency-pulsed magnetic fields (1.5 mT peak) increased the intracellular levels of a number of proteins by at least a factor of 2. A subsequent study revealed that as little as 15-min of exposure to a 60 Hz, 1.1 mT sine-wave magnetic field can enhance the intracellular level of σ^{32} mRNA (Cairo et al. 1998). Gross colony morphology effects and slowed growth after exposure to ELF-EMFs were reported in a study by Strašák et al. (2002).

Nawrotek et al. (2014) have noted that the application of 50 Hz rotating magnetic fields (RMF) significantly increased the rate of growth in two species, *E. coli* and *S. aureus*, with the largest effect seen within the exponential phase, or approximately 150 min into their exposure. It was also found that 50 Hz RMF applications can modulate a variety of bacterial functions in various species, such as metabolic activity, biofilm formation and overall growth dynamics (Fijalkowski et al. 2014). These studies taken together demonstrate the variety of biological processes susceptible to electromagnetic stimuli, in particular growth dynamics, and emphasize the importance of the spatial–temporal component of the applied field.

* Corresponding author at: Department of Biology, Laurentian University, Sudbury, Ontario, Canada P3E 2C6. Tel.: +1 705 675 4824; fax: +1 705 671 3884.
E-mail addresses: lx_tessaro@laurentian.ca (L.W.E. Tessaro), mpersinger@laurentian.ca (M.A. Persinger).

http://dx.doi.org/10.1016/j.micres.2014.12.008
0944-5013/© 2015 Elsevier GmbH. All rights reserved.

In the present study, we exposed four bacterial strains for a total of 12 h to one of six magnetic field conditions using nutrient broth as a growth medium. The purpose of the present study was to investigate what effects the different magnetic field conditions may have upon the rate of bacterial growth. This is pertinent given the prevalence of electromagnetic fields in our everyday environments. Here we demonstrate a differential, although transient, effect of specific field strengths and patterns upon specific strains of bacteria and their observed rates of growth as inferred by optical density measurements.

Materials and methods

Bacterial cultures

Bacterial cultures were obtained from American Type Culture Collection (ATCC), Manassas, VA, USA. Four bacterial strains were used; *Staphylococcus epidermidis* (ATCC 14990), *Staphylococcus aureus* (25923) as Gram-positive bacteria and *Serratia marcescens* (264), *E. coli* (11303) as Gram-negative bacteria. These species were selected to have two representative Gram-positive and Gram-negative cultures. In addition, all species are considered to be part of the human microbiome, thus commonly interact with the magnetic fields that humans are exposed to. Strains of mentioned bacteria were cultured with nutrient agar from which stock nutrient broth cultures can be prepared. Nutrient broth powder (Oxoid CM00001) and agar powder (Fischer A360-500) were obtained from Fischer Scientific (Mississauga, ON).

Broth media were prepared in 1 L batches and dispensed in 10 mL aliquots in 20 mL Pyrex-brand test tubes (No. 9825), with 1 cm diameter and autoclaved with plastic caps on at 125 °C for 30 min, and stored at 4 °C with until required. Stock cultures were started 24 h prior to inoculation of experimental test tubes to calculated CFUs of: (a) 6.17×10^9 CFU/mL for *S. epidermidis*; (b) 1.2×10^7 CFU/mL for *S. aureus*; (c) 1.0×10^7 CFU/mL for *S. marcescens*; and (d) 2.8×10^7 CFU/mL for *E. coli*. Stock cultures were started in 10 mL test tubes, then 10 μL of those cultures inoculated into each of the experimental condition samples after the 24 growth period. The same stock culture was used to inoculate each condition at the beginning of each trial to ensure the same experimental conditions. Bacterial species were kept at 37 °C (±2 °C) for the full 12 h exposure.

Optical density assay

Fresh bacterial cultures for the exposures were initiated in 10 mL, 1 cm diameter Pyrex test tubes in autoclaved nutrient agar broth. The level of growth was measured utilizing the optical density of the respective broth cultures compared to a non-inoculated broth culture (blank/control), that is a aliquot of broth media that has not been inoculated with any bacteria. Optical density measurements were taken every hour for 12 h at 600 nm using a Spectronic 20 Spectrophotometer (Milton Roy Company).

Magnetic field exposure

The prepared strain samples were exposed to one of six magnetic field conditions. Three were variations of a complex, pulsed frequency-modulated field ("Thomas pulse") that has been shown to affect analgesic thresholds in mice or cell-relevant reactions in variety of invertebrates and vertebrates (Thomas et al. 1997; Martin et al. 2004a,b). We also utilized strong, static magnets with an intensity of 5000 G, a set of these magnets rotated counterclockwise, and a magnetic field generator termed the 'Resonator' with an intensity of 250 μT.

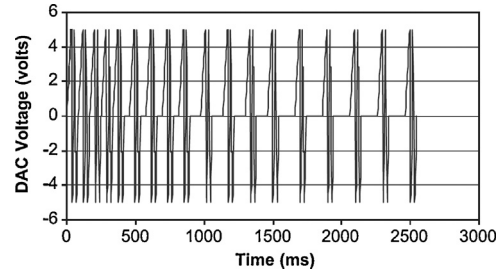

Fig. 1. The 'Thomas' pattern electromagnetic field waveform utilized in this study. The vertical axis denotes the voltage values converted from numbers between 0 and 255 (where 127 = 0 V). The horizontal axis denotes the temporal changes of the pattern generated by the numbers. The duration of each number (or point) was either 3 ms or 1 ms. The pattern itself contains a total of 850 points.

For the time-varying magnetic field (Thomas) the generator was a 30 Ω Helmholtz coil comprised of a 39 cm by 39 cm box wrapped by 305 m of 30-gauge AWG (American Wire Gauge) insulated copper wire as described by Hu et al. (2010). The pattern was generated by converting a row of 849 numbers between 0 and 255 into voltages. Values above 127 were positive polarity while values below 127 were negative polarity. These values then generated voltages between −5 V and +5 V using a Zenith-386 computer (Model ZF-148-41, Zenith Data Systems, Benton Harbor, MI, USA). A digital to analogue converter was utilized along with specialized software to convert the numbers to the appropriate current necessary to generate the field. Both the software and digital-to-analogue converter were custom built by our technologist, Prof. Stanley Koren.

The Thomas field is a frequency-modulated pattern that begins with an initial presentation of 25 Hz (Fig. 1). Thereafter, the frequency slows to 6 Hz. At the centre of the coil, bacteria were subjected to an average field strength of 3.8 μT (ranging from 2.1 to 5.5 μT). Bacteria not receiving the magnetic field exposure (i.e. shams) were placed in an identical non-active Helmholtz coil for 12 h housed within a separate identical incubator. The sham cultures were started at the same time as the experimental conditions to ensure all other experimental variables were controlled for excluding the magnetic field presentation.

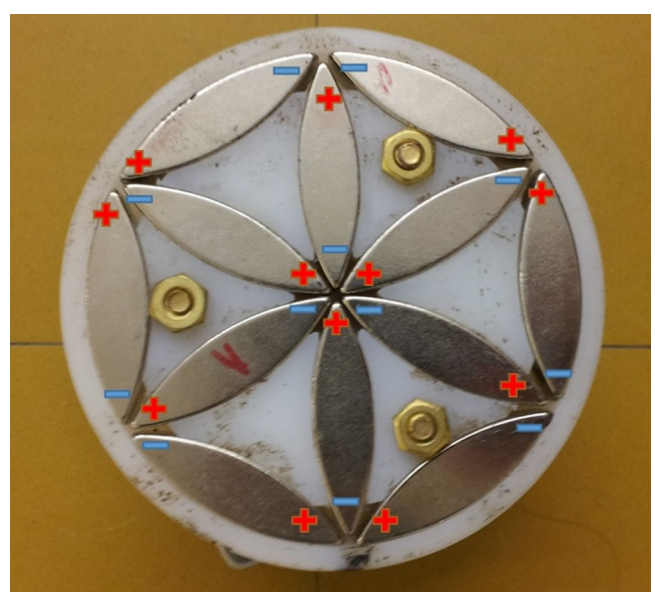

Fig. 2. Cross-sectional view of the arrangement of the 'puck' magnets. The 12-ovoid neodymium magnets are arranged in what is known as the 'Tree of Life' organization, based upon ideals of 'Sacred Geometry', such as the Golden ratio. The positive and negative signs denote polarity of the magnets. Areas where multiple poles are in contact had an intensity of 4800 G, with the centre of the magnets 1000 G.

Download English Version:

https://daneshyari.com/en/article/2092249

Download Persian Version:

https://daneshyari.com/article/2092249

Daneshyari.com