

1. Trends of regulation in Japan

In Japan, the regulations of MF for protection of general public from short term health effects such as nerve stimulation were enforced for electric utility company and railway business operator recently. Power frequency (50Hz or 60Hz in Japan) MF emitted from Substation, transformer, power line, etc. was the target of these regulations.

Table Summary of ELF-MF regulations in Japan

Responsible ministry	Ministry of Economy, Trade and Industry	Ministry of Land, Infrastructure, Transport and Tourism
Type	Ordinance of the Ministry	Ordinance of the Ministry
Name of Ordinance	Ministerial Ordinance setting technical standards concerning electrical facilities and its interpretation	Ministerial Ordinance to Provide the Technical Standard on Railway and its interpretation
Target MF	Power frequency (50/60Hz)	Power frequency (50/60Hz)
Limit value	200 μ T	— (200 μ T is noted in interpretation)
Target facilities	Electric installations such as Substation, Power line, Transformers, etc except consumer	Fixed electric installations such as substation, catenary, electric devices, etc.
Measurement method	Adopted IEC62110	Adopted IEC62110 and IEC/TS62597

2. Evaluation of biological effects by exposure to MF

Besides the recommendation of short term effect, the WHO also recommends further research to elucidate scientific uncertainty about health effects of long term exposure. In fact, there are a few reports on biological effects of various EMFs that exist in railway systems, such as intermediate frequency (IF) MFs that are related to variable-frequency drive (VFD) installations in rolling stock and combined MFs with different frequencies. To respond this, we've been performed biological studies for years.

For example in our study, mutagenicity, that related to cause cancer, and hormonal effect by 21kHz IF-MF were evaluated. Mutagenicity was evaluated in *in vitro* micronucleus assay (MN) using Chinese hamster lung derived CHL/IU cell that is a part of the standard test battery for genotoxicity in the OECD guideline. For evaluation of hormonal effect, MCF-7 cell (human breast adenocarcinoma cell line) modified with an estrogen response genetic element (luciferase) as a reporter of gene expression was used. The cells exposed to 21 kHz, IF-MF up to 3.9mT (144 times higher than reference level for public in the ICNIRP guideline) for up to 4 days. Frequency of micronucleus was analyzed by microscopic observation and hormonal effect was determined as

expression of luciferase in the cellular extract measured by its chemiluminescence assay.

As the result, there is no significant effect by exposure to 21 kHz, up to 3.9mT IF-MF in both biological assays. Therefore, with taking into account these results and practical situation that will be much weaker MFs than experimental conditions exist where person can be in railway environment, it would suggest that IF-MFs in railway have no or extremely small mutagenic and hormonal potential that will be not able to be detected in general biological assays. Thus, it should not be considered that IF-MFs in environment have any adverse health effect. Other data in strong DC MF, 50Hz and their combined exposure condition will be presented at the conference.

3. Evaluation of dosimetry

For plausible safety evaluation of MFs should include clarification of MF that are generated in our environment and estimation of the induced current/voltage using the numerical model of human whole body. So far, we studied two types of numerical human model are used to construct model condition in an electric train car. One model is heterogeneous whole-body voxel models of Japanese adult male (Taro) and female (Hanako) that developed by National Institute of Information and Communications Technology (NiCT) and the other is homogeneous numerical human model Quete (OGIS-RI, Japan). Two types of magnetic field source were examined. One is the magnetic field by line current that considered the power cable under the floor. The other is the magnetic field by magnetic dipole that considered the equipments such as reactor. Induced current was calculated by impedance method [5]. In this calculation, voxel size was 2mm and amount of voxels ranged 24,243,520 to 52,115,960. In addition, the compliance of “reference level” of ICNIRP guideline for these magnetic fields was evaluated. Maximum or average magnetic field density to expose human body was normalized to the “reference level” and calculated the induced current in head and trunk. These data compared with basic restriction (2 mA/m^2) of ICNIRP1998 guideline.

Standing and seated position was examined in Quete. In each position, induced current concentrated at bending region such as ankle, knee, crotch, armpit. On the other hand, strong induced current was observed in organs that contained high water content such as cerebral fluid and blood within bladder in inhomogeneous model Taro and Hanako. However, the density of induced current in inhomogeneous model is lower than homogeneous model Quete. Highest density of induced current was observed in the magnetic field of line current because attenuation of the magnetic field lower than that of magnetic dipole. Induced current by normalized average/maximum magnetic field

to “reference level” of ICNIRP guideline shows approx. 10^{-1} mA/m² (average) or approx. 10^{-2} mA/m² (maximum) in head and trunk. These results suggest that allowance in basic restriction of ICNIRP guideline will be 10 times (average) and 100 times (maximum) respectively. Evaluation of induced voltage is under evaluation.

Acknowledgement

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EMF issue in Railway systems

- Evaluation of Biological Effects and Trends of Regulation in Japan -

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The World Health Organization (WHO) published an Environmental Health Criteria monograph of Extreme Low Frequency Field (EHC, No. 238) in 2007. In the EHC, they recommended that the international exposure guidelines should be adopted by policy makers. To deal with this, Japan has recently introduced regulations for protecting public people based on the ICNIRP guideline from short term health effect of power frequency MFs that is generated by electric installations in power utility and railway systems.

On the other hand, WHO also recommends further research to elucidate scientific uncertainty about health effects of long term exposure. In fact, there are a few reports on biological effects of various EMFs that exist in railway systems, especially about intermediate frequency (IF) MFs that are related to variable-frequency drive (VFD) installations in rolling stock. To respond this, we've been performed biological studies.

In our study, mutagenicity, that related to cause cancer, and hormonal effect by 21kHz IF-MF were evaluated. Mutagenicity was evaluated in *in vitro* micronucleus assay (MN) using Chinese hamster lung derived CHL/IU cell that is a part of the standard test battery for genotoxicity in the OECD guideline. For evaluation of hormonal effect, MCF-7 cell (human breast adenocarcinoma cell line) modified with an estrogen response genetic element (luciferase) as a reporter of gene expression was used. The cells exposed to 21 kHz, IF-MF up to 3.9mT (144 times higher than reference level for public in the ICNIRP guideline) for up to 4 days. Frequency of micronucleus was analyzed by microscopic observation and hormonal effect was determined as expression of luciferase in the cellular extract measured by its chemiluminescence assay.

As the result, there is no significant effect by exposure to 21 kHz, up to 3.9mT IF-MF in both biological assays. Therefore, with taking into account these results and practical situation that will be much weaker MFs than experimental conditions exist where person can be in railway environment, it would suggest that IF-MFs in railway

have no or extremely small mutagenic and hormonal potential that will be not able to be detected in general biological assays. Thus, it should not be considered that IF-MFs in environment have any adverse health effect.

In addition, trends in Japanese regulation of MFs and future agenda in railway systems will be presented at the conference.

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Theme

-Systems and Information: Electro Magnetic compatibility (EMC)

Evaluation of Health effect of intermediate frequency magnetic field

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Objectives: Considering human health effect related to railway environment is important to provide reassurance and safety for customers and workers. It is known that zero to several tens kHz magnetic fields (MF) are generated in rolling stock. However, there are still a few studies on health effects of MF with frequency of kHz which is named intermediate frequency (IF), which are generated by several devices such as inverter on electric train. In this study, the health effect of the IF-MF was evaluated by *in vitro* study using mammalian cell.

Method and Result: So far, we have newly developed an IF-MF exposure apparatus for *in vitro* study, which cells can be exposed to 21 kHz up to 3.9 mT continuously under appropriate incubation condition. In this study, the effect of the IF-MF on early phase of mammalian development using murine embryonic stem (ES) cell. Twenty-one kHz and 3.9 mT IF-MF was exposed to cells for 10 days when murine ES cell formed embryonic body (EB) and differentiated into embryonic tissues (Fig. 1).

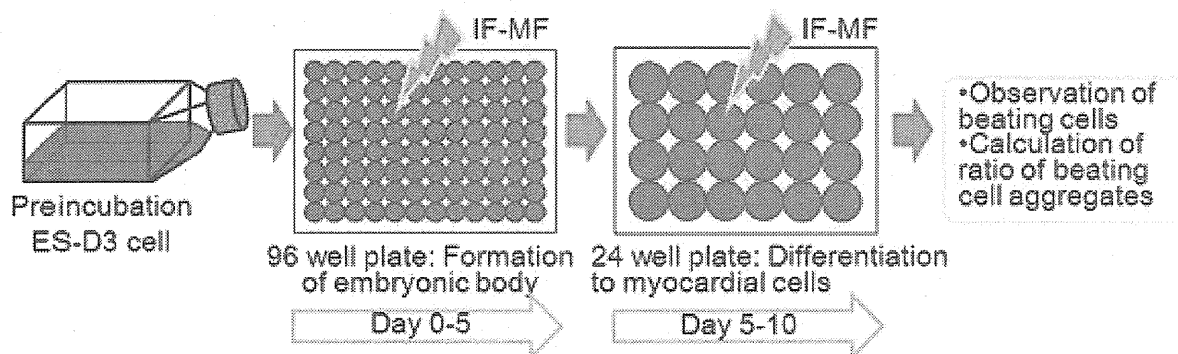


Fig. 1 Schematic of IF-MF exposure to ES cells

In this experiment, contracting myocardial cell was an index of differentiation. On the day 10th, each cell aggregate that was originated from an EB was observed by optical microscope and calculated the ratio of the number of the cell aggregates including contracting myocardial cell to that of all cell aggregates. As a result, there was no significant difference in the ratio of cell aggregate including myocardial cell between sham and IF-MF exposed cells (Fig. 2).

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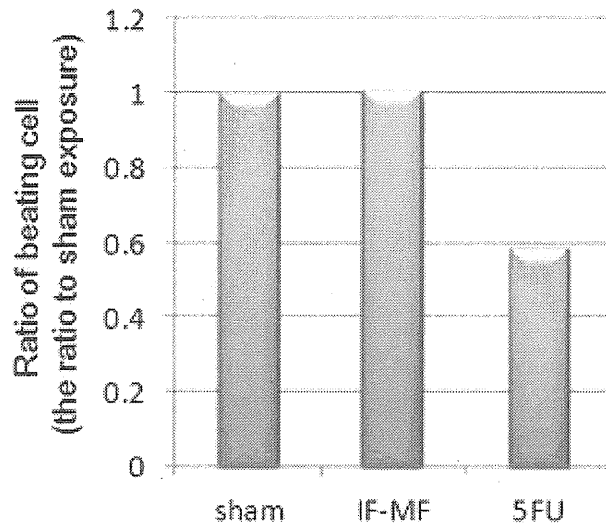


Fig. 2 Result of cell differentiation in 3.9 mT IF-MF exposure. The ratio of beating cell aggregation to all of cell aggregation was calculated. Each bar is indicated as ratio to sham exposure. 5FU (5-fluorouracil) was positive control.

Impact: These results indicated that IF-MF did not affect the process of differentiation to myocardial cell of murine ES cell under the conditions in this study. Namely, this result suggests that IF-MF would not affect on early development of fetus, since the magnetic flux density of the IF-MF generated in railway environment is lower than the one used in this study. This finding will contribute to explain the safety of MF in railway environment.

Implementation / What's new: The regulation to protect human for power frequency magnetic field (MF) (50 and 60 Hz), which are generated from railway facilities, has been introduced in Japan since August 2012. The other frequency range has not been regulated yet, however, evaluation of health effect is necessary for future countermeasure.

Novel: There is no report to investigate the effect of IF-MF on the differentiation of murine ES cell.

International Collaboration: Welcome.

Acknowledgment: This work was supported in part by Research on Health Security Control, Health and Labour Sciences Research Grants (08150668).

中間周波磁界の全身亜慢性ばく露影響 に関する研究

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Study on the biological effects of Long term and whole body Exposure to the Intermediate frequency magnetic Field

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Abstract: There exists the public concerns on health effect about the magnetic field from IH cooking hob, which generates intermediate frequency (IF) magnetic field (MF). Due to the lack of the evidences of biological effects, we explored the effects of IF-MF to the experimental animals. Male Sprague-Dawley rats (4 weeks old) were divided into 3 groups; cage-control, sham, 3.8mT exposure group, respectively. IF-MF at 21kHz were exposed to the animal fixed in the acrylic holder. Exposure was done for 1hour/day and 14 consecutive days. At the 15th day, the whole blood was collected and sent to the analysis. Biochemical and the hematological parameters in the blood were analyzed. We also evaluated the immunotoxicity by several methods. Results indicate that there is no effect to these parameters.

Keywords: Immune Toxicity, Intermediate frequency magnetic field, rat

1. Introduction

磁界の生体作用は、低周波領域では神経や筋の刺激作用が支配的であり、高周波領域では生体の組織で電力エネルギーが吸収されることによって生じる温度上昇（熱作用）が支配的であると言われる。

中間周波（intermediate frequency, 以下 IF と略）電磁界は、これらの間に挟まれる周波数帯、すなわち 300Hz から 100kHz 程度の周波数の電磁界であり、刺激作用を持つ低周波領域と、熱作用を持つ高周波領域の中間に属する。これまで、この周波数領域においては、工業用の誘導加熱等を除けば、生活基盤を支える技術には応用されてこなかったため、その生体影響に関する研究についても余り行われてきていなかった。しかしながら近年、オール電化の推進に伴い、中間周波電磁界を使用する IH 調理器の普及が進んでいる。IH 調理器では基本的に 20kHz～50kHz の電磁界を誘導加熱に利用するものである。しかしながらこの周波数帯の電磁界に関しては、これまで研究対象としての関心が低く、十分な科学的な知見が存在していない。そのため、2007年に発刊された WHO の超低周波電磁界に関する環境保健クライテリア (EHC238) では以下の通りに記載され、研究の推進が必須とされている¹⁾。「この領域のデータが欠落している現状を考慮し、すべてを包括する必要事項として、通常は 300Hz～100kHz とされる中間周波電磁界に関する更なる研究が必要である。健康リスク評価に必要とされる知識ベースの極少数しか集まっておらず、既存の研究の多くは結果が一貫していないので、更なる具体化が必要である。健康リ

スク評価のための十分な IF データベースを構成するための一般的な要件には、ばく露評価、疫学研究、ヒト実験室研究、動物および細胞 (in vitro) 研究が含まれる」(下線は加筆)

以上を背景に、本研究では最終的に IF 電磁界の動物に対する影響の有無を明らかにすることを目的とする。本研究では、動物用全身均一ばく露装置を用いて、1日1時間のばく露を連続14日間行った際の影響について、血液生化学指標、血球分析指標の検討を行い、同時に免疫系の指標に対する検討を行った。免疫系の指標としては、①免疫毒性試験において非特異的免疫機能の評価項目の一つに挙げられるナチュラルキラー (NK) 細胞活性測定、②顆粒球の遊走能・食食能、③T 細胞のサブセット解析を行った。

2. 方法

2.1 動物とばく露条件

本研究では、全身均一ばく露装置を用いて、ばく露影響を検討した。動物は、Sprague Dawley (SD) ラット (日本エスエルシー) を実験に供した。ばく露は平均磁束密度で、3.8mT とした。実験開始時点で、4週齢のラットを用いて、ばく露の際にはアクリル製の保定用筒に入れて1日1時間のばく露を連続する14日間行い、15日目に採血および脾臓の摘出を行い分析に供した。なお、実験においてはケージコントロール群、シャムばく露群、3.8mT ばく露群の3群を作成した。

2.2 分析用血液の採取

採血の際は血液生化学分析用にヘパリンリチウム含有微量採血管（ヘルスウェーブ社）、及び血球分析用に EDTA 含有微量採血管（ヘルスウェーブ社）にそれぞれ 300 μ l 程度ずつ分注し、残りの血液は後述する顆粒球調整およびリンパ球タイピング用に 50mM EDTA を含む PBS 1 容量に血液が 9 容量になるよう混和した。

2.3 血液生化学の分析

ヘパリンリチウム含有微量採血管に採取した血液は直ちに動物用・生化学自動分析装置 VS2 (Avaxis 社) を用いて分析した。

2.4 血球分析

EDTA 含有微量採血管に採取した血液は直ちに動物用・自動血球計測装置 HM2 (Avaxis 社) を用いて分析を行った。

2.5 ナチュラルキラー細胞活性測定

摘出した脾臓から調整したリンパ球浮遊液を NK 画分を含むエフェクター細胞として使用した。また、マウスリンパ腫細胞 YAC-1 (理化学研究所バイオリソースセンターより提供) をターゲット細胞として非 RI 法により NK 活性を測定した。

2.6 顆粒球走化性の測定

測定に用いる顆粒球は Percoll の不連続密度勾配遠心分離法により調製した。走化性は、細胞動態解析装置 EZ-TAXIScan (GEヘルスサイエンス社) を用いて検討を行った。

2.7 顆粒球貪食能の測定

上述した方法に従って回収した顆粒球を用いて、貪食能を測定した。100 倍に希釈した蛍光ビーズ (Fluoresbrite Microspheres, 1.0 μ m YG) と顆粒球を混和し、37 $^{\circ}$ C で 1 時間浸透しながらインキュベートした。その後、細胞をフローサイトメーター Cytomics FC500 (ベックマン・コールター社) で解析を行った。

2.8 T 細胞サブセットの検出

溶血させたのちの細胞懸濁液に PE (Phycoerythrin) 標識抗-CD3 抗体 (0.2 μ g), FITC 標識抗-CD4 抗体 (0.2 μ g), PE-Cy7 (Phycoerythrin-Cy7) 標識抗-CD8 抗体 (0.2 μ g) を加え抗体の結合反応を氷上で 30 分行った後に PBS で洗浄した。染色細胞はフローサイトメーター FC500 (ベックマン・コールター社) で検出を行った。

2.9 統計処理と倫理的配慮

統計処理はいずれも一元配置分散分析によって行い、有意差が認められた場合は Dunnett 法により、ケージコントロール群との比較を行った。統計処理は、SPSS を用いた。

本研究の実施にあたっては、国立保健医療科学院動物実験委員会の承認を得た。

3. 結果と考察

3.1 血液生化学・血球分析

1 日 1 時間のばく露を 14 日間連続して行い 15 日目に採血して分析をした。その結果、一部の指標において、統計的には有意差を観察したが再現性が十分に得られていないため、生物学的な意味を持たない可能性がある今後さらに検討が必要である。

3.2 ナチュラルキラー細胞活性

NK 活性を検討した結果、磁界ばく露による有意

な影響を認めなかった。

3.3 顆粒球走化性、貪食能

Percoll の不連続密度勾配遠心分離法によって分離した顆粒球の走化性について検討を行った。本研究においては、顆粒球の移動を 30 分間、30 秒おき計 61 枚の画像に納め、それを AVI ムービーに変換し、画像解析ソフトで細胞の輝度を追跡する方法により細胞の平均移動速度を求めた。通常、走化性物質が添加されない場合は、移動速度が低い細胞が多くを占めるのに対して、走化性物質 fMLP の存在下では細胞の移動速度が上昇する。しかしながら、移動速度の変化などに磁界ばく露が有意な影響を与えることはなかった。

また、貪食能について検討を行ったところ、ばく露による影響は認めなかった。

3.4 T 細胞サブセットの検出

本研究では、電磁波をばく露したラットにおいて T 細胞由来の免疫機構の異常の有無を確認するため、T 細胞表面マーカーである CD3/CD4/CD8 を用いたフローサイトメリーにより、CD4/CD8 比の検出、CD4⁺/CD8⁺ (double positive) および CD4⁺/CD8⁻ (double negative) を呈する T 細胞の検出を試みた。末梢血中の CD3 陽性細胞における CD4/CD8 比 CD4⁺/CD8⁺ (double positive) および CD4⁺/CD8⁻ (double negative) を呈する T 細胞の検出を試みた。その結果、長期磁界ばく露がこれらの数値に有意な影響を与えることはなく、T 細胞の分化には磁界ばく露が影響を与えることはないと考えられた。

4. 結論

本研究においては、21 kHz で最大 3.8mT の磁界を発生することが可能な装置を用いて、ラットに 1 日 1 時間、連続 14 日間のばく露を行い、磁界ばく露の影響を調べ、体重、血液生化学、血球分画の各指標に健康に影響を与えるような変化が見られないことを確認した。また同様に一部の免疫毒性の指標について検討し、NK 活性、顆粒球の走化性・貪食能、T 細胞サブセット (CD4・CD8) に関しては影響を与えないことを示した。

5. 謝辞

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Reference

[1] WHO Environmental Health Criteria 238 (2007): Extremely Low Frequency (ELF) Fields. WHO, Geneva, Switzerland

The Effects of 21 kHz Intermediate Frequency Magnetic Fields on Blood Properties and Immune Systems in Juvenile Rats

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1. INTRODUCTION

The use of induction heating (IH) cooking hob makes our life more convenient. On the other hand, there exist public concerns on possible health effect about the intermediate frequency (IF) magnetic fields (MF) from IH cooking hob. Although there are many studies on the biological effects of electromagnetic fields exposure to extremely low frequency and radio frequency, there are very few studies on IF-MF. Therefore, WHO recommended to study the biological effects of IF-MF exposure in the Environmental Health Criteria monograph No.238 [1]. In this study, we explored the effects of IF-MF to the experimental animals by using newly developed exposure apparatus which emit high intensity of IF-MF at 21 kHz [2]. Following the IF-MF exposure to rats, we analyzed the biochemical and the hematological parameters of the peripheral blood and several immune functions.

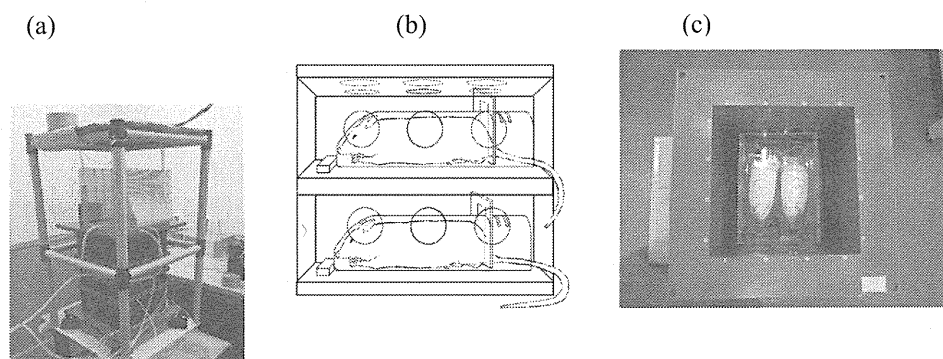
2. MATERIALS AND METHODS

Animals and IF-MF exposure

Male Sprague-Dawley rats (Japan SLC Inc., Shizuoka, Japan) were used. For IF-MF exposure, we used an apparatus which we originally developed [2]. The apparatus was able to generate IF-MF at 21 kHz up to 3.8 mT. Uniform magnetic intensity was obtained in a space of 15 x 15 x 15 cm inside of the coil. To avoid thermal effect from the coil, we applied a water-loop cooling circuit during the IF-MF exposure. Dosimetry calculation showed that mean induced electric density were 4.7 [V/m], which is corresponding to 1.7 fold higher than the value of basic restriction for general public of ICNIRP guideline [3].

IF-MF at 21 kHz was exposed to the animals under fixed condition in an acrylic holder (Natsume Seisakusho Co. Ltd.). For short term exposure, rats (6-7 week old) were divided into 4 groups; cage-control, sham, 2 mT and 3.8 mT exposure group (n=11-14, each). Exposure to IF-MF was done at 1hour/day for 3 consecutive days. For long term exposure, rats (4-5 week old) were divided into 3 groups; cage-control, sham and 3.8 mT exposure group (n=11-12, each). Exposure to IF-MF was done

at 1 hour/day for 14 consecutive days (2 weeks). On the next day of the last exposure, the whole blood and spleen of animals were collected and subjected to the analysis. The protocol of animal study was approved by the committee of animal experiment in National Institute of Public Health, Japan.



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Analysis

To explore biological effects of IF-EMF exposure, biochemical and the hematological parameters of blood were analyzed. For the biochemical analysis, 14 parameters were analyzed using a biochemical analyzer (VetScan VS2, Avaxis Inc., USA). For hematological analysis, 22 parameters were measured by use of a fully-automated 5-part differential cell counter (VetScan HM5, Avaxis Inc., USA).

For the cytotoxic assay, natural killer cell fraction prepared from spleen was used as effector cell. Cultured YAC-1 cell (mice lymphoma) was used as target cell. The cytotoxic activity was assayed by using CytoTox 96 Non-Radioactive Cytotoxicity Assay kit (Promega Co., USA). Experimental procedure was basically followed by the standard protocol.

For the phagocytotic assay, granulocytes were fractionated from peripheral blood by density separation method using Percoll (GE Healthcare Bioscience Co., USA). Granulocytes were co-incubated with FITC-conjugated polystyrene beads for 1 hour at 37 °C. After washing granulocytes, cells were analyzed by flowcytometer (Cytomics FC500, Beckman Coulter, Inc., USA).

T cell population was detected by flow-cytometric analysis with three antibodies, which were added to the bloods and splenocyte as followed: fluorescein isothiocyanate conjugated anti-rat CD4 antibody, phycoerythrin anti-rat CD8 antibody, and phycoerythrin-cyanine-7 conjugated anti-rat CD3 antibody (eBioscience Inc., San Diego, CA, USA). As for all CD3 positive cell, the ratios of CD4-CD8+ (Killer T cell), CD4+CD8+ (double positive), CD4-CD8- (double negative), and CD4+CD8- (Helper T cell) were compared among groups. Data obtained from FC500 were analyzed with CXP software (Beckman Coulter Inc., Miami, FL, USA).

One-way ANOVA was applied for statistical analysis by using SPSS software.

3. RESULTS AND DISCUSSION

In biochemical data, significant differences were observed in total protein concentration and glucose level for short term exposure, in amylase activity and glucose level for long term exposure, when compared to cage control group, respectively. However, these differences do not attribute to the intensity of IF-MF exposure because the data did not show dose-dependent relationship. Hematological data did not show any significant differences. Under short and long term exposure conditions, immune function such as cytotoxic activity and phagocytotic activity did not show any effects. We also analyzed T cell population in peripheral blood. Populations of Helper T cell, double positive, double negative, and Killer T cell after long term exposure did not show any significant difference (Table). Overall, no significant effect were detected under our experimental conditions.

Table: Results of T-cell subset analysis in a peripheral blood after long term exposure

	cage control (n=11)	Sham(n=12)	3.8mT(n=12)
CD4/CD8 ratio	1.82 ± 0.16	2.03 ± 0.34	2.00 ± 0.28
CD8+/CD4+ (%)	1.68 ± 0.41	1.54 ± 0.21	1.53 ± 0.40
CD8-/CD4- (%)	0.73 ± 0.73	0.77 ± 0.78	0.77 ± 0.43

4. CONCLUSIONS

In this study, we reported the biochemical, hematological and immunological effects of 21 kHz IF-MF exposure to rats' whole body. To generate high magnetic density, we developed an exposure apparatus for this purpose. The apparatus can generate maximum 3.8 mT, which is 141 times higher than the reference level for general public of ICNIRP guideline [3]. Exposure to IF-MF was done at 1hour/day for three consecutive days or 14 consecutive days (2 weeks), however, all of results were negative. Therefore we could conclude that IF-MF did not show any toxicity under these experimental conditions. However, the exposure condition in this study was limited, therefore, further study will be needed.

ACKNOWLEDGEMENT

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Lack of teratological effects in pregnant rats being locally exposed to their abdomen of intermediate frequency magnetic fields

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Sparse investigations have been conducted to assess the exposure effects of intermediate frequency magnetic fields (IF-MFs).

In this study, we teratologically evaluated them in the pregnant rats using exposure apparatus which can locally expose high intensity of IF-MFs to the abdomen. Abdominal exposure to high intensity of IF-MFs during organogenesis did not show any significant reproducible teratogenicity under this experimental condition.

Long Abstract

Introduction:

The Environmental Health Criteria No.238 published by the World Health Organization requires the need for biological studies on hazard identification and health risk assessment of IF-MFs. The induction heating cooking hob emits such IF-MFs (20-90 kHz) and is recently wide spreading in some countries including Japan. When people use the IH cooking hob, abdomen is close to the hob. Therefore, reproductive and developmental effects are one of the main public concerns. Although it is reported that there are lack of teratological effects in rats due to 20 or 60 kHz MFs exposure [1], rats are whole-body exposed with relatively low intensities. In this study, we evaluated teratological effects of abdominal local exposure at higher level of IF-MFs during the period of organogenesis of rats.

Materials and Methods:

Both sexes of Sprague Dawley rats (Japan SLC Inc.) were purchased at the age of 11 and 9 weeks, respectively. After one week habituation, females were checked optimal day for mating with a vaginal impedance checker, and mated with male if their impedance value are enough high. In the morning of the next day, vaginal smear was checked to identify copulated females as gestation day 0. Pregnant rats were randomly divided into three groups (n=20 each): exposure, sham exposure and cage control group, respectively. For the abdominal exposure, we used originally developed apparatus, which have the spiral coil and emit sinusoidal MFs with 21 kHz [2]. We set MFs intensity to 10.3 mT at the center of abdominal surface. Exposure (or sham exposure) was done for 1hr/day from gestation day 7 up to 17. During exposure, dams were fixed in an acrylic holder except cage control group. On the gestation day 20, their fetuses were excised and weighed. The number of live fetuses, dead fetuses, and implantation sites were recorded. Half of fetuses were placed in Bouin's fixative and examined for external and internal abnormalities. Other half of fetuses were eviscerated then fixed in alcohol, and their skeletons were stained with Alizarin red S and Alcian Blue 8GX to examine skeletal abnormality. All teratological evaluations were conducted in a blind fashion. All experimental procedures complied with the ethical guidelines for animal experiments at the National Institute of Public Health, Japan.

Results and discussion:

In this study, we examined numeral dosimetry using a pregnant rat model of gestation day 16 rat. Induced electric field of each fetus ranged between 0.611 to 5.74 V/m (mean 3.01V/m) depending on the spatial position to the spiral coil. The mean value is higher than the basic restriction to general public exposure (2.83 V/m at 21 kHz) of ICNIRP guidelines[3].

Regarding dams, no significant difference among groups was observed in the hematological and blood chemistry examinations at the gestation day 20. Total 767 fetuses from 60 dams were eviscerated and subjected to teratological examination. No significant difference was observed in dams' body weight, average weight of fetuses, number of fetuses per dam and number of implantation sites. The incidence of external, visceral, and skeletal malformations in the fetuses also did not indicate significant differences among the groups.

In conclusion, high level of 21kHz IF-MFs which emits high induced electric field compared to ICNIRP guidelines, does not show teratogenicity under the present experimental conditions.

Acknowledgement

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The 21 kHz Intermediate Frequency Magnetic Fields do not affect on Blood Properties and Immune Systems in Juvenile Rats

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The use of induction heating (IH) cooking hob makes our life more convenient. On the other hand, there exists public concerns on possible health effect about the intermediate frequency (IF) magnetic fields from IH cooking hob. Although there are many studies on the biological effects of electromagnetic fields exposure to extremely low frequency and radio frequency, there are very few studies on IF-MF. Due to the lack of the science based evidences of IF-MF, we aimed to clarify the effects on immune system of intermediate frequency of magnetic fields (IF-MF) to the experimental animals.

In this study, we used the exposure apparatus, which we originally developed previously. This exposure apparatus could generate uniform IF-MF which can expose to rat's whole body without any increase of temperature at inside of coil. Male Sprague-Dawley rats were divided into 3 groups; cage-control, sham, 3.8 mT exposure group, respectively. IF-MF at 21 kHz was exposed to the animals under fixed conditions in an acrylic holder. Exposure was started from 4 week-old and done at 1hour/day for 14 consecutive days. On the 15th day following the exposure, biochemical and hematological parameters in the blood were analysed. We also examined the effects to the immunological functions such as cytotoxic activity and phagocytic activity. Furthermore, we analysed the T-Lymphocyte Helper/Suppressor Profile (CD4:CD8 ratio) in which the percentage of CD3 positive lymphocytes in the blood positive for CD4 (T helper cells) and CD8 (a class of regulatory T cells) were counted and compared.

Although it was found that some significant differences in blood biochemistry, these differences do not attribute to the intensity of IF-MF exposure because the data did not show dose-dependent relationship. Hematological data did not show any significant differences. Immune function such as cytotoxic activity and phagocytic activity did not show any effects. We also analyzed T cell population in peripheral blood. Populations of Helper T cell, double positive, double negative, and Killer T cell after long term exposure did not show any significant difference. Overall, results indicate that there is no health effects to the observed parameters under our experimental condition, even high magnetic flux density (3.8 mT; 141 times higher than the reference level to general public of ICNIRP guideline 2010) was exposed to the animals. Therefore, we could conclude that IF-MF did not show any toxicity under these experimental conditions. However, the exposure condition in this study was limited, therefore, further study will be needed.

Choose one category.

2. Biological effects

