The effects of natural magnetic fields on biological systems: Evidence from planaria, sunflower seeds and breast cancer cells

by

Victoria Hossack

A thesis submitted in partial fulfillment
of the requirements for the degree of
Master of Science (MSc) in Biology

The Faculty of Graduate Studies
Laurentian University
Sudbury, Ontario, Canada

THESIS DEFENCE COMMITTEE/COMITÉ DE SOUTENANCE DE THÈSE
Laurentian Université/Université Laurentienne
Faculty of Graduate Studies/Faculté des études supérieures

Title of Thesis Titre de la these	The effects of natural magnetic fields on biological systems: Evidence from planaria, sunflower seeds and breast cancer cells	
Name of Candidate Nom du candidat	Hossack, Victoria	
Degree Diplôme	Master of Science	
Department/Program Département/Programme	Biology	Date of Defence Date de la soutenance January 16, 2019

APPROVED/APPROUVÉ

Thesis Examiners/Examinateurs de thèse:

Dr. Blake Dotta
(Co-Supervisor/Co-directeur de thèse)

Dr. Rob Lafrenie
(Co-Supervisor/Co-directeur de thèse)

Dr. Michael Persinger (†)
(Supervisor/Directeur de thèse)

Dr. Peter Ryser
(Committee member/Membre du comité)

Dr. Bryce Mulligan
(External Examiner/Examinateur externe)

Approved for the Faculty of Graduate Studies
Approuvé pour la Faculté des études supérieures
Dr. David Lesbarrères
Monsieur David Lesbarrères
Dean, Faculty of Graduate Studies
Doyen, Faculté des études supérieures

ACCESSIBILITY CLAUSE AND PERMISSION TO USE

(†) Deceased

Abstract

Natural magnetic fields include the Earth's magnetic field and biomagnetic fields produced by living organisms. Earth has a static magnetic field whose intensity is constantly fluctuating and has deviations called geomagnetic storms. In multiple experiments, we found that geomagnetic storms that occurred either on the same day as the experiment, or four days before the experiment had a significant impact on the subsequent behaviour of planaria and photon emissions of germinating sunflower seeds. However, the effects were most evident in only a subset of their populations. Consistent with this idea, a small proportion of planaria seemed able to detect the presence of a weak, patterned electric field in a maze. We also found that when humans practised healing intentionality on breast cancer cultures, they had a subtle influence on the photon emission from the cells. These results demonstrate the effectiveness of natural magnetic fields even if their influences evade our awareness.

Keywords: electric field, magnetic field, geomagnetic storm, planaria, photon, cancer, experimental design, plant, endogenous, circadian, weather, neuroscience

Acknowledgements

First, I would like to thank Dr. Dotta. He has had a great undertaking over the past 6 months, but despite this he was still able to find the time to help me finish my master's, which I could not have done without him. I would like to thank Dr. Lafrenie as he was instrumental in this thesis, especially with the cell culture experiments; by providing the cells for us to begin, and then providing them again and again as we continued to have contaminations. Thank you Dr. Ryser for showing me how to run experiments with plants, in this thesis and beyond. I would also like to thank my external reviewer, Dr. Mulligan, for the helpful comments.

I would like to thank the NRG, you guys mean the world to me. What a great and strange experience it has been. Thank you for all of the collaboration and chats, both enthusiastic and argumentative were extremely beneficial in developing this thesis. I need to thank Professor Vares for all of the help with the photon data processing, his matlab wisdom saved 100s of hours. And LT, for being so supportive; I cannot imagine making it through this difficult time without you.

Most importantly, thank you Dr. Persinger. Words really cannot express how much he impacted me. I would like to thank him for inspiring me, challenging me, and showing me what it means to be an open minded rational thinker. Thank you Dr. Persinger for caring about me, being so supportive and kind and for believing in me. With pride, I am going to carry the tools you helped me develop and all of the wonderful and joyous memories, with me for the rest of my life.

Table of Contents

List of Tables

List of Figures

Abbreviations

Abbreviation	Meaning
°C	degrees Celsius
ECG	electrocardiography
EEG	electroencephalography
ELF	extremely low frequency
EMF	electromagnetic field
Hz	hertz
Λ	lambda
MCF-7	female human breast epithelial cancer cell line
MCG	magnetocardiography
MEG	magnetoencephalography
Ω	ohm
PMT	photomultiplier tube
rpm	rotations per minute
SD	standard deviation
SEM	standard error of the mean
SPD	spectral power density
T	Tesla
UV	ultraviolet
V/m	Volts per meter
W/m^2	Watts per meter squared

Chapter 1 – Introduction

From a young age, people are always taught that we have 5 senses: touch, taste, hearing, smell and sight. These systems all operate on the basic principle of sensory cells detecting an exogenous stimulus that they are specialized for, this information is then relayed to the cerebral cortex for the individual to become aware of. It is a common fallacy that most people assume our world consists of what we are able to perceive using the five senses. Many things that are not detectable by our five senses do exist. For example, a myriad of additional weather variables are continuously present in our environment. People are aware of temperature changes, precipitation, humidity and wind speeds because these are all easily perceived. However, people cannot easily perceive changes in the barometric pressure. We know it exists and that low pressure is associated with thunderstorms, but we do not have any specialized sensory cells that can detect this change in pressure. This does not mean that changes in barometric pressure cannot still have an influence on our behaviour. For example, decreases in barometric pressure are associated with restlessness, water retention that leads to arthritic pain (Persinger, 1980), decreased mood scores (Persinger & Levesque, 1983) and increased suicide rates in Japan (Tada et al., 2014).

Two other weather-related variables that we cannot easily perceive are atmospheric electricity and the Earth's electromagnetic field. Atmospheric electricity describes the potential difference generated between the negatively charged surface of the Earth and the positively charged upper atmosphere (Persinger, 1980). There are diurnal variations in field intensity with minimums between 02:00 and 04:00 local time, and seasonal variations with minimums during December-January and maximums during

July-August (Persinger, 1980). The intensity of the atmospheric electricity ranges from 120V/m (volts per meter) to 150V/m (volts per meter), which increases up to 10,000V/m during thunderstorms and 1,000V/m during falling snow (Persinger, 1980). The biological effects associated with an increased potential difference are similar to those described for barometric pressure. This is not surprising as the increase in potential difference just before a thunderstorm is coupled with a decrease in barometric pressure (Persinger, 1980).

Earth's Magnetic field (the geomagnetic field)

The Earth's static magnetic field is thought to be generated by the relative rotation of the liquid iron core at its centre (Press & Siever, 1974). The magnetic north and south poles of the Earth represent the dipoles of the magnetic field, with flux lines travelling from the south pole to the north pole (Sears & Zemansky, 1964). The greater the density of flux lines, the higher the intensity of the magnetic field, therefore because the flux lines converge at the poles, the geomagnetic field at the poles have an intensity of about 70μT, whereas the magnetic field at the equator is about 25μT (Persinger, 1980). The geomagnetic field also has a time varying component with frequencies that fall in the extremely low frequency (ELF) range. A frequency of approximately 7.8Hz, corresponds to a wavelength of the ELF that approximates the circumference of the Earth, which produces a resonance between the Earth and the ionosphere (Persinger, 1975b). Earth's magnetic field can be influenced by solar wind, a stream of plasma that originates from the Sun. This actually compresses the geomagnetic field, increasing its intensity

(Persinger, 1980), and subsequently produces a diurnal variation, with the lowest intensities during the night (Persinger, 1980; Liboff, 2013).

There are multiple indices that can be used to describe the shifts in the intensity of the Earth's magnetic field during a geomagnetic storm. The most well-known is the kp index, a semi logarithmic scale that was invented in 1938 (Bartels, Heck & Johnston, 1939). The Ap index is derived from the kp index and has units of nanoTesla. The values reported for these indices are derived as averages from 13 observatories around the globe (Rostoker, 1972). They represent the largest deviation in the horizontal and declination component of the field. The vertical component of the field used to be included but was removed as it was always disturbed the least (personal communication, Dr. Claudia Stolle, Professor of Geomagnetism, Head of Section 2.3 Geomagnetism at GFZ German Research Centre for Geosciences, Helmholtz Centre Potsdam). The dst index represents the ring current around the Earth and is derived from observatories near the equator (Rostoker, 1972). The AA (antipodal) index is derived from two observatories, one in the northern hemisphere in the UK and one in the southern hemisphere in Australia (Mayaud, 1972).

Biological effects of geomagnetic storms

In humans, geomagnetic storms have been associated with several abnormal behaviours including: increased psychotic episodes (Freidman et al., 1963), decreases in mood scores for males (Persinger, 1975a), increased psychotic depression in males (Kay, 1994), increased vestibular experiences (Persinger & Richards, 1995), increased out of body experiences (Persinger, 1995a), increased death rate for epileptics (Persinger, 1995b), and increased suicide rates for males in Japan (Tada et al., 2014). These results and many others have been summarized for the reader in Table 1. General trends that emerge from the table are that complex human behaviours, such as out of body experiences (Persinger, 1995a) and vestibular experiences (Persinger & Richards, 1995) have non-linear relationships with geomagnetic storm intensity. This was something that was predicted by Dr. Persinger (1995a). Additionally, responses that are more simple, such as hormone levels (O'Connor & Persinger, 1996), seizures and deaths in epileptic rats (Bureau & Persinger, 1992; Persinger, 1995b), and analgesia in rats (Galic & Persinger, 2007) displayed linear relationships with geomagnetic storm intensity. Another consistent finding is that for most of these studies there was a threshold of ~20nT in deviation for the response to occur (Table 1).

Table 1. Correlational data of biological responses to geomagnetic disturbances.

Behaviour	Response	Index	Threshold	Trend	Timing of storm, related to event	Citation
Human						
Psychiatric hospital admissions	Increase	Ap			14-35 days before admission	Friedman, Becker & Bachman, 1963
Mood	Decreased mood in males	AA		Linear	1-3 days before measurement	Persinger, 1975a, Persinger & Levesque, 1983
Telepathic experiences	Increases during transient quiet periods	AA			Occurred on days of quiet activity that followed storm conditions (V-shaped)	Persinger, 1985b
Bereavement hallucinations	Increase in hallucinations during transient quiet periods	AA			Occurred on days of quiet activity that followed storm conditions (V-shaped)	Persinger, 1988
Telepathy in dream states	Accuracy increase during quiet geomagnetic periods	AA	20nT		Occurred on days of quiet activity that followed storm conditions (V-shaped)	Persinger & Krippner, 1989

Temporal lobe signs and depersonalization experiences	Increase in signs in young adult males	AA	30nT	Linear	Day after birth	Hodge & Persinger, 1991
Out of body experiences while being exposed to weak magnetic fields	Increase in out of body experiences in individuals with complex partial epileptic-like signs	AA	16-45nT	Non-linear	Same day	Persinger, 1995a
Sudden death in individuals with epilepsy	Increase	AA	50nT		Days that exceeded 50nT in one month	Persinger, 1995b
Vestibular experiences while being exposed to weak magnetic field	Increase in vestibular experiences	AA	15-20nT	Non-linear	Same day	Persinger & Richards, 1995
Hormone levels in complex partial epileptic female	Increase in thyroxine but not cortisol or prolactin	AA	20-25nT	Linear	Same day	O'Connor & Persinger, 1996
Sudden infant deaths	Increase	AA		Non-linear	Number of days per month with increased geomagnetic storm intensities	O'Connor & Persinger, 1997
Births	Males born during higher activity than females	AA			High activity 3 days before to day of	Persinger & Hodge, 1999

Likelihood of having an epileptic seizure in adults	Increased	AA		Linear	Within 1 day of birth	Persinger & O'Connor, 1999
Melatonin metabolite	Decreased	AA	20nT		1-2 days before measurement	Burch, Reif & Yost, 1999
Religious experiences beside open pit magnetite mine	Increase number of experiences after period of high activity	AA			Occurred on days of quiet activity that followed storm conditions (V-shaped)	Suess & Persinger, 2001
Plane crashes from pilot or electronic errors	Increased	AA			Same day	Fournier & Persinger, 2004
Hospital admissions for psychotic depression	Increase in males	AA			Peak 8-14 days following storm	Kay, 2004
Sensed presence when exposed to weak magnetic fields	Increase	Ap	15-20nT	Linear	Same day	Booth, Koren & Persinger, 2005
Human electroencephalographic activity	Decrease in gamma and theta power in right frontal lobe and increase in theta power in left and right temporal and parietal lobes	Atmospheric power		Linear	During the measurement	Mulligan, Hunter & Persinger, 2010

Accuracy of remote viewing	Decrease	Kp		Linear	During the task, or the variability in Kp for the day of	Scott & Persinger, 2013
Coherence between posterior temporal lobes	Increases	Kp	2	Non-linear	Time of measurement	Saroka et al., 2014
Suicide rates in Japan	Increase in males (Increase in suicides in males and females were also associated with decreased barometric pressure)	Kp			Used monthly mean K index	Tada et al., 2014
Hormone levels in Svalbard (one of the most northern cities)	Increase cortisol June and October; decreased T3 in June; no difference for T4	Kp and Ap			Day of measurement	Breus, Boiko & Zenchenko, 2015
Rat						
Day time activity on wheel	Increase	A			~ day before	Persinger, 1976
Mortality in epileptic rats	Increase	AA	20nT	Linear	1-2 days previous	Bureau & Persinger, 1992
Nocturnal ambulation	Decrease	AA		Linear	0-3 days before measurement	Bureau & Persinger, 1992

Latencies of seizure onset	Decrease	AA	20nT	Linear	Same day	Bureau & Persinger, 1995
Sudden death in epileptic rats	Increase	AA			Same day	Persinger, 1995b
Number of seizures in epileptic rats	Increase	AA	50nT		Same day and day before	Persinger, 1995b
Analgesia	Decrease	Ap	15-20nT	Linear	3 days before	Galic & Persinger, 2007
Planaria						
Group mortality	Increase	Kp	6		1 day before to same day	Murugan et al., 2015

These studies all show correlations between environmental electromagnetic exposure, in particular geomagnetic storms, and changes in behaviour. However, as with any correlational findings, one of the limitations is that correlation does not mean causation. When studying effects of different weather conditions, multiple weather variables will change simultaneously (Persinger, 1980), making it difficult to isolate the driving factors. That is why experimental manipulation is important to verify some of the relationships found above. Most importantly, in terms of geomagnetic field research, experiments with synthetic magnetic fields that were patterned to imitate a geomagnetic storm have been conducted many times (Persinger et al., 2005; Mulligan et al., 2012; Gang et al., 2013; Mekers et al., 2015). For example, increased mortality in epileptic rats was shown to occur after the incidence of geomagnetic storms with a threshold of 50nT, a trend also found in the death of humans with epilepsy (Bureau & Persinger, 1992; Persinger, 1995b). When a synthetic magnetic field patterned after geomagnetic storms was applied nocturnally to epileptics rats, there was a significant increase in mortality within 24 hours, most substantially when the field intensity was 50nT (Persinger et al., 2005). Increased death was also found in Daphnia magna after exposure to magnetic fields that were digitized recordings of a geomagnetic storm (Krylov et al., 2014). In another instance, exposure of rats to the same magnetic field pattern was used to produce a significant increase in the percent seizure display (Persinger, 1996; Michon & Persinger, 1997), supporting the identified correlational relationships (Persinger, 1995b). Another replicated finding was the change in electroencephalographic activity that predominantly occurred in the right frontal, temporal and parietal lobes theta activity (Mulligan et al., 2010). An experimentally applied geomagnetic storm field produced similar changes in right parietal theta activity (Mulligan & Persinger, 2012). This has also been shown for increase in the low frequency component of heart rate variability found in participants after exposure to a synthetic magnetic field patterned after a geomagnetic storm (Caswell et al., 2014), which replicated some of the relationships found in correlation with natural geomagnetic storms (Dimitrova et al., 2013; McCraty et al., 2017).

Biological effects of applied electromagnetic fields

The EMFs appear to be the most effective when they are designed to mimic the electrical activity associated with physiologic processes. For example, a magnetic field that was patterned after a burst-firing neuron in the limbic system (Richards et al., 1993) has been to shown to reduce clinical symptoms of depression in individuals who sustained closed head injuries (Baker-Price & Persinger, 1996) and reduce psychometric signs of depression in normal individuals (Corradini & Persinger, 2013). Exposure to this field pattern increases the pain threshold in rats (Martin et al., 2004) and acts on opioid receptors with similar effectiveness to morphine (Fleming et al., 1994; Murugan et al., 2014).

Potential role of endogenous EMFs

Bioelectricity refers to the cells ability to create its own, endogenous electrical current often by the movement of specific ions across the cell membrane. Bioelectricity is thought to be used by cells and tissues in processes such as communication within themselves and between each other. These endogenous fields exist not just around neurons and muscle cells, but are associated with epithelial cells as well (Levin, 2009), and is critical for wound repair in epithelial tissues (Zhao, 2009). Organs that contain cells that are specialized for bioelectric discharges can also generate their own electromagnetic fields. For example, the bioelectric discharges present in the heart (McCraty, 2015) and the brain (Xiang et al., 2009) are routinely measured using ECG/MCG (electrocardiography/magnetocardiography) or EEG/MEG (electroencephalography/magnetoencephalography), respectively. These electromagnetic fields may have a role in social interactions (Liboff, 2016; Liboff, 2017). McDonnell (2014) has posited that how humans interact with each other may in part be determined by unique electromagnetic patterns that each of us can generate. Liboff (2016) estimated that human brains can generate magnetic field intensity fluctuations of around 100nT. It has been demonstrated experimentally that an individual who practices meditation and Reiki can induce changes of up to ~12nT in the direction of the horizontal component of the Earth's field while imagining white light (Persinger et al., 2013).

How EMFs may influence experiments

Geomagnetic storms may behave as an additional magnetic field stimulus that can interact with the bioelectric field generated by activity of cells in the organism which would potentially alter the baseline state of the organism and consequently alter the behavioural responses being measured. This would consequently alter the response of the organism during the experiment. Alternatively, changing the background magnetic field intensity may alter the efficacy of an applied magnetic field (Persinger, 1985a). Every sensory modality has a Weber fraction (1),

$$(1) \qquad \text{Weber fraction} = \Delta S/S$$

Where, ΔS is the change in background intensity at which a stimulus can be detected and S is the absolute intensity of the background stimulus (Persinger, 1985a). The sensory modalities all have a ratio to describe their different levels of sensitivity; it has been proposed that a ratio for magnetic sensitivity could exist as well (Persinger, 1985a). Therefore, since geomagnetic storms alter the background intensity of the Earth's endogenous field they may alter an organism's sensitivity to magnetic fields. This may explain the results of an experiment which demonstrated differences in the effects of magnetic fields on plant growth, depending on whether geomagnetic storms occurred during the experiment (Rakosy-Tican et al., 2005). More evidence for this idea comes from Blackman et al. (1985) who found that altering the background intensity of the horizontal component of the geomagnetic field would predict if exposure to an alternating current magnetic field was able to induce calcium influx.

Individuals that are significantly influenced by geomagnetic storms are usually members of sensitive populations, either having anomalous cardiac or psychiatric behaviours (Freidman et al., 1963; Persinger, 1995b; Kay, 2004). This indicates that altered electrical lability in an individual is an important component in predicting their responsiveness to a geomagnetic storm. While organisms that are used in biological experiments, such as planaria, are all bred and fed in the same manner, the normal distribution still applies. Therefore, each unit would have a different level of sensitivity to geomagnetic storms. This means that they aren't all identical, and in experiments sometimes it won't be the whole population that produces a different response to geomagnetic storms, but only a portion of the population. In the statistical analysis, this effect may be obscured if all of the planaria are averaged together, and that's why it's important to also compute a variability score between organisms in a given condition and given trial, as that may be a more sensitive indicator of an effect.

References

Baker-Price, L. A., & Persinger, M. A. 1996. Weak, but complex pulsed magnetic fields may reduce depression following traumatic brain injury. Perceptual and Motor Skills, 83: 491-498.

Bartels, J., Heck, N. H., & Johnston, H. F. 1939., The three-hour-range index measuring geomagnetic activity, Terrestrial Magnetism and Atmospheric Electricity, 44(4), 411–454, doi: 10.1029/TE044i004p00411.

Blackman, C. F., Benane, S. G., Rabinowitz, J. R., House, D. E., & Joines, W. T. 1985. A Role for the magnetic field in the radiation-induced efflux of calcium ions from brain tissue in vitro. Bioelectromagnetics, 6: 327-337. doi:10.1002/bem.2250060402

Booth, J. N., Koren, S. A., & Persinger, M. A. 2005. Increased feelings of the sensed presence and increased geomagnetic activity at the time of the experience during exposures to transcerebral weak complex magnetic fields. International Journal of Neuroscience, 115:7, 1053-1079, DOI: 10.1080/00207450590901521

Breus, T. K., Boiko, E. R., & Zenchenko, T. A. 2015. Magnetic storms and variations in hormone levels among residents of North Polar area – Svalbard. Life Sciences in Space Research, 4:17-21.

Burch, J. B., Reif, J. S, & Yost, M. G. 1999. Geomagnetic disturbances are associated with reduced nocturnal excretion of a melatonin metabolite in humans, Neuroscience Letters, 266(3): 209-212.

Bureau, Y. R. J. & Persinger, M.A. 1992. Geomagnetic activity and enhanced mortality in rats with acute (epileptic) limbic lability. International Journal of Biometeorology, 36(4):226-232.

Bureau, Y. R. J. & Persinger, M.A. 1995. Decreased latencies for limbic seizures induced in rats by lithium-pilocarpine occur when daily average geomagnetic activity exceeds 20 nanoTesla. Neuroscience Letters, 192(2): (142-144). https://doi.org/10.1016/0304-3940(95)11624-6

Caswell, J. M. Singh, M., & Persinger, M. A. 2016. Simulated sudden increase in geomagnetic activity and its effect on heart rate variability: Experimental verification of correlation studies. Life Sciences in Space Research, 10:47-52.

Corradini, P. L., & Persinger, M. A. 2013. Brief cerebral applications of weak, physiologically-patterned magnetic fields decrease psychometric depression and increase frontal beta activity in normal subjects. Neurology & Neurophysiology, 4:175. doi:10.4172/2155-9562.1000175.

Dimitrova, S., Angelov, I., & Petrova, E. 2013. Solar and geomagnetic activity effects on heart rate variability. Natural Hazards, 69: 25. https://doi.org/10.1007/s11069-013-0686-y

Fleming, J. L., Persinger, M. A., & Koren, S. A. 1994. Magnetic pulses elevate nociceptive thresholds: Comparisons with opiate receptor compounds in normal and seizure-induced brain-damaged rats. Electro-and Magnetobiology, 13: 67-75. https://doi.org/10.3109/15368379409030699.

Fournier, N. M., & Persinger, M. A. 2004. Geophysical Variables and Behavior: C. Increased geomagnetic activity on days of commercial air crashes attributed to computer or pilot error but not mechanical failure. Perceptual and Motor Skills, 98(3_suppl), 1219–1224. https://doi.org/10.2466/pms.98.3c.1219-1224

Friedman, H., Becker, R. O., & Bachman, C. H. 1963. Geomagnetic parameters and psychiatric hospital admissions. Nature, 200:626-628.

Galic, M. A., & Persinger, M. A. 2007. Lagged association between geomagnetic activity and diminished nocturnal pain thresholds in mice. Bioelectromagnetics, 28: 577-579. doi:10.1002/bem.20353

Hodge, K. A., & Persinger, M. A. 1991. Quantitative increases in temporal lobe symptoms in human males are proportional to postnatal geomagnetic activity: verification by canonical correlation. Neuroscience Letters, 125(2): 205-208.

Jenrow, K. A., Smith, C. H., & Liboff, A. R. 1996. Weak extremely low frequency magnetic field-induced regeneration anomalies in the planarian *Dugesia tigrina*, Bioelectromagnetics, 17, 467-474.

Kay, R. 1994. Geomagnetic storms: Association with incidence of depression as measured by hospital admission. British Journal of Psychiatry, 164(3), 403-409. doi:10.1192/bjp.164.3.403

Krylov, V. V., Zotov, O. D., Klain, B. I., Ushakova, N. V., Kantserova, N. P.,Znobisheva, A. V., Izyumov, Y. G., Kuz'mina, V. V., Morozov, A. A., Lysenko, L. A., Nemova, N. N., & Osipova, E. A. 2014. An experimental study of the biological effects of geomagnetic disturbances: The impact of a typical geomagnetic storm and its constituents on plants and animals. Journal of Atmospheric and Solar-Terrestrial Physics. 110. 10.1016/j.jastp.2014.01.020.

Levin, M. 2009. Bioelectric mechanisms in regeneration: unique aspects and future perspectives. Seminars in Cell and Developmental Biology, 20(5): 543-556.

Liboff, A. R. 2013. Why are living things sensitive to weak magnetic fields? Electromagnetic Biology and Medicine, 33(3), 241-245. doi:10.3109/15368378.2013.809579

Liboff, A. R. 2016. A human source for ELF magnetic perturbations. Electromagnetic Biology and Medicine, 35:4, 337-342, DOI: 10.3109/15368378.2015.1107841

Liboff, A. R. 2017. The electromagnetic basis of social interactions. Electromagnetic biology and medicine, 36:1-5. 10.1080/15368378.2016.1241180.

Martin, L. J., Koren, S. A., & Persinger, M. A. 2004. Thermal analgesic effects from weak, complex magnetic fields and pharmacological interactions. Pharmacology, Biochemistry and Behaviour, 78(2): 217-227.

Mayaud, P.-N. 1972. The aa indices: A 100-year series characterizing the magnetic activity. Journal of Geophysical Research, 77(34), 6870–6874. doi:10.1029/ja077i034p06870

Mekers, W. F. T., Murugan, N. J., & Persinger, M. A. 2015. Introduction of planaria as a new model for multiple sclerosis research: Evidence from behavioural differences in cuprizone treated planaria exposed to patterned magnetic fields. Journal of Multiple Sclerosis (Foster City) 2:156. doi:10.4172/2376-0389.1000156

McCraty, R. 2015. Science of the heart: Exploring the role of the heart in human performance (Vol. 2). HeartMath. Boulder Creek, California.

McCraty, R., Atkinson, M., Stolc, V., Alabdulgader, A. A., Vainoras, A., & Ragulskis, M. 2017. Synchronization of human autonomic nervous system rhythms with geomagnetic activity in human subjects. International Journal of Environmental Research and Public Health 14, 770.

McDonnell, A. 2014. The sixth sense-emotional contagion; Review of biophysical mechanisms influencing information transfer in groups. Journal of Behavioral and Brain Science, 4:342-374.

Michon, A. L., & Persinger, M. A. 1997. Experimental simulation of the effects of increased geomagnetic activity upon nocturnal seizures in epileptic rats. Neuroscience Letters, 224(1): 53-56.

Mulligan, B. P., Hunter, M. D., & Persinger, M. A. 2010. Effects of geomagnetic activity and atmospheric power variations on quantitative measures of brain activity: Replication of the Azerbaijani studies. Advanced Space Research, 45:940-948

Mulligan, B. P., & Persinger, M. A. 2012. Experimental simulation of the effects of sudden increases in geomagnetic activity upon quantitative measures of human brain activity: Validation of correlational studies. Neuroscience Letters, 516(1):54-56.

Murugan, N. J., Karbowski, L. M., & Persinger, M. A. 2014. Weak burst-firing magnetic fields that produce analgesia equivalent to morphine do not initiate activation of proliferation pathways in human breast cancer cells in culture. Integrative Cancer Science and Therapeutics, 1(3): 47-50.

Murugan, N. J., Karbowski, L. M., Mekers, W. F., & Persinger, M. A. 2015. Group planarian sudden mortality: Is the threshold around global geomagnetic activity ≥K6?. Communicative & integrative biology, 8(6), e1095413. doi:10.1080/19420889.2015.1095413

O'Connor, R. P., & Persinger, M. A. 1996. Brief communication: Increases in geomagnetic activity are associated with increases in thyroxine levels in a single patient: Implications for melatonin levels. International Journal of Neuroscience, 88: 243-247.

O'Connor, R. P., & Persinger, M. A. 1997. Geophysical Variables and Behavior: LXXXII. A strong association between sudden infant death syndrome and increments of global geomagnetic activity—possible support for the melatonin hypothesis. Perceptual and Motor Skills, 84(2): 395–402. https://doi.org/10.2466/pms.1997.84.2.395

Persinger, M. A. 1975a. Lag responses in mood reports to changes in the weather matrix. International Journal of Biometeorology, 19(2):108-114.

Persinger, M. A. 1975b. Geophysical models for parapsychological experiences. Psychoenergetic Systems, 1(2), 63-74.

Persinger, M.A. 1976. Day time wheel running activity in laboratory rats following geomagnetic event of 5–6 July 1974. International Journal of Biometeorology, 20: 19. https://doi.org/10.1007/BF01553167.

Persinger, M. A. 1980. The Weather Matrix and Human Behaviour. Praeger Publishers. New York, New York.

Persinger, M. A., & Levesque, B. F. 1983. Geophysical Variables and Behavior: XII. The weather matrix accommodates large portions of variance of measured daily mood. Perceptual and Motor Skills, 57(3), 868–870. https://doi.org/10.2466/pms.1983.57.3.868

Persinger, M. A. 1985a. Letter: Classical psychophysics and ELF magnetic field detection. Journal of Bioelectricity, 4:2, 577-584, DOI: 10.3109/15368378509033272

Persinger, M. A. 1985b. Geophysical variables and behavior: XXX. Intense paranormal experiences occur during days of quiet, global, geomagnetic activity. Perceptual and Motor Skills, 61(1), 320-322

Persinger, M. A. 1988. Increased geomagnetic activity and the occurrence of bereavement hallucinations: Evidence for melatonin-mediated microseizuring in the temporal lobe? Neuroscience Letters, 88(3):271-274

Persinger, M. A., & Krippner, S. 1989. Dream ESP experiments and geomagnetic activity. Journal of the American Society for Psychical Research, 83(2), 101-116.

Persinger, M. A., & Richards, P. M. 1995. Vestibular experiences of humans during brief periods of partial sensory deprivation are enhanced when daily geomagnetic activity exceeds 15-20 nT. Neuroscience Letters, 194:69-72.

Persinger, M. A. 1995a. Out-of-body-like experiences are more probable in people with elevated complex partial epileptic-like signs during periods of enhanced geomagnetic activity: a nonlinear effect. Perceptual and Motor Skills, 80(2):563-569. doi: 10.2466/pms.1995.80.2.563.

Persinger, M. A. 1995b. Sudden unexpected death in epileptics following sudden, intense, increases in geomagnetic activity: Prevalence of effect and potential mechanisms. International Journal of Biometeorology, 38: 180. https://doi.org/10.1007/BF01245386.

Persinger, M.A. 1996. Enhancement of limbic seizures by nocturnal application of experimental magnetic fields that simulate the magnitude and morphology of increases in geomagnetic activity. The International Journal of Neuroscience, 86 (3-4): 271-80.

Persinger, M. A., & Hodge, K. A. 1999. Geophysical Variables and Behavior: LXXXVI. Geomagnetic activity as a partial parturitional trigger—are male babies more affected than female babies? Perceptual and Motor Skills, 88(3): 1177–1180. https://doi.org/10.2466/pms.1999.88.3c.1177

Persinger, M.A., McKay, B.E., O'Donovan, C.A., & Koren, S. A. 2005. Sudden death in epileptic rats exposed to nocturnal magnetic fields that simulate the shape and the intensity of sudden changes in geomagnetic activity: An experiment in response to Schnabel, Beblo and May. International Journal of Biometeorology, 49: 256. https://doi.org/10.1007/s00484-004-0234-2

Persinger, M.A., Dotta, B.T., Saroka, K.S., & Scott, M.A. 2013. Congruence of energies for cerebral photon emissions, quantitative EEG activities and ~ 5 nT changes in the proximal geomagnetic field support spin-based hypothesis of consciousness. Journal of Consciousness Exploration and Research. 4. 1-24.

Press, F., & Siever, R. 1974. Earth (J. Gilluly & A. O. Woodford, Eds.). San Francisco, California: W. H. Freeman and Company.

Rakosy-Tican, L., Aurori, C., & Morariu, V. 2005. Influence of near null magnetic field on in vitro growth of potato and wild Solanum species. Bioelectromagnetics, 26: 548-557. doi:10.1002/bem.20134

Richards, P. M., Persinger, M. A., & Koren, S. A. 1993. Modification of activation and evaluation properties of narratives by weak complex magnetic field patterns that simulate limbic burst firing. International Journal of Neuroscience, 71: 71-85.

Rostoker, G. 1972. Geomagnetic indices. Review of Geophysics, 10(4), 935–950, doi: 10.1029/RG010i004p00935.

Saroka, K. S., Caswell, J. M., Lapointe, A., & Persinger, M. A. 2014. Greater electroencephalographic coherence between left and right temporal lobe structures during increased geomagnetic activity. Neuroscience Letters, 560: 126-130

Sears, F. W., & Zemansky, M. W. 1964. University Physics (3rd ed.). Reading, Massachusetts:Addison-Wesley.

Suess, L. A., & Persinger, M. A. 2001. Geophysical Variables and Behavior: XCVI. "Experiences" attributed to Christ and Mary at Marmora, Ontario, Canada may have been consequences of environmental electromagnetic stimulation: Implications for religious movements. Perceptual and Motor Skills, 93(2), 435–450. https://doi.org/10.2466/pms.2001.93.2.435

Tada, H., Nishimura, T., Nakatani, E., Matsuda, K., Teramukai, S., & Fukushima, M. 2014. Association of geomagnetic disturbances and suicides in Japan, 1999-2010. Environmental health and preventive medicine, 19(1), 64-71.

Xiang, J., Liu, Y., Wang, Y., Kotecha, R., Kirtman, E. G., Chen, Y., Huo, X., Fujiwara, H., Hemasilpin, N., deGrauw, T., & Rose, D. 2009. Neuromagnetic correlates of developmental changes in endogenous high-frequency brain oscillations in children: A wavelet-based beamformer study. Brain Research, 1274, 28-39. doi:10.1016/j.brainres.2009.03.068

Zhao, M. 2009. Electrical fields in wound healing-an overriding signal that directs cell migration. Seminars in Cell & Developmental Biology, 20:674-682.

Chapter 2- Seasonal and lunar variability in planaria mobility, and influences from natural fluctuations in the Earth's geomagnetic field

Abstract

There are many behaviours that show seasonal and lunar cycles that are evolutionarily advantageous for the organism. Occasionally when organisms are kept in controlled conditions, they maintain these cycles, indicating the potential existence of endogenous rhythms, or a sensitivity to environmental factors that are not yet known and can penetrate buildings. Seasonal and lunar cyclicity has been demonstrated in planaria (*Dugesia tigrina*). A yearlong experiment was conducted where weekly measurements were made of planaria mobility to investigate seasonal or lunar cycles, as well as any sensitivity to geomagnetic storms. A statistical interaction of season by lunar phase interaction was found, where planaria showed significantly more movement when there was a new moon but only in one half of the Earth's orbit. The change in planaria behaviour was associated with a change in background photon counts, a possible cosmic source is discussed. It was also found that the occurrence of geomagnetic storms were associated with outlier behaviour in some of the planaria. The outlier behaviour is similar to what's been demonstrated in sensitive human populations, indicating there may be common mechanisms.

Introduction

Cycles are pervasive in the behaviours of biological organisms (Gauquelin, 1967). Cycles can occur over a period of hours, termed ultradian, such as hunger (Wuorinen & Borer, 2013). Circatidal rhythms are behaviours that alternate with the tides (about 12 hours long), such as the mobility of the mangrove cricket (Takekata et al., 2014). Cycles that occur over a period of one day are termed circadian, one of the simplest examples being the human sleep cycle (Mongrain et al., 2004). There are also lunar cycles, such as the spawning of the grunion (Walker, 1949; Carson, 1950; Gauquelin, 1967). Circannual rhythms are behaviours that occur once a year, such as mating in deer (Gaspar-López et al., 2010), or that show annual variability such as hair growth and thyroid activity (Gauquelin, 1971).

Studies have demonstrated that when placed in a light- and temperature-controlled environment, biological organisms can maintain cyclicity in their behaviour. Some studies have concluded this as proof that these cycles are regulated by endogenous mechanisms (Gwinner & Dittami, 1990), while others concluded they are being driven by exogenous variables (Brown et al., 1955; Spruyt, Verbelen, & De Greef, 1987; Moraes et al., 2012). Either is possible and both may contribute to these effects.

Frank Brown Junior demonstrated that planaria could orient themselves according to the Earth’s static magnetic field but that this behaviour was sensitive to the lunar phase, except during the summer months (Brown, 1962). He also demonstrated that during the summer months planaria displayed a peak in sensitivity to weak gamma radiation (Brown, 1963). In the wild, planaria have been observed to be most abundant when the water temperatures range from 13-25°C and are sparse or can no longer be found during the winter months, presumably because of the temperature drop (Stokely et al., 1965). In addition to the obvious temperature changes, there are also yearly variations in atmospheric electricity with minimum intensities in the summer months (Persinger, 1980) and circannual variations in geomagnetic storms (Rostoker, 1972; Persinger, 1980).

Planaria are small fresh water flat worms from the phylum Platyhelminthes. Their central nervous system consists of bilateral cephalic ganglia (joined by an anterior commissure) and ventral nerve cords (Marsal et al., 2003). There are two ways in which planaria can move. The first is by using cilia present along the length of their bodies which they use to glide along a surface (Nishimura et al. 2007). If these cilia are removed, the planaria can then use their longitudinal muscles to inch along a surface (Nishimura et al., 2007; Nishimura et al., 2011).

This experiment was designed to evaluate any change in planaria mobility behaviour that occurred across the different seasons that might parallel what has been observed previously (Brown, 1962; Stokely et al., 1965). Lunar phases and geomagnetic activity were included because of the research that has shown planaria to be sensitive to these variables (Brown, 1962; Mulligan et al., 2012; Gang et al., 2013; Murugan et al., 2015). It begun with an analysis on planaria that were in control conditions from a variety of previous experiments, which indicated that the current experiment was warranted.

Methods

Planaria

Brown planaria (*Dugesia tigrina*) were obtained from Boreal Biological Supplies. They were acclimatized to lab conditions and housed in President's Choice spring water at 4°C. Planaria were fed calf liver once a week and were not used within three days of being fed.

Measurement

Three to five planaria were observed once a week in an open field paradigm. First, they were transferred from their housing container kept in the fridge to a petri dish and allowed to acclimate to room temperature for ~15 minutes before they were observed consecutively in the open field. The open field consisted of a 10 cm petri dish containing 20 mL of spring water, placed on top of a piece of 0.5 cm grid paper. The planaria were allowed 5 minutes to roam freely in the dish and the number of gridlines that they crossed were counted. Data was collected weekly beginning on February 5, 2017 and finished February 4, 2018; there were a total of 5 weeks during this time when there was no data collected.

Statistical Analysis

The mean and standard deviation (SD) of the number of gridlines crossed were determined for all of the planaria tested for each day of observation. When investigating seasonal and lunar cycle effects, each week represent one sample in the analysis. Another analysis was completed on the occurrence of planaria that were outliers. These outliers were determined by computing a z-score for each planaria in the experiment, any planaria with z-scores greater than 2 were considered outliers. The weeks were then coded as containing an outlier or not, so that again each week represented one sample in the analysis. Geomagnetic storm activity was quantified using the Ap index, a linear measures of the disturbance and has base units of nanoTesla (nT) (Rostoker, 1972). All statistical analyses were completed with SPSS.

Results

Statistical analysis revealed that planaria movement was similar in winter and spring so measurements in those seasons were grouped into one condition and fall and summer measurements were grouped into another. The moon phases were also grouped, where measurements during the new moon and third quarter were grouped together and measurements during the first quarter and full moon were grouped together. A two-way ANOVA determined a significant interaction between the grouped seasons and grouped moon phases for the mean number of gridlines crossed [$F(1,46)=6.19$, $p=0.017$; Figure 1], but not for the SD between the planaria observed on one day ($p>0.05$). In the post-hoc analysis the alpha cut-off was raised to 0.10 in order to show the group differences that were driving the interaction. Tukey's post-hoc showed that the new moon/third quarter planaria in the fall/summer moved 1.8 fold more than the first quarter/full moon planaria in the same season group ($p=0.055$) and also moved 1.75 fold more than the new moon/third quarter planaria in the winter/spring ($p=0.073$).

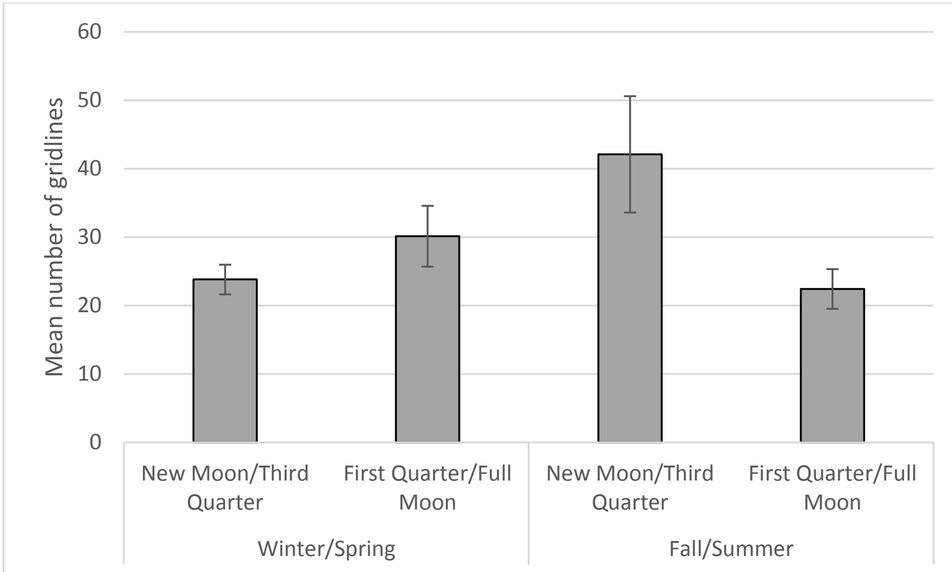


Figure 1. Average number of gridlines crossed by planaria. Error bars represent the standard error of the mean (SEM).

For the analysis on the presence of outliers the geomagnetic storm index values, represented by the AP-index for the day of observation and the 6 preceding and succeeding days were entered into the database. A discriminant analysis that used the geomagnetic data was able to discriminate between the days of observation with an outlier (N=9) and days without an outlier (N=38). Variables that entered were the AP indices from 4 days before the day of observation and 4 days after the day of observation [Wilk's $\Lambda=0.676$, $X^2(2)=17.2$, $p<0.001$]. The function had a canonical correlation of 0.569 and explained 76.6% of the original cases, and 74.5% of the cross validated cases. To determine what the differences were between the AP values between the outlier vs. non-outlier days, a within-subjects MANOVA was used on just these two days with the presence of an outlier as the independent variable. A significant between subjects effect was found [$F(1,45)=21.3$, $p<0.001$; partial eta squared=0.32; Figure 2] with no significant within subject effects (p>0.05). A oneway ANOVA determined that both 4 days pre and 4 days post were elevated when there was an outlier vs when there was not [$F(1,46)=9.45$, $p=0.004$ and $F(1,46)=7.94$, $p=0.007$, respectively].

If fluctuations in geomagnetic intensity can increase the incidence of outliers, this suggests that exposure to geomagnetic fluctuations can promote large variations in movement for individual planaria. The presence of these outliers would contribute to the overall means and may have influenced the season- and moon- phase interaction presented above. Therefore, that analysis was repeated using the AP-index as a covariate. The AP-index from 4 days before measurement was a significant covariate [$F(1,46)=4.25$, $p=0.045$, $B=0.582$] as was the AP-index from 4 days after the measurement [$F(1,46)=7.46$, $p=0.009$, $B=0.664$], however neither removed the significance of the original interaction.

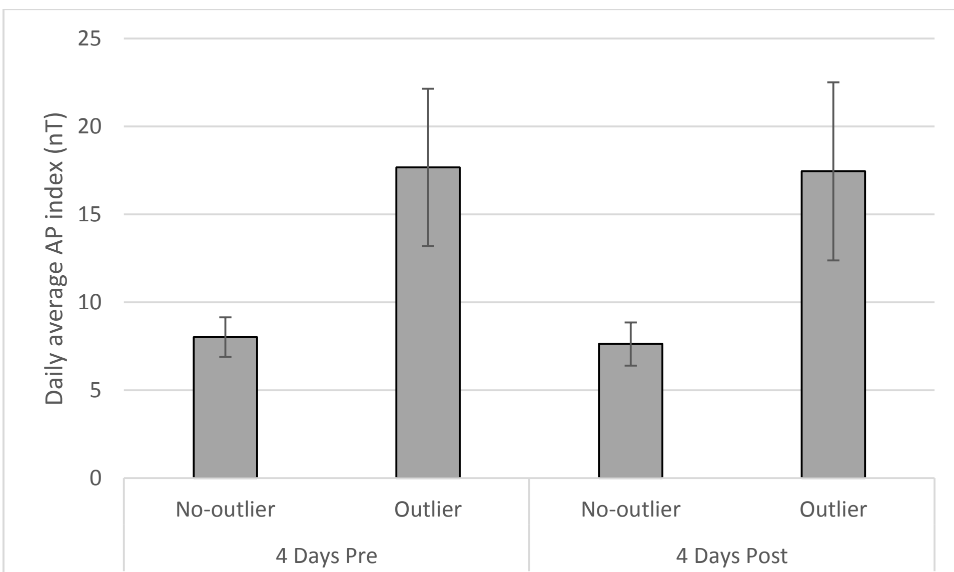


Figure 2. Intensity of daily average AP index (nT) 4 days before and 4 days after the day of observation. Outlier refers to weeks that had a planaria that moved more than 2

standard deviations above the grand mean. There were 9 weeks with an outlier and 38 weeks with none. Error bars represent SEM.

Discussion

Planaria mobility showed an interaction between moon phase and season of observation. This was only apparent when the data recorded in the winter and spring were grouped into one variable and the data recorded in the summer and the fall were grouped into one variable. Additionally, the data recorded in the new moon and third quarter were grouped together, as was the data recorded during the full moon and first quarter. For example, during the new moon/third quarter in the fall/spring is that the sun and moon are both on the same side as the Earth. An image was created to demonstrate the arrangement of the Sun, Moon and Earth during the group that was driving the interaction (Figure 3). The interaction was being driven by the planaria movement during the fall/summer (June 21st – December 20th), in one half of the Earth's orbit (Figure 3). The interaction with lunar phase was not seen in the other half of the Earth's orbit (December 21st – June 20th). This indicates that there was something unique occurring when Earth was traveling between June 21st and December 20th. The interaction of planaria movement with seasonal and lunar position suggests a possible astronomical influence.

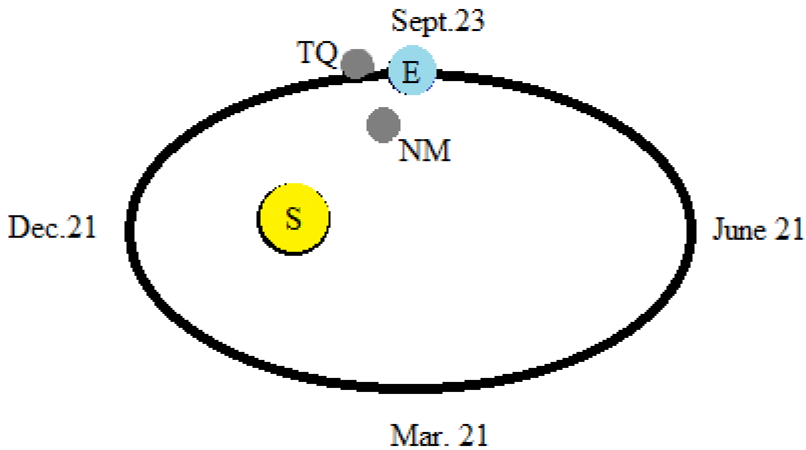


Figure 3. Schematic of approximate arrangement of Sun, Earth and Moon during highest gridline counts of planaria. S=Sun, E=Earth, NM=New Moon and TQ=Third Quarter. The galactic center would be on the same side of the Sun as the Earth is in this picture. This view is looking down at the solar system, where the Earth is moving counter-clockwise around the Sun and the Moon is moving clockwise around the Earth.

Dr. Persinger (2015) demonstrated a reliable change in background photon counts over the course of a year that showed peak numbers when the Earth was closer to the galactic center (at Fall Equinox, September 22) and annual lows when the Earth was farthest away (at Spring Equinox, March 20). He found it to result in a change of 10^{-12} W/m^2, which corresponded to double the number of photons (Persinger, 2015). One source of electromagnetic radiation from the galactic center is gamma radiation (van Eldik, 2015). Frank Brown Junior (1964) has demonstrated that planaria are sensitive to weak intensity gamma radiation and that this sensitivity is variable over the course of the year. However, when gamma rays come into contact with the atmosphere, they breakdown into an air shower of electrons and positrons, some of which will produce Cherenkov light, which is predominantly in the UV and blue light spectrum (Hinton, 2011;

van Eldik, 2015). These wavelengths of light fall into the wavelength sensitivity range of the photomultiplier tube (280 to 850nm) used by Dr. Persinger (2015). Additionally, studies have shown that when exposed to UV and blue light, planaria display an increase in movement not found with other wavelengths (Paskin et al., 2014; Murugan PhD thesis, 2017). The change in planaria behaviour seems to have been influenced by the circannual variation in background photon counts as related to Earth's distance from the galactic center (Persinger, 2015).

The analysis of the incidence of statistical outliers in planaria movement showed that on the days when the outliers were present there was a greater amount of geomagnetic storm activity before and after the day of observation, with intensities in the range of 15-20nT. This suggests that some planaria have the potential to be outliers but that it was the storm activity that precipitated this behaviour. Increased planaria locomotion has been demonstrated after being exposed to a magnetic field patterned after a geomagnetic storm (Gang et al., 2013).

These results demonstrate that there is some process that contributes to the planaria's mobility that is sensitive to geomagnetic storms and that this sensitivity is only present in a small proportion of a treatment-naïve population. In humans, geomagnetic storms have been associated with abnormal behaviours such as increased psychotic episodes (Freidman et al., 1963), decreases in mood scores of males (Persinger, 1975), psychotic depression in males (Kay, 1994), increased vestibular experiences (Persinger & Richards, 1995), increased out of body experiences (Persinger, 1995a), increased deaths in epileptics (Persinger, 1995b), and increased suicide rates of males in Japan (Tada et al., 2013). While planaria mobility is not as complex as the indicated human behaviours, the fact that they all occur in association with increased geomagnetic storms could mean that they are all derived from similar mechanisms.

References

Barata, B. C., Chendo, I., Salta, M., Caixas de Sousa, R., Mendes, S. H., Ribeiro, R., Ribeiro, B., & Gamito, A. O. 2015. TTe sun, the moon and the mood: Seasonal associations between the lunar cycle and acute manic states. European Psychiatry, 30(Supplement 1), 563-563. doi:10.1016/S0924-9338(15)30446-6

Brown, F. A., Webb, H. M., & Bennett, M. F. 1955. Proof for an endogenous component in persistent solar and lunar rhythmicity in organisms. Proceedings of the National Academy of Sciences of the United States of America, 41(2), 93-100.

Brown, F. A. 1962. Responses of the planarian, *Dugesia*, and the protozoan, *Paramecium*, to very weak horizontal magnetic fields. *The Biological Bulletin*, 123(2): 264-281.

Brown, F. A. 1963. An orientational response to weak gamma radiation. *The Biological Bulletin*, 125(2):206-225.

Brown, F. A, & Park, Y. H. 1965. Phase-shifting a lunar rhythm in planarians by altering the horizontal magnetic vector. *The Biological Bulletin*, 129(1):79-86.

Carson, R. L. 1950. The Sea Around Us. Oxford University Press. New York, New York.

Friedman, H., Becker, R. O., & Bachman, C. H. 1963. Geomagnetic parameters and psychiatric hospital admissions. Nature, 200:626-628.

Gang, N., Parker, G. H., Lafrenie, R. M., & Persinger M. A. 2013. Intermittent exposures to nanoTesla range, 7 Hz, amplitude-modulated magnetic fields increase regeneration rates in planarian, International Journal of Radiation Biology, 89:5, 384-389, DOI: 10.3109/09553002.2013.754554

Gaspar-López, E., Landete-Castillejos, T., Estevez, J., Ceacero, F., Gallego, L., & García, A. 2010. Biometrics, testosterone, cortisol and antler growth cycle in iberian red deer stags (cervus elaphus hispanicus). Reproduction in Domestic Animals, 45(2), 243-249. doi:10.1111/j.1439-0531.2008.01271.x

Gauquelin, M. 1967. The Cosmic Clocks. Avon. New York, New York.

Gauquelin, M. 1971. How atmospheric conditions affect your health. New York: Stein and Day.

Gwinner, E., & Dittami, J. 1990. Endogenous reproductive rhythms in a tropical bird. Science, 249(4971): 906-908. doi: 10.1126/science.249.4971.906.

Kay, R. 1994. Geomagnetic storms: Association with incidence of depression as measured by hospital admission. British Journal of Psychiatry, 164(3), 403-409. doi:10.1192/bjp.164.3.403

Mongrain, V., Lavoie, S., Selmaoui, B., Paquet, J., & Dumont, M. 2004. Phase relationships between sleep-wake cycle and underlying circadian rhythms in morningness-eveningness. Journal of Biological Rhythms, 19(3), 248-257. doi:10.1177/0748730404264365

Moraes, T. A., Barlow, P. W., Klingelé, E., & Gallep, C. M. 2012. Spontaneous ultra-weak light emissions from wheat seedlings are rhythmic and synchronized with the time profile of the local gravimetric tide. Die Naturwissenschaften. 99. 465-72. 10.1007/s00114-012-0921-5.

Muñoz-Hoyos, A., Sánchez-Forte, M., Molina-Carballo, A., Escames, G., Martin-Medina, E., Reiter, R. J., Molina-Font, J. A., & Acuña-Castroviejo, D. 1998. Melatonin's role as an anticonvulsant and neuronal protector: Experimental and clinical evidence. Journal of Child Neurology, 13(10):501-509. https://doi.org/10.1177/088307389801301007

Mulligan, B., Gang, N., Parker G. H., & Persinger, M. A. 2012. Magnetic field intensity/melatonin-molarity interactions: Experimental support with planarian (Dugesia sp.) activity for a resonance-like process, Open Journal of Biophysics, 2(4): 137-143. doi: 10.4236/ojbiphy.2012.24017

Murugan, N. J., Karbowski, L. M., Mekers, W. F., & Persinger, M. A. 2015. Group planarian sudden mortality: Is the threshold around global geomagnetic activity ≥K6?. Communicative & integrative biology, 8(6), e1095413. doi:10.1080/19420889.2015.1095413

Murugan, N. M. 2017. The emission and application of patterned electromagnetic energy on biological systems. Doctor of Philosophy in Biomolecular Sciences, Laurentian University, Sudbury, Ontario.

Persinger, M. A. 1975. Lag responses in mood reports to changes in the weather matrix. International Journal of Biometeorology, 19(2):108-114.

Persinger, M. A. 1980. The Weather Matrix and Human Behaviour. Praeger Publishers. New York, New York.

Persinger, M. A., & Richards, P. M. 1995. Vestibular experiences of humans during brief periods of partial sensory deprivation are enhanced when daily geomagnetic activity exceeds 15-20 nT. Neuroscience Letters, 194:69-72.

Persinger, M. A. 1995a. Out-of-body-like experiences are more probable in people with elevated complex partial epileptic-like signs during periods of enhanced geomagnetic activity: a nonlinear effect. Perceptual and Motor Skills, 80(2):563-569. doi: 10.2466/pms.1995.80.2.563.

Persinger, M. A. 1995b. Sudden unexpected death in epileptics following sudden, intense, increases in geomagnetic activity: Prevalence of effect and potential mechanisms. International Journal of Biometeorology, 38: 180. https://doi.org/10.1007/BF01245386.

Persinger, M. A. 2015. Annual fluctuations (~10-12 W•m-2) in ground level photon power densities: Quantitative evidence for possible modulation from the galactic center. International Journal of Astrophysics and Space Science, 3(5): 70-73. doi: 10.11648/j.ijass.20150305.11

Rostoker, G. 1972. Geomagnetic indices. Review of Geophysics, 10(4), 935–950, doi: 10.1029/RG010i004p00935.

Spruyt, E., Verbelen, J., & De Greef, J. 1987. Expression of Circaseptan and Circannual Rhythmicity in the Imbibition of Dry Stored Bean Seeds. Plant Physiology, 84(3), 707-710.

Stokely, P. S., Brown, T. S., Kuchan, F., & Slaga, T. J. 1965. The distribution of fresh-water triclad planarians in Jefferson County, Ohio. The Ohio Journal of Science, 65(6):305-318

Tada, H., Nishimura, T., Nakatani, E., Matsuda, K., Teramukai, S., & Fukushima, M. 2014. Association of geomagnetic disturbances and suicides in Japan, 1999-2010. Environmental health and preventive medicine, 19(1), 64-71.

Takekata, H., Numata, H., & Shiga, S. 2014. The circatidal rhythm persists without the optic lobe in the mangrove cricket *Apteronemobius asahinai*. Journal of Biological Rhythms, 29(1), 28-37. doi:10.1177/0748730413516309

van Eldik, C. 2015. Gamma rays from the Galactic Centre region: A review. Astroparticle Physics, 71:45-70. https://doi.org/10.1016/j.astropartphys.2015.05.002.

Walker, B. W. 1949. Periodicity of spawning by the grunion, *Louresthes tenuis*, an atherine fish. Doctor of Philosophy in Zoology, University of California, Los Angeles.

Wehr, T. A. 2018. Bipolar mood cycles and lunar tidal cycles. Molecular Psychiatry, 23:923-931.

Wuorinen, E. C., & Borer, K. T. 2013. Circadian and ultradian components of hunger in human non-homeostatic meal-to-meal eating. Physiology & Behavior, 122(Complete), 8-16. doi:10.1016/j.physbeh.2013.08.001

Chapter 3: Sensitivity of planaria to weak, patterned electric current and the subsequent interactions with fluctuations in the intensity of Earth's magnetic field

Abstract

Some species of fish show highly evolved mechanisms by which they can detect exogenous electric and magnetic fields. The detection of electromagnetic fields has been hypothesized to exist in humans, despite the lack of specialized sensors. In this experiment planaria were tested in a t-maze with weak electric current pulsed in one arm to determine if the planaria showed any indication of being able to detect it. It was found that a small proportion of the population seemed to be attracted to this current. Additionally, if the experiment was preceded by a geomagnetic storm, the planaria showed a linear increase in the variability of their movement in response to the presence of the weak electric field. Both of these results indicate that a subpopulation of planaria show some ability to respond to electric or magnetic fields.

Introduction

There are several species of fish that have specialized cells for detecting electric fields in their environment (Meyer et al., 2004; Salazar et al., 2013; Bellono et al., 2018). Most fish capable of eletro- or magnetoreception do so by producing a weak electromagnetic field of their own and detect fluctuations and disturbances of this endogenously produced field (Salazar et al., 2013). The electroreception receptors involved operate through well studied mechanisms, involving low voltage L-type calcium (Cav1.3) and potassium ion channels (Bellono et al., 2018). The Cav1.3 channels are present on a wide variety of other cells, not traditionally thought to have the capacity of electroreception, including dopamine secreting cells in the brain (Putzier et al., 2009; Liu et al., 2014) and cochlear cells in the ear canal responsible for hearing (Chen et al., 2012). While mammals don't live in a medium as electrically conductive as water, air is able to efficiently conduct magnetic fields (Persinger, 1980). Additionally, electromagnetic fields are generated by organs whose cells communicate with each other with bioelectric discharges caused by the movement of ions across the cell membrane, such as in the heart (McCraty, 2015) and in the brain (Xiang et al., 2009). Some experiments have indicated the potential for reception of disturbances of the Earth's magnetic or electric field. For example, the application of a magnetic field with the same intensity as the Earth's was used to classically condition sharks (Meyer et al., 2004). This detection may occur in mammals even if it does not enter conscious awareness or is attributed to a different cause. This potential interaction between environmental electromagnetic fields has already been demonstrated in the human brain, where the pattern of its electrical activity showed transient periods of coherence with the Earth's magnetic field (Pobachenko et al., 2006; Saroka et al., 2016).

Planaria are small, fresh water flatworms that have central nervous systems containing all of the classical neurotransmitters that are found in humans (Ribiero et al., 2005). Planaria have dopamine-synthesizing cells in their anterior regions that are critical for their mobility (Nishimura et al., 2007). They also have endogenous melatonin that shows a similar diurnal variation to humans (Itoh et al., 1999). Past research has shown interactions between exogenous melatonin and exposure to magnetic fields that imitate geomagnetic storms (Mulligan et al., 2012).

In the 1960's Frank Brown Junior demonstrated that planaria could reliably re-orientate themselves relative to the North-South compass direction, indicating they may be able to detect the Earth's static magnetic field (Brown, 1962; Brown & Park, 1965). The present experiment aimed to investigate electroreception in planarian flatworms. We did this by placing the planaria in a maze that had an electric field in one arm of the maze, and then observing the planaria's movement for indications of a preference – or avoidance – towards any specific arm.

Methods

Planaria

Planaria (*Dugesia tigrina*) were housed at 4 degrees Celsius in a refrigerator. They were fed calf liver weekly. Before any experimentation began they were given 10 minutes to acclimatize to room temperature. They were kept at all times in President's Choice spring water.

Electric field application

The electric field was generated by a homemade system consisting of a SainSmart ATMega2560 microcontroller that interfaced with a circuit built onto a breadboard that consisted of a diode, transistor, and 20kΩ resistor. The breadboard also had an R2R resistor ladder which allowed digital to analog conversion. The digital input originated from a Gateway laptop (model: NV53A) computer. From this laptop, Arduino software programming was used to construct the parameters of the field. The same laptop was used for the entire experiment.

The field used was patterned after a magnetic field called Thomas, patterned after a chirp in a communication system (Murugan & Persinger, 2014) (Figure 4). This pattern was created through the Arduino- microcontroller output system that would translate points ranging from 0-256 to a potential difference between +/ – 5V. The duration of each point was set to 3 msec (milliseconds) and the delay between the end of one pattern and the beginning of another was also set to 3 msec. The field was turned on before the planaria were placed in the maze or was left off for the entire duration. The latter group were considered as the sham condition.

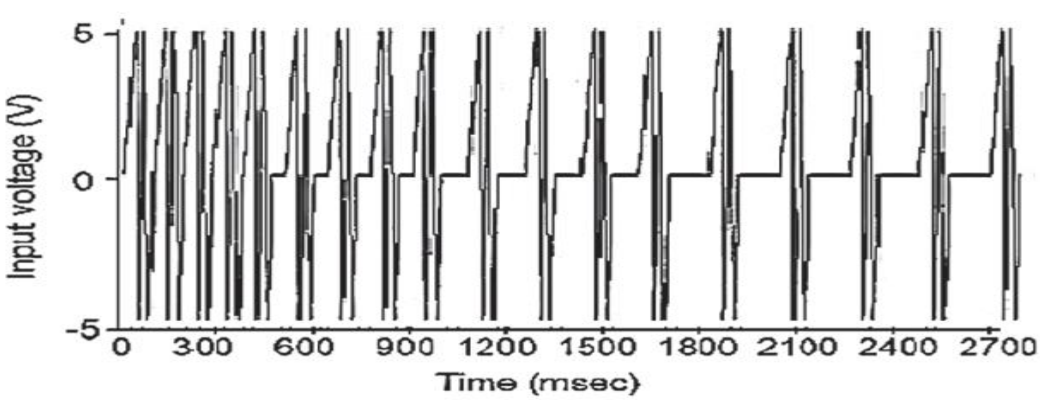


Figure 4. Thomas pattern, modeled after chirp features of a communication system (Murugan & Persinger, 2014).

A picture of the t-maze used can be seen in Figure 5. It was made from a rectangular plastic dish filled with paraffin wax, which had a "T" shape mould (credit: Dr. Nirosha Murugan). A drawing of the t-maze is found in Figure 6. A pair of electrodes (one positive and one negative) were placed opposite one another on Line B, in either Arm 1 or Arm 2. The electrodes were separated by about 1 cm and were able to form a circuit through the spring water.

Behavioural Measures

The planaria were placed in the bottom of the starting arm and allowed to roam freely in the maze for 5 minutes. The movement of the planaria were recorded for the amount of time it took them to leave the starting arm (cross Line C in Figure 6), which arms in the maze they went into, the amount of time it took for them to cross into each arm (cross either of the Line A's in Figure 6), and the total number of arms they visited. A planaria was considered to have crossed into an arm when its full body had crossed over the line.

Figure 5. A picture of the t-maze that was used for this experiment. It was created by filling a plastic dish with paraffin wax and molding the "T" shape into this. During testing it was filled with ~ 7mL of President's Choice spring water.

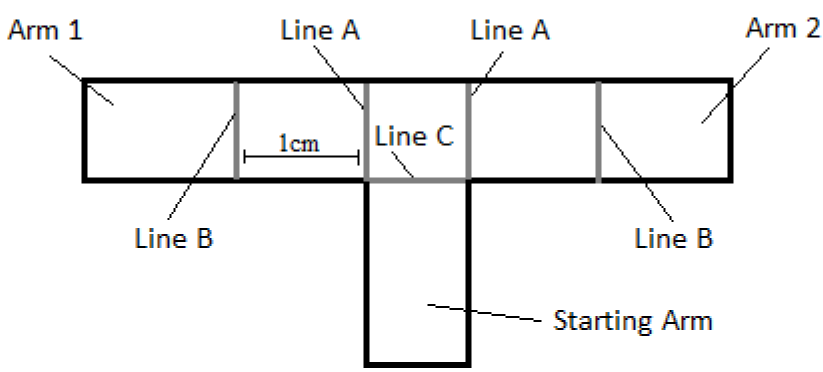


Figure 6. Drawing of the t-maze and the layout of all the arms. The electrodes were placed at Line B, in either Arm 1 or Arm 2. The planaria were placed in the bottom of the starting arm and allowed to roam free for 5 minutes. The arms that they visited and the time for them to cross Line C

Statistical Analysis

The mean and standard deviation (SD) of the indicated planaria movement variables were taken for each day of experiment in each condition (3-5 planaria per condition per day), so that one day of experiment for either sham or the field was represent by one mean value and one SD value. Analysis of variance (ANOVA) was used when looking for main effects between the presence of the electric field and the arm of the maze it was present in. Pearson's r and Spearman's rho were used to correlate planaria behaviour with geomagnetic storm activity. Geomagnetic storm activity was quantified using the Ap and AA indices; they are both linear measures of the geomagnetic disturbances and have base units of nanoTesla (nT) (Rostoker, 1972; Mayaud, 1972). All statistical analyses were completed with SPSS.

Results

There were no significant effects for the presence of the field in any of the variables of planaria movement in the maze ($p>0.05$). Over the course of the experiment, it was noted that occasionally a planaria would go up to one of the electrodes and make contact. This was noted 15 different times out of a total of 143 planaria in the study. Each time this occurred, the behaviour of the planaria was not the same; sometimes the planaria would glide over the electrode and seem unaffected, while at other times the planaria would convulse but stay close to the electrode and keep convulsing (even if the current was then turned off to allow the planaria to move away). A new variable was computed for the percentage of planaria that were noted to display this type of behaviour for each condition on each day. A two-way ANOVA with independent variables of presence of field and side of electrodes indicated a significant main effect for the presence of field [$F(1,35)=5.67$, $p=0.023$; omega squared=0.14; Figure 7].

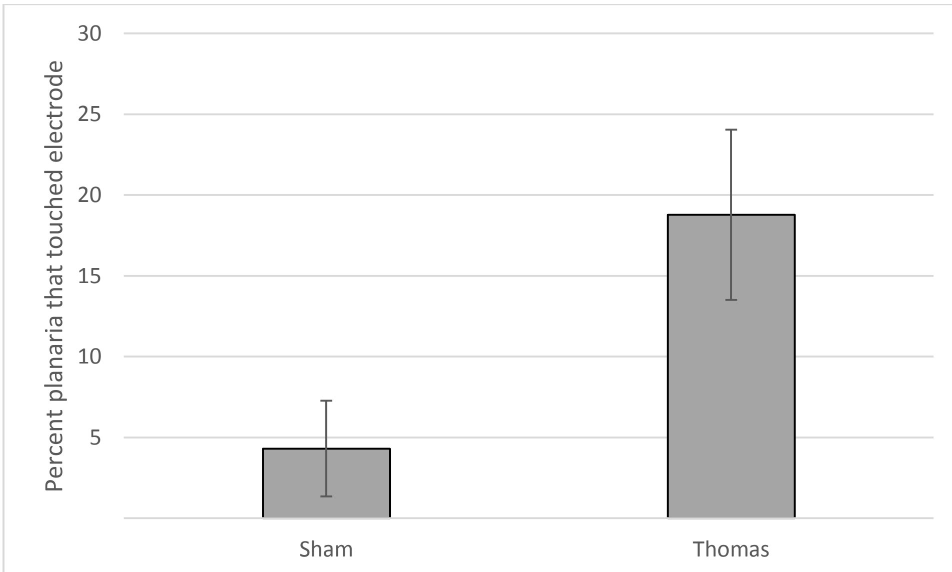


Figure 7. Percent of planaria per day of experiment that made contact with an electrode while in the t-maze. There were 19 days of experiments in the Thomas group and 17 days of experiments in the sham group. Error bars represent standard error of the mean (SEM).

It was noted that the phenomenon reported above did not occur homogenously across the different days of experimentation. A discriminant analysis was used to determine if there was any relation between the AP index of geomagnetic activity and occurrence of planaria making contact with an electrode in the Thomas condition. The days were coded in binary; the phenomena either occurred (N=9) or did not (N=10). The average daily AP index for the day of measurement and the preceding and succeeding 6 days were also recorded. Variables that entered into the model were the AP indices from

4 and 5 days after the presence of the behaviour [Λ=0.573, $X^2(2)=8.90$, p=0.012]. The function had a canonical correlation of 0.653 and explained 73.7% of the original and cross-validated cases. To determine the difference between the AP indices 4 and 5 days after the day of observation between the days when the phenomenon was present or absent, a within subjects MANOVA was used. There was a significant interaction between the presence of the phenomenon and the day of AP indices [F(1,17)=11.5, p=0.003, partial eta squared=0.40; Figure 8]. Paired t-tests showed that the AP indices from plus 4 days to plus 5 days were significantly different in the group where the phenomenon did not occur [t(9)=3.48, p=0.007]. There was no significant difference between the AP index measured between these two days when the phenomenon did occur [t(8)= -0.86, p=0.416].

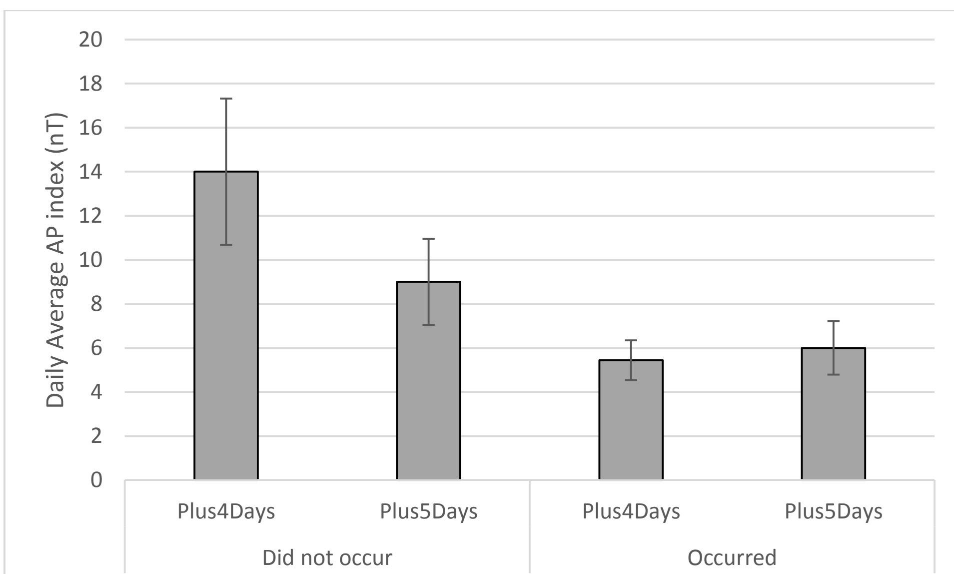


Figure 8. Daily average AP indices 4-5 days after the planaria were observed in the t-maze. Error bars represent SEM.

Correlational analysis was used to determine any relationships between the continuous variables (the amount of time it took for them to cross Line C or Line A and the number of arms they visited in the 5 minutes) and the AP indices. A correlation was considered significant if the Pearson correlation and Spearman correlation analysis were both statistically significant. There were no significant results when the entire dataset was analyzed, only when the groups of Thomas (N=19 days of experiments) and Sham (N=17 days of experiments) were analyzed separately were significant correlations found with the amount of movement variables. This was quantified as the total number of arms the planaria visited during the 5 minutes it was given in the t-maze. The mean and standard deviation were taken for the values in each condition for each day of experiment. Results are summarized in Table 2 for the mean number of arms visited and in Table 3 for the standard deviation of the number of arms visited. These tables contain the Pearson and Spearman values, if both these values were found to be significant then a Fischer's r to z test was used to determine if the Pearson's r was statistically different in the Thomas exposure group compared to the Sham group. The most significant differences were found in the standard deviation values, which are represented in Figure 9. Changes in variability were most likely driving the correlations found with the mean values.

Table 2. Pearson and Spearman correlation coefficients for the daily average of the total amount of arms visited with the AP indices of days surrounding the day of experiment.

Day	**Sham**	**Thomas**	**Fischer r to z score**
- 6 days	r= -.132 rho= -.156	r= .476* rho= .653*	z= -1.78; p=0.075
- 5 days	r= -.389 rho= -.354	r= .283 rho= .359	

- 4 days	r= -.373 rho= -.304	r= .465* rho= .449	
- 3 days	r= -.179 rho= -.098	r= .478* rho= .531*	z= -1.92; p=0.055
- 2 days	r= -.334 rho= -.181	r= .151 rho= .156	
- 1 day	r= -.467 rho= -.272	r= .128 rho= .036	
Day 0	r= -.338 rho= -.058	r= .111 rho= .112	
+ 1 day	r= .170 rho= .313	r= -.008 rho= .042	
+ 2 days	r= .013 rho= .128	r= -.218 rho= -.305	
+ 3 days	r= -.175 rho= -.137	r= -.386 rho= -.400	
+ 4 days	r= -.024 rho= .021	r= -.484* rho= -.374	
+ 5 days	r= .188 rho= .196	r= -.380 rho= -.352	
+6 days	r= .060 rho= .070	r= -.474* rho= -.235	

* = p<0.05

Table 3. Pearson and Spearman correlation coefficients for the daily standard deviation of the total amount of arms visited with the AP indices of days surrounding the day of experiment.

Day	**Sham**	**Thomas**	**Fischer r to z score**
- 6 days	r= -.220 rho= -.314	r= .763** rho= .692*	z= -3.35, p<0.001
- 5 days	r= -.208 rho= -.201	r= .509* rho= .461*	z= -2.11, p=0.035
- 4 days	r= -.215 rho= -.154	r= .737** rho= .561*	z= -3.18, p=0.002
- 3 days	r= -.218 rho= -.137	r= .587* rho= .556*	z= -2.44, p=0.015
- 2 days	r= .055 rho= .085	r= .173 rho= .250	
- 1 day	r= .128 rho= -.022	r= .223 rho= .225	
Day 0	r= .162	r= .254	

	rho= .034	rho= .324	
+ 1 day	r= .260 rho= .098	r= .039 rho= .048	
+ 2 days	r= .277 rho= .192	r= -.101 rho= -.356	
+ 3 days	r= .241 rho= .195	r= -.495* rho= -.529*	z= 2.15, p=0.032
+ 4 days	r= .200 rho= .322	r= -.555* rho= -.428	
+ 5 days	r= .106 rho= .270	r= -.523* rho= -.486*	z= 1.88, p=0.060
+6 days	r= .380 rho= .435	r= -.450 rho= -.288	

* = p<0.05
** = p<0.001

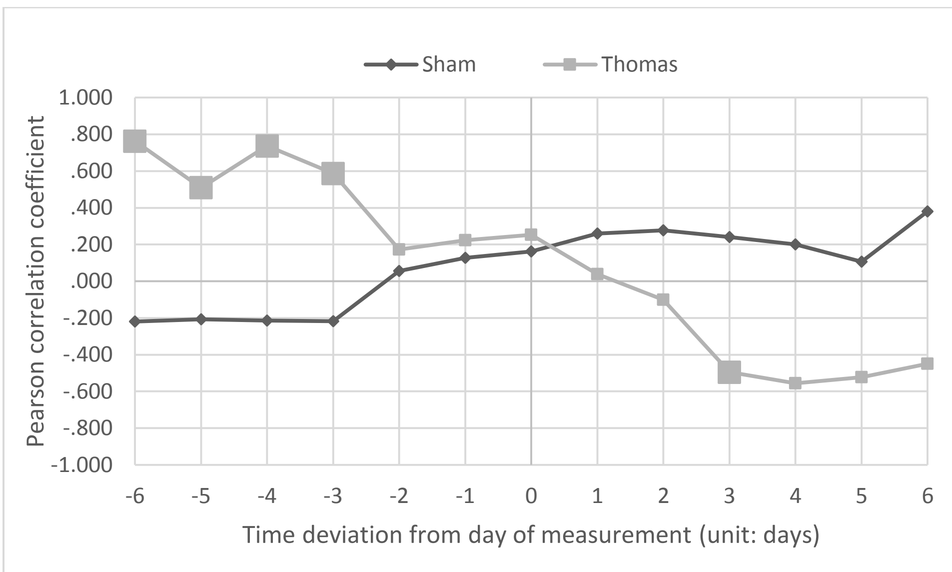
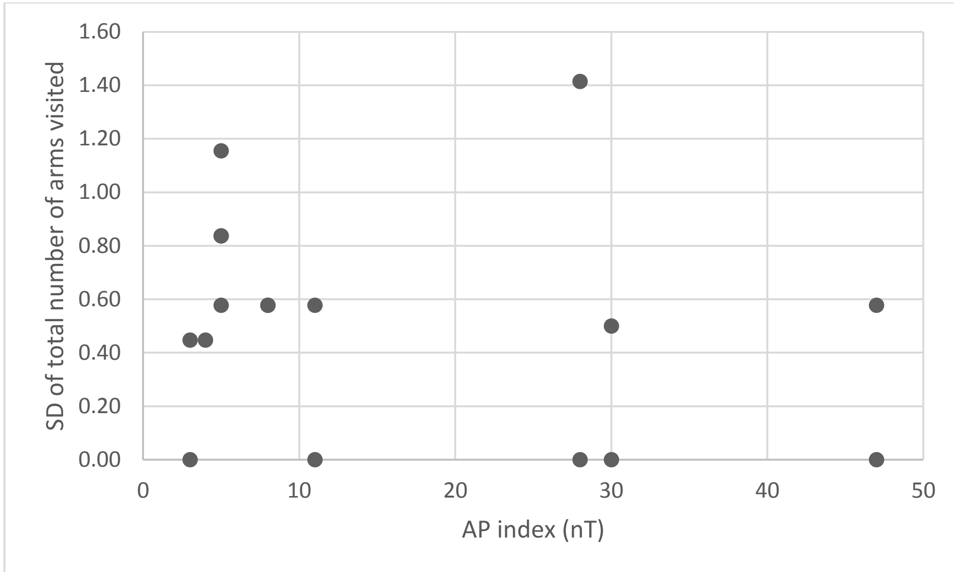
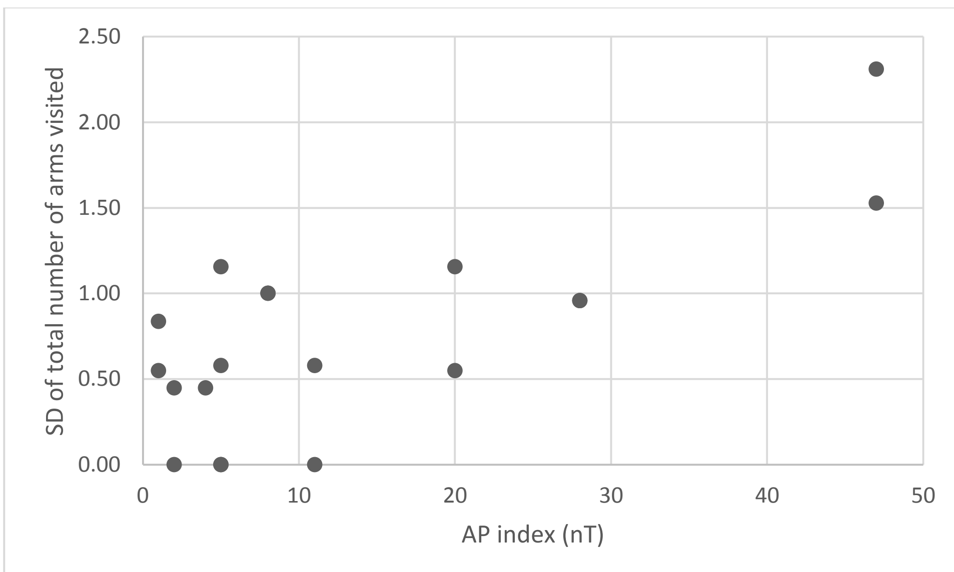


Figure 9. Pearson correlation coefficients of the average daily AP indices on days before and after the day of experiment, with the standard deviation of the total number of arms each planaria visited while in the t-maze. Extra-large data points in the Thomas condition were significant in Pearson's and Spearman's correlation tests and were more than 2 z-scores from their respective point in the Sham group.

The correlation between the standard deviation of the number of arms visited in the Thomas exposure group is evident in the scatterplots (Figure 10). Previous research investigating relationships with the geomagnetic storm indices (Bureau & Persinger, 1995) binned their data into 5 nT increments to investigate threshold effects. While there are not enough data points in this dataset for that type of analysis, it seems worthwhile to visually inspect the scatterplots for any conspicuous thresholds. Comparing Figure 10A and 10B demonstrates the difference in correlation coefficients shown in Figure 9. Figure 10B indicates that during quiet conditions the daily standard deviation in each condition was in the range of approximately 0-1.40, but started increasing at AP indices greater than 15 nT. Previous research in this laboratory studying the influence of the geomagnetic field determined threshold values to be 20nT (Persinger, 1988; Bureau & Persinger, 1992; Bureau & Persinger, 1995; Persinger & Richards, 1995; Persinger, 1995; O'Connor & Persinger, 1996; Galic & Persinger, 2007), however it is important to note that these studies used the AA indices of geomagnetic deviations. When the AA indices were entered into the current dataset, the potential 15 nT threshold that was found with visual inspection of the scatterplot, was now found at 20 nT (Figure 10C).


1.60
1.40
1.20
1.00
0.80
0.60
0.40
0.20
0.00
SD of total number of arms visited
0
10
20
30
40
50
AP index (nT)
A.


2.50
2.00
1.50
1.00
0.50
0.00
SD of total number of arms visited
0
10
20
30
40
50
AP index (nT)
B.

C.

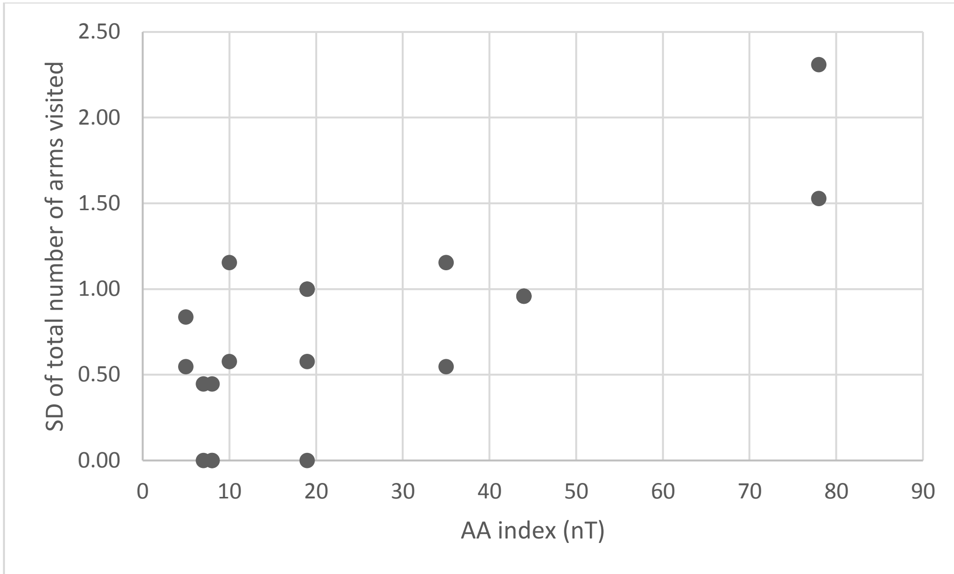


Figure 10. Correlation between the standard deviation (SD) in the total number of arms visited by the planaria and the geomagnetic storm indices 4 days before the experiment. A. In the sham-exposed planaria with the AP index. B. In the Thomas-exposed planaria with the AP index. C. In the Thomas treated planaria with the AA index.

Discussion

This experiment demonstrated that some planaria exhibited a positive electro-tatic behaviour towards the Thomas-patterned electric field. This was characterized by the planaria moving towards the electrodes and convulsing from being in such close proximity but not showing any behaviour indicating they tried to move away. Most stayed near the electrode, even when it was turned off. While the effect was significant, the average proportion of planaria that responded was small; about 18% of the planaria in the Thomas-exposure made contact with an electrode compared to about 4% in the sham condition. The discriminant analysis showed that this behaviour occurred more often when the day of observation was followed by quiet geomagnetic activity. This does not mean that the planaria's behaviour was able to influence the geomagnetic activity. It is more likely indicative of either the planaria responding to solar storms as they are happening, which take about 1.5-4 days to reach the Earth (Vladmirsky & Bruns, 2010). Or this may be the result of a low sample size not accurately representing the effect or statistical significance occurring from chance. However, if this behaviour in the planaria does represent a form of electroreception then it would make sense for this behaviour to be more likely to occur during quiet geomagnetic activity although it would be expected for that quiet activity to occur on the day of the experiment.

If there were environmental geomagnetic disturbances 3-6 days before the day of the experiment then the planaria exposed to the Thomas-electric field showed an increase in mobility as compared to those exposed to sham (Table 2). However, this effect was much more robust when looking at the standard deviation between the mobility of the planaria in one group for each day of experiment (Table 3; Figure 9). This indicates that some individual planaria were more affected than others. Geomagnetic disturbances and electric fields are both forms of electromagnetic fields (Persinger, 1980; Van Bladel, 2007). Exposure to the geomagnetic disturbances may have pre-treated the planaria so that they then responded differently to the patterned electric field. Delayed effects of geomagnetic disturbances or storms have been reported before and have been hypothesized to indicate that time was needed to generate the change in signalling pathways that led to the behaviour (Galic & Persinger, 2007). The effect in this experiment appeared to have a threshold of about 15 nT in the AP indices and about 20 nT in the AA indices (Figure 10B/C).

Significant trends in various behaviours have been correlated with geomagnetic AA indices in Dr. Persinger's laboratories indicate a threshold of 15-25 nT. These studies include: decreased latencies of seizure onset in rats (Bureau & Persinger, 1995), normal rat ambulation and seized rat mortality (Bureau & Persinger, 1992), thyroxine levels in a single patient (O'Connor & Persinger, 1996), reports of bereavement hallucinations (Persinger, 1988), vestibular experiences (Persinger & Richards, 1995), out of body experiences (Persinger, 1995), and decreased pain thresholds in rats (Galic & Persinger, 2007).

In the experiment that measured out of body experiences, Dr. Persinger (1995) predicted and confirmed that the response of complex human behaviours to increasing geomagnetic storm intensity would be non-linear. Non-linear trends were also present in the vestibular experiences (Persinger & Richards, 1995), and bereavement hallucinations (Persinger, 1988) with geomagnetic storm intensities. However, in the experiments with findings of geomagnetic correlations with thyroxine levels (O'Connor & Persinger, 1996), pain thresholds in rats (Galic & Persinger, 2007), ambulation in normal rats and mortality in seized rats (Bureau & Persinger, 1992) the trends were linear. This may be because these latter behaviours are simpler compared to the complex human behaviour described above. These simpler behaviours may have had linear trends if the effects were being driven by primarily one factor, such as a neurotransmitter level.

Geomagnetic storms have been shown to influence circulating levels of melatonin (Burch et al., 1999). Planaria not only have melatonin, but it demonstrates a circadian rhythm similar to mammals (Itoh et al., 1999). Melatonin is known to have anti-convulsant properties and is thought to reduce the amount of aberrant electrical activity in the brain (i.e. help prevent seizures). (Muñoz-Hoyos et al., 1998). Decreased levels of melatonin may also result in an increase in the amount of circulating dopamine (Bureau & Persinger, 1992). Planaria mobility requires a functioning dopaminergic system, including dopamine synthesizing cells in their anterior region (Nishimura et al., 2007). Acute exposure to dopamine antagonists reduce the mobility of planaria (Raffa et al., 2001), whereas acute exposure to dopamine agonists results in convulsions in hyperkinesia's in the planaria (Venturini et al., 1989). Perhaps the results found above may have resulted from decreased melatonin resulting in increased dopamine; those two changes combined may have resulted in excitotoxicity of the dopamine synthesizing cells in the planaria, and the increased mobility was a result of a partially regenerated dopaminergic system that was sensitive to the weak electric fields used in this experiment.

References

Bellono, N. W., Leitch, D. B., & Julius, D. 2018. Molecular tuning of electroreception in sharks and skates. Nature letters, 558:122-126.

Brown, F. A. 1962. Responses of the planarian, Dugesia, and the protozoan, Paramecium, to very weak horizontal magnetic fields. The Biological Bulletin, 123(2): 264-281.

Brown, F. A, & Park, Y. H. 1965. Phase-shifting a lunar rhythm in planarians by altering the horizontal magnetic vector. The Biological Bulletin, 129(1):79-86.

Burch, J. B., Reif, J. S, & Yost, M. G. 1999. Geomagnetic disturbances are associated with reduced nocturnal excretion of a melatonin metabolite in humans, Neuroscience Letters, 266(3): 209-212.

Bureau, Y. R. J. & Persinger, M.A. 1992. Geomagnetic activity and enhanced mortality in rats with acute (epileptic) limbic lability. International Journal of Biometeorology, 36(4):226-232.

Bureau, Y. R. J. & Persinger, M.A. 1995. Decreased latencies for limbic seizures induced in rats by lithium-pilocarpine occur when daily average geomagnetic activity exceeds 20 nanoTesla. Neuroscience Letters, 192(2): (142-144). https://doi.org/10.1016/0304-3940(95)11624-6

Chen, J., Chu, H., Xiong, H., Chen, Q., Zhou, L., Bing, D., Liu, Y., Gao, Y., Wang, S., Huang, X., & Cui, Y. 2012. Expression patterns of CaV1.3 channels in the rat cochlea, Acta Biochimica et Biophysica Sinica, 44(6): 513–518, https://doi.org/10.1093/abbs/gms024

Galic, M. A., & Persinger, M. A. 2007. Lagged association between geomagnetic activity and diminished nocturnal pain thresholds in mice. Bioelectromagnetics, 28: 577-579. doi:10.1002/bem.20353

Itoh, M. T., Shinozawa, T., & Sumi, Y. 1999. Circadian rhythms of melatonin-synthesizing enzyme activities and melatonin levels in planarians. Brain Research, 830(1): 165-173. https://doi.org/10.1016/S0006-8993(99)01418-3.

Liu, Y., Harding, M., Pittman, A., Dore, J., Striessnig, J., Rajadhyaksha, A., & Chen, X. 2014. Cav1.2 and Cav1.3 L-type calcium channels regulate dopaminergic firing activity in the mouse ventral tegmental area. Journal of neurophysiology, 112(5), 1119-30.

Mayaud, P.-N. 1972. The aa indices: A 100-year series characterizing the magnetic activity. Journal of Geophysical Research, 77(34), 6870–6874. doi:10.1029/ja077i034p06870

McCraty, R. 2015. Science of the heart: Exploring the role of the heart in human performance (Vol. 2). HeartMath. Boulder Creek, California.

Meyer, C. G., Holland, K. N., & Papastamatiou, Y. P. 2004. Sharks can detect changes in the geomagnetic field. Journal of the Royal Society, Interface, 2(2), 129-30.

Mulligan, B., Gang, N., Parker G. H., & Persinger, M. A. 2012. Magnetic field intensity/melatonin-molarity interactions: Experimental support with planarian (Dugesia sp.) activity for a resonance-like process, Open Journal of Biophysics, 2(4): 137-143. doi: 10.4236/ojbiphy.2012.24017

Muñoz-Hoyos, A., Sánchez-Forte, M., Molina-Carballo, A., Escames, G., Martin-Medina, E., Reiter, R. J., Molina-Font, J. A., & Acuña-Castroviejo, D. 1998. Melatonin's role as an anticonvulsant and neuronal protector: Experimental and clinical evidence. Journal of Child Neurology, 13(10), 501–509. https://doi.org/10.1177/088307389801301007

Murugan, N. J., & Persinger M. A. 2014. Comparisons of responses by planarian to micromolar to attomolar dosages of morphine or naloxone and/or weak pulsed magnetic fields: Revealing receptor subtype affinities and non-specific effects. International Journal of Radiation Biology, 90(10): 833-840.

Nishimura, K., Kitamura, Y., Inoue, T., Umesono, Y., Sano, S., Yoshimoto, K., Inden, M., Takata, K., Taniguchi, T., Shimohama, S., & Agata, K. 2007. Reconstruction of dopaminergic neural network and locomotion function in planarian regenerates. Developmental Neurobiology, 67: 1059-1078. doi:10.1002/dneu.20377

O'Connor, R. P., & Persinger, M. A. 1996. Brief communication: Increases in geomagnetic activity are associated with increases in thyroxine levels in a single patient: Implications for melatonin levels. International Journal of Neuroscience, 88: 243-247.

Persinger, M. A. 1980. The Weather Matrix and Human Behaviour. Praeger Publishers. New York, New York.

Persinger, M. A., & Richards, P. M. 1995. Vestibular experiences of humans during brief periods of partial sensory deprivation are enhanced when daily geomagnetic activity exceeds 15-20 nT. Neuroscience Letters, 194:69-72.

Persinger, M. A. 1988. Increased geomagnetic activity and the occurrence of bereavement hallucinations: Evidence for melatonin-mediated microseizuring in the temporal lobe? Neuroscience Letters, 88(3):271-274.

Persinger, M. A. 1995. Out-of-body-like experiences are more probable in people with elevated complex partial epileptic-like signs during periods of enhanced geomagnetic activity: a nonlinear effect. Perceptual and Motor Skills, 80(2):563-569. doi: 10.2466/pms.1995.80.2.563.

Putzier, I., Kullmann, P. H., Horn, J. P., & Levitan, E. S. 2009. Cav1.3 channel voltage dependence, not Ca2+ selectivity, drives pacemaker activity and amplifies bursts in nigral dopamine neurons. The Journal of Neuroscience: The official journal of the Society for Neuroscience, 29(49), 15414-9.

Raffa, R. B., Holland, L. J., & Schulingkamp, R. J. 2001. Quantitative assessment of dopamine D2 antagonist activity using invertebrate (planaria) locomotion as a functional endpoint. Journal of Pharmacological and Toxicological Methods, 45(3): 223-226.

Ribeiro, P., El-Shehabi, F., & Patocka, N. 2005. Classical transmitters and their receptors in flatworms. Parasitology, 131: S19-S40.

Rostoker, G. 1972. Geomagnetic indices. Review of Geophysics, 10(4), 935–950, doi: 10.1029/RG010i004p00935.

Salazar, V. L., Krahe, R., & Lewis, J. E. 2013. The energetics of electric organ discharge generation in gymnotiform weakly electric fish. The Journal of Experimental Biology, 216: 2459-2468.

Saroka, K. S., Vares, D. E., & Persinger, M. A. 2016. Similar spectral power densities within the Schumann resonance and a large population of quantitative electroencephalographic profiles: Supportive evidence for Koenig and Pobachenko. PLoS ONE 11(1): e0146595. https://doi.org/10.1371/journal.pone.0146595

Van Bladel, J. G. 2007. Electromagnetic fields. IEEE Press, Hoboken, New Jersey.

Venturini, G., Stocchi, F., Margotta, V., Ruggieri, S., Bravi, D., Bellantuono, P., & Palladini, G. 1989. A pharmacological study of dopaminergic receptors in planaria. Neuropharmacology, 28(12): 1377-1382.

Vladimirsky, B. M., & Bruns, A. V. 2010. Cosmic weather, physical-chemical systems, and the technosphere. Izvestiya Atmospheric and Oceanic Physics. 46. 935-951. 10.1134/S0001433810080037.

Xiang, J., Liu, Y., Wang, Y., Kotecha, R., Kirtman, E. G., Chen, Y., Huo, X., Fujiwara, H., Hemasilpin, N., deGrauw, T., & Rose, D. 2009. Neuromagnetic correlates of developmental changes in endogenous high-frequency brain oscillations in children: A wavelet-based beamformer study. Brain Research, 1274, 28-39. doi:10.1016/j.brainres.2009.03.068

Chapter 4 – Seed germination and photon emissions following exposure to a rotating magnetic field

Abstract

A multitude of experiments have applied magnetic fields to plants or seeds and found a variety of different and sometimes contradicting results. We have a magnetic field generating device called the Chrysalis resonator, which has been shown to influence the brain activity of human participants, the photon emissions from bacteria, and photon emissions from mammalian cell cultures and from water itself. In this experiment sunflower seeds (*Helianthus annus*) were allowed to begin germination and then exposed to either the field generated by the Chrysalis resonator or a sham condition. Their growth and photon emissions were taken over the next 5 days. It was found that the seeds showed less germination 48 hours after exposure and significantly higher photon emissions when 3 seeds were measured together in a dish, but not if 2 seeds or 1 seed were measured. These result seemed to indicate that the seeds may have become more sensitive to the presence of neighbouring seeds. The photon emissions results were also significantly impacted by the external weather conditions.

Introduction

The characteristics of magnetic field treatments previously used to treat plants are highly variable, and so are the results (Maffei et al., 2014). Some consistent findings seem to be reduced growth when high frequency (GHz) magnetic fields such as from cell phones or Gunn generators (produce fields in the microwave frequencies) are used (Vian et al., 2016). While the high frequency magnetic fields in Vian et al. (2016) are man-made and produce decreases in plant growth, there have been other magnetic fields that have similar frequencies to powerlines and electrical outlets (50-60 Hz) that have shown positive effects in growth (Naz et al., 2012; Aleman et al., 2014) and negative (Mroczek-Zdyrska et al., 2016). A review of experiments that investigated the effects of reduced ambient geomagnetic field (either using a Faraday cage-like device or active shielding) also found there was a trend of reduced growth (Belyavskaya, 2004). One study (Rakosy-Tican et al., 2005) decreased the intensity of the X-component of Earth's magnetic field, and found that these conditions could either increase or decrease plant growth depending on the geomagnetic storms conditions.

Studies that apply low intensity magnetic fields that have frequencies that converge on the Schumann frequency seem to increase plant growth measures (Namba et al., 1995; Betti et al., 2011; Radhakrishnan & Kumari, 2013; Huang et al., 2018). For example, one study (Radhakrishnan & Kumari, 2013) used a 1500 nT field (~20 times weaker than Earth's static field) at frequencies of 0.1 to 100 Hz to pretreat seeds, and found that exposure to a 10 Hz field was the most effective at increasing germination, water absorption, and electrical conductivity of the seed leachates. The changes in the electrical conductivity were associated with a more acidic pH and could indicate that the magnetic field-treated seeds had an altered ion exchange with their external environment (Radhakrishnan & Kumari, 2013). Increased germination was found with exposure to a 400-500 μT field (~10 times stronger than Earth's static field) that was applied at frequencies of 1 to 1000 Hz, with 10 Hz having the largest increase in germination (Namba, Sasao, & Shibusawa, 1995). Recently, researchers applied a magnetic field of 300μT at 7.83Hz (the Schumann frequency) and found an increase in germination compared to controls (Huang et al., 2018). Experiments utilizing static magnetic fields that are in the milliTesla intensity range show a high variability of results with findings of decreased growth (Vashisth & Nagarajan, 2010) and increased growth (Ćirković et al., 2017).

Biological organisms emit photons (Galle et al., 1991; Albrecht-Buehler, 2005; Fels, 2009; Cifra et al., 2011; Dotta et al., 2012; Persinger et al., 2015; Dotta et al., 2014; Dotta et al., 2016; Fels, 2017), as do plants (Bernard & Williams, 1951; Kuzin & Surbenova, 1995; Yan, 2006; Gallep & dos Santos, 2007; Sun et al., 2010; Moraes et al., 2012; Gallep et al., 2014; Footitt et al., 2016; Ćirković et al., 2017). One study that measured photon emissions from seeds, found that they could manipulate the number of photons by altering the temperature and humidity, factors involved in the onset of germination (Footitt et al., 2016). They also dissected the seeds and measured the separated sections and determined that the source of photon emissions were specifically from the inner layer of the seed coat, which surrounds the seed embryo, indicating that it may be involved in signalling the seed to begin germinating (Footitt et al., 2016). Photon emissions have also been demonstrated as a method of communication between organisms (Kuzin & Surbenova, 1995; Albrecht-Buehler, 2005; Fels, 2009; Cifra, Fields, & Farhardi, 2011; Fels, 2017).

The current experiment was designed to investigate the effect of a magnetic field on the growth of germinating seeds, their individual photon emissions as well as any photon emissions that may be involved in inter-seed communication.

Methods

Seed germination preparation

Sunflowers (*Helianthus annus*) were obtained from the gardening section of a local department store (subtype, Russian mammoth). For each trial, 108 seeds were split between two solutions of 25 mL of a 5% bleach solution and submerged for 5 minutes. The bleach was then washed with tap water. The seeds were put into 100 mm petri dishes with 10 mL of President's Choice spring water, with 18 seeds per dish. In each trial there were three dishes in each condition.

Rotating magnetic field device

The rotating magnetic field device is known as the Chrysalis resonator. It consists of columns of solenoids arranged in a circle. This model was similar but not identical to the one described previously (Tessaro et al., 2015). When active, it had a peak frequency of 113 Hz, an intensity of ~0.75 gauss and a speed of between 3000-4000 r.p.m (rotations per minute).

Procedure

After the seeds were placed into their dishes, three of these dishes were placed directly on the Chrysalis resonator for 1 hour to either be exposed to the field ON or to the sham condition (field OFF but with the fan running). After the first condition was complete there was an hour duration before the next condition. When not being exposed and during germination, the seeds were placed on flat surfaces in the dark. Daily measurements were taken for five days to determine if each seed had begun to germinate and to measure the length of the root/stem that had emerged from the shell. Statistical analysis of the length measurements were completed only on the seeds that had begun germination.

After these measurements there were photon measurements taken on a photomultiplier tube (PMT). To do this, 6 seeds were picked from each condition that were within a range of seedling length measurements that had been taken that day (Table 4). These were then put into three 35mm dishes; in the first dish there was one seed, in the second dish there were two seeds and in the third dish there were three seeds. Each of these dishes contained 0.5 mL of fresh spring water, this was to reduce the amount of stress that may have been occurring by measuring these seeds.

Light emissions were measured on a photomultiplier tube (PMT) housed in a black-painted wooden box that was covered in black towels. Samples were measured three times in one minute intervals at a sampling rate of 50 Hz. For statistical analysis, the average of the second and third recording was used for analysis. The first recording was not used because after each sample was placed on the PMT there would be a visible decrease in the number of photons being measured over the recording. Variables that are used to represent the photon emissions are the mean and the standard deviation of each recording. An example of a recording can be seen in Figure 11, from this the mean of all the points was computed as well as the standard deviation of all the points.

Table 4. Approximate range of root/stem lengths of seeds that were chosen from each condition to be used for photon measurements.

Day of Measurement	Range of seedling length (in millimetres)
1	0-1
2	1-2
3	2-7
4	9-30
5	30-80

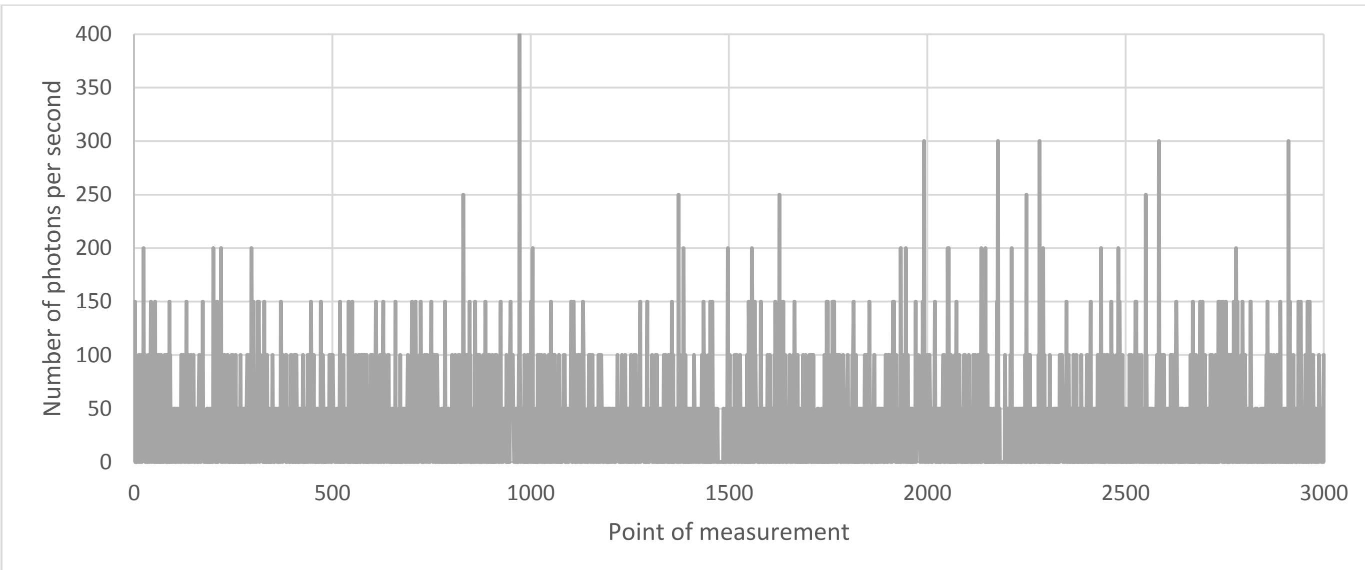


Figure 11. Example of 1 minute recording of seeds on photomultiplier tube. One measurement was taken every 20 milliseconds (sampling rate of 50 Hz).

Results

<u>*Germination*</u>

Seeds were germinated in 100 mm petri dishes, with 18 seeds in one dish. The percent number of seeds in each dish that germinated were calculated for each day for both the magnetic field exposure and sham condition. The average was taken of the three dishes in each condition. There was a significant interaction between day of measure and the field condition [$F(4,16)=4.51$, $p=0.013$; partial eta squared=0.53; Figure 12]. There was a significant decrease in the proportion of seeds which germinated for the magnetic field condition compared to the sham condition. Paired t-tests for each condition showed the interaction comes from difference in slope between day 1 and day 2 for the two conditions, evident in the disparity between the t-statistics (Table 5). This indicates that the effect on germination rate wasn't evident until the day 2 measurement, which was taken ~48 hours after exposure.

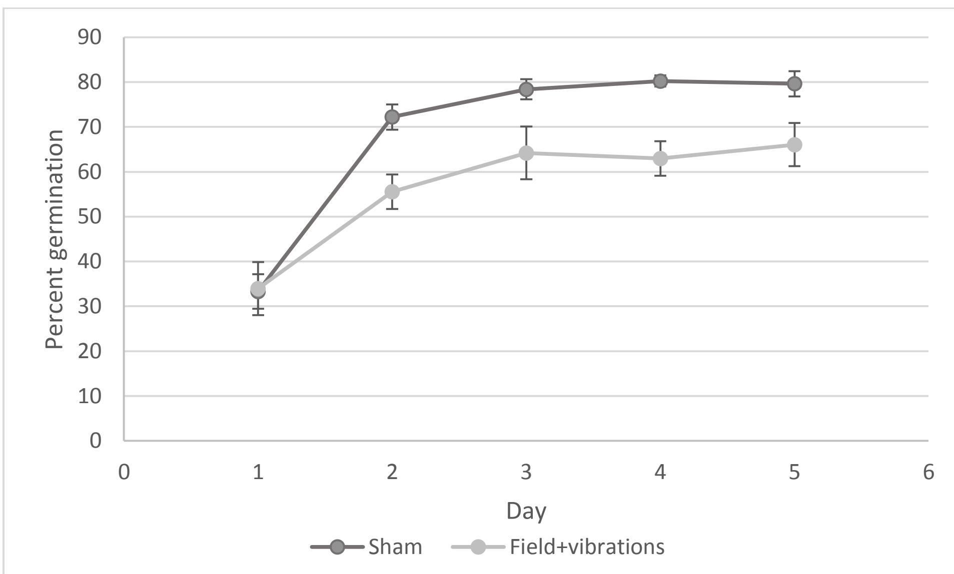


Figure 12. The percent seeds that had germinated over 5 days in the dark, resting in spring water. Seeds begun germination at Day 0 and were subsequently exposed to one of the conditions. Error bars represent SEM.

Table 5. Paired t-tests between the 5 days of measurement for the different magnetic field conditions. Values represent the t-statistic which had 2 degrees of freedom.

	Day 1 to 2	Day 2 to 3	Day 3 to 4	Day 4 to 5
Sham	18.2*	1.25	1.73	0.378
Field	9.71*	2.14	0.555	2.50

* = p<0.05

Length of seedling was then measured for the seeds that had germinated. There was no significant difference between the field exposed of sham exposed seedlings [F(4,16)=0.45, p=0.770; Figure 13].

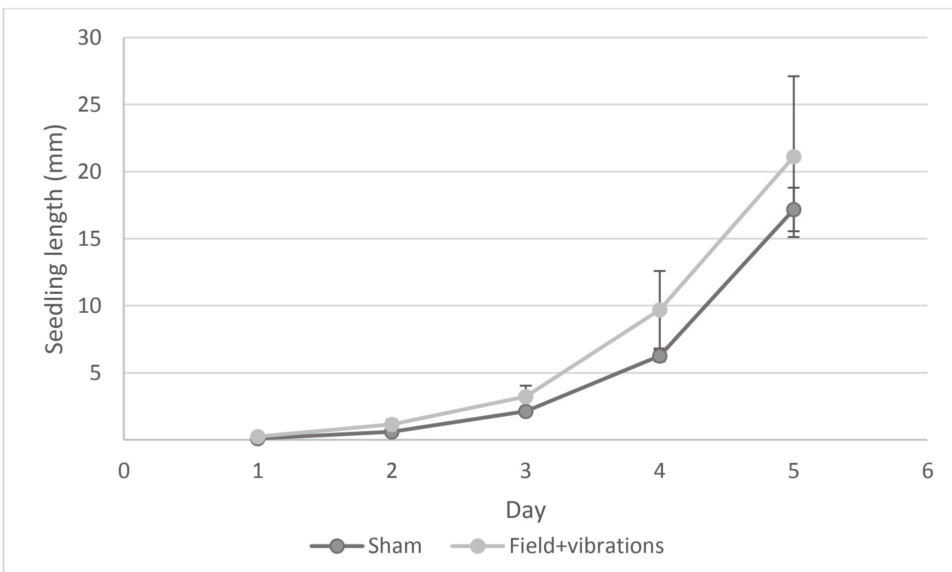


Figure 13. The length of seedling over the 5 days of germination in the dark. Seeds begun germination at Day 0 and were subsequently exposed to one of the conditions. Error bars represent SEM.

Photon

Initial results indicated no significant effects, however there was a large variability between the average number of photons between the different replicate of experiments [$F(2,17)=15.2$, $p<0.001$; Figure 14], as well as large differences in the standard deviation of the number of photons [$F(2,17)=27.0$, $p<0.001$; Figure 15]. In both variables, Tukey's post hoc test determined that the three replications of the experiment had different amounts of photon emissions with the second replicate was significantly higher than the first and third replicate ($p<0.05$).

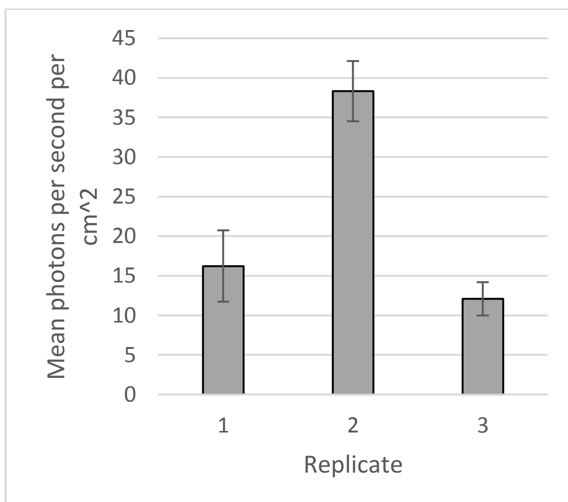


Figure 14. Difference across in mean photons in the three different replicates. Error bars represent the SEM.

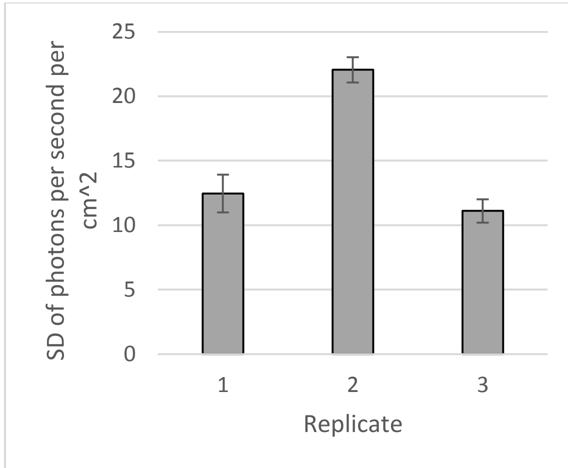


Figure 15. Difference across in standard deviation (SD) of photons in the three different replicates. Error bars represent the SEM.

Due to this large variability, two statistical methods were used for further analysis. The first was entering weather variables into the dataset, to see if controlling for any of these removed the variability. Second, within-subjects z-scores were used as well to confirm any results found with the weather variables.

Weather variables included in the dataset were the daily and hourly average of temperature, relative humidity and AP index. Also included were the hour of day the measurements were taken and the day of year of the replicate. Using a series of multivariate analysis of covariances (MANCOVAs) it was determined that most of the significant covariates were from the between subjects analysis (between the three replications) and not the within subjects analyses (within each single replication) (Table 6). Temperature and humidity explained the most variance when using the values for the hour of photon measurement, whereas the AP index explained the most variance when using the daily average values (Table 6). A regression analysis was used with all of the weather variables and the mean number of photons. The only variable that entered as a predictor was the daily average AP index [$F(1,17)=32.4$, $p=<0.001$, $r^2=0.648$; Table 7].

Table 6. F-values from multivariate analysis of covariance with weather variables from the hour of photon measurement or the daily average. Included are the effect sizes, represented as partial eta².

		Mean photons per second per cm²		SD photons per second per cm²	
Covariate	Statistic	Between – subjects	Within-subjects	Between - subjects	Within-subjects
Temperature of hour	F- statistic	46.3**	0.84	80.7**	2.83
	Partial eta²	0.81	0.018	0.88	0.057
Temperature, daily average	F- statistic	0.20	0.00	0.12	1.33
	Partial eta²	0.018	0.0001	0.011	0.028
Humidity of hour	F- statistic	47.6**	1.87	83.7**	4.60*
	Partial eta²	0.81	0.038	0.88	0.089
Humidity, daily average	F- statistic	4.48	1.00	5.64*	0.02
	Partial eta²	0.29	0.021	0.34	0.0005
Ap index of hour	F- statistic	4.31	11.6*	4.11	10.8*
	Partial eta²	0.28	0.20	0.27	0.19
Ap index, daily average	F- statistic	79.7**	4.93*	168.4**	3.81
	Partial eta²	0.88	0.095	0.94	0.075
	F- statistic	30.1**	3.23	35.2**	3.82

Hour of measurement	Partial eta^2	0.73	0.064	0.76	0.075
Day of year	F- statistic	0.66	Cannot compute	0.53	Cannot compute
	Partial eta^2	0.057		0.046	

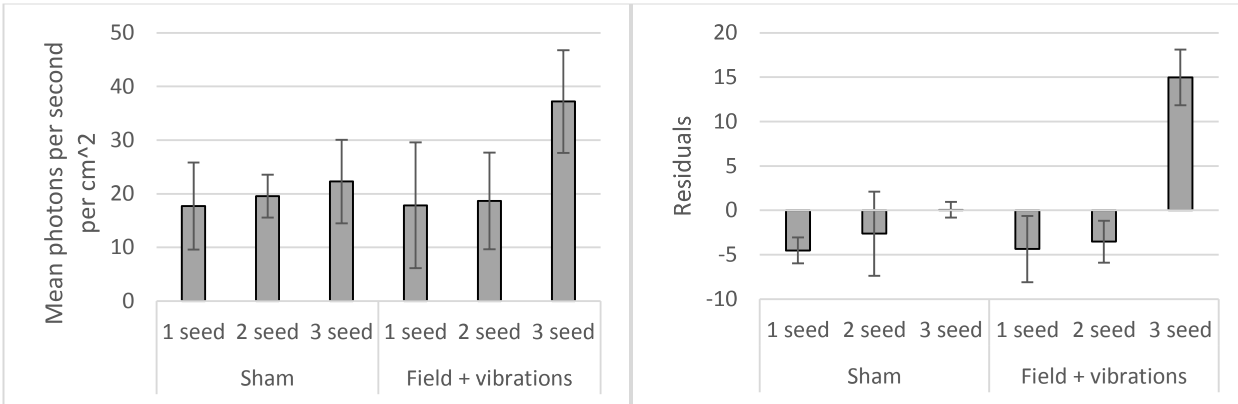
* $p<0.05$

** $p<0.001$

Table 7. Regression statistics of the daily AP index predicting the mean photons per second per cm^2.

Variable	Regression	Change in r^2	B	Std Err of B	Beta
Daily AP	0.82	0.65	6.53	1.15	0.818
Constant			-20.5	7.77	

The residuals from this analysis were saved and analyzed in a two-way ANOVA finding a significant main effect for the number of seeds [$F(2,17)=9.40$, $p=0.003$] and a significant two way interaction between the number of seeds and resonator condition [$F(2,17)=4.28$, $p=0.040$; Figure 16B]. Tukey's post-hoc test determined this was being driven by the 3 seeds group in the resonator condition being significantly higher than all other groups ($p<0.05$). The same trend of results was found with the standard deviation of photons, with the strongest effect being the 3 seeds in the resonator exposure condition (Figure 17).



A. B.

Figure 16. Mean photon emissions per second per cm^2 (assumed diameter of 2.5cm for PMT aperture) across the different conditions in a resonator experiment. A. Original values. B. Residuals after counting for daily average AP index. Error bars represent SEM.

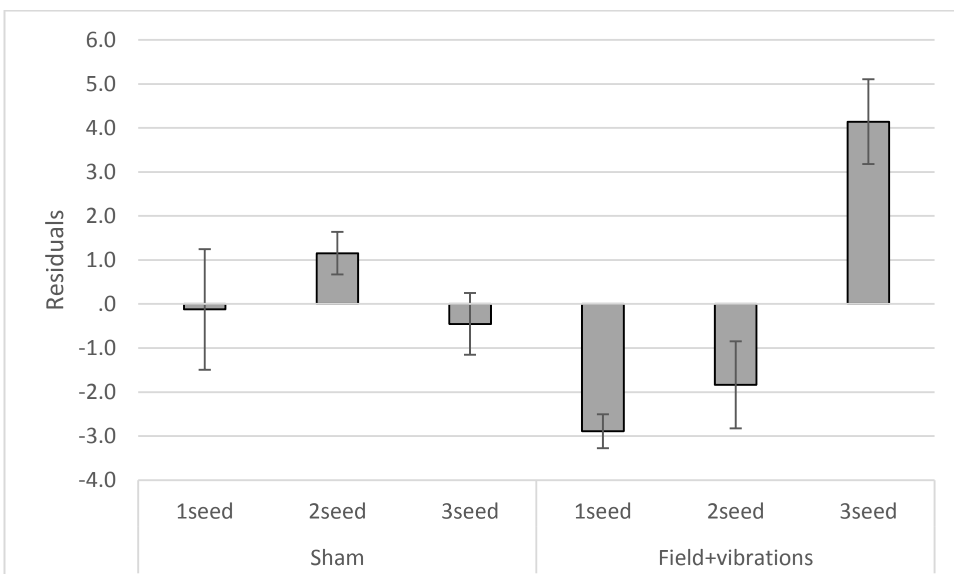


Figure 17. Residuals of the standard deviation of recording of photon emissions per second per cm^2 (assumed diameter of 2.5cm for PMT aperture) across the different conditions. Error bars represent SEM.

Another analysis was then carried out, using within subject z-scores, to see this would show the same result found above. For this analysis the dataset had to be re-organized. Such that the measurements were averaged over the 5 days into one value. The within-subject component beccame the number of seeds that were measured and the within-subject z-scores were computed. When a MANOVA was used on the mean number of photons, the effect for number of seeds was still present [$F(2,8)=5.58$, $p=0.030$], where paired t-tests showed that the 3 seed group was significantly greater than the 1 seed group [$t(5)=7.16$, $p=0.001$]; but the there was no longer an interaction with resonator condition [$F(2,8)=1.25$, $p=0.336$]. When this same analysis was used with the standard deviation of photons, there was no londer a significant main effect of number of seeds [$F(2,8)=3.51$, $p=0.080$] but the interaction between number of seeds and resonator condition was significant [$F(2,8)=4.67$, $p=0.045$]. Paired t-tests showed that none of the groups were significantly different in the Sham condition ($p>0.05$) but that in the Field +vibrations condition, the 3seed was group was significantly greater than the 2seed group [$t(2)=7.21$, $p=0.019$] and the 1seed group [$t(2)=19.3$, $p=0.003$]. These are the same differences found between groups as was found in the residual analysis for the standard deviation of photon recordings.

Discussion

Exposure to the dynamic field of the Chrysalis resonator (and its vibrations) caused approximately a 15% decrease in the number of seeds that germinated. This appeared in the measurements 48 hours after exposure, while there was no difference in germination

24 hours after exposure. Delayed effects of magnetic field exposure on germination have been previously reported (Ćirković et al., 2017).

The decrease in germination is an effect in this experiment similar to the high frequency man-made fields (Vian et al., 2016) or to environments that reduced the background intensity of the Earth's static magnetic field (Belyavskaya, 2004). Is it possible that these two broad categories of fields are reducing a developing seeds coherence or connection with the Earth's magnetic field? The phenomenon of this type of coherence has already been demonstrated in humans (Pobachenko et al., 2006; Persinger, 2014; Saroka et al., 2016), where the patterns of electrical activity in the brain correlated with the resonance frequencies of the Earth's magnetic field.

The interaction in photon emissions with number of seeds was evaluated with two different statistical techniques. When using residuals that had controlled for weather variables, the interaction was significant both in the mean photons and standard deviation of photon emissions over the measurement period. However, when using the within subject z-score methods, only the standard deviation interaction remained significant. This indicates that this variable demonstrated the greatest change from exposure to the dynamic condition of the Chrysalis resonator and that the changes in SD would be seen to a lesser extent in the mean values.

Photon communication in biological organisms has been demonstrated many times (Kuzin & Surbenova, 1995; Albrecht-Buehler, 2005; Fels, 2009; Cifra et al., 2011; Prasad et al., 2014). It has been demonstrated that germinating radish seeds that had been exposed to gamma irradiation could influence the germination rate of other radish seeds that had never been exposed (Kuzin & Surbenova, 1995 in Cifra et al., 2011). Indicating a potential for seed to seed communication through photon emission; when the seed germination rate was altered, these seeds may have been influenced by nearby seeds' through biophoton emission. Biophoton signalling has been previously implicated in the start of germination (Footitt et al., 2016), in this experiment, the resonator may have altered the biophoton signalling of the seeds and as a result interfered with their germination. In the present experiment, we found altered biophoton emission, but only when three germinating seeds were measured together, and not when one or two seeds were measured. One explanation is signal to noise ratio, where all of the seeds had altered biophoton emission, but three seeds were needed for the PMT to be able to detect the difference between conditions.

Another explanation for the results is that the increased photon emissions of the three seeds measured together was the result of a stress reaction in the seeds due to overcrowding. It has been previously demonstrated that changes in population density of biological organisms can alter their biophoton emission (Galle et al., 1991) and also influence the growth of organisms nearby (Fels, 2009; Fels, 2017). This could imply that the resonator induced the germinating seeds to be hyper sensitive to the presence of other seeds nearby. This may also explain the decrease in germination that was found, in that it was a response to increased population density, which could also explain why the decrease in germination was found 48 hours after the beginning of germination and not 24 hours after. There is an increased likelihood that the plants would be able to sense the presence of surrounding seeds at that time, either by their individual biophoton emission, contact or seed leachates.

References

Albrecht-Buehler, G. 2005. A long-range attraction between aggregating 3T3 cells mediated by near-infrared light scattering. Journal of Cell Biology 114, 493-502.

Aleman E. I., Mbogholi A., Boix Y. F., Gonzalez-Ohnedo J., Chalfun A. 2014. Effects of EMFs on some biological parameters in coffee plants (*Coffea arabica* L.) obtained by *in vitro* propagation. Polish J. Environ. Stud. 23, 95–101

Belyavskaya, N. A. 2004. Biological effects due to weak magnetic field on plants. Advances in Space Research, 34(7):1566-1574.

Bernard, B. S., & William, A. 1951. Light production by green plants, The Journal of General Physiology, 20(6):809–820.

Betti, L., Trebbi, G., Fregola, F., Zurla, M., Mesirca, P., Brizzi, M., & Borghini, F. 2011. Weak static and extremely low frequency magnetic fields affect in vitro pollen germination. TheScientificWorldJournal, 11:875-890. doi:10.1100/tsw.2011.83

Cifra, M., Fields, J.Z., & Farhadi, A. 2011. Electromagnetic Cellular Interactions. Progress in Biophysics and Molecular Biology, 105, 223-246.

Ćirković, S., Bačić, J., Paunović, N., Popović, T. B., Trbovich, A. M., Romčević, N., & Ristić-Djurović, J. L. 2017. Influence of 340 mT static magnetic field on germination potential and mid-infrared spectrum of wheat. Bioelectromagnetics, 38: 533-540. doi:10.1002/bem.22057

Dotta, B. T., Saroka, K. S., & Persinger, M. A. 2012. Increased photon emission from the head while imagining light in the dark is correlated with changes in electroencephalographic power: Support for Bókkon's biophoton hypothesis. Neuroscience Letters, 513(2): 151-154.

Dotta, B. T., Murugan, N. J., Karbowski, L. M., Lafrenie, R. M., & Persinger, M. A. 2014. Shifting wavelengths of ultraweak photon emissions from dying melanoma cells: their chemical enhancement and blocking are predicted by Cosic's theory of resonant recognition model for macromolecules. Naturwissenschaften, 101(2):87-94.

Dotta, B. T., Karbowski, L. M., Murugan, N. J., Vares, D. A. E., & Persinger, M. A. 2016. Ultra-weak photon emissions differentiate malignant cells from non-malignant cells in vitro. Archives in Cancer Research, 4: 2.

Fels, D. 2009. Cellular communication through light. PLoS ONE, 4(4): e5086. doi:10.1371/journal.pone.0005086.

Fels, D. 2017. Endogenous physical regulation of population density in the freshwater protozoan *Paramecium caudatum*. Scientific Reports, 7:13800. doi:10.1038/s41598-017-14231-0.

Footitt, S., Palleschi, S., Fazio, E., Palomba, R., Finch-Savage, W. E., & Silvestroni, L. 2016. Ultraweak Photon Emission from the Seed Coat in Response to Temperature and Humidity-A Potential Mechanism for Environmental Signal Transduction in the Soil Seed Bank. Photochemistry and photobiology, 92(5): 678-87.

Galle, M., Neurohr, R., Altmann, G., Popp, F. A., & Nagl, W. 1991. Biophoton emission from *Daphnia magna*: A possible factor in the self-regulation of swarming. Experientia, 47(5): 457-460.

Gallep, C. M., & dos Santos, S. R. 2007. Photon-counts during germination of wheat (Triticum aestivum) in wastewater sediment solutions correlated with seedling growth. Seed Science and Technology, 35(3):607-614.

Gallep, C. M., Moraes, T. A., Cervinková, K., Cifra, M., Katsumata, M., & Barlow, P. W. 2014. Lunisolar tidal synchronism with biophoton emission during intercontinental wheat-seedling germination tests. Plant signaling & behavior, 9(3), e28671.

Huang, P., Tang, J., Feng, C., Cheng, P., & Jang, L. 2018. Influences of Extremely Low Frequency Electromagnetic Fields on Germination and Early Growth of Mung Beans. IEEE International Conference on Consumer Electronics-Taiwan (ICCE-TW), Taichung, 2018, pp. 1-2. doi: 10.1109/ICCE-China.2018.8448809

Maffei, M. E. 2014. Magnetic field effects on plant growth, development, and evolution. Frontiers in Plant Science, doi:10.3389/fpls.2014.00445.

Moraes, T. A., Barlow, P. W., Klingelé, E., & Gallep, C. M. 2012. Spontaneous ultra-weak light emissions from wheat seedlings are rhythmic and synchronized with the time profile of the local gravimetric tide. Die Naturwissenschaften. 99. 465-72. 10.1007/s00114-012-0921-5.

Mroczek-Zdyrska, M., Kornarzynski, K., Pietruszewski, S., & Gagos, M. 2016. The effects of low-frequency magnetic field exposure on the growth and biochemical parameters in lupin (Lupinus angustifolius L.). Plant Biosystems, 151(3). doi:10.1080/11263504.2016.1186123.

Namba, K., Sasao, A., & Shibusawa, S. 1995. Effect of magnetic field on germination and plant growth. Acta horticulturae. 399, 143-148. doi: 10.17660/ActaHortic.1995.399.15

Naz, A., Jamil, Y., ul Haq, Z., Iqbal, M., Ahmad, M. R., Ashraf, M. I., Ahmad, R. 2012. Enhancement in the germination, growth and yield of Okra

(*Abelmoschus esculentus*) using pre-sowing magnetic treatment of seeds. Indian Journal of Biochemistry and Biophysics. 49, 211–214.

Persinger, M. A. 2014. Schumann resonance frequencies found within quantitative electroencephalographic activity: implications for Earth-Brain interactions. International Letters of Chemistry, Physics and Astronomy, 30: 24-32.

Persinger, M. A., Dotta, B. T., Karbowski, L. M., & Murugan, N. J. 2015. Inverse relationship between photon flux densities and nanotesla magnetic fields over cell aggregates: Quantitative evidence for energetic conservation. FEBS open bio, 5, 413-8. doi:10.1016/j.fob.2015.04.015

Pobachenko, S.V., Kolesnik, A.G., Borodin, A.S., & Kalyuzhin, V. V. 2006. The contingency of parameters of human encephalograms and Schumann resonance electromagnetic fields revealed in monitoring studies. Biophysics, 51(3):480-483. https://doi.org/10.1134/S0006350906030225

Radhakrishnan, R., & Kumari, B. D. 2013. Influence of pulsed magnetic field on soybean (Glycine max L.) seed germination, seedling growth and soil microbial population. Indian Journal of Biochemistry and Biophysics, 50(4): 312-317.

Rakosy-Tican, L., Aurori, C., & Morariu, V. 2005. Influence of near null magnetic field on in vitro growth of potato and wild Solanum species. Bioelectromagnetics, 26: 548-557. doi:10.1002/bem.20134

Saroka, K. S., Vares, D. E., & Persinger, M. A. 2016. Similar spectral power densities within the Schumann resonance and a large population of quantitative electroencephalographic profiles: Supportive evidence for Koenig and Pobachenko. PLoS ONE 11(1): e0146595. https://doi.org/10.1371/journal.pone.0146595

Sun, Y., Wang, C., & Dai, J. 2010. Biophotons as neural communication signals demonstrated by in situ biophoton autography. Photochemical & Photobiological Sciences, 9(3): 315-322. doi: 10.1039/b9pp00125e

Tessaro, L. W. E., 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, 172: 26-33.

Vashisth, Ananta & Nagarajan, Shantha. 2009. Effect on germination and early growth characteristics in sunflower (Helianthus annus) seeds exposed to static magnetic field. Journal of plant physiology. 167. 149-56. 10.1016/j.jplph.2009.08.011.

Vian, A., Davies, E., Gendraud, M., & Bonnet, P. 2016. Plant responses to high frequency electromagnetic fields. BioMed Research International, vol. 2016, Article ID 1830262, 13 pages. https://doi.org/10.1155/2016/1830262.

Yan, Yu. 2006. Biophoton Emission and Delayed Luminescence of Plants. Biophotonics:195-204. 10.1007/0-387-24996-6_15.

Chapter 5 – Influence of healing intentionality on cell growth and photon emissions

Abstract

Changes in the intensity of the geomagnetic field have been associated with a range of biological responses, as have exposure to magnetic fields. Unique individuals have demonstrated a capability to alter the intensity of the geomagnetic field close to their head when engaging in a state specific task. This experiment investigated the potential for healing intentionality to influence the growth and photon emissions by breast cancer cells. Five participants with varying experience in healing intentionality were volunteers. All trials were completed in our cell culture laboratory. A tray of cell cultures was placed in front of the individual, they were given no specific directions on how they should proceed and there were no time restrictions. A side study was completed with one of the participants, with alive and dead cancer cells. Results indicated differences in photon emissions of the alive and dead condition, as well as the appearance of a circadian rhythm in photon emissions for the cells. There appeared to be a small effect on photon emissions between the participants, but it was more related to their program of study as opposed to healing intentionality experience. These large main effects may have occluded any effect of healing that occurred, future studies should have better controlled trials and more replicates.

Introduction

Incidence rates of cancer continue to increase and yet there is no easy treatment for these individuals. While chemotherapy is effective in treating some cancers, it also decreases the quality of life. Exposure to specific weak, frequency-modulated magnetic fields have demonstrated promise in the inhibition of growth of cancer cells (Hu et al., 2010; Buckner et al., 2015). These fields are used at roughly the same intensity of the Earth's magnetic field (~25-70 μT). Exposure to these magnetic fields, and some other patterned magnetic fields have shown a wide range of effects, from reducing pain thresholds (Fleming et al., 1994; Martin et al., 2004) to inducing the feeling of the presence of a sentient being (Persinger & Healey, 2002). Geomagnetic storms, which are natural magnetic field disturbances, are of intensities in the range of 15-100 nT. This is ~1000 times weaker than the background intensity of the Earth's magnetic field, however they can still produce effects on biological organisms (Freidman et al., 1963; Bureau & Persinger, 1992; Richards & Persinger, 1995; Galic & Persinger, 2007; Mulligan et al., 2010; Murugan et al., 2015).

Unique individuals who have abilities such as remote viewing have unique brain activity, such as high occipital alpha (Persinger et al., 2002) and increased temporal theta and high frequency activity (Hunter et al., 2010). Sean Harribance (SH), who is able to engage in intuitive like states, describes an individual's memories just by looking at their picture, produces a decrease in the intensity of the horizontal component of the Earth's magnetic field during this process in the area around the right side of his head. This shift is of a magnitude at about 150 nT one centimeter away, and 5 nT one meter away from his head (Hunter et al., 2010). Similarly, a Reiki practioner was able to reduce the intensity of that same component 15 centimeters away from her head by an average of 7nT but up to 12nT, when imagining white light (Persinger et al., 2013).When SH's electrical activity from the parahippocampal region of his temporal lobe was recorded and digitized into a magnetic field, it was able to inhibit the growth of cancer (Karbowski et al., 2012).

In this experiment recruited 5 participants with a variety of experience in healing intentionality volunteered, to attempt to reduce the growth of cancer cells with the power of their mind.

Methods

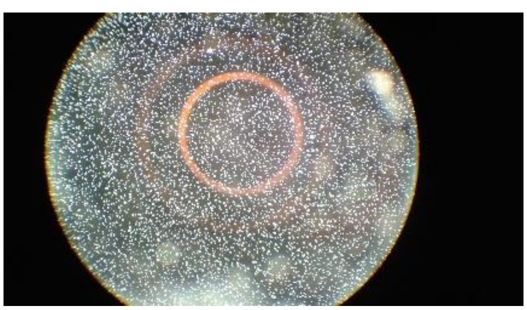
Cell culture

MCF7 breast cancer cells were maintained in DMEM media supplemented with 10% fetal bovine serum, 100U/ml penicillin G, 100μg/ml streptomycin sulfate and 250ng/ml amphotericin B. They were subcultured every 2-4 days in 150 mm cell culture dishes. Media was removed, 5 mL of 0.25% trypsin was added for 5 minutes, and then 5 mL of cell media was added, this solution was centrifuged then resuspended in media and split at a ratio of approximately 1:5. Cells were obtained from Dr. Carly Buckner, and Dr. Robert Lafrenie from the Health Sciences North Cancer Research Centre in Sudbury, Ontario. For experiments, cells were plated in 100 mm and 60 mm dishes for the cell growth measurements and 60mm for the biophoton measurements. Cells were cultured in complete media at 37°C and %5 CO_2.

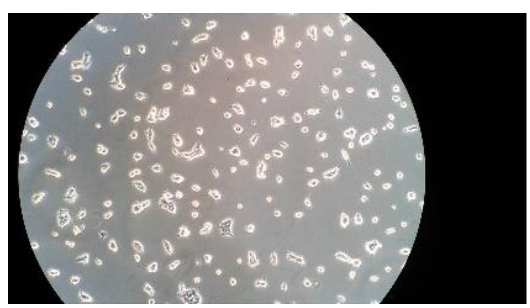
Measurements

Photon measurements: Measurements were taken with a photomultiplier tube (PMT) in a dark box in a dark room. The dark box is a wooden box that is ~ 1 foot cubed (with no top), painted black and covered in black towels. The PMT sits inside the box with its aperture pointed upwards. Cell plates are placed onto this aperture for measurement. The device was programmed to take measurements every 20msec which gave it a sampling rate of 50Hz. Four consecutive 1 minute measurements were taken, the 3rd measurement was used for analysis.

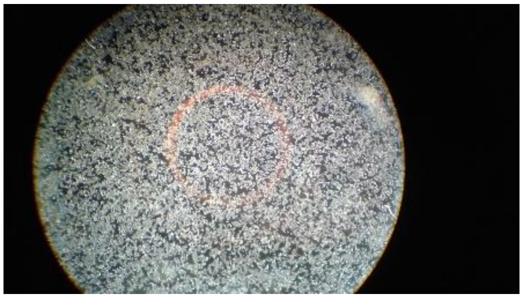
Microscope Pictures: Each plate of cells had two pictures taken of it, one at a magnification of 100x under a light compound microscope and the other under a broken inverted microscope. Because the inverted microscope is broken, I am unsure of what the magnification is, however, it is much lower than the other microscope, sample pictures are provided below (Figure 18). These pictures were taken with a camera phone through the ocular of the microscopes. To analyze the images they were converted to .jpeg files, then the image file size was obtained for each plate. This is a value that approximates complexity measure of the image, the higher the complexity the greater the file size would be.

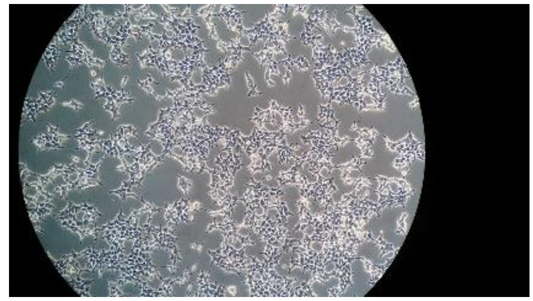


A. Low magnification, low cell density



B. High magnification, low cell density





C. Low magnification, high cell density D. High magnification, high cell density

Figure 18. Sample of pictures taken of cell plates.

Trypan Blue Exclusion Method: The media was removed from the cell dishes and replaced with 1 mL of 0.25% trypsin for 5 minutes. The cells were collected in this solution, transferred to 2 mL tubes and centrifuged for 10 minutes at 2000 g. The supernatant was removed and the cells were resuspended in PBS (phosphate buffered saline) with 20 µL of trypan blue. This solution was loaded into a haemocytometer and the clear (live) and blue-stained (dead) cells were counted.

Experiment 1: Cancer cell response to human intentionality

On day 0 of the experiment, the cells were harvested and subcultured onto 60 mm or 100 mm dishes. On day 1 the cells received their treatment, and then were assessed at two different time points of measurement. The first measurement was taken within 9 hours after treatment and the second was taken on day 2, 24-28hours after treatment. Cells were split so that the plates would be close to 100% confluent on day 2.

There were a total of five participants, each with a varying degree of experience in practicing healing intentionality (Table 8). Each time a participant came in for a treatment they entered the cell biology lab, took a seat in either a chair or a stool and filled out a mood score evaluation. When completed, they handed it to the experimenter who then took out four 60 mm dishes and two 100 mm dishes from the incubator that had been split the night before. The tray was placed in front of the individual and they were asked to indicate to the experimenter when they began their treatment and when they finished so that the duration of the treatment could be recorded. After the treatment they completed another mood score evaluation and after the participant left another tray of cells was placed on the bench in the same place as the treatment plates for the same duration.

The 60 mm dishes were used for measuring the light emission from the plates with the PMT and then counting the number of cells on the plate with the trypan blue exclusion method. The 100mm dishes were used for western blot analysis of the phosphorylation of ERK (extracellular regulated kinase) (data not shown). All of these measurements were taken at the first and again at the second time point.

Table 8. Demographics of participants in experiment.

Participant	Gender	Age	Healing Experience	Occupation
P01	Female		Professional	Practicing Shaman Healer
P02	Male	32	Novice to intermediate	Ph.D. Interdisciplinary Human Studies
P03	Male	26	Novice/beginner practitioner	PhD Biomolecular Sciences, forensics psychometrist and counsellor
P04	Male	25	Novice	M. Sc in Biology

P05	Female	32	Intermediate to professional, closer to professional	Human Studies PhD Program

<u>Experiment 2: Influence of Shaman healer on alive vs. dead cancer cells</u>

There was a side study completed with subject P01, she treated both healthy MCF7 breast cancer cells and also dead MCF7 breast cancer cells. The dead condition of cancer cells were plated and split in the same manner as the alive cells. On the day of the experiment, about 2 hours before the Shaman arrived, the dead cell condition was created by dumping out their media and adding a 3% (w/w) solution of hydrogen peroxide (H2O2) to the cells for 5 minutes. The cells were then centrifuged for 10 minutes at 2000 rpm, H2O2 was removed, and the cells were re-suspended in media. After treatment by the Shaman healer or a negative control trial photon measurements were taken that same day and 24 hours later. They were measured 4 times for 1 minute each at a sampling rate of 50Hz.

<u>Experiment 3: Circadian rhythm of biophoton emission of cell cultures</u>

MCF-7 cells were split on Day 0 into twelve 60mm culture dishes. Two plates per time point were measured at ~ Noon, 6pm and midnight on both Day 1 and Day 2. Taking measurements for two consecutive days controlled for cell count effects. Photons were measured as described above. Before any cells were put into the box, there were empty box photon measurements taken. This would indicate if any changes in photon counts in the cells were a result of a change in background photons.

Results

Cell counts

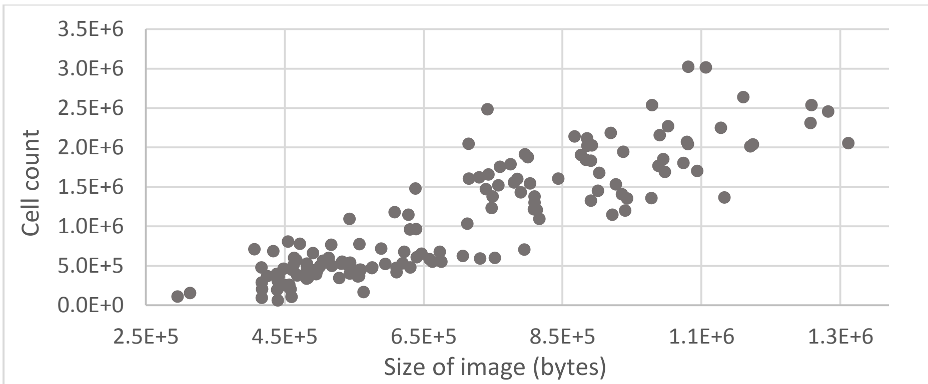
A three way analysis of variance (ANOVA) was used with independent variables of time point, participant and condition. Condition referred to either the plates that were treated by the participant or the plates that served as controls for each treatment. If there was a significant effect of the treatment on cell growth or survival then we would expect to see a significant interaction with the condition variable with participant. However, this was not the case ($p>0.05$).

Microscope Pictures

In the image size analysis we correlated the size of the image of each plate with the number of cells that were counted with the trypan blue exclusion method. There was a conspicuous relationship between the pictures taken at the higher magnification ($r= 0.869$, $p<0.001$; $rho=0.868$, $p<0.001$; Figure 19A). The pictures taken at the lower magnification did not show this same relationship ($r=0.013$, $p=0.904$; $rho=0.012$, $p=0.910$; Figure 19B). Because this measure of complexity may represent more than just number of cells, and perhaps how the cells organized themselves as they filled the plates, an analysis of variance was run with image size as the dependent and independents of participant, day and condition and covarying for number of cells, but this did not show any significant relationships ($p>0.05$).

Using the picture size analysis as a measure of complexity to measure number of cells is novel, and to the knowledge of the writer has not been previously demonstrated. More experiments need to be completed to confirm that this relationship exists, while also exploring the influence of cell size and shape may have on these values.

A.



B.

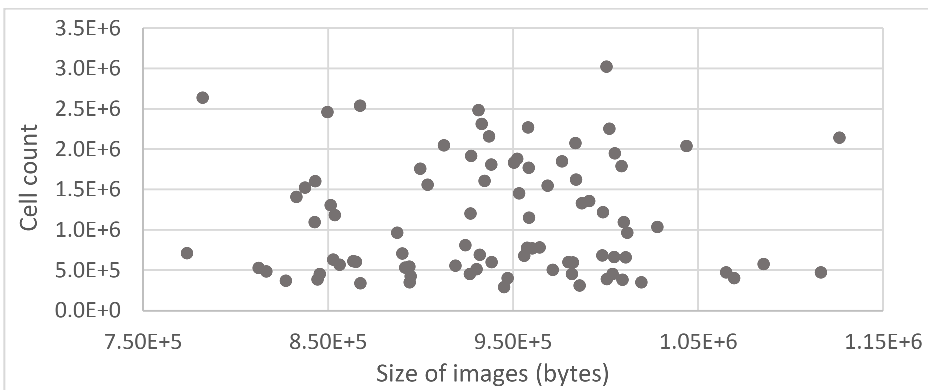


Figure 19. Corrleations between the size of the image taken of a cell plate and the number of cells that were subsequently counted with the trypan blue exclusion method. A. Images taken at higher magnification. B. Images taken at lower magnification.

Photon Measures

First, it should be stated that about halfway through the experiments there was a contamination of the MCF7 cell cultures and new cultures were obtained. When comparing negative controls from the MCF7s in the first half (Stock 1) of the experiment with the second half (Stock 2) of the experiment, there was a significant difference between the mean number of photons emitted.

An ANOVA determined there was an overall difference in the mean photon count between the two stock plates. To check to see if there was some change in the photomultiplier tube, the empty box measurements were included in the analysis. This produced an interaction between the stock plate and the presence of cells [$F(1,37) = 10.7$, $p = 0.003$; Figure 20]. Where the empty box measurements taken during the stock 1 (M=0.151, SEM=0.00471) were not significantly different from the empty box measurements taken during the stock 2 time period (M=0.164, SEM=0.0143). However, the control cells from the stock 1 time period (M=1.40, SEM=0.163) were significantly higher ($p<0.001$) than the stock 2 control cells (M=0.57, SEM=0.0918).

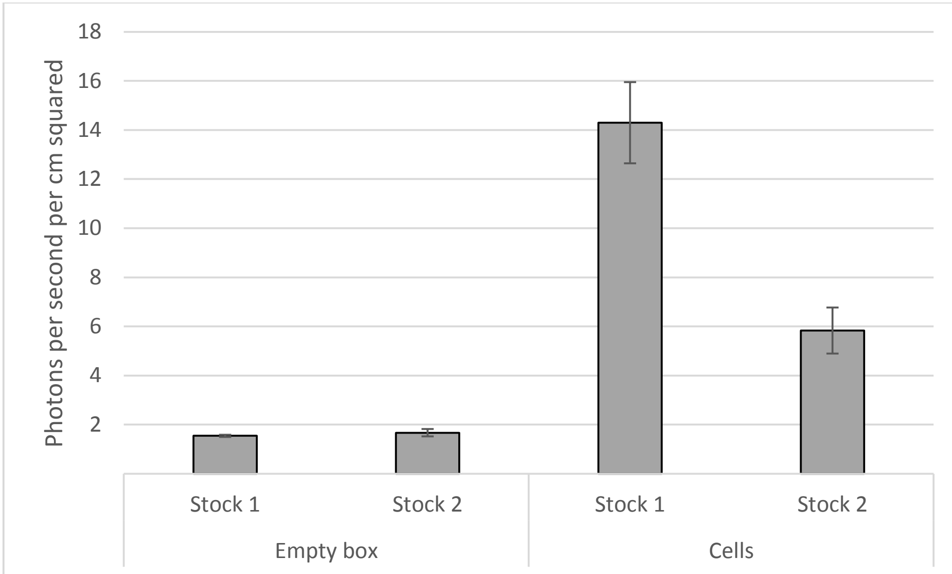


Figure 20. Photon emissions from stocks of MCF7s that were measured in two distinct periods of time show differences not seen in background measurements. Error bars represent SEM.

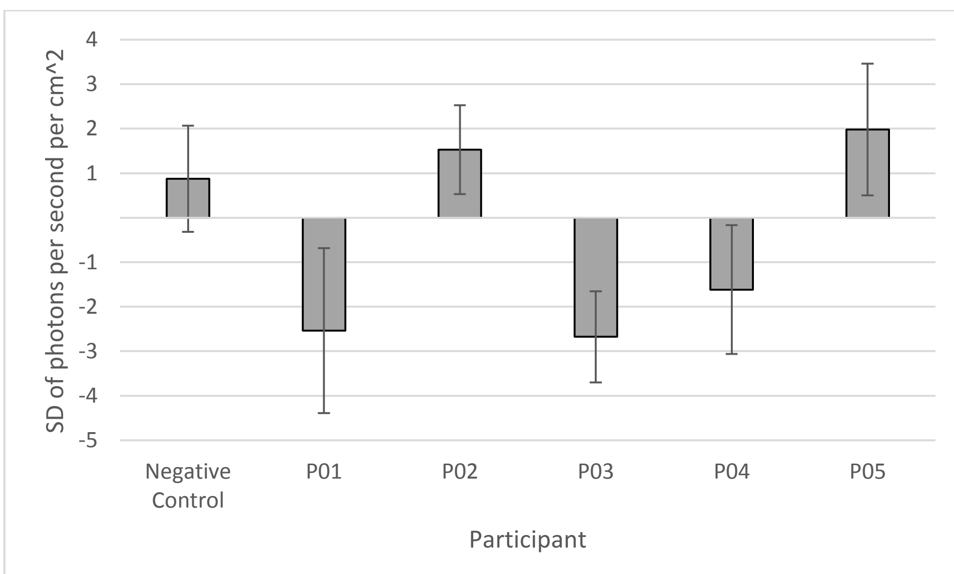
Another trend that was noticed in the raw data was an effect for time of day with photon emissions (see Experiment 3). And therefore, difference scores were computed using the control plates that were run after every treatment. These were computed in the following manner:

Difference score= treatment measurement-control measurement

Positive values indicate that the treatment plate had greater photon emissions than the control plate for that measurement and negative values indicate that the treatment plate had less than the control plate.

There were no significant effects for the difference scores in the mean number of photons ($p>0.05$), however for the difference scores in the standard deviation for the number of photons, a main effect for participant was approaching significance [$F(5,39)=1.96$, $p=0.116$, $\omega^2=0.25$; Figure 21A]. It was observed that the participants that are currently completing graduate degrees, that the human studies participants were trending in the same direction and the biomolecular/biology participants were trending in the same direction. When a new variable was created to group these participants together, the effect became significant [$F(3,39)=3.43$, $p=0.029$, $\omega^2=0.24$; Figure 21B]. You'll notice that the effect sizes between the non-significant and significant statisitcs were comprable. Tukey's post hoc's determined that the changes in biophotons produced in response to exposure to participants for the human studies group was significantly higher than those produced by exposure to the biomolecular/biology group ($p=0.041$).

A.



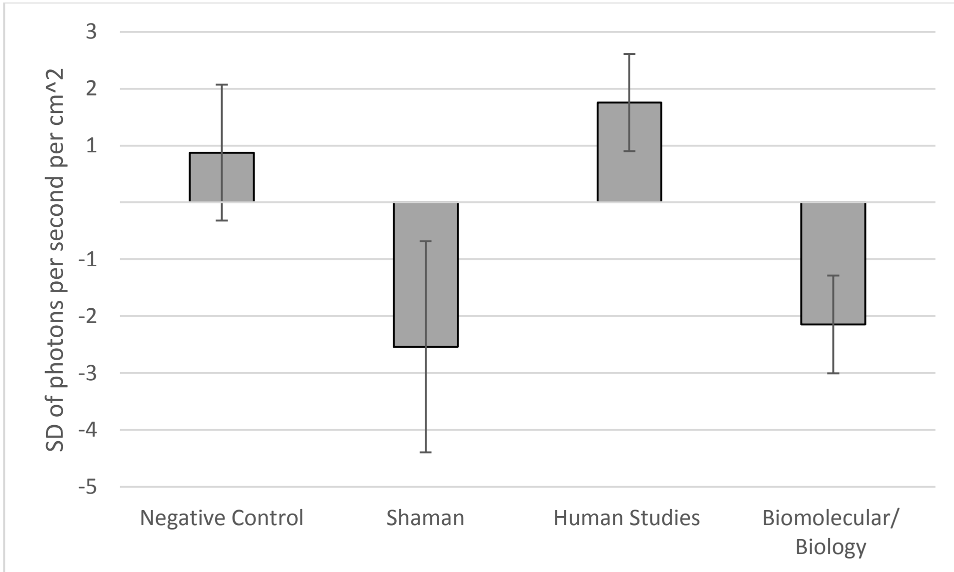
B. 

Figure 21. Difference scores in the standard deviation (SD) of photon emissions over a recording of cells treated by one of the participants or the negative control condition. A. The effect is not significant. B. Participants were grouped by degree of study, the effect became significant. Error bars represent SEM.

Experiment 2: Influence of Shaman healer on alive vs. dead cancer cells

The following analysis was conducted to determine photon differences between alive and dead cells. There was no significant difference for the mean photon emissions from the cells [$F(1,45)=0.305$, $p=0.584$]. A spectral analysis was completed on the photon recordings which created 0.1 Hz frequency bins between 0 and 25 Hz. These values were averaged into 1 Hz bins and used in a discriminant between the alive (N=24) and dead cells (N=22). Each sample represents the average of two cell culture measurements in the same condition treated on the same day. In the discriminant analysis, the frequency bins 3 Hz, 12 Hz, and 20 Hz all entered into the equation to discriminate between alive and dead cells [Wilk's Λ=0.668, $X^2(3)=17.2$, p=0.001]. The function [Function= 6.56*(3 Hz) – 9.24*(12 Hz) + 6.48*(20 Hz) – 3.28] had a canonical correlation of 0.58 and was able to correctly classify 71.7% of the cross-validated cases. These frequency bins were then analyzed with an analysis of variance with cell state (alive vs dead) as well as participant (Shaman vs negative control). The 3 Hz [F(1,45)=6.72, p=0.013) and 20 Hz [F(1,45)=6.01, p=0.019] bins were found to be significant between alive and dead, whereas the 12 Hz bin was not [F(1,45)=0.117, p=0.734]. Next, the 0.1Hz values that comprised the bins were analyzed as separate variables to determine which ones were driving the effect. For the 3Hz bin, it was 2.6Hz that was significantly different (p=0.014), with 2.3Hz approaching significance (p=0.051) (Figure 22). And for the 20Hz bin, it was 19.3Hz (p=0.009) and 19.7Hz (p=0.048) that were significantly different with 20.0Hz approaching significance (p=0.085) (Figure 22).

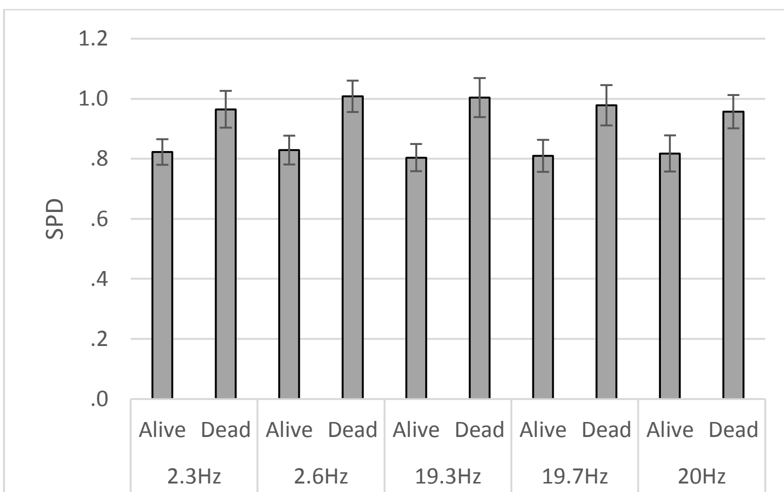


Figure 22. Spectral power density (SPD) values for frequency bins that discriminated between alive vs dead MCF7 breast cancer cells. Error bars represent SEM.

Experiment 3: Circadian rhythm of biophoton emission of cell cultures

A oneway ANOVA found a significant effect for time point [$F(5,17)=6.16$, $p=0.005$, $\omega^2=0.72$; Figure 23 (bars on the left)], where Tukey's post hoc test indicated that photon counts at noon on day 2 were significantly higher than 6pm on day 1 ($p=0.021$) and day 2 ($p=0.012$) as well as midnight on day 1 ($p=0.014$) and day 2 ($p=0.011$). There was no significant effect for time point in the empty box measurements [$F(5,17)=0.529$, $p=0.750$; Figure 23 (bars on the right)].

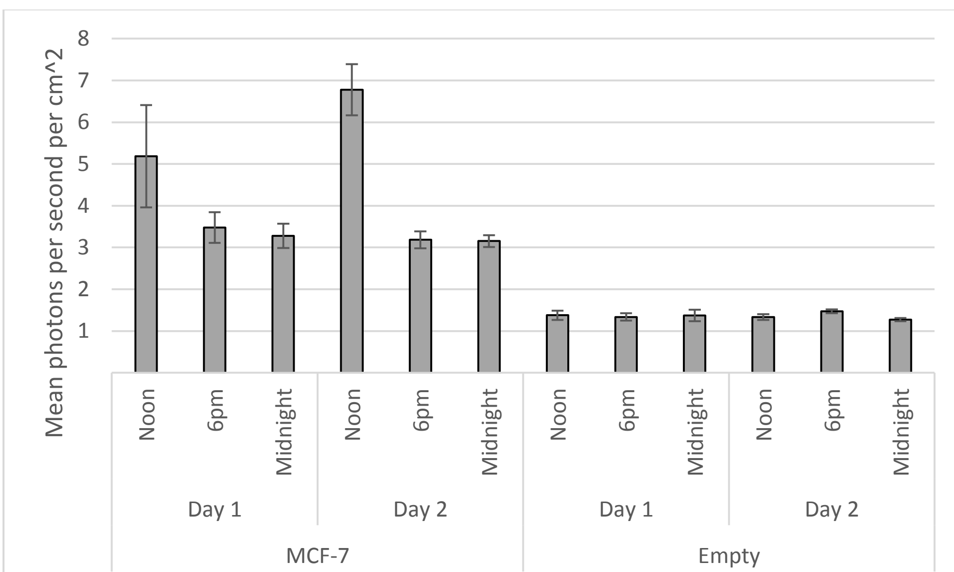


Figure 23. Changes in photons from MCF-7 cell cultures show circadian rhythm not seen in photon recordings of an empty box. Error bars represent SEM.

Discussion

While we did not observe any inhibition of cell growth in this experiment, this may have been due to the experimental design or the tool of measurement. In terms of design, perhaps we should have designed the treatments to be administered similar to how applied weak, magnetic fields have shown to be effective in slowing cancer growth. Our method of counting was the trypan blue exclusion method. Perhaps this tools is best for measuring robust treatments that show a large decrease in growth. Subtle changes with healing intentionality in growth have been found before with MCF7s however the method of measurement was the MTT assay (Smith & Laskow, 2000). Also, in that experiment the researchers cultured the cells so that they were in a stressed state, either with doxorubicin or by plating at high densities. This stressed state might be a closer approximation to disease *in vivo* which might be a requirement for healing intentionality to be effective.

There was a large difference in the photon emissions for the negative controls that were derived from two different stocks of MCF7 breast cancer cells. These differences might be due to the fact that these measurements were separated in time; measurements of the first stock plates took place in August, 2017, whereas measurements of the second stock plates took place in September-October, 2017. This may have resulted from something that occurred during the transport of the two stocks from the Cancer Centre to the University, they are separated by ~ 3km. It is possible that some event during transportation occurred that significantly impacted only the photon emissions of the cells but not their growth. Alternatively, if this reflects something different in the time periods of measurement, a primary candidate would be temperature, or even the presence of the

Shaman Healer, as she was in the lab almost every day during the end of July until mid-August. It was shortly after she left that the cells became contaminated and we needed to get a new stock. Finally, it is possible that over time there was a shift in the biological properties of the MCF7 breast cancer cells, and that this genetic drift is evident in biophoton emissions.

Difference scores needed to be taken to correct for the above effect found between cells stocks and the circadian rhythm effect found. The difference scores taken of the standard deviation in the photon emission recordings indicated a potential difference between cells from the treatment from the participant, but not related to their experience and skill level in healing intentionality, but related to the program of study. This may be an indirect representation of an intrinsic characteristic, or personality trait. Endogenous electromagnetic fields and patterns have previously been hypothesized to exist and have a role in social interactions that we are not aware of (McDonnell, 2014; Liboff, 2016; Liboff, 2017).

Comparing the spectral characteristics of the photon emissions of the alive vs dead cells found differences in the 2-3 Hz and 19-20 Hz range. Differences around 20 Hz are interesting because this frequency range has previously been associated with the plasma membrane of cancer cells (Persinger & Lafrenie, 2014). In photon measurements the 20 Hz frequency band typically has higher spectral power densities in cancerous cell lines compared to non-cancerous cell lines (Karbowski et al., 2015; Dotta et al., 2016). Perhaps, if the 20 Hz frequency is associated with the membrane, but does not require a functioning cell and arises from the components of the cell membrane, then it is increased in the dead cancerous cell vs alive cancerous because the dead form may represent an increased entropy state of the membrane, which cancer cells have been hypothesized to represent (Persinger & Lafrenie, 2014). Interestingly, Dotta et al. (2016) also found photon emission in the 2.6 Hz band was increased in the healthy tissue vs the cancerous tissue. If this frequency is involved with internal processes related to malignancy, then finding it also increased in dead cancer cells may be indicative of no cancer whatsoever, either dead or non-malignant cells.

In this experiment, we found that MCF-7 breast cancer cells emitted significantly more photons at noon than at 6pm and at midnight (Figure 23). The temperature in the dark room in this experiment should have been fairly uniform, as this data was collected in December (2017) and the greatest temperature change inside the room should have occurred shortly after 10:30pm (the time that they turn the heating off in the building, ref: maintenance worker). While it is possible that these differences were due to changes in the culture media, the effect is more evident on day 2, when there would be more cells in the dish to emit photons, indicating it's more likely this is due to circadian rhythms in the cells.

Are these circadian rhythms or are the cells responding to changes in the external environment? This data cannot be interpreted without taking into consideration the question just proposed, which is something that has been argued among scientists for decades. Have circadian rhythms been incorporated into our DNA and therefore are genetically conserved in cells despite being kept in a temperature- and humidity-controlled incubator? Or is the "machinery" responsible for circadian rhythms just sensitive to environmental variables which have their own circadian rhythms? For example, water uptake of germinating seeds kept in a temperature and humidity controlled incubator not only maintained seasonal variability but also had a correlation value of 0.75 with the temperature outside the building (Spruyt et al., 1987).

It should also be noted how odd it is to find differences in mean photon output of the cell dishes with circadian rhythms and with different stocks of the same cell line, but not when comparing alive vs dead cells. Why this is, is not known.

These experiments produced a few significant effects that, while are interesting in themselves, but did not directly answer the question: can healing intentionality from individuals who practice that as a profession, reduce the growth of cancer cells in culture? The significant results in this experiment of cells from different stocks, "personality" of individual, circadian rhythm in photon emissions, may as a whole indicate the complicated nature of the experimental method. Therefore, when studying subtle phenomenon, the variables that produce large main effects need to be controlled for and homogenized across trials, to increase the sensitivity of the measurements.

References

Buckner, C. A., Buckner, A. L., Koren, S. A., Persinger, M. A., & Lafrenie, R. M. 2015. Inhibition of cancer cell growth by exposure to a specific time-varying electromagnetic field involves t-type calcium channels. PLoS ONE, 10(4): e0124136.

Bureau, Y. R. J. & Persinger, M.A. 1992. Geomagnetic activity and enhanced mortality in rats with acute (epileptic) limbic lability. International Journal of Biometeorology, 36(4):226-232.

Dotta, B. T., Karbowski, L. M., Murugan, N. J., Vares, D. A. E., & Persinger, M. A. 2016. Ultra-weak photon emissions differentiate malignant cells from non-malignant cells in vitro. Archives in Cancer Research, 4: 2.

Fleming, J. L., Persinger, M. A., & Koren, S. A. 1994. Magnetic pulses elevate nociceptive thresholds: Comparisons with opiate receptor compounds in normal and seizure-induced brain-damaged rats. Electro-and Magnetobiology, 13: 67-75. https://doi.org/10.3109/15368379409030699.

Friedman, H., Becker, R. O., & Bachman, C. H. 1963. Geomagnetic parameters and psychiatric hospital admissions. Nature, 200:626-628.

Galic, M. A., & Persinger, M. A. 2007. Lagged association between geomagnetic activity and diminished nocturnal pain thresholds in mice. Bioelectromagnetics, 28: 577-579. doi:10.1002/bem.20353

Hu, J. J., St-Pierre, L. S., Buckner, C. A., Lafrenie, R. M., & Persinger, M. A. 2010. Growth of injected melanoma cells is suppressed by whole body exposure to specific spatial-temporal configurations of weak intensity magnetic fields.

International Journal of Radiation Biology, 86:2, 79-88, DOI: 10.3109/09553000903419932

Hunter, M. D., Mulligan, B. P., Dotta, B. T., Saroka, K. S., Lavallee, C. F., Koren, S. A., & Persinger, M. A. 2010. Cerebral dynamics and discrete energy changes in the personal physical environment during intuitive-like states and perceptions. Journal of Consciousness Exploration and Research, 1, 1179-1197.

Karbowski, L. M., Harribance, S. L., Buckner, C. A., Mulligan, B. P., Koren, S. A., Lafrenie, R. M., & Persinger, M. A. 2012. Digitized quantitative electroencephalographic patterns applied as magnetic fields inhibit melanoma cell proliferation in culture. Neuroscience Letters, 523(2):131-134.

Karbowski, L. M., Murugan, N. J., Dotta, B. T., & Persinge,r M. A. 2015. Only 1% melanoma proportion in non-malignant cells exacerbates photon emissions: Implications for tumor growth and metastases. International Journal of Cancer Research and Molecular Mechanisms, 1(2): doi http://dx.doi.org/10.16966/2381-3318.108

Liboff, A. R. 2016. A human source for ELF magnetic perturbations. Electromagnetic Biology and Medicine, 35:4, 337-342, DOI: 10.3109/15368378.2015.1107841

Liboff, A. R. 2017. The electromagnetic basis of social interactions. Electromagnetic biology and medicine, 36:1-5. 10.1080/15368378.2016.1241180.

Martin, L. J., Koren, S. A., & Persinger, M. A. 2004. Thermal analgesic effects from weak, complex magnetic fields and pharmacological interactions. Pharmacology, Biochemistry and Behaviour, 78(2): 217-227.

McDonnell, A. 2014. The sixth sense-emotional contagion; Review of biophysical mechanisms influencing information transfer in groups. Journal of Behavioral and Brain Science, 4:342-374.

Mulligan, B. P., Hunter, M. D., & Persinger, M. A. 2010. Effects of geomagnetic activity and atmospheric power variations on quantitative measures of brain activity: Replication of the Azerbaijani studies. Advanced Space Research, 45:940-948

Murugan, N. J., Karbowski, L. M., Mekers, W. F., & Persinger, M. A. 2015. Group planarian sudden mortality: Is the threshold around global geomagnetic activity ≥K6?. Communicative & integrative biology, 8(6), e1095413. doi:10.1080/19420889.2015.1095413

Persinger, M. A., & Richards, P. M. 1995. Vestibular experiences of humans during brief periods of partial sensory deprivation are enhanced when daily geomagnetic activity exceeds 15-20 nT. Neuroscience Letters, 194:69-72.

Persinger, M. A., & Healey, F. 2002. Experimental facilitation of the sensed presence: possible intercalation between the hemispheres induced by complex magnetic fields. The Journal of nervous and mental disease, 190 8, 533-41.

Persinger, M. A., Roll, W. G., Tiller, S. G., Koren, S. A., & Cook, C. M. 2002. Remote viewing with the artist Ingo Swann: Neuropsychological profile, electroencephalographic correlates, magnetic resonance imaging (MRI), and possible mechanisms. Perceptual and Motor Skills, 94(3), 927–949. https://doi.org/10.2466/pms.2002.94.3.927

Persinger, M.A., Dotta, B.T., Saroka, K.S., & Scott, M.A. 2013. Congruence of energies for cerebral photon emissions, quantitative EEG activities and ~ 5 nT changes in the proximal geomagnetic field support spin-based hypothesis of consciousness. Journal of Consciousness Exploration and Research. 4. 1-24.

Persinger, M. A., & Lafrenie, R. M. 2014. The cancer cell plasma membrane potentials as energetic equivalents to astrophysical properties. International Letters of Chemistry, Physics and Astronomy, 36:67-77

Smith, A. D, & Laskow, L. 2000. Intentional healing in cultured breast cancer cells. Proceedings of the Academy of Religion and Psychical Research, Annual Conference, 96-100.

Spruyt, E., Verbelen, J., & De Greef, J. 1987. Expression of Circaseptan and Circannual Rhythmicity in the Imbibition of Dry Stored Bean Seeds. Plant Physiology, 84(3), 707-710.

Chapter 6 – General Discussion

This dissertation demonstrates the importance of understanding our Earth's magnetic field. Not only can the geomagnetic field influence mental states, especially for sensitive populations, but the two planaria chapters in this thesis demonstrate that a portion of their population is sensitive as well. This may indicate common mechanisms between humans and planaria. One potential mechanism is that geomagnetic storms act by decreasing the amount of melatonin (Bureau & Persinger, 1992; Kay, 1994; Persinger, 1995b; Burch et al., 1999; Mulligan et al., 2012). Decreased melatonin metabolites after geomagnetic storms have been measured in humans (Burch et al., 1999). Melatonin is a known anti-convulsant (Muñoz-Hoyos et al., 1998) and its suppression has been hypothesized to explain the decreased latencies for seizure onset (Bureau & Persinger, 1995). Inhibition in melatonin synthesis could result in an increase in dopamine production (Bureau & Persinger, 1992). The planarian responses to increased geomagnetic disturbance was an increase in mobility. Antidopaminergic agents, such as antipsychotics, are known to decrease the mobility in planaria (Raffa, Holland, & Schulingkamp, 2001), which is consistent with this idea.

This thesis provides good cautionary data for scientists because of the way that the geomagnetic field can influence an experiment. An experiment conducted in the winter, may not produce the same results as an experiment conducted in the summer. The change in weather variables may be influencing the results, however, one could argue that being in a temperature-controlled room would eliminate this possibility. Scientists are still human, and are therefore still capable of believing that our world consists of what we are able to perceive. The geomagnetic field intensity shows variability over the course of a year, as does atmospheric electricity, as does barometric pressure, as does humidity and as does the frequency that geomagnetic storms occur. This is true as well for the time of day, it is well known that organisms have circadian rhythms, but much research has shown that these may be controlled by circadian rhythms in environmental variables, such as light cycle or tidal variations (Barlow et al., 2012; Moraes et al., 2014), which influence the intensity of the Earth's field (Persinger, 1980; Liboff, 2013). Yet, when discrepancies are found between trials or experiments, they are usually attributed to something in the immediate environment, such as equipment, carelessness, etc. Much like Skinner (1948) showed in his pigeon experiments, where the animals developed superstitious behaviour concerning food release. Because the food was released at random intervals, the pigeon would pair the release with whatever was occurring at the time. This makes consistency in completing experiments important, or if inconsistent results are found, then continually repeating the procedure, as this should elucidate the source of discrepancy.

The threshold for geomagnetic effects seems to consistently be around 20nT for most responses, if plugged into Weber's fraction (Persinger, 1985), with the background intensity of the Earth found in the Neuroscience research labs of ~ 50,000nT (Persinger et al., 2002), would produce a value of 0.04%. We may not have any specialized sensory cells for detecting magnetic fields, but there is still an unconscious detection/influence. This has been demonstrated previously with electroencephalographic activity of the brain (Mulligan et al., 2010), where the right hemisphere was more affected, this being the hemisphere that's typically involved with unconscious processing (Ottoson, 1987 referenced in Scott & Persinger, 2013).

The Weber ratio for sensitivity demonstrates the importance of background sensitivity (Persinger, 1985), this indicates that regions of higher latitude may have different sensitivity and therefore different responses to applied magnetic exposures, since the background intensity would be increased compared to lower latitudes (Persinger, 1980). Changes in background intensity altering effects in experiments with applied magnetic fields was demonstrated by Blackman et al. (1985). Perhaps this can explain the discrepancies in results that are found between studies that are conducted in different geographical locations. For example, O'Connor & Persinger (1996) found a linear increase in thyroxine (T4) levels in a patient with epilepsy, which was hypothesized to be due to a decrease in melatonin, and melatonin is known to inhibit the release of thyroid stimulating hormone. But this contrasts findings of individuals in Svalbard (one of the most northern cities in the world), where increased geomagnetic activity was positively associated with cortisol levels, negatively associated with triiodothyronine (T3) and no relationship with T4 (Breus et al., 2015). Interestingly both of these former relationships were season specific. These results contrast the results of O'Connor & Persinger (1996), but instead of concluding improper experimentation, this may be more indicative of differential sensitivity in different geomagnetic climates.

In another example, Bunevicius et al. (2017) reported a failure in replicating a previously found association between lunar phase and intracranial aneurysm ruptures. They also found two other large studies that also found no association of aneurysm ruptures with lunar phase. Their conclusion for the inconsistency was the use of inappropriate statistical methods. While this is possible, it's important to consider other factors as well, such as geomagnetic climate (as discussed above), potential seasonal interactions with lunar phase (as found in this thesis), and other yet to be discovered variables. In terms of statistical analysis, while including too many variables and finding statistical significance by chance needs to be avoided, sometimes researchers while investigating environmental variables will do the opposite and not consider the potential lag effects of weather variables. For example, Schnabel et al. (2000) sought to replicate the findings of Dr. Persinger (1995), which showed an increased likelihood of sudden unexpected deaths in epileptics during months of high geomagnetic activity. In their study they only included the Ap indices for the estimated time of death, and when they found no relationship, they concluded that therefore there was none. This is negligent, as in studies of changes in sensitive populations following geomagnetic storms there is often a temporal lag effect of anywhere from a few days to one month (Freidman et al., 1963; Persinger, 1995; Kay, 2004; Tada et al., 2014). Geomagnetic storms may reduce a threshold in these individuals and then it may be some other stressful event that precipitates the response. This indicates a potential for experimenter bias, which is analogous to the debate over the McConnell findings in the 1950's with learning in planaria. He showed that planaria took less time to learn a task if they were fed diced planaria that had already learned it (McConnell, 1962). Controversy over these results came about when some researchers failed to replicate the results (Walker, 1966). However, it was pointed out that this may not have had to do with an experimenter biasing their results to find significance, but rather that planaria are sensitive creatures and it may have been that the researchers who found negative results did not handle them properly (Travis, 1981). A well-designed experiment demonstrated that what McConnell found was true, but that it was the cannibalization that improved learning regardless of whether the planaria eaten had been conditioned (Hartry et al., 1964). However, this still demonstrates the importance of the researcher and the experimental design. The previous chapter of this thesis demonstrated the potential that cells will emit different photon emissions when treated by individuals as a function of their program of study. It also demonstrated the care that needs to be taken when planning experiments. The poorly planned experiment may be analogous to the mis-handling planaria hypothesis, which introduces too much variability to find subtle results.

This thesis demonstrates the importance of considering the weather matrix and how it can influence biological organisms in ways we do not yet fully understand. In the Weather Matrix and Human Behaviour, Dr. Persinger (1980) reports and discusses a series of experiments called the Hollander experiments. Briefly, these consisted of experiments conducted in a chamber, which could control temperature, humidity, barometric pressure, and concentrations of positive and negative ions in the air. They recruited participants who had previously suffered joint and arthritic pain to live in the chamber for 2-3 weeks. Over this time the participants were exposed to a multitude of combinations of these weather variables and would report subjective changes in joint pain. They found the weather conditions that produced the most joint pain were decreased barometric pressure and increased humidity. Before discussing applications, Dr. Persinger first points out that while the participants were blind to the weather conditions, the staff that interacted with the participants was not. He then outlines a mechanism by which these two variables would interact to produce the joint pain and the decreased attentional capacity which has also been associated with decreased barometric pressure (Persinger, 1980). However, at the end of this he states "The reader is reminded that the previous discussion is a story dressed in scientific terminology". I would like to steal this line and apply to the present thesis. Because while there seemed to be logical explanations for the results I found, much of how we interact with the electromagnetic medium of the Earth is unknown and therefore as always, the quantitative results have the most value.

References

Blackman, C. F., Benane, S. G., Rabinowitz, J. R., House, D. E., & Joines, W. T. 1985. A Role for the magnetic field in the radiation-induced efflux of calcium ions from brain tissue in vitro. Bioelectromagnetics, 6: 327-337. doi:10.1002/bem.2250060402

Breus, T. K., Boiko, E. R., & Zenchenko, T. A. 2015. Magnetic storms and variations in hormone levels among residents of North Polar area – Svalbard. Life Sciences in Space Research, 4:17-21.

Bunevicius, A., Gendvilaite, A., Deltuva, V. P., & Tamasauskas, A. 2017. The association between lunar phase and intracranial aneurysm rupture: Myth or reality? Own data and systematic review. BMC Neurology, 17 doi:10.1186/s12883-017-0879-1

Burch, J. B., Reif, J. S, & Yost, M. G. 1999. Geomagnetic disturbances are associated with reduced nocturnal excretion of a melatonin metabolite in humans, Neuroscience Letters, 266(3): 209-212.

Bureau, Y. R. J. & Persinger, M.A. 1992. Geomagnetic activity and enhanced mortality in rats with acute (epileptic) limbic lability. International Journal of Biometeorology, 36(4):226-232.

Bureau, Y. R. J. & Persinger, M.A. 1995. Decreased latencies for limbic seizures induced in rats by lithium-pilocarpine occur when daily average geomagnetic activity exceeds 20 nanoTesla. Neuroscience Letters, 192(2): (142-144). https://doi.org/10.1016/0304-3940(95)11624-6

Friedman, H., Becker, R. O., & Bachman, C. H. 1963. Geomagnetic parameters and psychiatric hospital admissions. Nature, 200:626-628.

Hartry, A. L., Keith-Lee, P., & Morton, W. D. 1964. Planaria: Memory transfer through cannibalism reexamined. Science, 146(3641), 274–275. doi:10.1126/science.146.3641.274

Kay, R. 1994. Geomagnetic storms: Association with incidence of depression as measured by hospital admission. British Journal of Psychiatry, 164(3), 403-409. doi:10.1192/bjp.164.3.403

McConnell, J. V. 1962. Memory transfer through cannibalism in planarians. Journal of Neuropsychiatry, 3:532-548

Mulligan, B. P., Hunter, M. D., & Persinger, M. A. 2010. Effects of geomagnetic activity and atmospheric power variations on quantitative measures of brain activity: Replication of the Azerbaijani studies. Advanced Space Research, 45:940-948

Mulligan, B. P., & Persinger, M. A. 2012. Experimental simulation of the effects of sudden increases in geomagnetic activity upon quantitative measures of human brain activity: Validation of correlational studies. Neuroscience Letters, 516(1):54-56.

Muñoz-Hoyos, A., Sánchez-Forte, M., Molina-Carballo, A., Escames, G., Martin-Medina, E., Reiter, R. J., Molina-Font, J. A., & Acuña-Castroviejo, D. 1998. Melatonin's role as an anticonvulsant and neuronal protector: Experimental and clinical evidence. Journal of Child Neurology, 13(10), 501–509. https://doi.org/10.1177/088307389801301007

O'Connor, R. P., & Persinger, M. A. 1996. Brief communication: Increases in geomagnetic activity are associated with increases in thyroxine levels in a single patient: Implications for melatonin levels. International Journal of Neuroscience, 88: 243-247.

Persinger, M. A. 1980. The Weather Matrix and Human Behaviour. Praeger Publishers. New York, New York.

Persinger, M. A. 1985a. Letter: Classical psychophysics and ELF magnetic field detection. Journal of Bioelectricity, 4:2, 577-584, DOI: 10.3109/15368378509033272

Persinger, M. A. 1995b. Sudden unexpected death in epileptics following sudden, intense, increases in geomagnetic activity: Prevalence of effect and potential mechanisms. International Journal of Biometeorology, 38: 180. https://doi.org/10.1007/BF01245386.

Persinger, M. A., Roll, W. G., Tiller, S. G., Koren, S. A., & Cook, C. M. 2002. Remote viewing with the artist Ingo Swann: Neuropsychological profile, electroencephalographic correlates, magnetic resonance imaging (MRI), and possible mechanisms. Perceptual and Motor Skills, 94:927-949.

Raffa, R. B., Holland, L. J., & Schulingkamp, R. J. 2001. Quantitative assessment of dopamine D2 antagonist activity using invertebrate (planaria) locomotion as a functional endpoint. Journal of Pharmacological and Toxicological Methods, 45(3): 223-226.

Schnabel, R., Beblo, M., & May, T. W. 2000. Is geomagnetic activity a risk factor for sudden unexplained death in epilepsies? Neurology, 54 (4):903-908; DOI: 10.1212/WNL.54.4.903

Skinner, B. F. 1948. 'Superstition' in the pigeon. Journal of Experimental Psychology, 38(2), 168-172. http://dx.doi.org/10.1037/h0055873

Tada, H., Nishimura, T., Nakatani, E., Matsuda, K., Teramukai, S., & Fukushima, M. 2014. Association of geomagnetic disturbances and suicides in Japan, 1999-2010. Environmental health and preventive medicine, 19(1), 64-71.

Travis, G. 1981. Replicating replication? Aspects of the social construction of learning in planarian worms. Social Studies of Science, 11(1), 11-32. Retrieved from http://www.jstor.org/stable/284734

Walker, D. R. 1966. Memory transfer in planarians: An artifact of the experimental variables. Psychonomic Science, 5: 357. https://doi.org/10.3758/BF03328437