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Direct suppressive effects of weak magnetic fields
(50 Hz and $16^{2/3}$ Hz) on melatonin synthesis in the pineal
gland of Djungarian hamsters (*Phodopus sungorus*)

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Abstract

In many investigations performed thus far on rodents, a suppression of melatonin synthesis was observed when animals were exposed to weak magnetic fields. However, among the several issues not yet resolved is the question whether the observed changes are caused by direct effects on the pineal gland or by indirect effects, e.g., at the level of the eyes. We, therefore, performed a series of experiments in which direct effects of weak magnetic fields were studied in isolated pineal glands of Djungarian hamsters (*Phodopus sungorus*). After sacrifice of animals during morning hours, pineal glands were removed and placed individually into glass chambers that were perfused with oxygenated buffer. Experiments ($n = 8$) lasted for 8 hours. Magnetic fields ($16^{2/3}$ or 50 Hz at 86 μ Tesla) were generated by Helmholtz coils, and were present during the entire period. In each experiment, 12 exposed, and 12 sham-exposed, pineal glands were placed individually in glass chambers which were surrounded by identical coils. Because of the design of the coils (bifilament) and the experimental setup (black box) the experimentator was not aware which coil was activated until the results were obtained. After 3.25 hours of adaptation, melatonin production was stimulated by isoproterenol (10^{-7} M) for 30 min. Eluted fractions were collected, and melatonin concentrations were measured by radioimmunoassay. Maximum melatonin production was achieved after 5-7 hours. In all experiments, maximum melatonin concentrations were lower in the exposed groups compared to the sham-exposed controls. Statistical analyses for each frequency showed significant suppressive effects at $16^{2/3}$ Hz ($P < 0.01$) and 50 Hz ($P < 0.001$). It is concluded that the suppressive effects of magnetic fields on the synthesis of melatonin are a result of primary mechanisms at the level of the pineal gland.

Introduction

It has long been suspected that magnetic or electromagnetic fields are a possible health risk. The available data of epidemiological studies, however, are rather inconsistent [Feychting and Ahlbom, 1995; [McCann et al., 1998](#); Moulder, 1998; [Repacholi, 1998](#); [McBride et al., 1999](#); [Repacholi and Greenebaum, 1999](#)]. In some cases, however, low frequency magnetic fields seem to be associated with increased risks for cancer and childhood leukemia [Dockerty et al., 1998; Hatch et al., 1998; Wartenberg, 1998]. The primary mechanism by which these fields may interact with biological systems is not yet clarified.

In this regard, the pineal gland and its hormone melatonin may play a central role. According to the melatonin hypothesis [[Stevens and Davis, 1996](#)], the primary effect of exposure is a reduction of the synthesis of melatonin which finally results in secondary endocrine effects which, in turn, may promote growth of cancer. Alternatively, melatonin's function as a potent radical scavenger may be a key which links melatonin with cancer. Thus, if the concentration of melatonin is low, higher amounts of oxygen radicals may lead to DNA damage which in turn would increase cancer risk [[Reiter et al., 1999](#)].

In fact, it has been frequently demonstrated that low frequency magnetic fields are able to affect the function of the pineal gland in animals and humans [[Lerchl et al., 1990](#); Grota et al., 1994; Kato et al., 1994; [Selmaoui and Touitou, 1995](#); [Mevisen et al., 1996](#); [Pfluger and Minder, 1996](#); John et al., 1998; Karasek et al., 1998; [Lerchl et al., 1998](#); [Wood et al., 1998](#); Burch et al., 1999; [Wilson et al., 1999](#)]. However, these studies often yielded inconsistent results even when the conditions of exposure seemed to be identical [[Truong and Yellon, 1997](#); [Löscher et al., 1998](#); [Reiter et al., 1998](#); Yellon and Truong, 1998].

There are only few studies addressing the question of whether magnetic fields may interfere with the pineal gland directly or indirectly, e.g., via the eyes [[Olcese et al., 1988](#)]. It was shown that the isolated pineal gland of rats responds to intermittent magnetic field exposure with a reduced melatonin content and lowered levels of N-acetyltransferase (NAT), the rate-limiting enzyme of the melatonin biosynthesis [[Richardson et al., 1992](#)]. Another experiment involving isolated

pinealocytes likewise revealed lowered melatonin concentrations in the culture media [[Rosen et al., 1998](#)].

The aim of the present investigation was threefold: first, the previous studies on direct effects on pinealocytes/pineal glands were tested at two important frequencies, $16^{2/3}$ Hz and 50 Hz. These frequencies correspond to those generated by household appliances and high-power transmission lines (50 Hz), and by the railway system ($16^{2/3}$ Hz), respectively. Second, the variability between the results of repeated experiments was analyzed; third, the production of melatonin in a continuous flow setup was the endpoint rather than the static concentration of melatonin within the glands. The latter point is important because melatonin in the pineal gland may be unchanged because of exposure while the secretion is suppressed, as reflected by serum concentration [Grota et al., 1994].

Material and methods

Organ preparation and culture

For each experiment, 24 pineal glands were used; they were divided into two groups of 12 (sham-exposed and exposed). The exposure system was located inside an incubator which was maintained at 37°C. For temperature accuracy, the incubator was activated 24 hr before the start of the experiment. Adult female Djungarian hamsters, born and raised in our breeding colony [Lerchl, 1995], were decapitated during morning hours (between 08:00 and 09:00 hr), the pineal gland and a small piece of attached skull was removed and placed into a glass tube (0.5 cm I.D., 1 cm length) which was closed with rubber stoppers at both ends. Krebs-Ringer buffer (Sigma, Deisenhofen, Germany), supplemented with tryptophan (10^{-4} M), was kept in the incubator for at least 12 hr prior to the experiment, and was pumped through the tubes at a flow rate of 0.4 mL/min. The buffer was gassed continuously with 95% CO₂ and 5% O₂. To prevent the occurrence of gas bubbles, the buffer was pumped through the tubes from the bottom. The eluates were collected in hourly intervals for 8 hr except for the second experiment at $16^{2/3}$ Hz (7 hr). After an initial period of 3.25 hr, the pineal glands were stimulated for 30 min with isoproterenol (10^{-7} M) to initiate melatonin synthesis.

Exposure system

The 12 glass tubes from each group were located inside the active area of Helmholtz coils (diameter 10 cm, distance 5 cm, Fig. 1). They were constructed with double filament wires (Fig. 2), and fastened to a black box which was connected to the generators. Inside the box, switches were turned on or off, so that one coil was active (current flow only through one filament), the other was inactive (current flow in opposite directions through both filaments). By this procedure, the inactive coil produced no magnetic field while any other parameter was identical between the two coils. The switches inside the box were set by a person that was not involved in the experiments. The box was locked, and only after the complete analysis of each data set was the code broken. The pineal glands were exposed or sham-exposed for the entire experimental duration.

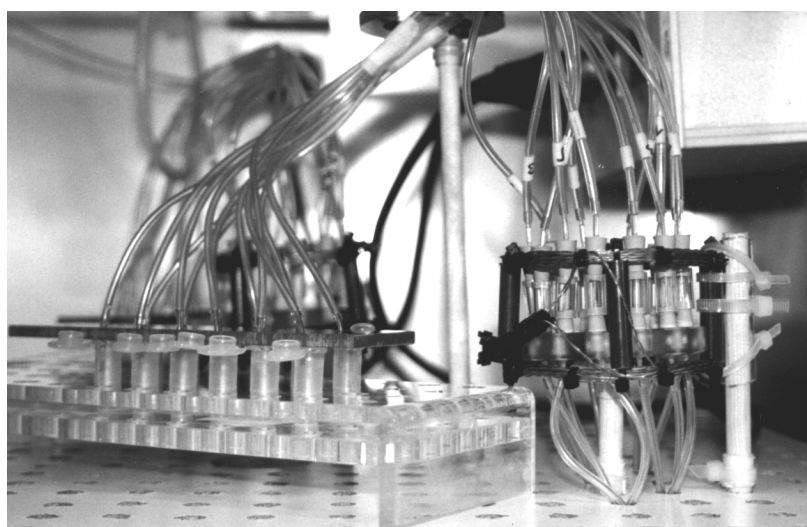


Fig. 1. Overview of the exposure system inside the incubator. From the bottom of the incubator (not visible), tubings are attached to the exposure chambers (right) through a wooden rack. Perifusates are collected in tubes (left).

At 300 mA, the active coil produced a magnetic field with a flux density of 86 μT (0.86 G). A function generator (FG 7000, ELV, Leer, Germany) was set at either $16^{2/3}$ Hz or 50 Hz at a rectangular waveform. The inductivity of the coils prevented rectangular changes of the magnetic field, which rather followed an exponential rise and decay. The variations of the flux densities at the different locations of the glass chambers were less than 5%. The stray field from both the

active coil, and from the incubator, were below 1 μT at the site of the sham coil. The static geomagnetic field inside the incubator was approximately 30 μT .



Fig. 2. Detailed view of the coil with the double filament wire. In active condition, current flow is in one filament only, while in inactive condition, current flows through both filaments in opposite directions, so that no magnetic field is produced.

Melatonin radioimmunoassay

The concentration of melatonin in the eluates was measured as described previously [[Lerchl and Schlatt, 1992](#)]. Briefly, 250 μL of sample were combined with 100 μL specific antiserum (Stockgrand Ltd., Surrey, UK) and 100 μL^3 H-tracer (Amersham, Braunschweig, Germany). After overnight incubation at 4°C, 500 μL charcoal suspension were added. Following another incubation for 15 min at 4°C, tubes were centrifuged for 20 min at 3000g and 4°C. The supernatant was combined with scintillation fluid, and counted in a β -counter. Concentrations of the unknown samples were calculated by comparison with a standard curve, ranging from 2 pg/mL to 500 pg/mL. Pooled control sera were analyzed with each assay. Intra-assay variations were less than 12%, and inter-assay variations were less than 15%. The high concentration of tryptophan in the buffer (10^{-4} M) did not cause any interference with the antiserum.

Statistical analysis

For each experiment, results between exposed and control groups were compared by two-sided t-tests for each time point. For each frequency, the results of all 4 experiments were analyzed by multifactor analysis of variance (MANOVA) with time, group and experiment as grouping factors. Data were considered significant at $p < 0.05$.

Results

In Fig. 3 and Fig. 4, the results of all experiments are shown. As expected, melatonin levels were low at the start of exposure. Stimulation with isoproterenol resulted in a transient increase in melatonin production, with peak levels observed between 5 and 7 hr after the experiment began.

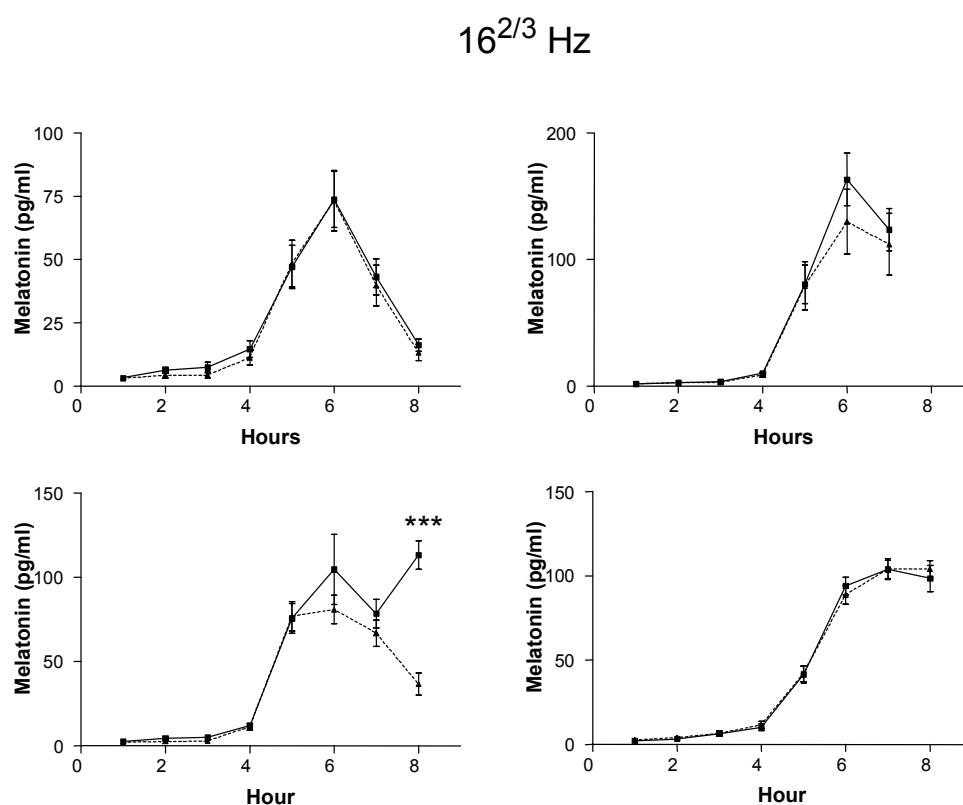


Fig. 3. Effects of exposure ($16^{2/3}$ Hz) on melatonin release from isolated pineal glands from Djungarian hamsters. Significant differences between the exposed (dashed lines) and the sham-exposed (solid lines), groups are indicated by asterisks: ***, $P < 0.001$. The analysis of all 4 experiments indicated a significant effect of exposure (MANOVA: $P < 0.01$).

Although the general features of melatonin synthesis patterns were comparable between exposed and sham-exposed groups, at most time points at both frequencies, exposure led to a suppression of melatonin release. In two experiments, significant differences were observed after stimulation. The MANOVA analyses revealed that exposure significantly reduced melatonin concentrations at $16^{2/3}$ Hz ($p < 0.01$) and 50 Hz ($p < 0.001$).

50 Hz

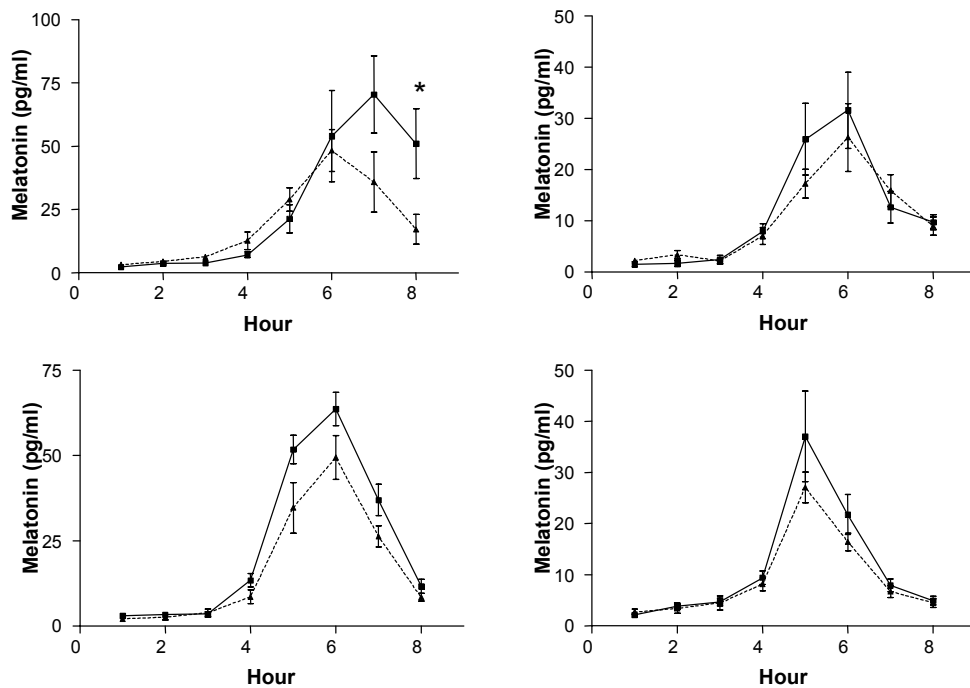


Fig. 4. Effects of exposure (50 Hz) on melatonin release from isolated pineal glands from Djungarian hamsters. Significant differences between the exposed (dashed lines) and the sham-exposed (solid lines), groups are indicated by asterisks: *, $P < 0.05$. The analysis of all 4 experiments indicated a significant effect of exposure (MANOVA: $P < 0.001$).

Discussion

The results of the present investigation show that low-frequency magnetic fields are able to reduce melatonin production in isolated hamster pineal glands. These data are, therefore, in good agreement with previous studies in rat pineolocytes [[Rosen et al., 1998](#)] and pineal glands [[Richardson et al., 1992](#)], despite some experimental differences. An important aspect of this study is the fact that instead of measuring the melatonin content of the glands, the production of melatonin was analysed. Hence, this situation more closely reflects the natural conditions.

It is quite interesting to note that despite the general tendency towards lower melatonin levels as a result of magnetic field exposure, in only two out of eight experiments were statistically significant differences found. This important finding has to be seen in the context of in vivo studies, where it is also very difficult to achieve replicable results, even when using the same species, and seemingly identical exposure conditions [[Yellon, 1994](#); [Truong and Yellon, 1997](#); [Löscher et al., 1998](#); [Reiter et al., 1998](#)]. Consequently, it appears logical to assume that there might be factors involved which have not yet been identified. Apart from meteorological influences for which effects on pineal physiology are unclear, there is one possible candidate: geomagnetic disturbances. It has recently been shown that geomagnetic variations of more than 30 nT resulted in severe reductions of the synthesis of urinary 6-hydroxymelatonin sulfate, the main metabolite of melatonin in humans [[Burch et al., 1999](#)]. Likewise, a depressive effect of annual geomagnetic field variations on melatonin synthesis has been found which may synchronize annual rhythms in melatonin production in rats [[Bartsch et al., 1994](#)].

The flux densities of the applied fields are beyond those actually measured for human exposure. However, according to the recent guidelines [[ICNIRP, 1998](#)], the exposure at $16^{2/3}$ Hz may be as high as 1500 μ T for occupational exposure, and 300 μ T for residential exposure, and at 50 Hz 500 μ T and 100 μ T, respectively. Thus, the effects as described here are of relevance for both occupational and public exposure limits.

In conclusion, our data demonstrate that the pineal gland itself responds to magnetic field exposure with a significant decrease in melatonin production. Hence, the eyes are not necessarily involved in the response to exposure, although they may be affected as well, as

shown previously [[Olcese et al., 1988](#); Olcese, 1990]. Moreover, the study shows for the first time that the melatonin production, as measured over 8 hr in a continuous flow system, is significantly diminished. The results of the present investigation support the hypothesis that the melatonin production in the pineal gland is suppressed by low-frequency magnetic fields.

Acknowledgments

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Literature cited

- BARTSCH, H., C. BARTSCH, D. MECKE, T. H. LIPPERT (1994) Seasonality of pineal melatonin production in the rat: possible synchronization by the geomagnetic field. *Chronobiol. Int.* 11:21-26.
- BURCH, J.B., J.S. REIF, M.G. YOST, T.J. KEEFE, C.A. PITRAT (1999) Reduced excretion of a melatonin metabolite in workers exposed to 60 Hz magnetic fields. *Am. J. Epidemiol.* 150:27-36.
- DOCKERTY, J.D., J.M. ELWOOD, D.C. SKEGG, G.P. HERBISON (1998) Electromagnetic field exposures and childhood cancers in New Zealand. *Cancer Causes Control* 9:299-309.
- FEYCHTING, M., A. AHLBOM (1995) Childhood leukemia and residential exposure to weak extremely low-frequency magnetic fields. *Environ. Health Perspect.* (103 Suppl.) 2:59-62.
- GROTA, L.J., R.J. REITER, P. KENG, S. MICHAELSON (1994) Electric field exposure alters serum melatonin but not pineal melatonin synthesis in male rats. *Bioelectromagnetics* 15:427-437.
- HATCH, E.E., M.S. LINET, R.A. KLEINERMAN, R.E. TARONE, R.K. SEVERSON, C.T. HARTSOCK, C. HAINES, W.T. KAUNE, D. FRIEDMAN, L.L. ROBISON, S. WACHOLDER (1998) Association between childhood acute lymphoblastic leukemia and use of electrical appliances during pregnancy and childhood. *Epidemiology* 9:234-245.
- ICNIRP (1998) Guidelines for limiting exposure to time-varying electric, magnetic, and electromagnetic fields (up to 300 GHz). *Health. Phys.* 74:494-522.
- JOHN, T.M., G.Y. LIU, G.M. BROWN (1998) 60 Hz magnetic field exposure and urinary 6-sulphatoxymelatonin levels in the rat. *Bioelectromagnetics* 19:172-180.
- KARASEK, M., M. WOLDANSKA-OKONSKA, J. CZERNICKI, K. ZYLINSKA, J. SWIETOSLAWSKI (1998) Chronic exposure to 2.9 mT, 40 Hz magnetic field reduces melatonin concentrations in humans. *J. Pineal Res.* 25:240-244.
- KATO, M., K. HONMA, T. SHIGEMITSU, Y. SHIGA (1994) Recovery of nocturnal melatonin concentration takes place within one week following cessation of 50 Hz circularly polarized magnetic field exposure for six weeks. *Bioelectromagnetics* 15:489-492.
- LERCHL, A. (1995) Breeding of Djungarian hamsters (*Phodopus sungorus*): influence of parity and litter size on weaning success and offspring sex ratio. *Lab. Anim.* 29:172-176.

- LERCHL, A., S. SCHLATT (1992) Serotonin content and melatonin production in the pineal gland of the male Djungarian hamster (*Phodopus sungorus*). J. Pineal Res. 12:128-134.
- LERCHL, A., K.O. NONAKA, K.A. STOKKAN, R.J. REITER (1990) Marked rapid alterations in nocturnal pineal serotonin metabolism in mice and rats exposed to weak intermittent magnetic fields. Biochem. Biophys. Res. Commun. 169:102-108.
- LERCHL, A., A. ZACHMANN, M.A. ALI, R.J. REITER (1998) The effects of pulsing magnetic fields on pineal melatonin synthesis in a teleost fish (brook trout *Salvelinus fontinalis*). Neurosci. Lett. 256:171-173.
- LÖSCHER, W., M. MEVISSSEN, A. LERCHL (1998) Exposure of female rats to a 100-microT 50 Hz magnetic field does not induce consistent changes in nocturnal levels of melatonin. Radiat. Res. 150:557-567.
- MCBRIDE, M.L., R.P. GALLAGHER, G. THERIAULT, B.G. ARMSTRONG, S. TAMARO, J.J. SPINELLI, J.E. DEADMAN, S. FINCHAM, D. ROBSON, W. CHOI (1999) Power-frequency electric and magnetic fields and risk of childhood leukemia in Canada. Am. J. Epidemiol. 149:831-842.
- MCCANN, J., F. DIETRICH, C. RAFFERTY (1998) The genotoxic potential of electric and magnetic fields: an update. Mutat. Res. 411:45-86.
- MEVISSSEN, M., A. LERCHL, M. SZAMEL, W. LÖSCHER (1996) Exposure of DMBA-treated female rats in a 50-Hz, 50 μ T magnetic field: effects on mammary tumor growth, melatonin levels, and T lymphocyte activation. Carcinogenesis 17:903-910.
- MOULDER, J.E. (1998) Power-frequency fields and cancer. Crit. Rev. Biomed. Eng. 26:1-116.
- OLCESE, J., S. REUSS, J. STEHLE, S. STEINLECHNER, L. VOLLRATH (1988) Responses of the mammalian retina to experimental alteration of the ambient magnetic field. Brain Res. 448:325-330.
- OLCESE, J.M. (1990) The neurobiology of magnetic field detection in rodents. Prog. Neurobiol. 35:325-330.
- PFLUGER, D.H., C.E. MINDER (1996) Effects of exposure to 16.7 Hz magnetic fields on urinary 6-hydroxymelatonin sulfate excretion of Swiss railway workers. J. Pineal Res. 21:91-100.
- REITER, R.J., D.X. TAN, B. POEGGELER, R. KAVET (1998) Inconsistent suppression of nocturnal pineal melatonin synthesis and serum melatonin levels in rats exposed to pulsed DC magnetic fields. Bioelectromagnetics 19:318-329.

- [REITER, R.J., D.X. TAN, J. CABRERA, D. D'ARPA, R. M. SAINZ, J.C. MAYO, S. RAMOS \(1999\) The oxidant/antioxidant network: role of melatonin. Biol. Signals Recept. 8:56-63.](#)
- [REPACHOLI, M.H. \(1998\) Low-level exposure to radiofrequency electromagnetic fields: health effects and research needs. Bioelectromagnetics 19:1-19.](#)
- [REPACHOLI, M.H., B. GREENEBAUM \(1999\) Interaction of static and extremely low-frequency electric and magnetic fields with living systems: health effects and research needs. Bioelectromagnetics 20:133-160.](#)
- [RICHARDSON, B.A., K. YAGA, R.J. REITER, D.J. MORTON \(1992\) Pulsed static magnetic field effects on in vitro pineal indoleamine metabolism. Biochim. Biophys. Acta 1137:59-64.](#)
- [ROSEN, L.A., I. BARBER, D.B. LYLE \(1998\) A 0.5 G, 60 Hz magnetic field suppresses melatonin production in pinealocytes. Bioelectromagnetics 19:123-127.](#)
- [SELMAOUI, B., Y. TOUITOU \(1995\) Sinusoidal 50-Hz magnetic fields depress rat pineal NAT activity and serum melatonin. Role of duration and intensity of exposure. Life. Sci. 57:1351-1358.](#)
- [STEVENS, R.G., S. DAVIS \(1996\) The melatonin hypothesis: electric power and breast cancer. Health Perspect. \(104 Suppl.\) 1:135-140.](#)
- [TRUONG, H., S.M. YELLON \(1997\) Effect of various acute 60 Hz magnetic field exposures on the nocturnal melatonin rise in the adult Djungarian hamster. J. Pineal Res. 22:177-183.](#)
- [WARTENBERG, D. \(1998\) Residential magnetic fields and childhood leukemia: a meta-analysis. Am. J. Public Health 88:1787-1794.](#)
- [WILSON, B.W., K.S. MATT, J.E. MORRIS, L.B. SASSER, D.L. MILLER, L.E. ANDERSON \(1999\) Effects of 60 Hz magnetic field exposure on the pineal and hypothalamic-pituitary-gonadal axis in the Siberian hamster \(*Phodopus sungorus*\). Bioelectromagnetics 20:224-232.](#)
- [WOOD, A.W., S.M. ARMSTRONG, M.L. SAIT, L. DEVINE, M.J. MARTIN \(1998\) Changes in human plasma melatonin profiles in response to 50 Hz magnetic field exposure. J. Pineal Res. 25:116-127.](#)
- [YELLON, S.M. \(1994\) Acute 60 Hz magnetic field exposure effects on the melatonin rhythm in the pineal gland and circulation of the adult Djungarian hamster. J. Pineal Res. 16:136-144.](#)

YELLON, S.M., H.N. TRUONG (1998) Melatonin rhythm onset in the adult siberian hamster: influence of photoperiod but not 60-Hz magnetic field exposure on melatonin content in the pineal gland and in circulation. *J. Biol. Rhythms* 13:52-59.