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Effect of occupational EMF exposure from radar at two different frequency bands on plasma melatonin and serotonin levels

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Abstract

Objective: **To delineate the effect of chronic electromagnetic field (EMF) exposure from radar on plasma melatonin and serotonin levels in occupationally exposed military personnel.**

Subjects and methods: **A total of 166 male military personnel participated in the study out of which only 155 joined for blood draw. They were divided into three sets: Control group ($n = 68$), exposure group I ($n = 40$) exposed to 8–12 GHz and exposure group II ($n = 58$) working with radar at 12.5–18 GHz frequency. The three groups were further split into two groups according to their years of service (up to 10 years and > 10 years) in order to investigate the effect of years of exposure from radar. Melatonin and serotonin levels were estimated by enzyme immunoassay in fasting blood samples collected from 06:00–07:00 h. EMF measurements were recorded at different locations using Satimo EME Guard 'Personal Exposure Meter' and Narda 'Broad Band Field Meter'.**

Results: **The group I exposed population registered a minor though not significant decrease in plasma melatonin concentration while the other group II exposed population registered statistically significant decline in melatonin concentration when compared with controls. Highly significant increase in plasma serotonin levels was found in exposure group II when compared to control whereas marginal non-significant rise was also registered in exposure group I in comparison to control. Exposure in terms of length of service up to 10 years did not produce any significant effect in the indoleamine levels in both the exposure groups when they were compared with their respective control groups. Whereas, length of service greater than 10 years was observed to decrease and increase respectively the melatonin and serotonin concentration significantly in exposure group II but not in exposure group I. However, correlation test did not yield any significant association between years of service and melatonin or serotonin levels respectively in both the exposure sets I and II. No significant association was observed between melatonin and serotonin levels as well.**

Conclusion: **The study showed the EMF ability to influence plasma melatonin and serotonin concentration in radar workers, significantly in 12.5–18 GHz range with service period greater than 10 years.**

Keywords: Melatonin, serotonin, electromagnetic fields, oxidative stress, radar band, microwave

Introduction

Recent technological breakthroughs and mushrooming usages of electronic gadgets have substantially mutated the ambient electromagnetic field (EMF) levels thereby leading to a new situation often termed as 'electromagnetic pollution or electro-pollution'. Electromagnetic pollution is an emerging environmental issue, which has gained serious concern. Despite the immeasurable benefits of EMF and promising results in wound healing as well as in treatment of cancers (Detlavs et al. 1996, Lai and Singh 2010), their effect on normal physiological processes remains a matter of concern. Literature is replete with EMF effects on health, physiology and well-being (Goldoni et al. 1993, Pfluger and Minder 1996, Reiter et al. 1998, Kliukiene et al. 2003, Mollerlokken and Moen 2008, Sokolovic et al. 2008, Garaj-Vrhovac and Orescanin 2009, Garaj-Vrhovac et al. 2011, Singh and Kapoor 2014).

Radio detection and ranging (RADAR) employing the radio frequency (RF) part of the EMF spectrum between 10 MHz to 300 GHz to geographically locate mobile and immobile objects, finds widespread applications in defense, aviation, navigation, weather forecasting and for other purposes. Since, these equipments utilize high power RF waves to produce high voltage and high frequency alternating electrical current, radar workers regularly experience these high EMF emissions. Studies have investigated the effect of EMF emissions from radar on oxidative stress (Garaj-Vrhovac et al. 2011), general health (Goldoni et al. 1993), fertility (Mollerlokken and Moen 2008) and cytogenetic damage (Garaj-Vrhovac and Orescanin 2009). Microwaves from radars or ionizing radiation released from electrical equipment generating the microwaves have also been implicated to increase the risks of hemolymphatic tumors in radar workers (Degrave et al. 2009). Hormones melatonin and serotonin, tryptophan derivatives produced chiefly from the pineal gland have also been documented to be affected by EMF (Lerchl et al. 1990, Rogers et al. 1995b, Pfluger and Minder 1996, Reiter et al. 1998, Burch et al. 1999).

Melatonin is well known as a potent broad-spectrum antioxidant (Reiter et al. 1995, Allegra et al. 2003), free

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radical scavenger (Reiter et al. 1999, Qi et al. 2001), anti-stress (Kirby et al. 1999) and anti-tumor agent (Conti and Maestroni 1995, Panzer and Viljoen 1997, Lissoni, 2000). Because of the capacity of melatonin to safeguard DNA from oxidative insults, its suppression may enhance the risks of cancer as DNA mutilation from free radicals is considered to initiate the process of mutagenesis in most of the tumors (Cerutti et al. 1994). Mutated melatonin levels hence could instigate carcinogenesis. Additionally, given their role in checking estrogen release as well as other cancer-supporting hormones (Cohen et al. 1978, Voordouw et al. 1992, Cagnacci 1996), melatonin inhibition by EMF either alone or in conjunction with light-at-night or other agents, could augment estrogen production thereby enhancing the threat of breast malignancy (Stevens and Davis 1996). The reported connections between job-related power frequency (50/60 Hz) electric and magnetic fields and cancers and leukemia like biological effects (Savitz 1993, Kheiffets et al. 1995) well below the current EMF standards have boosted the research in this field. The indoleamine, serotonin functions as an envoy for the nervous system, as a mood hormone in the brain and its suppression has been linked with depression, lethargy, inner agitation and numerous psychiatric problems. Serotonin undergoes acetylation reaction to generate melatonin and it is speculated that altered melatonin concentration may be the possible mechanism of EMF action and problems like insomnia, depression, suicide, chronic fatigue and various other subjective symptoms associated with EMF.

Despite decades of research, the central role played by EMF in pineal melatonin suppression is still controversial mainly due to inconsistent research findings as well as lack of dose-response studies and established bio-interaction mechanisms. Part of the inconsistency can be attributed to the varying EMF interaction with the biological system (Yellon 1994, Yellon and Truong 1998) and biological variables like sex and life stage (Fernie et al. 1999) of the bio-organism and, physical variables like frequency modulation (Penafiel et al. 1997), waveform (Zhou et al. 2014), intensity (Selmaoui and Touitou 1995), pulsed or continuous field (Rogers et al. 1995a, 1995b), direction of field (Kato et al. 1993, 1994a, 1994b), exposure duration (Selmaoui and Touitou 1995, Fernie et al. 1999) etc. Given the biological importance of melatonin and serotonin for living organisms and, to advance the knowledge base, more investigations are required in this regard. Very few studies have been conducted on the effect of frequencies in X band (8–12 GHz) and frequencies above this band utilized in radar. Military personnel working in a complex electromagnetic environment are exposed to comparatively higher EMF levels from radar because of their occupation. Hence, the need for intentional exposure of subjects to EMF for longer durations is eliminated for ethical reasons and as they form an occupational group, their exposures can be monitored rationally as well. Additionally, the chronic effects of radar emissions on melatonin and serotonin concentration have not been studied along with the effects of exposure duration in terms of years of service. As far as we know, this is the first study constructed to delineate the effects of chronic EMF exposure from radar at two different frequency bands (X and Ku band) on plasma melatonin and serotonin concentration in military personnel.

Subjects and methods

Study groups

The study population was derived from the Indian army comprising 166 active male volunteers selected randomly. The participants were divided into different groups: Control ($n = 68$), exposure group I ($n = 40$) and exposure group II ($n = 58$) depending upon their exposure to EMF from radar. Exposure group I worked with radar in X-band, at 8–12 GHz whereas volunteers from exposure group II had experience with radar in Ku band, at 12.5–18 GHz frequency. Subjects from the same station not exposed to EMF were included as control population. Of the 166 subjects, 155 personnel participated for blood collection. The study was conducted in September 2011 for all the three groups and all the subjects were posted at the same station at the time of study. The participants were explained with the details of the study design, which was approved by the Human Ethics Committee of the Institute and written informed consent was obtained. All the volunteers were asked to fill in a questionnaire comprising information pertaining to health history, occupational history, education level, diet, personal habits etc. Personnel with some history of the health problem or with brain surgery, stroke, post-trauma and with night shift before blood draw were excluded from the study. None of the volunteers reported any medicine intake, presence of hereditary or chronic disease. In both exposure groups, the workers had total duration of 8 h exposure per day. During the course of study, all participants had taken the same type of diet as per the army ration scale and were not undergoing any supplementation with any vitamin. Throughout the study period volunteers had followed daily 1 h physical training protocol as per army training schedule (slow running, body flexibility exercise, and pull ups) in the morning along with playing games (cricket, volley ball) in the afternoon session for 1 h. All the volunteers shared similar lifestyles, age and physical characteristics. The anthropometric and socio-demographic characteristics of these volunteers are summarized in Table I.

EMF exposure levels

The EMF levels measured in power density (W/m^2) were monitored with EME Guard personal exposure meter (Product No. 331 001A, Satimo, Brest, France) (frequency range 27 MHz to 40 GHz with upper and lower detection limit of 200 V/m and 5 V/m, respectively) and Broad Band Field Meter (NBM 520, Narda Safety Test Solutions GmbH, Pfullingen, Germany) (frequency range 100 KHz to 60 GHz). Measurements were undertaken inside the radar cabins and outside the radar at different distances of occupational exposures of the personnel. The power density of microwave radiation level inside the radar cabin and outside at various locations around the radar vehicle, where a worker of Group I worked during the course of normal duty ranged from 0.24–0.77W/m^2. Subjects of Group II were exposed to

Table I. Anthropometric and socio-demographic characteristics of the volunteers (plus-minus values are mean ± SD or *n* (%) unless otherwise specified).

Characteristics	Control group ($n = 68$)	Exposure group I ($n = 40$)	Exposure group II ($n = 58$)
Mean age (in years)	31.9 ± 6.0	27.9 ± 5.2	31.4 ± 4.0
Age range (in years)	21–48	20–45	20–43
Height (in cm)	170.27 ± 3.7	171.5 ± 3.6	172.5 ± 4.3
Weight (kg)	70.0 ± 7.3	69.3 ± 6.2	69.2 ± 6.4
BMI (kg/m^2)	23.7 ± 1.9	23.4 ± 2.1	23.3 ± 1.9
Exposure period, Range (in years)	12.2 ± 6.3, 1–28	8.3 ± 5.3, 1–26	11.5 ± 4.1, 1–23
Education			
Matriculation	58 (85.3)	34 (85)	31(53.5)
Higher Secondary	3 (4.4)		12 (20.7)
Graduation or above	7 (10.3)	6 (15)	14 (24.1)
Personal habits			
Smokers	20 (29.4)	14 (35)	15 (25.9)
Non-smokers	47 (69.1)	26 (65)	41 (70.7)
Alcohol	25 (36.8)	17 (42.5)	37 (63.8)
No alcohol	41 (60.3)	22 (55)	21 (36.2)
Tobacco	14 (20.6)	12 (30)	13 (22.4)
No tobacco	49 (72.1)	25 (62.5)	34 (58.6)
Vegetarian	20 (29.4)	14 (35)	15 (25.9)
Non-vegetarian	47 (69.1)	26 (65)	43 (74.1)
Marital status			
Married	58 (85.3)	27 (67.5)	48 (82.8)
Unmarried	9 (13.2)	12 (30)	9 (15.5)

microwave power density level of 0.1–15.6W/m^2 inside and outside the radar vehicle.

Chemicals and kits

Heparin (Product No. H0878-100KU, Sigma-Aldrich Chemicals, St Louis, MO, USA) was procured from Sigma-Aldrich Chemicals. Plasma melatonin levels were quantified using commercially available enzyme immunoassay kit from DRG International (Cat. No. EIA 1431, DRG International, Inc., USA) with an analytical sensitivity of 1.6 pg/ml with an intra- and inter-assay coefficient of variation ranging from 3.0–11.4% and 6.4–19.3%, respectively, and mean recovery of 102.4%. Briefly, the test principle is based on competitive ELISA in which the biotinylated and a non-biotinylated antigen contest for a set number of antibody binding sites. The concentration of unknown samples is reached by relating their enzymatic activity with a response curve obtained by utilizing known melatonin standards. Serotonin levels were measured by employing DLD, Diagnostika GMBH kit (Cat. No. EA602/96, DLD, Diagnostika GMBH, Adlerhorst, Hamburg). This assay is also based on the tenet of competitive ELISA involving the fight between acylated and solid phase bound serotonin both contesting for a limited number of antiserum binding spots. An inverse relation between the amount of antibody bound to solid phase serotonin and serotonin concentration of the unknown samples gives the desired serotonin levels. The sensitivity for serotonin ELISA was 5 ng/ml and the mean recovery was 107%.

Sample collection and processing

All the subjects were given necessary information beforehand for blood collection. Fasting early morning (06:00–07:00 h) blood samples were collected in heparin treated vials from the antecubital vein in resting condition. The collected blood samples were coded and subjected to similar treatment process. Remaining portion of blood was centrifuged at 1000 *g* for 15 min to separate out plasma and was stored in aliquots. All the samples were maintained at −20°C and transported to laboratory where these were kept at −80°C until assayed.

Metabolite quantification

Plasma melatonin and serotonin levels were quantified using the commercially available enzyme immunoassay kit following the manufacturer's instructions. Samples were run in duplicate and the absorbance was measured by microplate reader (Model Versa$_{max}$, Molecular Device, Sunnyvale, CA, USA). The experiment was carried out blindly in single series to eliminate intra-assay differences. Results were obtained in pg/ml and ng/ml of blood for melatonin and serotonin, respectively.

Statistical analysis

The values are expressed as mean ± SEM (standard error of mean). For each assay, values from duplicate samples were averaged to obtain one value point. Analysis of variance (ANOVA) test was used to analyze the group difference. When a significant effect was recognized, post hoc comparison was performed by employing Dunnett's multiple comparison test to see the difference in respect to the control group. Pearson correlation test was performed to find the strength of association between years of service and melatonin or serotonin along with the relation between melatonin and serotonin in both the exposure bands. Differences were judged significant at $p < 0.05$. The statistical software used for statistical analysis is GraphPad Prism version 5.00 (2007).

Results

Highly significant differences between the three groups were observed in connection with melatonin. While the group I exposed population registered a minor though not significant decrease in plasma melatonin concentrations, the group II

exposed population registered statistically significant decline in plasma melatonin concentrations when compared with controls (Control: 22.7 ± 2.11; Exposure group I: 21.2 ± 2.01; Exposure group II: 11.5 ± 0.6; $p < 0.05$; Figure 1a). Significant differences were also found among the three sets in respect to serotonin. Post hoc comparison revealed highly significant increase in plasma serotonin levels in exposure group II when compared to control (Exposure group II: 33.9 ± 3.3; Control: 21.4 ± 1.5; $p < 0.05$) whereas marginal non-significant rise was also registered in exposure group I in comparison to control (Exposure group I: 27.3 ± 1.9; Control: 21.4 ± 1.5; ns) (Figure 1b). Pearson correlation analysis did not reveal any significant association between years of service and melatonin or serotonin, respectively (Figure 3c–f). No significant correlation was found between the plasma melatonin and serotonin levels as well (Figure 3a, b). No significant group difference in serotonin levels was found in terms of radar exposure up to 10 years of service (Control: 26.7 ± 2.3; Exposure group I: 27.8 ± 2.3; Exposure group II: 34.7 ± 4.9; ns; Figure 2b) whereas; significant group variation was reached in respect to melatonin levels in the same service duration. Post hoc test however, reflected no statistically significant variation in melatonin concentration in both exposure group I and II when compared with their respective control group

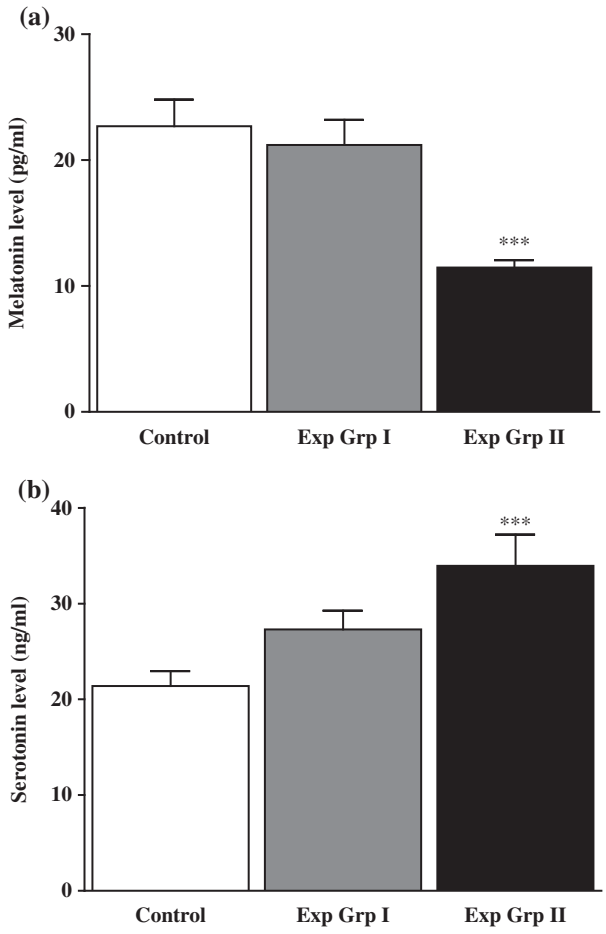

Figure 1. Showing the plasma melatonin (a) and serotonin (b) concentration in defense personnel exposed to EMF and control group (Values are mean ± SEM, error bars indicate the SEM for $n = 63$, $n = 40$, $n = 52$ for control, exposure group I and exposure group II, respectively, $p < 0.05$, * significance in regard to control).

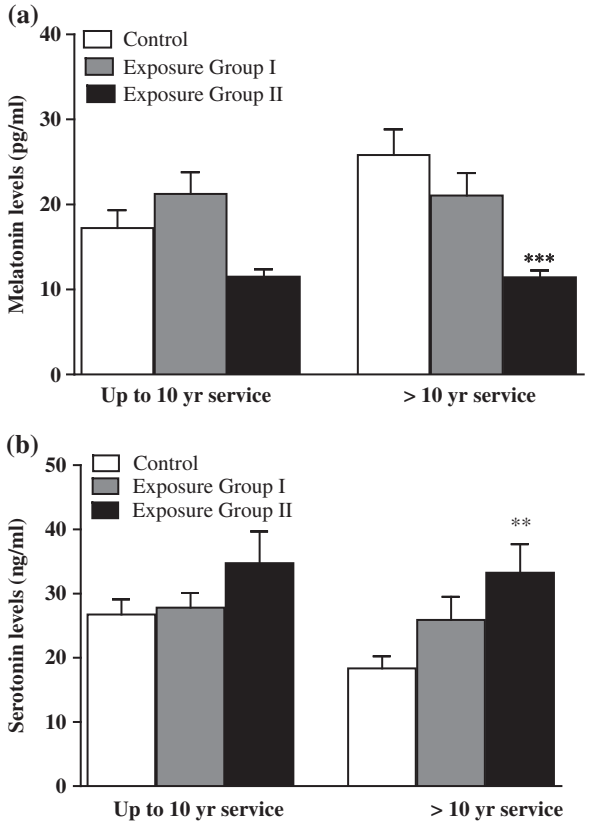

Figure 2. Showing the plasma melatonin (a) and serotonin (b) concentration in defense personnel exposed to EMF and the respective control group with up to 10 years and > 10 years of service duration (Values are mean ± SEM, error bars indicate the SEM for $n = 23$, $n = 30$, $n = 24$ for control, exposure group I and exposure group II, respectively, in up to 10 years of service category and $n = 40$, $n = 10$, $n = 28$ for control, exposure group I and exposure group II in > 10 years service category, respectively, $p < 0.05$, * significance in respect to control of the respective service category).

(Control: 17.2 ± 2.1; Exposure group I: 21.3 ± 2.5; Exposure group II: 11.5 ± 0.9; ns; Figure 2a). As regards, the effect of exposure period greater than 10 years of service on serotonin and melatonin levels, we found significant rise in serotonin (Control: 18.3 ± 1.8; Exposure group I: 25.9 ± 3.6; Exposure group II: 33.3 ± 4.4; $p < 0.05$; Figure 2b) and significant decline in melatonin levels (Control: 25.8 ± 3.0; Exposure group I: 21.1 ± 2.6; Exposure group II: 11.4 ± 0.8; $p < 0.05$; Figure 2a) in exposure group II but not in I in comparison to their respective reference groups.

Discussion

Despite the measured EMF levels found to be well within the acceptable limits of occupational exposure of 50W/m^2 for controlled environments (1.5–150 GHz) (International Commission on Non-Ionizing Radiation Protection [ICNIRP] 1998, 2002, Canada Safety Code 6, 2009, National Radiation Protection Board [NRPB] 2004), changes in pineal indoleamine concentrations with radar exposure in terms of both frequency band and years of service have been observed. The significantly depressed antioxidant level of melatonin in exposure group II signifies the potential of EMF exposure

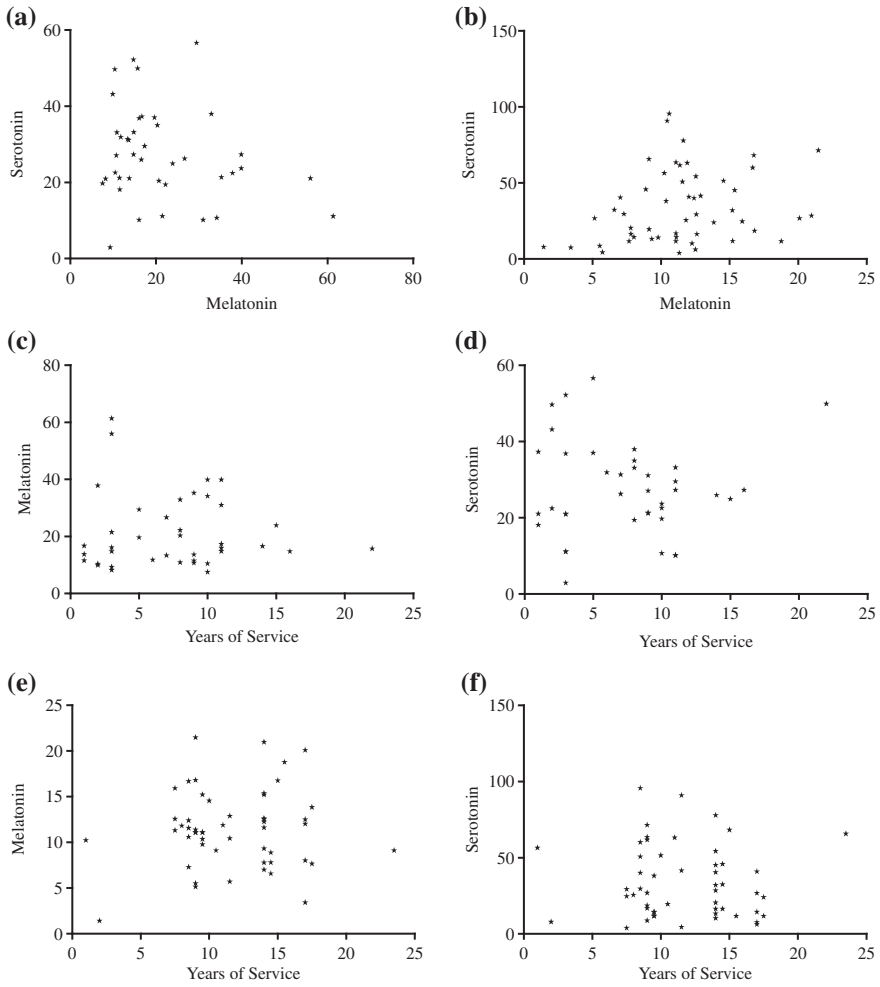

Figure 3. Correlation between plasma melatonin and serotonin levels in exposure group I (a) and exposure group II (b); correlation between years of service and melatonin (c) and serotonin (d), respectively, in exposure group I; correlation between years of service and melatonin (e) and serotonin (f), respectively, in exposure group II; correlation coefficient statistically insignificant $p < 0.05$.

combination at Ku frequency band and mean exposure period of 11.5 years in terms of length of service in inducing stress. At the same time, the slight fall registered in group I may be due to comparatively lower cumulative exposure both in terms of frequency band and length of service (mean 8.3 years) to which the group might have acclimatized as apparent by the non-significant difference when compared with the reference group. Correlation analysis however, did not yield any significant association between years of service and melatonin or serotonin levels in both the exposure sets I and II. No significant correlation was reached between the two indoleamines as well in both the exposure groups. As regards, the effect of exposure duration in terms of length of service, up to 10 years of service produced a noticeable but insignificant rise in melatonin in exposure group I and a decline in group II; whereas service period greater than 10 years caused suppression in both the groups though significant only in group II. Even though the results were insignificant in up to 10 years of service category for both the groups and only group I in the other service category, they do reveal the differential effect of EMF frequency band. Overall, the findings reflect the importance of both frequency and exposure duration in terms of service period in eliciting the effect and suggest that the effects produced are cumulative in nature. Also, it can be speculated that an adaptive response might set in some individuals but not all up to 10 years of occupational exposure from radar which when further extended may disrupt the equilibrium may be due to dysfunctional pineal or increased melatonin metabolism as a result of increased oxidative stress reported in various studies post EMF experience (Reiter et al. 1998). Previous studies have shown melatonin suppression post EMF exposure, which then reaches the normal level as the biological system, tries to acclimatize to a new environmental stress (Wilson 1986). However, with prolonged exposure the system may not be able to maintain the baseline levels. Decreased melatonin levels have also been previously reported both in experimental animals as well as in humans post EMF exposure however under differing exposure paradigms (Rogers et al. 1995b, Selmaoui and Touitou 1995, Pfluger and Minder 1996, Reiter et al. 1998, Burch et al. 1999). At the same time, our results are in contradiction to the investigations reporting rise in melatonin (Kumlin et al. 2005, Dyche et al. 2012) as well as the studies observing no change (Lee et al. 1995, Graham et al. 2001, Cocco et al. 2005). The latter effect however, has been found in exposure group I (registering minor but not significant difference in melatonin level), which is consistent with the previous findings as well. Other studies

reporting no alteration in melatonin concentration (Yellon and Truong 1998, Bortkiewicz et al. 2002, Gobba et al. 2006) with parallel investigations observing increase or decrease in melatonin levels reflect the inconsistency in this field of investigation. This variation in the interaction of EMF and hence its influence on the living system points towards the complex bio-interaction mechanism and biological sensitivity towards EMF. However, which combinations work when is yet not clear and additional investigations are warranted in this regard to reach meaningful conclusions.

EMF has been registered to cause oxidative stress in marine radar operators (Garaj-Vrhovac et al. 2011). At the same time, it has been well recorded to provide protection against EMF induced oxidative stress damage (Lai and Singh 1997, Koylu et al. 2006, Sokolovic et al. 2008). Recent investigations have further documented the cytoprotective role of melatonin against free radicals and oxidative stress induced damage in cell lines via suppression of apoptotic events as a result of stimulation of the phosphatidylinositol-3-kinase/protein kinase B (PI3K/Akt) signaling pathway (Lee et al. 2006, Kong et al. 2008). This PI3K/Akt pathway is recognized as one of the cellular survival mechanisms (Maddika et al. 2007) against oxidative stress induced apoptosis (Kimura et al. 2009, Bai et al. 2013). In the absence of any accepted causal mechanism of action so far, the fall in melatonin levels registered in our study might be better explained by the increased oxidative stress resulting in enhanced melatonin uptake by the tissues to cope up oxidative insult. To add, magnetic fields have been proposed to lengthen the life span of free radicals as well (Roy et al. 1995) which could hence, increase the demand of antioxidants like melatonin thereby diminishing its blood levels. In yet another of our studies (unpublished results Singh and Kapoor), we have also observed alterations in some oxidative stress biomarkers indicative of oxidative stress damage due to EMF exposure. The rise in serotonin levels also validates EMF stress as melatonin diminution corresponds well with serotonin rise in both exposure groups though non-significant in exposure group I, and is in accordance with previous results (Lerchl et al. 1990, 1991). Serotonin rise was also observed in both the service categories and exposure groups in comparison to their respective control groups however, significant results were reached only in the higher exposure category in terms of both frequency band and exposure duration. In this connection, significant suppression of N-acetyl transferase (NAT) activity, a rate-limiting enzyme in melatonin synthesis due to magnetic field exposure has also been reported (Lerchl et al. 1990, Yaga et al. 1993, Selmaoui and Touitou 1995) which may result in serotonin increase (Burchard et al. 1998) and melatonin diminution. In support, the melatonin forming enzyme Hydroxyindole O-methyl transferase (HIOMT) has been documented to be affected too (Henshaw and Reiter 2005). Hence, the observed serotonin rise in our study in both the exposure groups could be a consequence of the altered NAT activity in the pineal gland; however, this needs further investigation and for now, the exact reason is difficult to speculate.

The lack of any accepted biophysical mechanism up until now to relate the health-relevant changes in biological system to EMF experience complicates our understanding and puts this topic on top research priority. Moreover, the inhomogeneous nature of studies conducted at different times, for different exposure durations and at different locations in different animal models, renders the comparison among other studies complicated as well (Henshaw and Reiter 2005). Furthermore, the body undergoes inconsistent changes in respect to physiological functions when exposed to stress and these alterations are a function of the precise characteristics of the real stress condition (Pacak et al. 1995). The variation found in our investigation in respect to other studies as well as in exposure group I and II could therefore be attributed to differences in EMF exposure characteristics like frequency, exposure duration etc., and individual sensitivity to some extent. Lack of epidemiological and dose-response studies at the frequency investigated makes the comparison of our findings difficult in addition to the inter- and intra-variability in human melatonin levels. Additionally, melatonin levels are a function of factors like light exposure, exercise, food intake, alcohol and drug consumption, smoking etc. In our study, we also presumed the sunlight and artificial light exposure pattern to be alike in all the participants. On one hand, the presumption for sunlight exposure is quite agreeable as the participants belonged to the same area and military group; the night time exposure to simulated light needs to be considered. Despite the obvious effects of visible light exposure on melatonin production (Stevens and Rea 2001, Erren et al. 2003), night time simulated light exposure and light use have been registered to have no effect on 6-OHMS concentration assessed in urine samples of a Canadian population (Levallois et al. 2001). Nonetheless, forthcoming investigations concerning EMF exposure and melatonin production should charily account for sunlight and simulated light exposure. Moreover, the participants belonged to the same occupational group and military set-up, their lifestyle, diet, and exercise protocols were the same with similar physical characteristics. Further, the studies by Bartsch et al. (1994, 2010, 2012a, 2012b) have indicated the possible influence of the natural environment with regard to changes in solar activity through the 11-years sunspot cycle which might cause geomagnetic disturbances by altering the direction and intensity of the horizontal magnetic field component and thereby controlling the upshots of man-made EMF experiences on health, survival and melatonin rhythm as well. The authors have therefore stressed the importance of year and season in endocrine research regardless of the homogenous environmental conditions negligence of which may lead to contradictory findings. Since, we performed our study in the same year and month and at the same location, the role of these natural influences hence, could be eliminated. The disagreement with previous studies with regard to melatonin concentration can thus be attributed to some extent to the differing seasons or months as well in which the experiments were conducted. However, the puzzling results obtained at the two different frequency bands in our study depict the complexity of bioelectromagnetics research in terms of EMF bio-interaction and highlight the deficiency in our understanding of EMF influences on the human body. Apart from this, EMF exposure sources other than radar like proximity to cellular phone base stations,

electricity power stations or television broadcast or radio relay stations etc. were also assessed through the questionnaire and none of them were found to exist near the military set-up. Since, the experimenter was not aware of the control and exposed status of the samples and the experiment was performed blindly hence it also rules out the possibility of one of the confounders. As far as limitations are concerned, EMF exposure levels were assessed by spot measurements at the frequently visited occupational sites as opposed to long-term personal monitoring by dosimeters supposed to be the superior method of exposure measurement (Hutter et al. 2006). Nevertheless, this method has been utilized in previous study (Cocco et al. 2005) and the employment of spot measurements have been found to have lowest amount of inaccuracy (Eskelinen et al. 2002). In addition, it is voluntarily better accepted by the participants (Cocco et al. 2005) and is economically less expensive for epidemiological studies in comparison to long-term personal monitoring. The absence of night blood sampling in our study for the assessment of nocturnal indoleamine levels is also a limitation but due to practical reasons, we had to restrict our study along with the exclusion of multiple sampling for identifying the rhythmic changes.

Conclusion

In light of the observed alterations in melatonin and serotonin found in both the frequency bands of radar and service category though, significant only in the higher frequency band and in greater than 10 years of service duration, our study does imply the EMF potential to alter the plasma indoleamine levels in radar workers. The results need further corroboration; hence, the results should be interpreted with caution. Given the significance of these pineal secretions for organisms, further studies with better EMF characterization and standardization are crucial. In this regard, future studies should target occupational groups with cohort or cross-sectional studies with more time-point measurements in order to find the pattern of melatonin and serotonin response with EMF experience. Upcoming studies should also address the effect of EMF on all the components of melatonin biosynthesis in order to concretize the findings in addition to taking into account possible confounders. For the time being, precautionary approach should be adopted and unnecessary exposures should be checked, along with suitable protective measures where such exposures are unavoidable and considerably high.

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Declaration of interest

The authors report no conflicts of interest. The authors a lone are responsible for the content and writing of the paper.

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