Low-Frequency Magnetic Field Exposure System for Cells Electromagnetic Biocompatibility Studies

Zuzana Judakova *, Ladislav Janousek, Roman Radil and Lucia Carnecka

Department of Electromagnetic and Biomedical Engineering, Faculty of Electrical Engineering and Information Technologies, University of Zilina, 01026 Zilina, Slovakia; ladislav.janousek@feit.uniza.sk (L.J.); roman.radil@feit.uniza.sk (R.R.); lucia.carnecka@feit.uniza.sk (L.C.)

* Correspondence: zuzana.judakova@feit.uniza.sk

Abstract: The advancement in science and technology has resulted in the invention and widespread usage of many electrical devices in the daily lives of humans. The exponential use of modern electronic facilities has increased electromagnetic field exposure in the current population. Therefore, the presented article deals with designing, constructing, and testing a new applicator system developed for cells electromagnetic biocompatibility studies. The applicator system is intended for studying the non-thermal impacts of low-frequency magnetic field on cell cultures growth. Main attention is focused on increasing the capacity of the applicator and effectivity of the experiments. The key idea is to reach high level of the magnetic field homogeneity in an area of interest and the temperature stability during the biocompatibility studies. The applicator system is designed based on numerical simulations and its construction, measurements, and properties evaluation are also reported for proving the applicator’s functionality. The new applicator allows performing five parallel experiments at the same time under the same conditions. The simulation together with the experimental results confirm that the magnetic field homogeneity reaches 99% in the area of interest and the maximum temperature instability is lower than 2% during the experiments. The effectiveness of new applicator is tested and proved during preliminary experiments with Saccharomyces Cerevisiae cells. The observed effects of MF exposure represent maximal stimulation of 74% and maximal inhibition of 49%. The reason why MF with the same parameters induces inhibition in one sample and stimulation in the other will be the subject of further research.

Keywords: low-frequency electromagnetic field; biocompatibility; applicator system; coil; ion parametric resonance theory; YPD medium

1. Introduction

This article aims to design, develop, construct, and verify a new exposure system to study eukaryotic cells’ low-frequency electromagnetic biocompatibility.

The research on the electromagnetic field (EMF) effects on living cells in the low frequency (LF) and extremely low-frequency (ELF) range is a worldwide longtime discussed topic. Even though the thermal effects are relatively well understood nowadays, there are still more questions than answers around nonthermal impacts. An impressive amount of research works conducted within this area during the last few decades [1–6] succeeded at least in terms of the necessity for nonthermal effects consideration when elucidating the response of the biological system to external EMF. In the LF range, the biological effects are usually investigated at weak levels, thus not likely to cause the thermal ones.

One of the significant challenges in investigating the LF EMF nonthermal biological impacts is the lack of precise results in epidemiological and laboratory studies, pointing to a reliable mechanism of action between the LF EMF and living cells. The problem, whether electric field (EF) or the magnetic one (MF) is responsible for the biological effects of LF EMF in the nonthermal area, is not yet clarified. Still, some researchers point to the threshold for...
an external EF to elicit a biological effect at the cellular level around $10^{-4}$ V·m$^{-1}$ [7]. On the other hand, the safety action values for power frequency (50 Hz) EF irradiation of workers are [8] set between 1 and $2 \times 10^4$ V·m$^{-1}$. The same standard also defines action values for magnetic flux density specified for the frequency mentioned above in the range of 1–6 mT. The wide gap between the safety standards determined by policy documents and presented experimental results present sufficient space for research focused on nonthermal biological effects and possible health risks associated with long-term exposure to LF EMF.

Several theories that describe the effects of weak LF MF on channel proteins in the cell membrane are discussed in scientific circles: the ion cyclotron resonance (ICR) theory, proposed by Liboff [9]; the ion parametric resonance (IPR) theory [10,11]; the theory of free radical pairs [12,13]. These three theories refer almost exclusively to the nonthermal action of the LF MF. New remarks and comments on these theories are constantly emerging. Despite decades of intensive work and expert discussions in this area, there is still no generally accepted theory for the action of LF EMF on cells.

The biological effect of EMF depends on several field parameters such as intensity, range of frequencies used, duration of action as well as the properties of the organism, i.e., dimensions, weight, water content, state of the body, age and others; the possible adaptation and evolution cannot be forgotten in the long term [14]. The first published reports on the effects of EMF on biological objects come from 1971 [15] (there are 2311 references to studies that point to the biological effect of EMF, the oldest study from 1910 on the physiological effects of alternating magnetic field), 1975 [16] (selective calcium removal from cell membranes by EMF), and 1979 [17] (indicates the possible effect of the magnetic field generated by electrical lines on the incidence of children leukemia). Current research of potential associations between exposure to EMF and various health outcomes is mainly concerned with the study of the possible mechanisms of EMF action on biological structures, the impact of EMF on the efficacy of anticancer drugs [18–20] and the efficacy of calcium channels, the connection of EMF and childhood leukemia [21,22], various types of malignancies, as well as the protective and therapeutic effects of EMF [23], drug and gene delivery [24]. The study is focused on the new exposure system for studying the LF MF nonthermal effects on cells. Such studies require a large set of experiments carried out under defined conditions. At each experiment, two data sets are collected: irradiate samples and the control ones. It is thus beneficial to irradiate as many samples as possible during one experiment. In such a case, homogeneity of the LF MF within irradiated samples plays an important role. Additional conditions concern constant temperature during experiments within irradiated and control samples, shielding of control samples and cultivation, especially temperature stability and microbiological purity. Those conditions should be adjusted purposefully due to the influence of these parameters on the system response. The new exposure system for the cells’ electromagnetic biocompatibility studies is designed based on numerical simulations considering the above mentioned requirements, real materials, and dimensions of a commercial incubator employed for the purpose. The system is composed of two identical LF MF applicators capable of performing five parallel experiments at a time. The numerical design is followed by applicators’ construction and testing, which are presented and discussed further in the article.

2. Design of Exposure System

The exposure system design assumes the use of two identical applicators of MF. Using two applicators will ensure the same cultivation conditions for exposed and control samples. One applicator serves as a source of MF; the second one represents a state without intervention/negative control. Furthermore, the design assumes the use of an incubator maintaining a constant temperature, Erlenmeyer flasks with liquid yeast-cell culture, shielding, and a system for additional temperature monitoring. The applicator of the exposure system suitable for LF MF irradiation should meet the following demands:

- The area of interest (AOI) with uniform exposure—the homogeneity of the MF represented by magnetic flux density field (B-field) at a minimum level of 98%;
Identical cultivation conditions for exposed and control samples;

The possibility to perform at least five experiments in parallel—five exposed and five control samples, to enhance the number of results for statistical evaluation and thus reduce the time required for experiments;

The applicator geometries optimized for an inner dimension of the incubator Q-Cell 240;

Sufficient airflow in the incubator for both control and exposed samples;

Safe handling with samples.

Due to the above-defined requirements, a simple long cylindrical coil seems to be the most suitable choice for the applicator. Other groups reported the employment of Helmholtz coil for in vitro [25] or in vivo experiments [26], two flattened orthogonal coils [27], exposure system sXc-ELF composed of a set of square Helmholtz coils [28], also solenoid coil for in vitro experiments [29,30], a system of coupling coils with adjustable transfer distance [31], tetra-coil [32], and many more setups presented in [33–37]. Most of the mentioned systems are designed for irradiation of only one sample or multiple samples with a minimal volume using a microplate and do not consider the inhomogeneity of the B-field.

To enhance the identical cultivation conditions for exposed and control samples, the laboratory incubator Q-cell 240 is chosen. The incubator maintains a stable temperature with an accuracy of 0.5 °C via forced air circulation in the chamber. The inner dimensions of the incubator 1315 × 504 × 320 mm³ (height × width × depth), determine the maximal size of the coils. Therefore, to ensure sufficient air circulation in the coil cavity, the maximal possible length of the coil is 1000 mm.

The inner diameter of the coil is determined by the pursuit to reach a high level of the magnetic field homogeneity—the smaller the diameter of the coil, the higher the homogeneity—and concerning the sample size (Erlenmeyer bank) and safe handling of them. The experiment used flasks with a volume of 100 mL, a height of 105 mm, and a diameter of 64 mm. Therefore, the inner diameter of the coil is set to 135 mm. The conceptual drawing of the cylindrical coil design is shown in Figure 1.
The coil should cover the entire space of the samples to achieve the necessary uniformity of B-field within the cell culture samples; therefore, the edge samples should be located 100 mm from the respective edge of the coil. The level height of the culture medium placed in the bank is also considered to determine the position from the edge of the coil.

The exposure coil is proposed to be driven by sinusoidal electric current to produce a high enough B-field within the irradiated volume but not affecting thermal conditions. For this purpose, tightly wound wire with a thickness of 1 mm and sufficient current load capacity is chosen, resulting in coils with 2000 turns each.

3. Numerical Simulations of Exposure System

Simulations are performed using two commercially available simulation software—CST Studio Suite and COMSOL Multiphysics to compare the results of numerical simulations of the identical model concerning the B-field distribution and its homogeneity in the AOI [38]. Each AOI is in real experiments Erlenmeyer flask filled with yeast extract-peptone-dextrose (YPD) medium with beer yeast cells Saccharomyces Cerevisiae. Within the simulations, the AOI is represented by a cylinder with dimensions of 68 mm in width, 40 mm in height, and a volume of 145 cm$^3$ and is filled with distilled water, Figure 2. Considering the percentage of distilled water in the YPD medium—95%, its substitution for distilled water, in terms of dielectric material properties, in the simulations, is permissible.

![Figure 2. Components included in the numerical model.](image)

Primary numerical simulations were realized to adjust the appropriate parameters of the system, where it was investigated how the input parameters influence the magnetic flux density distribution and its homogeneity [39].

The model includes two identical applicators—the exposed one and the control one, shielding between the coils, a metal incubator and ten samples representing the AOI, all depicted in Figure 2. The shielding with dimensions 1000 × 160 mm$^2$ (height × width) is placed exactly in the middle between the coils. The distance between the edges of coils is 174 mm.

The internal walls of the incubator Q-cell 240 are made from plastic. It is not known which kind of material is behind the internal plastic wall, so the simulations are made for the worst situation. Thus, the case behind the internal plastic wall is made from iron. The incubator manufacturer did not provide a technical drawing of the internal composition of
the incubator with a description of the material used. Therefore, the incubator is represented as a container with dimensions $1315 \times 504 \times 320 \text{mm}^3$ (height $\times$ width $\times$ depth) in the numerical simulation.

The walls of the incubator are modelled by a metal sheet with a thickness of 2 mm; the shielding is created from a metal sheet with a thickness of 3 mm. The electromagnetic parameters of the metal sheets at a frequency of 50 Hz are the ones from the simulation software library:

- $\varepsilon_r = 1$
- $\mu_r = 1000$
- $\sigma = 1.04 \times 10^7 \text{S} \cdot \text{m}^{-1}$.

This type of metal sheet is usually used in the manufacturing of refrigerators.

Ninety evaluation points, nine in each AOI, are considered for calculating the homogeneity of B-field (HBF). The B-field as RMS is calculated at these evaluation points and based on these values the HBF is calculated in the exposure coil for each sample separately:

$$HBF = \frac{B}{B_{C4}} \times 100 \text{ [%]}$$

(1)

where $B_{C4}$ is the value of B-field exactly in the middle of the coil, sample C, point 4 (p4), Figure 2, since the reference value $B_{C4}$ also represents the highest value of the B-field from all evaluation points, the value of $B$ is substituted by the minimum value within a particular sample, so HBF represents the worst case of each sample’s homogeneity.

The whole numerical space is discretized using a tetrahedral mesh in the CST Studio, and the low-frequency domain solver employing a magnetoquasistatic equation is applied. Built-in function Symmetry Planes cannot be applied; the boundary condition in XY, YZ, and YX planes are set to the open ones. For this reason and due to available computing capacity, the simulations are performed with the accuracy of $10^{-3}$. However, simulation complexity and computational capacity limit the simulation accuracy.

In COMSOL, the model is created in a 3D spatial layout discretized using an automatic generated tetrahedral fine mesh. Then, the multiphysical interface electromagnetic heating and the frequency-transient study are applied.

The values of HBF calculated in CST Studio and COMSOL in the exposure coil are listed in Table 1. The results show that the applied B-field uniformity of the AOI of three middle samples B, C, and D in the exposure coil is better than 99.16%, in sample A is minimum homogeneity of 92.55%, in sample E is the homogeneity of 92.75%. Therefore, the three middle samples would be a suitable solution for the research focused on electromagnetic biocompatibility studies at a cellular level. The two edge samples should be considered in further experiments as the non-homogeneous application of B-field. However, the existence of two sets of samples—homogeneous and inhomogeneous—can offer interesting and important results, namely how the less homogeneity of the applied field affects cell growth.

Table 1. The homogeneity of B-field at evaluation points in CST Studio and COMSOL in exposure coil.

| Sample/Software | A       | B       | C       | D       | E       |
|-----------------|---------|---------|---------|---------|---------|
| CST             | 95.61   | 92.55   | 99.77   | 99.71   | 99.88   |
| CM              | 99.71   | 99.71   | 99.98   | 100     | 99.16   |
| CST             | 99.16   | 99.72   | 95.13   | 92.75   |
| CM              |         |         |         |         |         |

Since the control samples represent a state without intervention, the main objective for the control coil is to reach the magnitude of the B-field lower than the Earth’s magnetic flux density value of $39 \pm 5 \mu T$, measured at the original experimental sample’s location. The geomagnetic field measurement is performed with a Vernier Labquest3 measuring instrument incorporating a Vernier MG-BTA probe. The maximal values of B-field RMS values obtained by simulation in CST and COMSOL are listed in Table 2. The highest value
of the B-field appeared in sample A at the level of 11.16 µT, which represents approximately 29% of the earth’s magnetic field value.

### Table 2. The maximal value of B-field at evaluation points in CST and COMSOL in control coil.

| Sample/Software | CST   | CM   | CST   | CM   | CST   | CM   | CST   | CM   | CST   | CM   |
|-----------------|-------|------|-------|------|-------|------|-------|------|-------|------|
| B [mT]          | 11.16 | 8.12 | 8.89  | 6.13 | 7.89  | 5.54 | 8.65  | 5.52 | 10.58 | 6.63 |

The results from both simulation programs are comparable in the center of the exposure coil and control coil, but values from the upper edge of the exposure coil differ significantly. The RMS values of the B-field obtained from both simulation programs differed in all evaluating points at most at 84 µT (4.94%) in the exposure coil and 4 µT (47.75%) in the control coil. The difference in the obtained values is acceptable and is caused by a different calculation method of the applied software programs, finite integration technique vs finite element method. The CST Studio is used for further verification of the EMF parameters of the exposure system.

Figure 3 shows the variation of B-field distribution in the solenoid on the axis at the input current of 0.7 A versus distance of the sample’s position in the coil cavity. The exposure coil meets the homogeneity criteria only in the three central samples, B, C, and D. The control coil meets the criteria of the maximum value of the B-field in all five samples.

![Figure 3. Magnetic flux density distribution along the axis for exposure and control coil, data from CST Studio Suite.](image-url)

The variation of E-field distribution in the coil on the axis at the current input of 0.7 A versus distance of the sample’s position in the coil cavity is shown in Figure 4. The
E-field induced by the magnetic field (MF) generated by the exposure coil is of the order of $10^{-4}$ V·m$^{-1}$ in both coils. In the peripheral parts of the samples, there is a sharp increase and decrease in the monitored parameter due to the induced eddy currents in the samples, Figure 5.

![Electric field distribution in exposure and control coil](image)

**Figure 4.** Electric field distribution in exposure and control coil, data from CST Studio Suite.

The E-field induced in the cell culture medium by the MF generated by the developed coil is in the order of $10^{-4}$ V·m$^{-1}$. The reported threshold for an E-field to elicit a biological effect is estimated to be around $10^{-4}$ V·m$^{-1}$ for neutrophil cells [8] and used in research into umbilical cord blood lymphocytes [40]. Thus, it is questionable if the EF generated by the new exposure system can be neglected, and the results obtained for biocompatibility studies can be referred to be caused by the MF.

On the other hand, the E-field in the presented exposure system is 10–11 orders of magnitude lower than the E-field across a 7.5 nm thick cell membrane [41] with a typical transmembrane potential of 50–120 mV, which turns out to be 6.67 MV·m$^{-1}$. Therefore, for the E-field calculation, the yeast cell is considered as a spherical capacitor according to:

$$E = \frac{U}{R_2} \frac{R_2}{R_2 - R_1}$$

where $U$ is the transmembrane potential, considered as 50 mV, $R_2$ is the external diameter of the cell representing the external electrode, and $R_1$ is the internal diameter of the cell representing the internal electrode. The $R_1$ value is calculated as a difference between $R_2$ and the thickness of the yeast cell membrane with a value of 7.5 nm. According to the calculation described above, the E-field induced by the new exposure system in the cell culture medium should not affect the processes in the cell membrane.
In the next step of simulations, the frequency change’s effect on the of the B-field’s homogeneity is verified. Eight frequencies are used: 50, 100, 500, 1000, 1500, 2000, 2500, 3000 Hz. The results show that the change in frequency does not affect the homogeneity of the B-field.

The temperature field distribution is evaluated using COMSOL in the samples after two, four, six, and eight hours from the start of the experiment, while the initial temperature is set to 30 °C. A graphical representation of the spatial temperature distribution in the AOI after eight hours of irradiation is shown in Figure 6. Any temperature variation and notable temperature differences between exposure and control AOIs are observed at a current of 0.7 A and a magnetic flux density of maximal value of 2.4 mT for the eight hours duration of the experiment.

The temperature distribution is uniform. The exposure coil does not cause temperature increase due to Joule heating of the content in the samples. The proposed exposure system is suitable for cell electromagnetic biocompatibility research studies.

The crucial findings from the simulations are that there is a negligible influence of the iron shielding and of the incubator’s iron walls on the exposure level or homogeneity.
The construction of a designed exposure system for cell electromagnetic biocompatibility studies consists of constructing a support system for Erlenmeyer flasks and of the construction of coils, the exposure one and the control one. The construction is followed by measuring of the magnetic flux density of both coils for verification of the numerical results.

The resistance to temperature, acids, alkalis, alcohol, and inflammability is considered when selecting the material of the coil support structure. Polytetrafluoroethylene (PTFE) meets the specified requirements—it has high thermal stability from $-200^\circ C$ to $+260^\circ C$, is chemically inert, nonflammable, and tolerated by living organisms [42].

The location of the irradiated samples is defined by exact positions by a support system (SpS). The SpS is designed and modelled in Inventor\textsuperscript{\textregistered} 3D CAD software. Therefore, it is necessary to make two support systems for each coil separately. The individual parts of the SpS are made of polyethylene (PE). Since the PTFE has a high surface tension, the SpS made from PE can be easily inserted into the coil cavity. The whole construction of SpS consists of four guide rails and five shelves. The SpS should secure safe handling with the yeast samples in the Erlenmeyer flasks.

After the suitable simulation of the exposure system, technical documentation is prepared for processing the material and producing a supporting structure of the coils. The supporting structure of the coil is made of PTFE, composed of parts—semi-finished products in the form of eight hollow cylinders. The purchased semi-finished parts are processed on a lathe to the required size and then assembled into the final shape. Enamelled copper wire of 1 mm diameter in a total length of 912 m for each coil is wound onto the coils supporting structure using the lathe. The completed pair of coils is shown in Figure 7. The location of the irradiated samples is defined on exact positions by a support system (SpS), also presented in Figure 7. The parts of the SpS are not permanently connected, the construction can be divided into individual parts and possibly modified. The SpS also provides easy and safe handling of the samples.
The magnetic flux density measurement is performed using the Narda NBM-550 EMF analyzer with the EPH-50D probe in the center of each coil for each sample position in the incubator with the door closed. The incubator contains the control coil, the exposure coil, and shielding at the time of measurement. The exposure coil is powered by an arbitrary wave generator RIGOL DG4162; the signal is amplified by a HUBERT KEYSIGHT Infinii Vision MSO-X 3012A linear amplifier while the driving current is controlled by a 6 1/2 local digital multimeter Aligent 34401A. The exposure coil is powered by $I = 0.7\, \text{A}$ (time course of sine shape, RMS), $f = 50\, \text{Hz}$. The conductors are attached to the coil through a drain channel at the rear of the incubator, which is used to drain excess water when the incubator is used for cooling so that the integrity of the incubator is not compromised.

The measured data are compared with the results obtained from the numerical simulation at the evaluation curve corresponding to the size of the probe.

The measured data of magnetic flux density are compared with the values obtained from the simulations, Figure 8. The percentage difference between the measured and simulated values does not exceed 11%, Figure 9.

The maximum of measured B-field RMS values in the control coil is $18.47\, \mu\text{T}$, while the exposure coil is powered by $I = 0.7\, \text{A}$ (time course of sine shape, RMS), $f = 50\, \text{Hz}$, with the incubator’s door closed. The measured magnetic flux density inside the control coil is 90 times lower compared to the value measured in the same position in the exposure coil.

For further using the exposure system, the values obtained by measurement are considered the reference values. A calibration curve is constructed to correctly determine the value of current required to obtain the desired value of magnetic flux density.
5. Cell Electromagnetic Biocompatibility Experiment

To examine the effect of LF MF on cells and verify the functionality of the proposed exposure system, 12 paired experiments are performed. The biological material used in the experiment is a pure strain, BY4741 yeast cells Saccharomyces cerevisiae. The yeast cells are obtained from the Slovak Academy of Science in Bratislava and refrigerated at 4°C in a Petri dish.

The theoretical basis of the experiment comes from Lednev’s IPR theory. The principle is in applying the MF generated by a parallel combination of static $B_{DC}$ and time-varying $B_{AC}$ MF. The theory of IPR suggests the nonthermal effect of MF on ions in living cells such that a protein-bound ion of mass $m$ and charge $q$ can be considered as a harmonic oscillator [12]. The correct application of the theory presupposes the application of time-varying MF with a frequency corresponding to the cyclotron frequency of the target bound ion, calculated according to the formula:

$$ f = \frac{1}{n} \frac{1}{2\pi m} q B_{DC} $$

(3)
where $q$ is the electric charge of the target ion, $m$ is the molecular mass of the target ion, and $B_{DC}$ is the value of static magnetic flux density—the geomagnetic field in this case. The target is calcium ion $^{40}\text{Ca}^{2+}$ with a cyclotron frequency of 29.89 Hz. According to the IPR, the maximal biological effect should be reached at the $B_{AC}/B_{DC}$ ratio of 1.8. For the earth’s magnetic field value of 39 $\mu$T measured at the location of the experiment’s realization, the corresponding value of $B_{AC}$ is 70.2 $\mu$T to maintain the ratio.

The exposure coil is powered by an arbitrary wave generator RIGOL DG4162; the signal is amplified by a HUBERT KEYSIGHT Infinii Vision MSO-X 3012A linear amplifier. The driving current is controlled by a digital multimeter Agilent 34401A. The exposure coil is powered by $I = 79.5$ mA (time course of sine shape, RMS), $f = 29.89$ Hz. The multimeter is connected for the whole experiment duration to monitor the value of the supply current continuously.

The control coil is without a power supply, and it is shielded. The control coil represents the state without intervention, and its aim is to show the difference in the results between the natural process of multiplication and the growth of yeasts affected by the MF.

The YPD medium is used in the experiment; the solution consists of 95% distilled water, 2% peptone, 2% dextrose, 1% yeast extract. After the medium is prepared, a 24 h pre-cultivation period is followed, starting with the inoculation of yeast cells into a flask with 20 mL of prepared YPD. The yeast medium is placed on a shaker and cultured for 24 h at 180 rpm at an ambient room temperature of 23 $^\circ$C. Pre-cultured yeast solution with a volume of 0.05 mL is pipetted into each of ten prepared Erlenmeyer flasks, 5 control, and 5 exposed samples containing 25 mL of medium. The flasks are closed with cotton-wool plugs to ensure air access. The samples containing cultured cells in the YPD medium are placed into a coil cavity and then moved into the incubator. The incubator ensures the maintenance of a stable temperature of 30 $^\circ$C during the whole experiment. The temperature is continuously measured in both cavities of the incubator using temperature sensors DS18B20. The time of exposure to the MF is 8 h.

To quantify the effect of applied MF, the numbers of cells in exposed vs control samples are compared. Cells are counted before and after each experiment in each sample using the Bürker chamber. The cells are photographed using Axiocam ERc 5 s in an inverted microscope Zeiss Primovert. The procedure for determining the effect of MF using the count of cells is depicted in Figure 10.

![Figure 10. The procedure for determining the effect of EMF.](image-url)
If X = 1, the cells in exposure and control coil grow comparable. If the parameter X is more than 1, the excitation/growth stimulation effect of the applied field appeared. If the parameter X is less than 1, the inhibition effect of the applied field appeared.

The realization of experiments corresponds to the usual laboratory practice with an emphasis on accuracy. To be able to repeat and verify the experiments, an experimental protocol incorporating a manual for implementing these experiments is developed. In addition, the necessary parameters are recorded for further comparison or repetition: the conductivity and temperature of the YPD medium before and after the experiment, the temperature during the whole experiment, and the photos of samples on the Bürker chamber of each sample before and after the experiment, and the count of cells of each sample before and after the experiment.

The results of experiments for all samples with respect to parameter X are reported in Table 3. The observed effects of MF exposure in the first experiment represent, for samples D, C, and A stimulation 16%, 23%, 2%, and for samples E and B inhibition 43%, 35%. The reason why MF with the same parameters induces inhibition in one sample and stimulation in the other will be the subject of further research.

**Table 3. The results of the experiments with respect to the parameter X.**

| Experiment Number | E  | D  | C  | B  | A  |
|-------------------|----|----|----|----|----|
| 1.                | 0.57 | 1.16 | 1.23 | 0.65 | 1.02 |
| 2.                | 0.66 | 1.44 | 1.43 | 1.46 | 1.82 |
| 3.                | 0.80 | 1.01 | 0.82 | 1.13 | 0.98 |
| 4.                | 0.61 | 0.62 | 0.44 | 1.47 | 0.64 |
| 5.                | 0.96 | 1.30 | 1.54 | 1.40 | 0.96 |
| 6.                | 1.20 | 0.85 | 0.88 | 0.52 | 1.60 |
| 7.                | 1.74 | 1.47 | 1.19 | 1.47 | 1.65 |
| 8.                | 1.14 | 1.02 | 1.09 | 0.51 | 0.89 |
| 9.                | 1.01 | 0.94 | 1.24 | 0.80 | 0.52 |
| 10.               | 0.83 | 1.57 | 0.58 | 1.49 | 0.90 |
| 11.               | 0.77 | 1.07 | 0.94 | 1.81 | 0.88 |
| 12.               | 1.86 | 1.09 | 1.17 | 1.53 | 1.66 |

6. Results and Discussion

The exposure system allows exposure to homogeneous LF MF of 10 samples, of which five control/shielded and five are exposed, with yeast Saccharomyces cerevisiae simultaneously, while maintaining the same temperature conditions for both groups—exposure and control one has been developed. The exposure system is designed using numerical simulations. The key parameters are measured using constructed facility to confirm the design.

The magnetic flux density field, electric field, and total current density field distributions are characterized along with the magnetic flux density field homogeneity and overall exposure uncertainty. The applied B-field uniformity of three middle samples in the exposure coil is better than 99.16%, and the maximum magnetic flux density in the control coil does not exceed 18.47 µT (0.75% of the maximum value of magnetic flux density in the exposure coil). The level of homogeneity of the three middle samples can be considered outstanding [43,44], and the homogeneity of samples A and E is satisfactory [45]. As for the value of the magnetic flux density field, these results cannot be confronted with the available data because other authors do not state its value; it has not been measured. The situation for the control samples is generally described as the control samples are not exposed to the field or the control samples are shielded. The impact of the switched-off apparatus was not considered [46–48]. To the shading of the B-field generated by the exposure system theoretically to zero, it would be necessary to use high-quality shading,
e.g., MuMetal. However, it would result in the shading of the terrestrial field. This state is not desirable because it does not represent natural conditions. The natural external EMF, such as the terrestrial E-field and B-field, or the atmospheric EMF—Schumann resonances are vital/necessary for any cell’s, microorganism’s, animal’s, and human’s physiological functioning [49].

The functionality of the developed exposure system is tested and confirmed during the 12 experiments, each lasting eight hours. The coil current and temperature in the cavities are continuously monitored during the experiments. The experiments are based on the IPR theory. The effect of applied MF is determined based on the number of cells, as explained in the previous section. Cell numbers are determined by direct counting in a hemocytometer, the Bürker chamber. Results of these experiments have shown significant differences between the control and exposed group of samples, Table 3. Mild inhibition and excitation effects of the applied MF are observed. Other groups reported an inhibitory effect on the growth of yeast cells [45,50], no effect [51], or both effects depending on B-field magnitude [52]. The questions of how the MF affects the yeasts or cells in general, what frequencies, B-field magnitudes, or their combinations are responsible for inhibition, or acceleration growth are far from answered. This area needs systematic, wide, and in-depth research as well. To increase the validity of experiments performed using this exposure system, it is desirable to use several evaluation methods for cell culture growth [46].

In the time domain of the experiment, the provided exposure system offers an additional benefit. For example, in the previously used experimental set-up [53] with one exposed and one control sample, the time required to perform five experiments correctly was 128 h, including pre-cultivation. Using the developed exposure system presented within this article, the time required to perform the same number of experiments, including pre-cultivation, is reduced to 32 h, resulting in significant time savings and thus effectivity enhancement.

It is also appropriate to point out the possible disadvantages of the proposed system. The proposed system is primarily intended for use in the area of low frequencies and low intensities of MF. The higher intensity MF could introduce a specific limitation to the presented system as the maximal current carrying capacity limits the coil design due to the copper wire. The solution that increases the MF in the sample area is to use a thicker wire and the Helmholtz configuration. It is questionable whether more accurate results can be reached using the multipurpose system (for low and high intensities) or a specific system designed directly for low and extremely low intensities. Due to the total cost of producing two coils and two SpS in the amount of 2153 €, it is more advantageous to use two different systems designed separately for low and extremely low intensities and the high intensities of the MF.

As mentioned in Section 2, comparable systems for EMF irradiation are designed for irradiation of only one sample or multiple samples with a minimal volume using a microplate and do not consider the inhomogeneity of the B-field. Other systems offer possible advantages of lower costs in terms of materials use and power consumption. However, they cannot be compared in terms of homogeneity.

Temperature changes are important experimental confounders in the research with yeast in general. Therefore, a stable and good controlled temperature is essential for the credible interpretation of results. To achieve minimum temperature fluctuation, a commercially available incubator is used. The incubator maintains a stable temperature of 30 °C. The temperature of the internal incubator environment (air temperature) is additionally monitored continuously during in vitro experiments by the temperature measurement system. The temperature measurement system consists of two temperature sensors, DS18B20, connected to the Arduino platform. The temperature is recorded on an SD card and shown on an OLED display every 10 min. Figure 11 shows the incubator air temperature course for eight hours lasting experiment; each point represents the average temperature from 12 experiments.
The low temperature of 24 °C occurring at the start of each experiment is caused due to the opening of the incubator door and manipulation with samples. During the 8 h lasting experiment, the door remains closed. The temperature difference stabilizes after 4 h ($\Delta T_{260}$) from the start of the experiment, where the temperature difference between control and exposed samples is less than 0.5 °C, and it is no longer increasing.

The temperature of the YPD medium in exposed and control samples is measured before and after each experiment using the conductometer PC 70 Vio. The average YPD temperature difference between control and exposed samples A-E in 12 experiments before and after the experiment is shown in Figure 12.

**Figure 11.** Temperature profile during the experiment, an average of 12 experiments.

**Figure 12.** The average YPD temperature difference between control and exposed samples before and after the experiment.
The average temperature difference between the exposed and control samples does not exceed 0.5 °C in both temperature sensors and conductometer. Achieved temperature differences are sufficient for the credible interpretation of yeast research results [54].

To the best of our knowledge, the system developed within this work, using live Saccharomyces cerevisiae cells intended for controlled LF MF exposure investigations based on the IPR theory, has not been described elsewhere and hence constitutes a new tool for cell electromagnetic biocompatibility studies. In addition, the system can be applied in both health risk assessment and investigations of claimed therapeutic effects of LF MF.

Further research using the presented system will be aimed at performing a sensitivity analysis of the frequency response of the biological system in the form of yeast Saccharomyces Cerevisiae according to the IPR theory.

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