

Establishing a Mechanism for the Effects of Specific Patterned Electromagnetic Fields at the
Molecular Level Using Fragmented Bacteria

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Abstract

Electromagnetic fields (EMF) are a physical property resulting from the movement of charged particles, have elicited behavioral changes. Changes at the microscopic level have yet to be observed. In the present study, the objective was to determine if EMFs have an effect on biological matter and to determine the mechanism producing the change. Bacterial species were an ideal candidate for this type of research, as their rapid growth permitted extensive experimentation. Four separate species of bacteria (*Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Serratia marcescens*) were lysed to destroy their cellular integrity, exposed to one of three EMF conditions (Sham, Thomas-EMF, LTP-EMF) for 60 minutes, and analyzed using spectroscopic techniques. The effects of the EMFs were ascertained by analyzing the absorbance and fluorescence of biological matter pre and post treatment. Results demonstrated that there was approximately a 10-15% increase in absorbance for solutions exposed to an EMF condition compared to sham. The results indicate that the EMF exposure had no significant impact on the fluorescence of the biological matter within the solution. Biological matter from the different bacterial species had a significant impact on their fluorescence. Implications for these results regarding the theory of abiogenesis shall be discussed.

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All organisms inhabiting our planet, from the largest animals to single cell bacteria, are immersed within the Earth's natural magnetic field (Dotta & Rouleau, 2014). Other artificially made electromagnetic fields (EMFs) are also being generated in everyday life from power lines, smartphones, computers, cars, etc. Studies have shown that EMFs produce an extensive range of effects on biological systems which are dependent on the nature of the field being generated as well as the biological system itself (Fleming, Persinger, & Koren, 1994; Fojt, Strašák, Vetterl, & Šmarda, 2004; Mach & Persinger, 2009; Murugan, Karbowski, Lafrenie, Persinger, & Aegerter, 2013; Tessaro & Persinger, 2013). These effects include changes in perception, learning, memory, growth, cognition and more. Many of these studies report beneficial effects for these biological systems (Mach & Persinger, 2009) (Martin et al., 2005) (Tessaro & Persinger, 2013); however others have reported severe disruptions in these processes causing detrimental consequences (Fojt et al., 2004) (Murugan et al., 2013). They all suggest different hypotheses in regards to their mechanisms of action but none has provided conclusive evidence in support of any one mechanism.

The purpose of this study is to determine if EMFs interact with biological systems at the molecular level of discourse. Reports of changes in function by EMFs should be indicative of a change in structure since structure dictates function (Persinger, 2001). Studies, as mentioned above, have shown that EMFs interact with these biological systems; however they could only provide hypotheses regarding them. They do not mark or demonstrate any underlying mechanism at a smaller level of discourse. Therefore the following fundamental question also arises; how do specific EMFs interact within biological life forms? More specifically, the

purpose was to examine biological changes at the molecular level which may be causing these effects given that structure determines function. In order to explore these aspects, an understanding of the physics delineating EMFs will explain how they apply to biological systems.

Electromagnetism

Electromagnetism is the phenomenon of electromagnetic forces produced by electrically charged particles (Fahidy, 2002). The electric component refers to the electrically charged particles while the magnetic portion demonstrates the attraction of opposing charges or repulsion of identical ones. An EMF is a physical property resulting from the movement of these charged particles (Fahidy, 2002). These fields are further described by Maxwell's equations which explain how the fields change in space by their sources (both the electrical and magnetic components) and how they propagate (Misner & Wheeler, 1957). Faraday's Law stipulates that the electric field circulates around the alternating magnetic field, that is, the electrically charged particles will move depending on the forces of attraction and repulsion similar to a magnet pulling or pushing another magnet (Barnes & Maekawa, 2007). Ampere's Law states that the magnetic field circulates around the alternating electric field (Griffiths, 1999). In other words, the forces of attraction and repulsion will differ if the electrically charged particles are moving within an area (Griffiths, 1999). These fields can be either static, where the intensity of the field remains unchanged as a function of time, or they can be dynamic, marking a fluctuation in intensity as a function of time (Pilla, 2006). As mentioned before, their effects are species and pattern dependent.

EMFs are virtually ubiquitous in modern urban environments (Knave, 2001). Their sources include but are not limited to those produced by power lines and modern technological devices such as smartphones, iPods, televisions, vehicles, etc. (Knave, 2001). Biological systems also produce their own electromagnetic fields (Black & Robinson, 1990). The rationale is that all life forms are composed of the fundamental elements carbon, hydrogen, oxygen and nitrogen (Woodard & White, 1986). Every chemical element is associated with an electric charge, as subatomic particles that comprise elemental atoms are also charged protons (positive), electrons (negative) and neutrons (no charge) (Parr, Ayers, & Nalewajski, 2005). Many biochemical processes involve the movement not only of charged macromolecules, but of individual protons and electrons as well, generating biological EMFs (Fahidy, 2002). For example, during an action potential, protein channels within the axon membrane of the neuron change conformation to allow the influx of sodium ions into the neuron and efflux of potassium ions (Mueller & Rudin, 1968). These movements of ions create a fluctuation in electric charges and magnetic force, therefore generating an EMF (Fahidy, 2002). The heart also produces its own special EMF (Black & Robinson, 1990). Much like action potentials, the muscles of the heart also exhibit electrical activity via their pacemaker cells (Woodard & White, 1986). Ions flow through the membrane, thus creating electric potentials which allow the muscles to contract in order to pump blood from the heart to the body (Woodard & White, 1986). These all illustrate the importance of electromagnetism as a fundamental property of life.

EMFs on Biological Systems

A significant amount of literature is now available which show that the effects of EMFs on living organisms are dependent on the species, and the particular behavior studied, as well as the pattern and intensity of the EMF. In a study by Fleming et al. (1994), it was shown that male

Wistar rats exposed to a pulsed EMF for 30 minutes showed a decrease in their perception of pain. This study demonstrated that the Burst-X, an EMF simulating the pattern of the amygdala's neuronal activity, produced effects equivalent to 4 mg/kg of morphine. This indicated that EMFs could initiate effects similar to their pharmaceutical counterpart.

A study by Martin et al. (2005) involved the exposure of male Wistar rats to the Thomas pattern EMF for varying durations of time and compared their pain threshold to controls (sham field). Rats exposed to the field pulse pattern for 30 minutes and tested 4 hours later marked a significant difference in latency time compared to their baseline thus delaying their perception of pain. The researchers hypothesized a change in the neuronal activity of the claustrum via the secondary messenger nitric oxide.

In another study, Mach and Persinger (2009) exposed groups of either normal or brain damaged rats to either a sham or LTP-EMF during a water maze behavioral task. Brain damage was caused by lithium-pilocarpine induced seizures. The brain damaged animals showed a significant improvement in the behavioral task compared with controls when exposed to LTP-EMF during the task. This would implicate changes in the synaptic structure of neurons associated with learning and memory.

Tessaro and Persinger (2013) revealed that EMF may increase the regeneration rate of bisected planarian flatworms. Planaria were exposed to the Thomas-EMF for durations ranging from 0 minutes to 180 minutes after being segmented at the level of the pharynx. Results showed that the field exposure had an optimal regeneration rate following 45 minutes of exposure. They hypothesized the activation of the p38-mitogen activated protein kinase (p38-MAPK) and heat shock protein 70 (hsp70) pathways to induce cell regeneration.

Murugan et al. (2013) demonstrated that EMFs can also be harmful to an entire organism. They subjected planaria to the Thomas-EMF for 6.5 hours for four days, followed by an exposure to a simulated geomagnetic storm for an additional 6.5 hours on the 5th day. Results showed a 100% mortality rate 24 hours subsequent to the geomagnetic field exposure. Switching the order did not produce dissolution of planaria, thus showing that the effect was very sensitive to the order of the EMF. They postulated a weakening of protein structure which may have caused an overwhelming influx of calcium into the cell provoking cell death.

As discussed above, EMFs produce a large diversity of effects. In order to determine their potential mechanism at the molecular level, a cellular model will be required for experimentation. Bacteria will make for a perfect model in providing such an insight.

Bacteria

Bacteria are prokaryotic unicellular microorganisms and are considerably smaller than human eukaryotic cells (Gitai, 2005). These organisms are easy to manipulate due to their rapid reproduction rate (Gitai, 2005). Like the eukaryotic cell, they possess a membrane which encompasses their cytoplasm and regulates the flow of substances leaving and entering the cell (Gitai, 2005). They are devoid of a “true” nucleus in the sense that they lack a nuclear envelope to contain and protect their genetic material (Gitai, 2005). Many of the most fundamental biochemical processes are identical across eukaryotic and prokaryotic cells, known as genetic conservation. With respect to the current study, all organic matter and living organisms are comprised of the same essential components of matter. These major structural and molecular similarities make bacteria a powerful tool in studying fundamental properties (Gitai, 2005).

Fojt et al. (2004) exposed three different strains of bacteria (*Escherichia coli*, *Leclercia adecarboxylata* and *Staphylococcus aureus*) to an extremely low frequency sinusoidal wave EMF for 12 minutes. They diluted the bacterial solutions and plated them on a Petri dish for exposure. Subsequent to the exposure, they were incubated for 24 hours prior to counting the number of colonies. They noticed a 60% decrease in colony forming units (CFU), or the amount of bacteria on the dish. Their hypothesis denotes the alterations in the permeability of charges within the cell membrane however it has not been tested.

Current Study

The objective of the present study is to determine if EMFs have a gross effect at the macromolecular level of biological matter. Previous literatures have all hypothesized changes in pathways which led to the differences observed. However, a definitive link between changes in molecular structure with biological function in response to EMFs has been made. Since structure dictates function (Persinger, 2001), determining if molecular changes of the cell occur subsequent to the EMF exposure could show how these fields produce their effects, since cells are the building blocks for all life forms.

This study is essential in expanding the knowledge of how EMFs exhibit these effects. If EMFs can affect the structure of molecules, then maybe we can develop an EMF to promote proper molecule structures. Examples of such structures are protein channels imbedded within the cell membrane. These can create irregular ion flux by protein dysfunction when it is misfolded. Correcting the structure would theoretically restore proper functioning of the protein thus fixing the malfunction. This may also help in the opposite, where destruction is required for improvement. A classic example is that of cancer. Understanding how electromagnetic fields

affect biological systems may provide insight on developing a tool to counteract cancerous growth and eliminating it from the affected area.

Hypotheses

There was a hypothesized main effect for EMF exposure. Fojt et al. (2004) denoted a decrease in bacterial reproduction subsequent to an EMF exposure possibly due to the changes in the cell membrane. Since an EMF constitutes a movement of charged particles and the bacteria are composed of matter with charges, then the changes in electromagnetic forces of attraction and repulsion should produce changes in the properties of the biological matter due to the EMF's influence on surrounding matter.

There was a hypothesized main effect for bacteria. Fojt et al. (2004) showed a decrease in bacterial reproduction for all three species of bacteria; however the rate of change was different. Since every bacterium has a different structural makeup, they will respond differently to the EMFs. *Escherichia coli* and *Serratia marcescens* have a double membrane compared to *Staphylococcus aureus* and *Staphylococcus epidermidis* single membrane with a thicker layer which may affect the degree of change.

Lastly, there was an expected interaction in that the EMFs will produce different effects depending on the bacterium. As mentioned in the literature review, EMFs produced remarkably different effects depending on the organism it was applied to. Martin et al. (2005) applied the Thomas field to rats and showed a delay in response time to pain whereas Tessaro and Persinger (2013) demonstrated that planarian worms had an increase in regeneration after being exposed to the same patterned field. The matching field being applied to two distinct species marked

different effects. The same should occur here because each bacterium has structural properties that are different from one another.

Method

Apparatus and Materials

This experiment required the utilization of a spectrophotometer. The spectrophotometer's mechanism involves shining a ray of light through a solution (Rendina, 1971). When the light hits regions containing fragments, it gets absorbed (Rendina, 1971). However, light that is not absorbed will transmit through the solution and be detected by the detector which will project an absorbance value compared to a blank solution (for this experiment, it is a tube containing the same solution as bacteria without any bacterial debris being present) (Rendina, 1971). An increase in absorbance is a correlate of aggregation as it implies less light is travelling through to the detector and therefore being absorbed by a structure becoming larger (Ruban, Horton, & Young, 1993)

Another instrument used in this experiment was the Olis Rapid-Scanning Monochromator spectrofluorometer. This device operates similarly to the spectrophotometer in regards to shining a ray of light to the solution (Rendina, 1971). However this light shines at a specific excitation wavelength to detect specific properties of the biological matter within the solution (Rendina, 1971). For this experiment, the excitation wavelength was set at 280nm, a wavelength of light that is absorbed by the amino acid tryptophan (Bronk & Reinisch, 1993). Afterwards, the light absorbed is emitted. The detector collects the rays emitted and produces a spectrofluorescent analysis of the fluorescent counts (photon emission) within a specific range set by a computer

software (Rendina, 1971). The range was set between 300nm-400nm because the peak emission wavelength of tryptophan is approximately 350nm (Dalteria et al., 1986).

EMF Exposure

The fields were produced using Helmholtz coil wrapped around plastic crates, a Zenith-386 computer with a specialized software and a digital to analogue (DAC) converter. The Thomas field was generated by converting a row of 849 numbers between 0 and 255 into the appropriate current required to generate it. All values above 127 had a positive polarity whereas values below 127 had a negative polarity. The voltages ranged from -5V to +5V with an average intensity of 38mG (milliGauss). Each pattern of the Thomas field lasted 2.55s with each point value being presented for 3ms. The time between pattern presentations was also 3ms. The LTP field was generated using the same equipment. However, 225 numbers between 127 and 256 were converted, providing strictly with positive polarity and ranged between 0V to +5V. Each point value was presented 1ms apart and the entire pattern was shown once every 4s. The average intensity was approximately 3mG. The groups receiving the Sham-field condition were placed within the same Helmholtz coil without any of the equipment running.

Procedure

The first independent variable that was utilized in this experiment was bacteria, measured in colony forming units (CFU, amount of bacteria present in the solution). This IV consisted of four different species of bacteria: *Escherichia coli* (4.0×10^9 CFU/mL), *Staphylococcus epidermidis* (7.0×10^9 CFU/mL), *Staphylococcus aureus* (3.0×10^9 CFU/mL) and *Serratia marcescens* (1.0×10^9 CFU/mL). These four bacterial species are commonly found in the human flora (Sears, 2005). *Escherichia coli* and *Serratia marcescens* are Gram-negative bacteria

(presence of an outer membrane) whereas *Staphylococcus aureus* and *Staphylococcus epidermidis* are Gram-positive (single membrane with a thickened peptidoglycan layer) (Hugenholtz, 2002).

The second independent variable that was explored was the EMF pattern. For this experiment, two different EMFs were employed: the Thomas pattern, which was shown to induce analgesic effects in rats (Mach & Persinger, 2009) and played an important role in the destruction of an entire organism (Murugan et al., 2013), and the LTP pattern that marked enhanced learning in brain damaged rats (Tessaro & Persinger, 2013). The Thomas field is described as a continuous field of 3ms pulses at 25Hz (Hertz) for the first 200ms followed by a reduction to 6Hz for the last 500ms (Murugan et al., 2013). The LTP field, on the other hand, consisted of a 5ms pulse followed by four 5ms pulses in rapid succession (10ms in between each) 150ms later (Tessaro & Persinger, 2013).

The first dependent variable that was observed in the current study was the biological matter's optical density. This current study will look at determining the amount of light travelling through the solution which is related to the nature of the biological matter in the solution. This will be measured using a spectrophotometer to determine the opaqueness before and after the electromagnetic field exposure. This will quantify any differences in the biological matter's optical density.

The second dependent variable that was analyzed was the fluorescent counts (the amount of photons emitted) of the bacterial species. This will be measured using the Olis Rapid-Scanning Monochromator spectrofluorometer to identify any specific changes tryptophan.

Bacteria were prepared 24 hours in advance prior to any experimentation. In order to prepare the solutions, 100 μ L of the stock solutions (concentrations ranging in the range of 10^9 bacteria/mL) were placed in 10mL of nutrient broth contained in a 15mL centrifugation tube sealed by a red cap. They were then placed in an incubator with a temperature of 37°C in order to maximize the conditions for the bacteria to reproduce. After 24 hour incubation, they were centrifuged at 22 000 G (13 000 rpm) for 15 minutes to induce fragmentation due to the pressure associated with the rapid spinning. Further fragmentation was induced via mechanical shearing using 30-gauge and 26-gauge needles. The small opening increased the pressure within the needle when force was applied to eject the bacteria, thus causing fragmentation. The fragmentation is crucial in order to ascertain that the bacteria are in an inactivated state. Lysed solutions were then placed in the Helmholtz coil and randomly exposed to the Sham-field, Thomas field or LTP field conditions for 1 hour. Spectrophotometry and spectrofluorometry measurements were performed to acquire baseline measurements both pre and post EMF exposure. This procedure was conducted in triplicate.

Results

Spectrophotometer analysis

The results of this experiment support the concept that EMFs affect biological matter. Results of the two-way analysis of variance (ANOVA) of absorbance values by species by EMF conditions indicated that the EMF conditions had a significant main effect on the magnitude of the difference in absorbance values ($F(2,32) = 8.140$, $p < .05$, $\eta^2 = 0.356$). Tukey's *post hoc* test revealed that the Sham field condition ($M=-0.0017$, $SE=0.00207$) was significantly different from both Thomas-EMF ($M=0.0392$, $SE=0.00773$) and LTP-EMF ($M=0.0283$, $SE=0.00683$)

conditions (Appendix, Figure 1). Generally, the electromagnetic field conditions displayed an increase in absorbance compared to initials and were not significantly different from one another. There was no significant main effect for the bacteria variable ($F(3,32) = .367, p > .05$). Similarly, there was no significant interaction between bacteria and EMF condition ($F(6,29) = .258, p > .05$).

Spectrofluorometer analysis

The results of this experiment demonstrate that EMFs do not affect biological matter properties associated with the amino acid tryptophan. Results of the two-way analysis of variance of fluorescent counts by species by EMF indicated that fluorescent counts increased following exposure to an EMF compared to sham, however it was not significant ($F(2,33) = 1.310, p > .05$), meaning the EMF condition did not produce a significant change in the fluorescence of the biological matter. There was no significant impact of the bacterial species on the difference in fluorescent counts ($F(3,32) = 1.026, p > .05$), meaning the difference in bacterial species did not produce a significant difference in fluorescent counts. The interaction between bacteria and EMF was also not significant ($F(6,29) = .312, p > .05$).

The results of a two-way analysis of variance of the fluorescent counts by species by EMF condition at every 1nm increment of the emission wavelength (300-400nm) following EMF exposure were significant. There was a main effect caused by species on the fluorescent counts starting at 340nm ($F(3,32) = 3.058, p < .05, \eta^2 = 0.269$) and remained significant to 400nm ($F(3,32) = 13.223, p < .05, \eta^2 = 0.606$) (Appendix, Figure 2). This effect was also present prior to EMF exposure.

Discussion

As expected, the results of this experiment show that EMFs have an influence on biological matter. All biological matter from the four species of bacteria displayed an increase in absorbance following a one hour exposure to an EMF whereas the sham field did not. In other words, more light was being absorbed by the solution of biological matter for the field conditions, therefore less light was travelling through to the detector and the solution became cloudier after the EMF exposure. As Ruban et al. (1993) indicated, an increase in absorbance is an indicator of aggregation, where smaller structures draw closer to one another and form larger complexes. These larger structures absorb more energy which would explain the increase in absorbance.

The spectrofluorescent analysis showed that there was an increase in photon emission associated with the amino acid tryptophan following an exposure to an electromagnetic field compared to sham; however the increase was not significant. This entails that the integrity of biological matter containing tryptophan did not change significantly. On the other hand, when comparing the fluorescence at every 1nm increment of the emission wavelength (300-400nm) before and after EMF exposure, there is a significant difference between fluorescent counts of the biological matter from the different species of bacteria starting at 340nm, within range of the peak emission of tryptophan (Dalteria et al., 1986). This serves as validation that the species of bacteria are structurally different. According to Figure 2, *Escherichia coli* and *Staphylococcus epidermidis*, Gram-negative and Gram-positive respectively, more closely resemble each other whereas *Serratia marcescens* and *Staphylococcus aureus*, also Gram-negative and Gram-positive respectively, show similarities despite differences in their membrane structures. This could be attributed to the differences of bacterial concentration as *Escherichia coli* and *Staphylococcus*

epidermidis contained higher concentrations than *Serratia marcescens* and *Staphylococcus aureus* resulting from their diverse growth rates (Tessaro, Murugan, & Persinger, 2015).

If aggregation is occurring and yet there is no significant change in biological matter structure, what could be the mechanism of this phenomenon? Let's consider the amount of energy stored within a volume from an EMF denoted by the equation $E = (B^2/\mu) \times V$ where E is the amount of energy stored in Joules, B is the strength of the EMF in Tesla (3.0×10^{-6} T for LTP-EMF), μ is the magnetic permeability constant ($4\pi \times 10^{-7}$ N/A²) and V is the volume of the solution (1×10^{-5} m³). This equation yields 1.15×10^{-10} J which, when divided by the total number of bacteria within the entire solution ($\sim 10^{10}$) equals $\sim 10^{-20}$ J, a very important neuromolecular quantum (Persinger, 2010). This value is within the range of the amount of energy observed for the formation of new biochemical bonds and to preserve intermolecular forces (Persinger, 2010). Therefore that amount of energy could be responsible for the formation of new biochemical bonds which could lead to aggregations that are maintained by the intermolecular forces. That quantum is also associated with an increase in the viscosity of water (Persinger, 2014). Ghauri and Ansari (2006) performed an experiment where they demonstrated an increase in the viscosity of water subsequent to the exposure of an EMF. This increase in viscosity implies the water is becoming thicker and more compact, thus bringing the molecules closer together. They hypothesized that the EMF increased the amount of hydrogen bonds, a type of intermolecular force between polar molecules, caused by a hydrogen atom with a partially positive charge and an atom of a separate molecule with a partially negative charge. This could explain the increase in absorbance values following the stimulation of an EMF occurring in the current study.

Implications

The results of this study suggest that EMF played a vital role in the aggregation of biological matter. Miller (1953) devised an experiment to simulate Earth's atmosphere billions of years ago. In his study, he placed water containing Earth's primitive oceans in a closed system composed of glass and boiled it to evaporate the water which created its primitive atmosphere. He then generated a spark to simulate lightning. The evaporated water would travel through a condenser and condensate into a collecting trap. After noticing the collected water transformed pink, he performed chromatography to identify the substance immersed inside. This substance was recognized as amino acids which he hypothesized to be the first abiogenic molecules to appear on Earth. In other words, they were the first molecules required for natural processes of life to be formed from non-living matter, also known as the theory of abiogenesis. The present study can relate to it since aggregates of biological matter were formed in a solution using an EMF. The EMF would serve as the closed system (Earth's geomagnetic field) in order to allow the biological matter to remain within proximity of each other. Water would serve as the medium required to bring these molecules together through an increase in viscosity induced by the EMF. Therefore without the EMF surrounding the Earth, the molecules could have easily escaped the atmosphere and scattered across the universe.

This study also demonstrates that EMFs interact with all that is made of water. All life on Earth is composed of water as all biological systems are made of cells which contain water (Woodard & White, 1986). The human body is composed of approximately 65% water (Mitchell, Hamilton, Steggerda, & Bean, 1945). The human brain is composed of 73% water (Mitchell et al., 1945). Seeing as EMF affect properties of water, then these changes would in turn affect

cellular processes involved with water. Therefore this study implies that EMFs produce their effects on biological systems by affecting the water that composes them.

Limitations and Future Directions

The present study indicates certain limitations. Although the degree of fragmentation for the bacteria was confirmed to be 100% in the first trial, it was not re-evaluated for the remainder of the experiment. The metabolic activity of non-fragmented bacteria could have influenced the absorbance values. There could have also been cross contamination as the same syringe and needles were employed to induce fragmentation via mechanical shearing. Despite washing the syringe and needles with distilled water after each bacterium, traces of them could have remained within these tools.

Further studies should be conducted to confirm the results of the present study. Alternatively, employing the geomagnetic field would confirm the relevance of this study to the theory of abiogenesis. Performing this very same experiment and using phase contrast microscopy to visualize the forming of aggregates would evaluate the validity of water as the mechanism behind these effects. Finally, replicating Miller's study (1953) and adding the geomagnetic field would confirm the role that the Earth's EMF played in the creation of biogenic molecules.

The present study has demonstrated that specific patterned EMFs are capable of inducing changes in biological matter. It is hypothesized that EMF initiate their effects through water, an essential medium present in all life. Further studies are required in order to expand our understanding of their interaction.

References

- Barnes, S. E., & Maekawa, S. (2007). Generalization of Faraday's law to include nonconservative spin forces. *Physical review letters*, *98*(24), 246601.
- Black Jr, W. C., & Robinson, S. E. (1990). Analysis of biological signals using data from arrays of sensors. U.S. Patent No. 4,977,896. Washington, DC: U.S. Patent and Trademark Office.
- Bronk, B. V., & Reinisch, L. (1993). Variability of steady-state bacterial fluorescence with respect to growth conditions. *Applied spectroscopy*, *47*(4), 436-440.
- Dalterio, R. A., Nelson, W. H., Britt, D., Sperry, J., Psaras, D., Tanguay, J. F., & Suib, S. L. (1986). Steady-state and decay characteristics of protein tryptophan fluorescence from bacteria. *Applied spectroscopy*, *40*(1), 86-90.
- Dotta, B. T., & Rouleau, N. Y. (2014). Electromagnetic Fields as Structure-Function Zeitgebers in Biological Systems: Environmental Orchestrations of Morphogenesis and Consciousness. *Name: Frontiers in Integrative Neuroscience*, *8*, 84.
- Fahidy, T. Z. (2002). The effect of magnetic fields on electrochemical processes. *In Modern aspects of electrochemistry* (pp. 333-354). Springer US.
- Fojt, L., Strašák, L., Vetterl, V., & Šmarda, J. (2004). Comparison of the low-frequency magnetic field effects on bacteria *Escherichia coli*, *Leclercia adecarboxylata* and *Staphylococcus aureus*. *Bioelectrochemistry*, *63*(1-2), 337-341.
- Ghauri, S. A., & Ansari, M. S. (2006). Increase of water viscosity under the influence of magnetic field. *Journal of Applied Physics*, *100*(6), 066101-066101.

- Gitai, Z. (2005). The new bacterial cell biology: moving parts and subcellular architecture. *Cell*, *120*(5), 577-586.
- Griffiths, D. J., & Reed College. (1999). Introduction to electrodynamics (Vol. 3). Upper Saddle River, NJ: Prentice hall.
- Hugenholtz, P. (2002). Exploring prokaryotic diversity in the genomic era. *Genome Biology*, *3*(2), 1-8.
- Knave, B. (2001). Electromagnetic fields and health outcomes. *Annals of the Academy of Medicine, Singapore*, *30*(5), 489-493.
- Mach, Q. H., & Persinger, M. A. (2009). Behavioral changes with brief exposures to weak magnetic fields patterned to stimulate long-term potentiation. *Brain research*, *1261*, 45-53.
- Martin, L. J., Koren, S. A., & Persinger, M. A. (2005). Thermal analgesic effects from weak, complex magnetic fields: critical parameters. *Electromagnetic Biology and Medicine*, *24*(2), 65-85.
- Miller, S. L. (1953). A production of amino acids under possible primitive earth conditions. *Science*, *117*(3046), 528-529.
- Misner, C. W., & Wheeler, J. A. (1957). Classical physics as geometry. *Annals of physics*, *2*(6), 525-603.
- Mitchell, H. H., Hamilton, T. S., Steggerda, F. R., & Bean, H. W. (1945). The chemical composition of the adult human body and its bearing on the biochemistry of growth. *Journal of Biological Chemistry*, *158*(3), 625-637.
- Mueller, P., & Rudin, D. O. (1968). Action potentials induced in biomolecular lipid membranes.

- Murugan, N. J., Karbowski, L. M., Lafrenie, R. M., Persinger, M. A., & Aegerter, C. M. (2013). Temporally-Patterned Magnetic Fields Induce Complete Fragmentation in Planaria. *PLoS ONE*, *8*(4), e61714.
- Parr, R. G., Ayers, P. W., & Nalewajski, R. F. (2005). What is an atom in a molecule?. *The Journal of Physical Chemistry A*, *109*(17), 3957-3959.
- Persinger, M. A. (2001). The neuropsychiatry of paranormal experiences. *Neuropsychiatry*, *13*(4).
- Persinger, M. A. (2010). 10-20 Joules as a neuromolecular quantum in medicinal chemistry: an alternative approach to myriad molecular pathways?. *Current Medicinal Chemistry*, *17*(27), 3094-3098.
- Persinger, M. A. (2014). Quantitative Convergence between Physical-Chemical Constants of the Proton and the Properties of Water: Implications for Sequestered Magnetic Fields and a Universal Quantity. *Physics and Astronomy*, *2*, 1-10.
- Pilla A. (2006). Mechanisms and therapeutic applications of timevarying and static magnetic fields. In: Barnes F, Greenbaum B, editors. Handbook of biological effects of electromagnetic fields. 3rd edition. Boca Raton, FL: CRC Press
- Rendina, G. (1971). Experimental methods in modern biochemistry.
- Ruban, A. V., Horton, P., & Young, A. J. (1993). Aggregation of higher plant xanthophylls: differences in absorption spectra and in the dependency on solvent polarity. *Journal of Photochemistry and Photobiology B: Biology*, *21*(2), 229-234.
- Sears, C. L. (2005). A dynamic partnership: celebrating our gut flora. *Anaerobe*, *11*(5), 247-251.

Tessaro, L. W., & Persinger, M. A. (2013). Optimal durations of single exposures to a frequency-modulated magnetic field immediately after bisection in planarian predict final growth values.

Bioelectromagnetics, 34(8), 613-617.

Tessaro, L. W., Murugan, N. J., & Persinger, M. A. (2015). Bacterial growth rates are influenced by cellular characteristics of individual species when immersed in electromagnetic

fields. *Microbiological Research*.

Woese, C. R., Kandler, O., & Wheelis, M. L. (1990). Towards a natural system of organisms: proposal

for the domains Archaea, Bacteria, and Eucarya. *Proceedings of the National Academy of*

Sciences, 87(12), 4576-4579.

Woodard, H. Q., & White, D. R. (1986). The composition of body tissues. *The British journal of*

radiology, 59(708), 1209-1218.

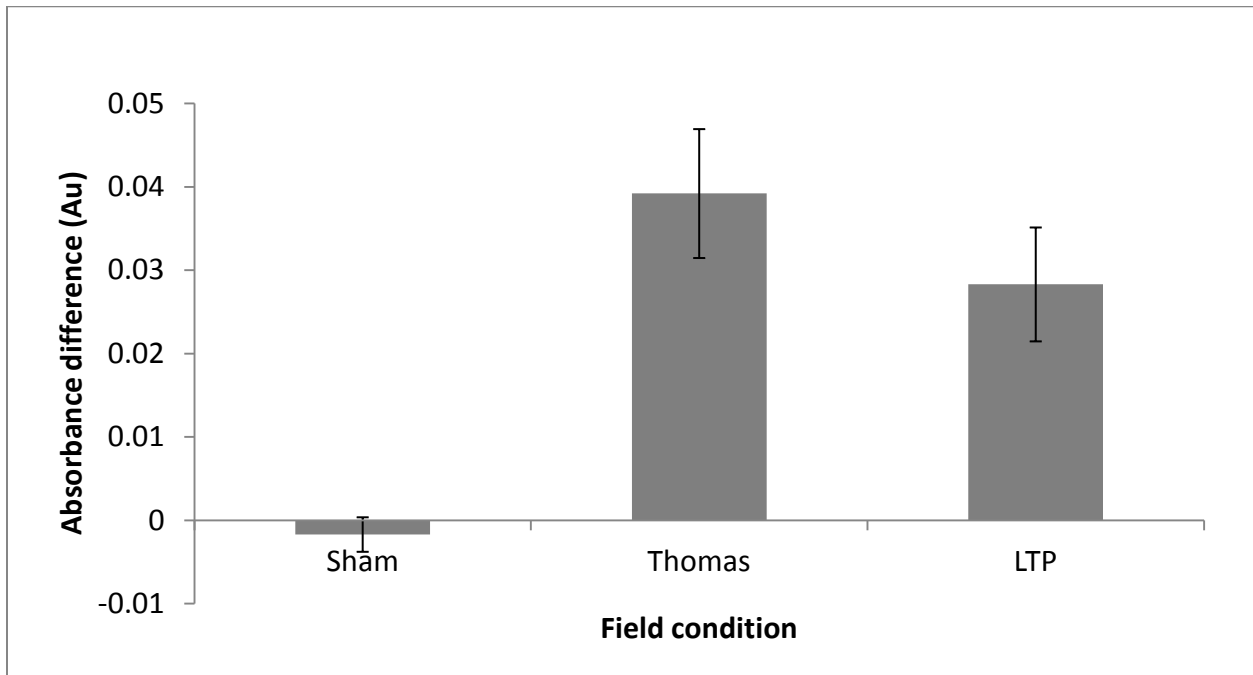
Appendix

Figure 1. Average change in absorbance values following a one hour exposure to a field condition. Error bars represent the standard error of the means.

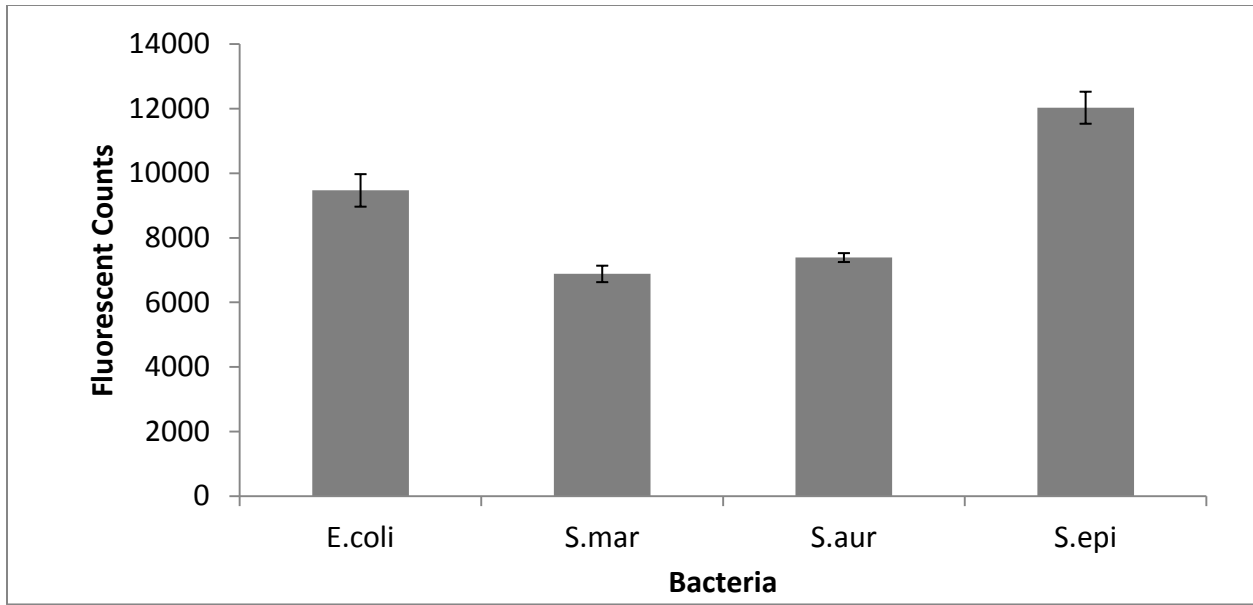


Figure 2. Average fluorescent counts at 390nm following a one hour exposure to a field condition. Error bars represent standard error of the means.