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"Silent Signals, Visible Effects: Behavioural Disruption by Low-Frequency EMFs in *Drosophila jambulina*"

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Abstract

The fruit fly *D. jambulina* serves as a valuable model organism for investigating the molecular pathways underlying numerous human conditions, including cancer, heart disease, and neurological disorders. This study assessed the effects of 50 Hz low-frequency electromagnetic field exposure on key behavioural traits of *D. jambulina*, including larval crawling, locomotion, climbing, courtship behaviour and aggression. Flies were exposed to EMFs using a Holzmolt coil for short- and long-term durations. Behavioural assays were conducted using standard methods such as crawling grid tracking, the RING test, and timed interaction observations. Exposure to LF-EMF significantly reduced larval movement and adult climbing ability in a time-dependent manner, with males showing greater sensitivity. Courtship behaviours declined progressively with longer exposure, indicating reproductive disruption. Aggression initially increased, especially in males, but later declined due to likely neuromodulator depletion.

Overall, LF-EMF exposure impaired neural and behavioural functions in *D. jambulina*, with effects linked to oxidative stress, neurotransmitter disruption, and possible sex-specific resilience. These findings suggest *D. jambulina* is a useful model for studying EMF-induced neurotoxicity and environmental stress.

Keywords – *D. jambulina*, LF-EMF, Aggressive behaviour, larval crawling activity, Climbing assay, Courtship behaviour, neuromodulators.

Introduction

Electromagnetic fields are invisible areas of energy, commonly associated with the use of electrical power and various forms of natural and man-made lighting. Among these, low-frequency electromagnetic fields, particularly those at 50 Hz generated by household and industrial power lines, have become increasingly pervasive due to widespread technological advancement (Roosli,2008). Although non-ionizing in nature and considered relatively weak compared to ionizing radiation, LF-EMFs can interact with biological systems in subtle but potentially significant ways, especially under prolonged or repeated exposure (Blank and Goodman, 2009).

Drosophila species, due to their short generation time, well-characterized genetics, and physiological similarities to higher organisms, have long served as effective model organisms for toxicological and environmental studies (Pandey and Nichols, 2011). Among these, *D. jambulina*, a lesser-known yet ecologically relevant species in South Asia, has recently gained attention for investigating environmental stressors, owing to its adaptability and sensitivity to genetic and behavioural perturbations (Sharma and Ranganath, 1987).

Several studies have reported that exposure to EMFs can induce oxidative stress, behavioural alterations, reproductive dysfunction, and genotoxic effects in *D. melanogaster* and other related species (Liu et al., 2015; Ayra-Pardo et al., 2022). LF-EMF exposure has been linked with increased reactive oxygen species (ROS) production, which in turn may damage DNA, proteins, and lipids, thereby impairing normal cellular functioning (Akdag et al., 2013). Moreover, behavioural assays have shown changes in locomotion, courtship and aggression in flies exposed to EMFs, suggesting a potential neuromodulator effect (Weisbrot et al., 2003).

However, limited research has been conducted on *D. jambulina* in the context of LF-EMF exposure, and its physiological, behavioural, and genetic responses remain largely unexplored. Given the species ecological significance and its potential as a model organism, it is imperative to investigate how chronic or acute exposure to LF-EMFs might affect its biology. Such investigations not only help in understanding fundamental mechanisms of EMF interactions with living organisms but also contribute to environmental risk assessment, especially in areas with high electromagnetic pollution.

The present study aims to evaluate the effects of 50 Hz LF-EMF exposure on various biological parameters of *D. jambulina*, including behavioural patterns, specially focus on larval crawling activity, locomotor activity, climbing activity, courtship behaviour and aggression behaviour, this research seeks to uncover how electromagnetic pollution influences insect health and provides insights that may be extrapolated to other organisms, including humans.

Research Methodology

EMF model

The 50 Hz electromagnetic field exposure model is a widely accepted experimental setup used to study the biological effects low-frequency, non-ionizing radiation. In this model, alternating current (AC) at 50 Hz is passed through a Holzmolt wire coil, generating a controlled and uniform EMF. This model has two modules first module consist an EMF chamber with winding copper wire and the second module is electronic module which regulate the EMF frequency. This simulates real-world exposure scenarios like those near power lines or electrical appliances. Organisms such as *D. jambulina* are exposed within this coil chamber to assess the effects on various biological endpoints, including behaviour, oxidative stress, reproduction, and genetic integrity. The system offers precise control, reproducibility, and environmental relevance, making it a valuable tool for both acute and chronic EMF exposure research.

Drosophila stock rearing and handling.

Collected of *D. jambulina* strain from the National Centre for Drosophila, Department of Zoology, University of Mysore, Karnataka, India. Took caution during all fly manipulation, sedation, and relocation procedures to reduce physical trauma and death rates. Daily observations were conducted to assess fly viability, behavioural patterns, and developmental progress. Any containers displaying contamination indicators or signs of dehydration were immediately removed and replaced with fresh ones.

Laboratory stocks of *D. jambulina* were maintained in a controlled insectary environment with standardized conditions of 25 ± 1 °C temperature, 60–70% relative humidity, and a 12:12 h light–dark photoperiod. drosophila was reared in standard cylindrical plastic vials (25 mm diameter × 95 mm height) containing 12 mL of freshly prepared Drosophila culture medium. Additionally, 200 mL glass bottles were employed for mass cultures, each provided with 25 mL of food medium.

Drosophila Food

The nutritional quality and consistency of the rearing medium are critical for the healthy development, fecundity, and experimental reproducibility in Drosophila studies. The standard food medium used for *D. jambulina* consisted of a carbohydrate-rich and protein-balanced substrate designed to support all stages of the life cycle.

Table 1. The food was prepared using the following ingredients (per Liter of medium).

Sr. No.	Ingredients	Amount of ingredients	Nutritional significance
1	Semolina	100 g	Provides carbohydrates and bulk
2	Yeast (active dry)	12 g	Primary source of protein, vitamins, and micronutrients
3	Jaggery	80 g	Additional energy source
4	Agar	8 g	Solidifying agent
5	Methylparaben (Nipagin)	1.5 g in 5 mL of 95% ethanol	Antifungal preservative
6	Propionic acid	4.4 mL	Prevents Mold and bacterial contamination
7	Distilled water	1 litre	Used for mixing ingredients

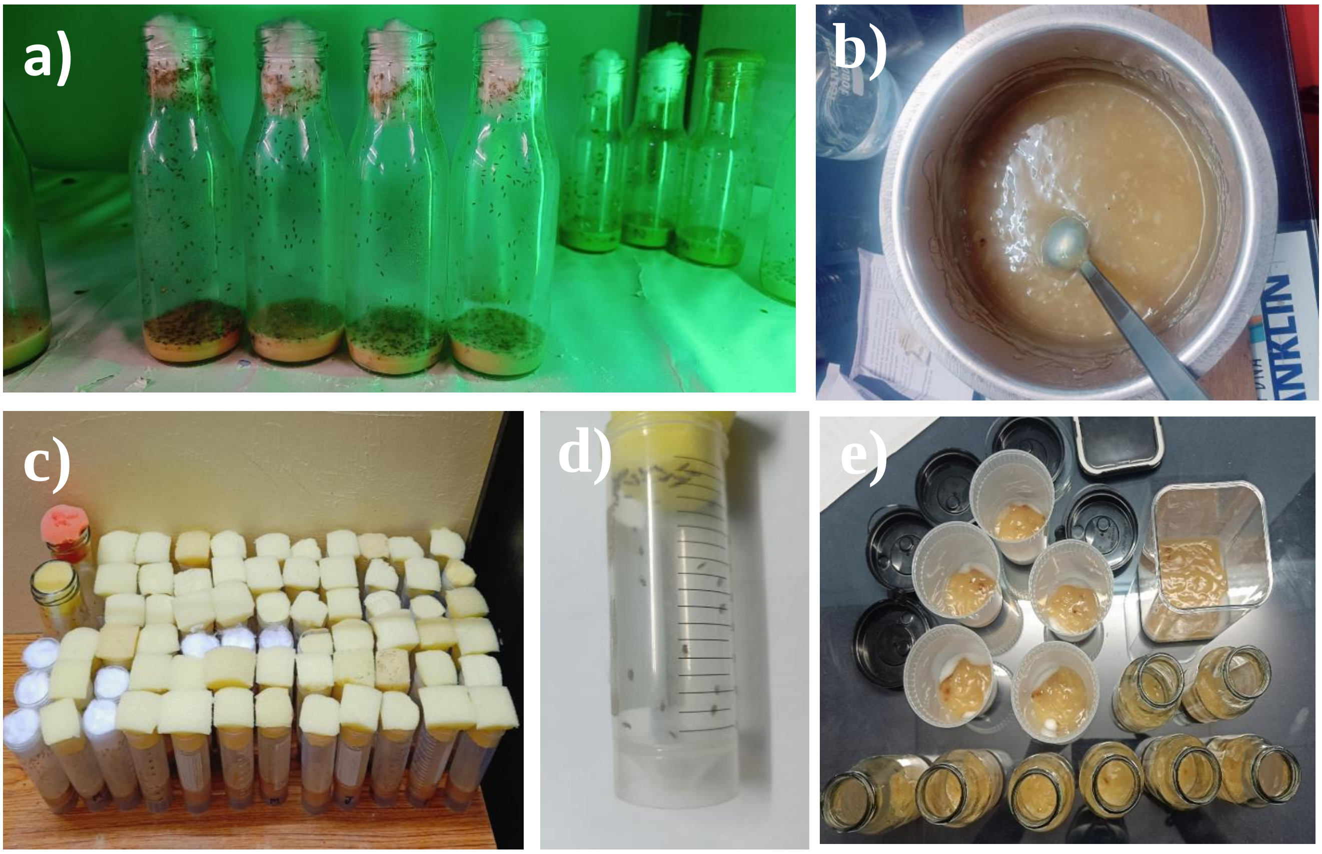


Figure 1.a) D. jambulina stock culture maintained under controlled laboratory conditions. (b) Preparation of standard semolina-agar food medium for Drosophila culture. (c) Experimental cultures derived from stock for behavioural assays. (d) Experimental vial containing D. jambulina used in the climbing (RING) assay. (e) Pouring prepared food medium into different types of vials for culture maintenance and experiments.

A. ***Larval crawling assay***

The larval crawling assay was used to evaluate the effects of (LF-EMF) exposure on the locomotor behaviour of *D. jambulina* third instar larvae. Larvae were first floated using 20% sucrose solution and collected using a pipette. After exposure to short-term or long-term LF-EMF, individual larvae were transferred with a wet paintbrush to the center of a moistened Petri dish placed over graph paper. (Nichols, C.D., Pandey, U.B. 2012) A thin layer of water prevented pupation and allowed smooth movement. Crawling behaviour was recorded for at least 10 seconds of continuous, uninterrupted movement. Trials were repeated until the desired number of valid recordings was obtained.

B. ***Locomotor assay***

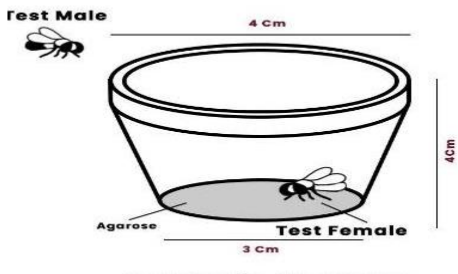
Larval locomotion in *D. jambulina* is influenced by environmental factors such as temperature and nutrient levels. This study examines whether (LF-EMF) also affect larval movement, specifically peristaltic contractions. this assay same as larval crawling assay. Third instar larvae were exposed to LF-EMF for short (15–60 min) and long (2–8 hr) durations. Peristaltic contractions were counted under a microscope for 1 minute per larva. The assay was repeated for multiple larvae per group.

C. ***Climbing Assay / RING Assay.***

Newly eclosed adult *D. jambulina* were collected, anesthetized, and maintained in standard food vials (30 flies per vial), then allowed to recover at room temperature for 3 days. For assays, groups of 10 unanaesthetised flies were transferred to vials of a RING apparatus and exposed to LF-EMF in an EMF chamber for short-term (15–60 min) or long-term (2–8 h) durations, followed by a 1 h acclimation. The RING apparatus was positioned 1 m in front of a mobile phone with fixed zoom and focus. To initiate climbing, the apparatus was tapped three times, and photographs were taken 3 s post-tap to record climbing behaviour. Each group was tested 5–6 times with 1 min intervals between trials (Gargano et al., 2005). Climbing height was quantified from images using a scale, and mean values per vial were calculated. Data from all exposure groups were statistically analysed to evaluate LF-EMF effects on climbing performance **(**Pandey and Nichols, 2011).

D. ***Courtship behaviour/ mating frequencies.***

Courtship in Drosophila is an innate, gene-regulated behaviour controlled by genes such as fruitless (fru) and double sex (dsx), is influenced by sensory cues and environmental factors (Villella and Hall, 2008; Spieth, T. ,1974) studies suggest that LF-EMF can affect neural and behavioural responses, including courtship (Weisbrot et al., 2003). This study investigates LF-EMF effects on *D. jambulina* courtship. Virgin males and females were collected within 2 hours of eclosion, aged separately for 5 days, and exposed to 50 Hz LF-EMF for short-term (8–32 h) or long-term (2–8 d) durations. Courtship was tested by pairing one male and one female in a clean transparent chamber, observing behaviours such as initiation and copulation for 10 minutes. Ten pairs were examined per group, with materials including ether, culture medium, mating chambers, stopwatch, and optionally a camera. Chambers were sterilized with 70% ethanol between trials to remove scent cues.



E. ***Aggressive behaviour***

Aggression is a vital, evolutionarily conserved behaviour important for survival and reproduction. In recent years, drosophila has become a powerful model for studying the genetic and neural basis of aggression due to its well-developed genetic tools (Kravitz and Fernandez, 2015). Environmental stressors, such as social isolation, are known to increase aggressive behaviours, while social enrichment tends to suppress it (Kim et al., 2018; Agrawal et al., 2020). Adult flies were collected from laboratory stock and distributed into standard food vials, with approximately 10 flies per vial. These vials were allowed to acclimate at room temperature for 1 hour. Following acclimation, the flies were exposed to LF-EMF for varying durations short-term (15, 30, 45, 60 minutes) and long-term (75, 90, 105, 120 minutes). After exposure, flies were observed for aggressive interactions, and behavioural responses were recorded and quantified as percentages.

Result And Discussion

1. **Larval Crawling Activity.**

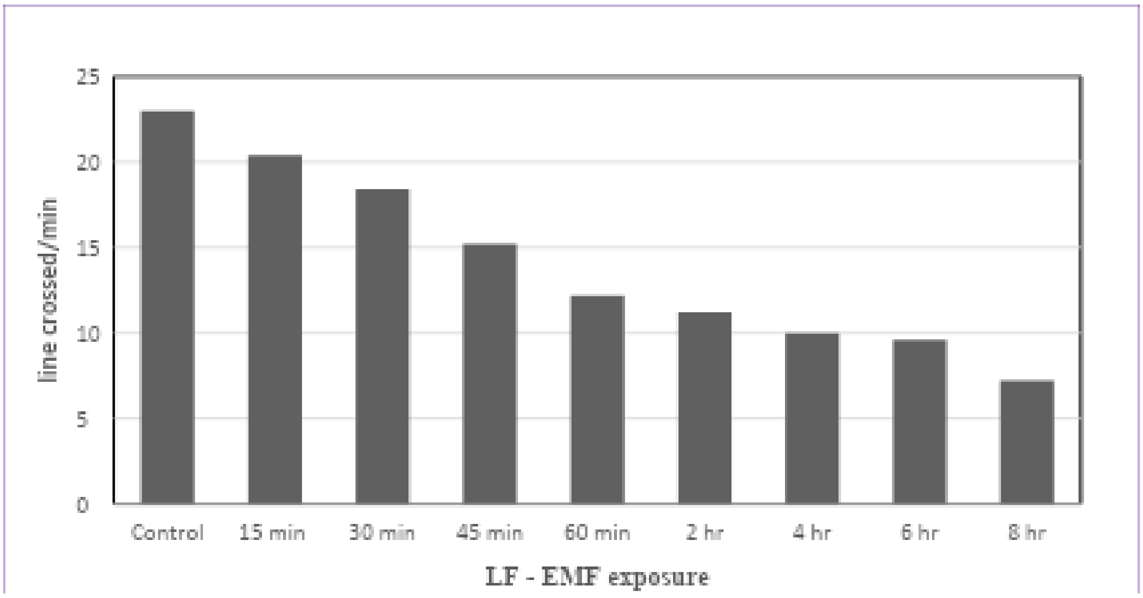


Table 2. rapid iterative negative geotaxis (RING) Activities in D. jambulina.

Exposure to 50 Hz LF-EMFs causes a clear dose-dependent decline in larval crawling of D. jambulina. Control larvae displayed normal activity (23 lines/min), while performance declined progressively with exposure, reaching severe impairment at 8 h (7.2 lines/min). ANOVA ($p < 0.0001$) and Tukey's tests confirmed significant reductions, especially beyond 60 min. The impairment likely arises from LF-EMF–induced disruption of neural circuits, oxidative stress, and mitochondrial dysfunction (Pall et. al., 2013), consistent with reports in D. melanogaster showing ROS elevation, lipid peroxidation, and locomotor deficits under LF-EMF exposure (Sepehrimanesh et al., 2014; Siddique and Naz, 2022). These findings suggest cumulative neurotoxic effects of LF-EMFs on locomotor function.

2. **Locomotor Activity.**

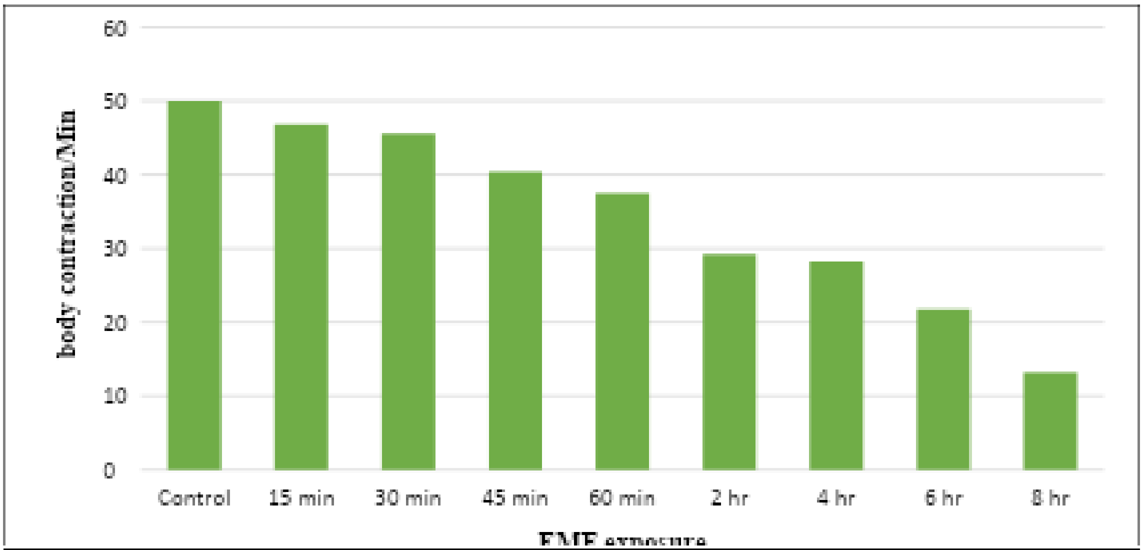
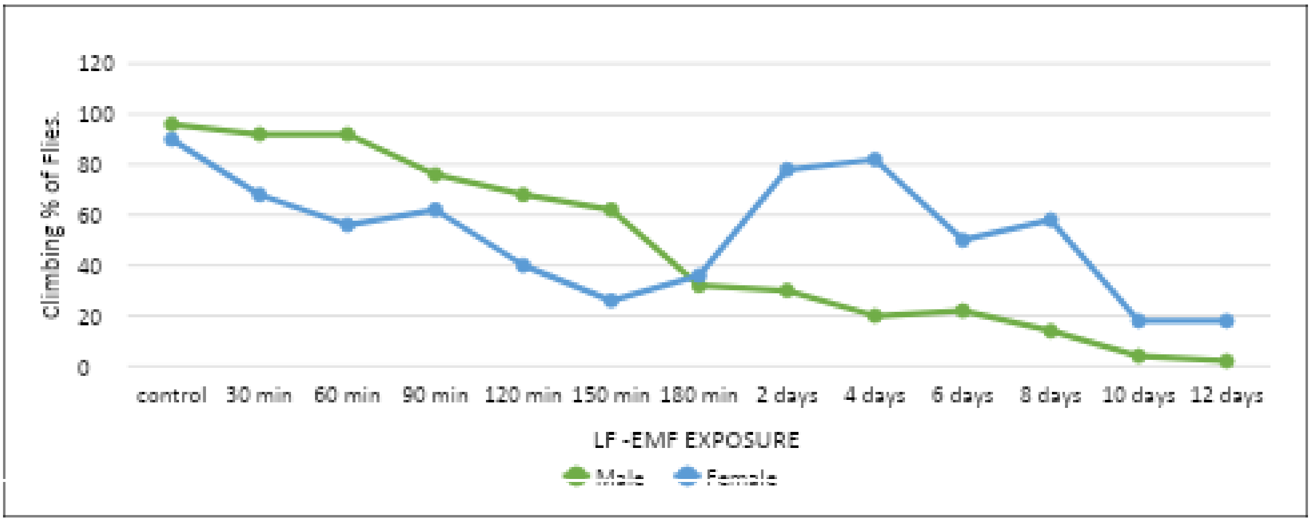


Table 3. Locomotor Activity under LF-EMF Exposure.

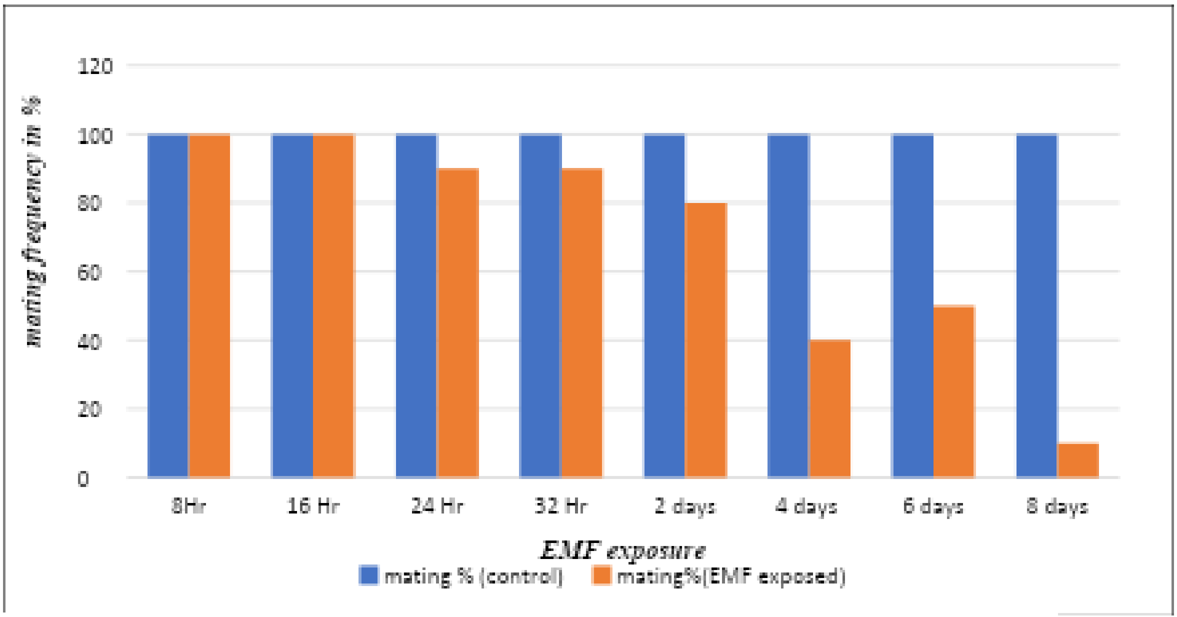
One-way ANOVA demonstrated a highly significant impact of LF-EMF exposure on larval locomotor activity in *D. jambulina* ($F (8, 36) = 168.78$, $p < 0.0001$). Post hoc comparisons using Tukey's test revealed that short-term exposures (15–30 min) produced moderate reductions in body contractions per minute, whereas prolonged exposures (2 h) induced a pronounced and statistically significant decline. The most severe impairment was observed in the 8 h exposure group, where mean body contraction frequency was reduced by approximately 74% relative to the control.

3. **Climbing (RING) Activity.**



Exposure to 50 Hz LF-EMFs significantly impairs dopaminergic-dependent climbing (negative geotaxis) in *D. jambulina*, likely through oxidative stress, disrupted neurotransmission, and motor neuron damage (Sepehrimanesh et al., 2014; Balmori, 2015; Misa-Agustiño et al., 2015). While females exhibit a transient recovery at 2–4 days, possibly reflecting hormonal or antioxidant buffering, males display a progressive decline, consistent with sex-specific vulnerability (Pall et al., 2013; Fedele, G. et al. 2014). The sustained reduction in climbing ability over 6–12 days suggests cumulative neurotoxicity involving mitochondrial dysfunction, neuronal degeneration, and potential genetic alterations that compromise locomotor regulation.

4. **Courtship Behaviours/ Mating frequencies.**



Chronic exposure to 50 Hz LF-EMFs causes a progressive, time-dependent decline in mating frequency in D. jambulina, paralleling findings in D. melanogaster where EMFs reduced copulation duration, delayed mating success, and altered sexual behaviour (Panagopoulos et al., 2010). This impairment likely arises from neurophysiological disruption of dopaminergic and serotonergic circuits (Pall, 2013), oxidative stress leading to neuronal and reproductive tissue damage (Sepehrimanesh et al., 2014), and possible interference with pheromone-mediated signalling that affects receptivity and courtship drive (Singh et al., 2022). The contrast between stable controls and declining EMF exposed groups, along with greater variability under exposure, underscores the sensitivity of reproductive behaviour to LF-EMF-induced stress.

5. **Aggressive Behaviour**

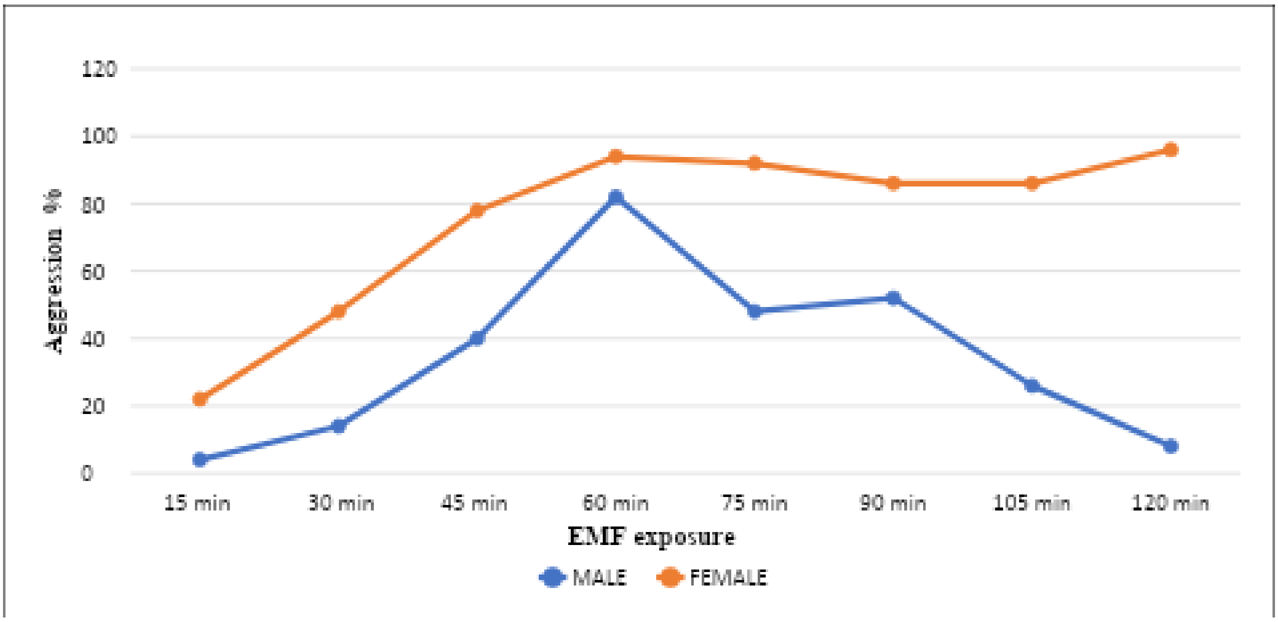


Table 6. Aggressiveness of D. jambulina.

Exposure to 50 Hz LF-EMFs elicits sex-dependent aggression responses in *D. jambulina,* acting as a neurological stressor that rapidly activates serotonergic, dopaminergic, and octopaminergic pathways central to aggression (Panagopoulos et al., 2010). Females showed higher baseline aggression and greater stability, likely due to stronger octopaminergic activity and oxidative stress tolerance, males exhibited a sharp aggression peak at 60 min followed by decline, suggesting neural fatigue, oxidative damage, or neurotransmitter depletion. Prolonged exposure (90–120 min) further reduced male aggression, reflecting behavioural exhaustion or neurotoxicity. These findings are consistent with reports that LF-EMFs disrupt circadian regulation, locomotion, and social behaviours in Drosophila (Siddique and Naz, 2022).

Conclusion

This study demonstrates that 50 Hz low-frequency electromagnetic field (LF-EMF) exposure significantly impairs the behavioural and physiological functions of *D. jambulina* in a dose- and time-dependent manner. EMF exposure disrupted larval crawling and locomotion, impaired adult climbing ability, reduced courtship, and mating success, and altered aggression patterns effects likely mediated by oxidative stress, neurotransmitter imbalance, and neuromuscular dysfunction.

Sex-specific responses were observed, with females showing temporary behavioural resilience, possibly due to hormonal or antioxidant defences. The progressive decline in behavioural performance suggests cumulative neurotoxicity and metabolic stress.

These findings highlight *D. jambulina* as a sensitive bioindicator for environmental EMF stress and underscore the broader ecological risks posed by increasing electromagnetic pollution. Further molecular studies are recommended to explore underlying mechanisms and long-term impacts.

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Conflict Of Interest

The author declares that there is no conflict of interest regarding the publication of this research work.

References

1. Agrawal, P., Kao, D., Chung, P., and Looger, L. L. (2020) The neuropeptide Drosulfakinin regulates social isolation-induced aggression in Drosophila. Journal Experimental Biology.2: jeb207407. https://doi.org/10.1242/jeb.207407
2. Akdag, M. Z., Dasdag, S., Ulukaya, E., Uzunlar, A. K., and Kurt, M. A. (2013). Effects of extremely low-frequency magnetic field on caspase activities and oxidative stress values in rat brain. *Biological Trace Element Research*, 152(3), 324–329.https://doi.org/10.1007/s12011-013-9624-5
3. Ayra-Pardo, C., Sierra, C. V., and Arencibia, D. I. (2022). Behavioral and molecular responses of *Drosophila melanogaster* to electromagnetic fields: A systematic review. *Electromagnetic Biology and Medicine*, 41(1), 1–15.
4. Balmori, A. (2014). *"Electromagnetic pollution from phone masts. Effects on wildlife."* Pathophysiology, 22(2), 117–123.
5. Blank, M., and Goodman, R. (2009). Electromagnetic fields stress living cells. *Pathophysiology*, 16(2-3), 71–78. https://doi.org/10.1016/j.pathophys.2009.01.006
6. Fedele, G. et al. (2014). An electromagnetic field disrupts negative geotaxis in Drosophila via a CRY-dependent pathway. Nat. Commun. 5:4391 doi: 10.1038/5391
7. Gargano, J.W., Martin, I., Bhandari, P., and Grotewiel, M.S. Rapid iterative negative geotaxis (RING): a new method for assessing age-related locomotor decline in Drosophila. Exp. Gerontol. 40(5), 386-95 (2005) https://doi.org/10.1080/15368378.2022.2032055
8. Kim, Y. K., Saver, M., Simon, J., et al., (2018) Repetitive aggressive encounters generate along-lasting internal state in Drosophila melanogaster males. Proceedings National Academy Sciences USA.115: 297-303. https://doi.org/10.1073/pnas.1716612115
9. Kravitz, E. A. and Fernandez, M. de la P. (2015) Aggression in Drosophila. Behavioral Neuroscience. 5: 549–563. https://doi.org/10.1037/bne0000089
10. Liu, Y., Li, X., Zhang, Z., Zhou, J., Zhang, X. (2015). Exposure to 50 Hz magnetic field induces oxidative stress in *Drosophila melanogaster*. *International Journal of Radiation Biology*, 91(12), 1057–1064. https://doi.org/10.3109/09553002.2015.1106025
11. Misa-Agustiño, M.J., et al. (2015). *"Oxidative stress and behavioral changes in zebrafish exposed to ELF-EMF."* Environmental Toxicology and Pharmacology, 40(3), 1090–1096.
12. Nichols, C.D., Pandey, U.B. (2012) Methods to assay Drosophila behavior. J. Vis. Exp. e3795, DOI: 10.3791/3795.
13. Pall, M.L. (2013). *"Electromagnetic fields act via activation of voltage-gated calcium channels to produce beneficial or adverse effects."* Journal of Cellular and Molecular Medicine, 17(8), 958–965. https://doi.org/10.1111/jcmm.12088
14. Panagopoulos, D.J., et al. (2010). *"Bioeffects of mobile telephony radiation in relation to its intensity, distance and duration of exposure." International Journal of Radiation Biology*, 86(5), 345–357. https://doi.org/10.3109/09553000903567961
15. Panagopoulos, D.J., et al. (2015). *"Mechanisms of EMF effects on animal systems: Stress protein activation and ROS generation." Reviews on Environmental Health*, 30(2), 107–117.

16. Pandey, U. B., and Nichols, C. D. (2011). Human disease models in *Drosophila melanogaster* and the role of the fly in therapeutic drug discovery. *Pharmacological Reviews*, 63(2), 411–436. https://doi.org/10.1124/pr.110.003293
17. Roosli, M. (2008). Radiofrequency electromagnetic field exposure and non-specific symptoms of ill health: A systematic review. *Environmental Research*, 107(2), 277–287. https://doi.org/10.1016/j.envres.2008.02.003
18. Sepehrimanesh, M., et al. (2014). *"Effects of 50 Hz electromagnetic field exposure on the antioxidant defense system and enzyme activity in the brain of male rats."* Electromagnetic Biology and Medicine, 33(2), 139–143. https://doi.org/10.3109/15368378.2013.801351
19. Sharma, G. P., and Ranganath, H. A. (1987). *Drosophila jambulina*: A suitable species for cytogenetic and evolutionary studies. *Genetica*, 75(2), 145–152.
20. Siddique, Y.H. and Naz, F. (2022). *"EMF exposure in Drosophila: effects on motor activity, oxidative stress and genetic damage."* Genetika, 54(1), 347–357.
21. Singh, R., Yadav, R., and Siddique, Y.H. (2022). *"Electromagnetic field-induced reproductive and behavioral effects in Drosophila: A review." Genetika*, 54(1), 347–357.
22. Spieth, T. (1974) Courtship behaviour in *Drosophila*, Annual Review Entomology, 19:385405
23. Villella, A., and Hall, J. C. (2008). Neurogenetics of courtship and mating in *Drosophila*. *Advances in Genetics*, **62**, 67–184. https://doi.org/10.1016/S0065-2660(08)00603-2
24. Weisbrot, D., Lin, H., Ye, L., Blank, M., and Goodman, R. (2003). Effects of mobile phone radiation on reproduction and development in *Drosophila melanogaster*. *Journal of Cellular Biochemistry*, 89(1), 48–55. https://doi.org/10.1002/jcb.10416