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Experimental Investigation of Electromagnetic Activity of Yeast Cells at Millimeter Waves

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The direct detection of oscillations in biological cells may deliver strong evidence for Fröhlich's theory of coherent excitation in biological systems. Some results of measurement at millimeter waves published in the literature are mentioned. A laboratory equipment for the measurement of mm waves excitations in yeast cells is described. The measurement system is controlled by computer and measured data are stored in the computer memory for evaluation and for statistical processing. Some results of the evaluation are given.

Keywords Microtubule Electromagnetic Activity; Millimeter Wave Measurement; *Saccharomyces cerevisiae*.

Introduction

Direct measurement of the electromagnetic fields generated by living cells may deliver strong support for Fröhlich's theory of coherent oscillations in biological systems [1]. We have exerted in our laboratory in recent years some experimental effort to realize direct measurements of cellular fields in rather broad frequency range. Results of our experimental investigation in the 8–9 MHz frequency region are described in Pokorný [2]. Our interest is attracted now by the direct measurement of mm wave fields of living cells.

The published results of investigation in this region are rather contradictory. Grundler et al. [3] observed resonant interactions of the external fields in the 42 GHz region with yeast cells (frequency dependent growth of yeast cells during exposure over 3 growth cycles). Nonthermal effects at 40 and 50 GHz on living cells are described in Deviatkov [4]. Measurements of Gandhi et al. [5] did not disclose the

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spectral resonances of biological samples and the authors concluded that spectral resonances in the mm-wave region reported by others likely are in error.

Yeast Cells

We assume that electromagnetic field generated by subcellular structures has character of the near zone field and could be measured in the near vicinity of the cell. As a model system for experimental investigation we have used cold-sensitive β-tubulin mutants *tub2-401* of yeast cells *Saccharomyces cerevisiae*. At the restrictive temperature (below 14°C), the mutant *tub2-401* has no microtubules. The yeast cells are at the boundary between the G2 and the M phase. After recovery of the arrested mutant *tub2-401* at the permissive temperature (above 25°C) microtubules were polymerized and mitotic spindles were formed (at about 20 min from the initial time). After 40 min of cultivation the majority of cells were in the anaphase A and some of them were in the anaphase B. Within 60 min of cultivation, the mitotic spindles of the majority of cells were in the anaphase B and some of them completed it. These data were verified by fluorescent microscopy [2].

Laboratory Equipment

We designed laboratory equipment for the measurement of living cells activity in the 8 mm-wave region. A block diagram of the measurement system of electromagnetic oscillations of living yeast cells is shown in Fig. 1. The sensor is formed by a fin-line structure. A unilateral fin-line is mounted in the E-plane in the middle of a rectangular waveguide housing. The planar circuit on the fin consists of a slot line printed on a thin dielectric substrate by using the thin-film photolitographic technique. Broadband transition between rectangular waveguide and fin-line is provided by a tapered fin-line section. A part of the fin-line is not covered by the upper part of the waveguide housing, which enables us to put a cuvette with suspension with cells on the slot of the slot line. The open end of the slot line is terminated by a surface mount chip resistor the value of which matches the impedance of the fin-line at 40 GHz. Photograph of the fin-line sensor is shown in Fig. 2. The sensor with cells is screened against spurious fields.

We also designed another sensor formed by a rectangular waveguide resonator operating in the TE_{102} mode, coupled through an iris with the preamplifier (Fig. 3).

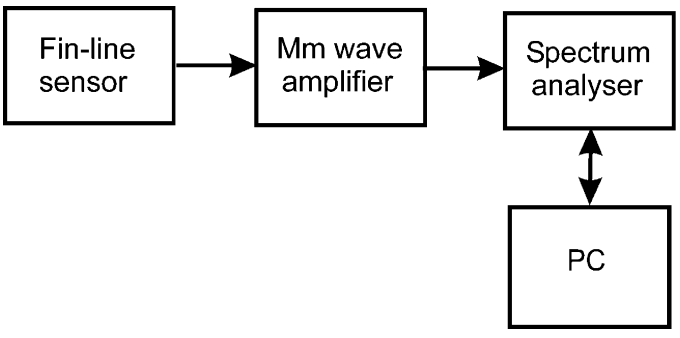


Figure 1. Block diagram of the laboratory equipment.

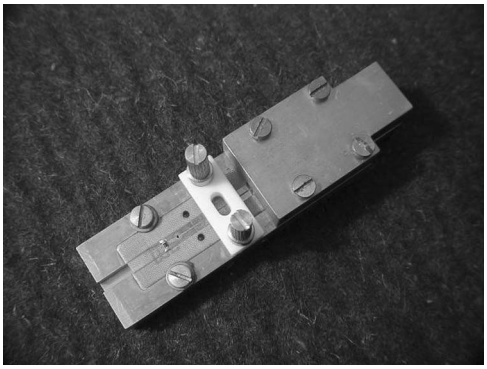


Figure 2. Photograph of a fin-line sensor with a cuvette.

The suspension with cells is inserted in a tip of a pipette, which penetrates through a hole in the E plane to the waveguide resonator in the plane of maximum E field.

Preamplifier Spacek SL406-34-4 (Spacek, Santa Barbara, California, USA), operating in the frequency band 37–43 GHz, is used followed by the spectrum analyzer Agilent E4448A. Measured sensitivity of the system for 300 Hz analyzer resolution bandwidth is −140 dBm. The frequency band 4 MHz at 41–42 GHz is divided into 20 subbands of 200 kHz and is scanned during 60 s. The system is controlled by a computer and measured data are stored in the computer memory for evaluation and for statistical processing. During measurement the temperature of the sensor with cell suspension and the temperature of

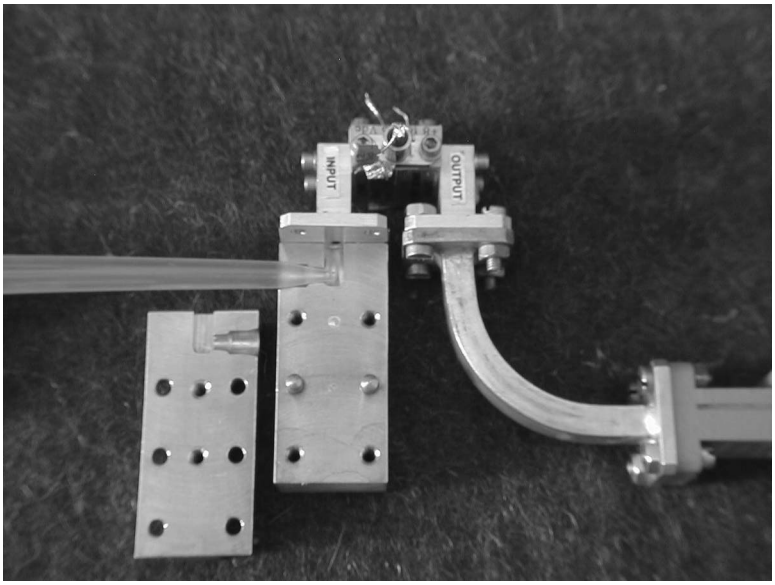


Figure 3. Photograph of a rectangular waveguide resonator as a sensor followed by the preamplifier.



Figure 4. Photograph of the laboratory arrangement for measurement of signals generated by yeast cells at mm waves.

the mm wave amplifier are kept at 28 ± 0.2°C and 32 ± 0.2°C, respectively, using a computer controlled system. The National Instruments NI PCI-4351 computer-based digitizer with platinum resistor sensors is used for temperature measurement and stabilization.

Figure 4 shows a photograph of the whole laboratory arrangement.

Measurement Results

Figure 5 shows measured signal in the subband 41.6996–41.6998 GHz (reproduced from the data stored in the computer memory). Critical level for evaluation is about 5.5 dB above the average level of the noise (detector of the maximum values is used). Figure 6 shows the evaluated signal in the frequency range 41.698–41.702 GHz (sum of power of the spectral lines exceeding the threshold level in 5 min intervals as a function of time). Average values from 10 experiments are shown. The circles connected with full lines (B) represent the evaluated data measured on synchronous cells of the mutant *tub2-401*. The squares connected with dashed lines (N) denote the same yeast cells cultivated at 30°C (nonsynchronous yeast cells). The triangles connected with dotted lines (S) denote data of sucrose solution. These results were obtained with the fin-line sensor.

The data measured on the *tub2-401* mutant revealed higher activity in 4 time periods that seem to coincide with the rearrangement of the microtubules in the

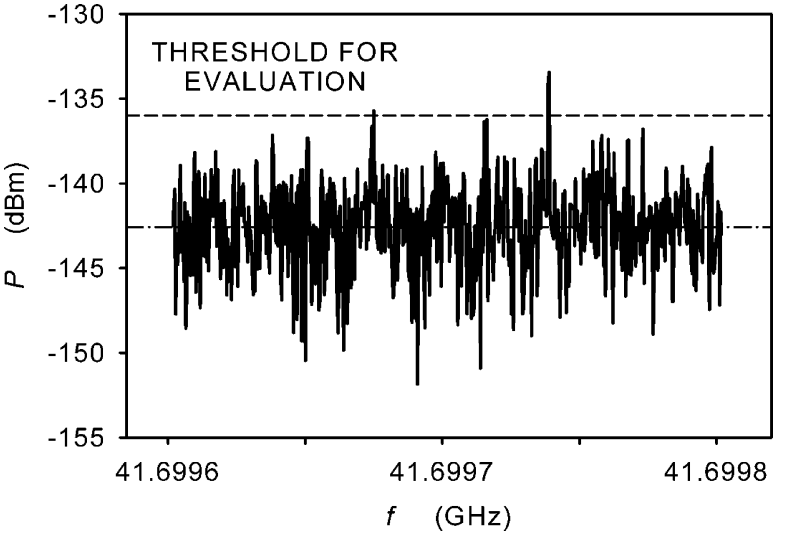


Figure 5. Measured signal in one of the 200 kHz subbands in the 42 GHz region.

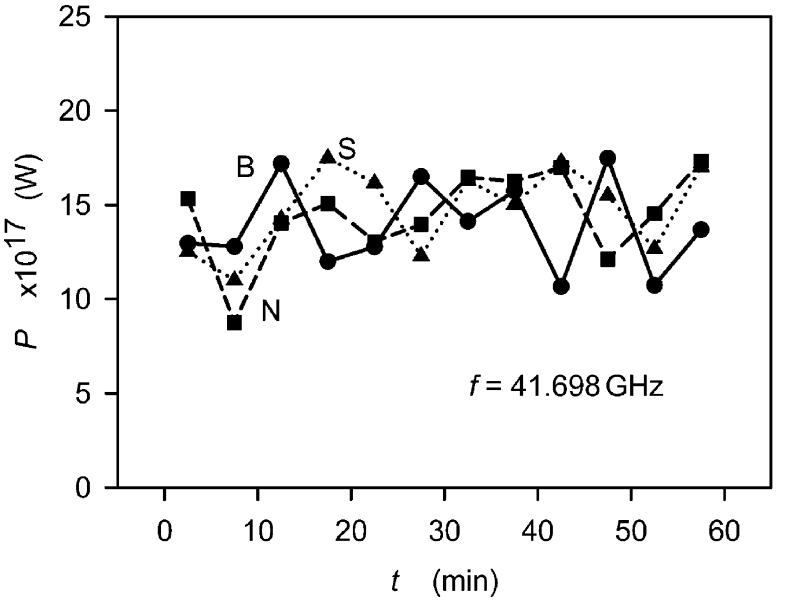


Figure 6. Evaluated signal in the frequency range 41.698–41.702 GHz as a function of time. Data were measured with the fin-line sensor. The solid line represents data measured on synchronous cells; the dashed line denotes nonsynchronous cells; the dotted line denotes sucrose solution.

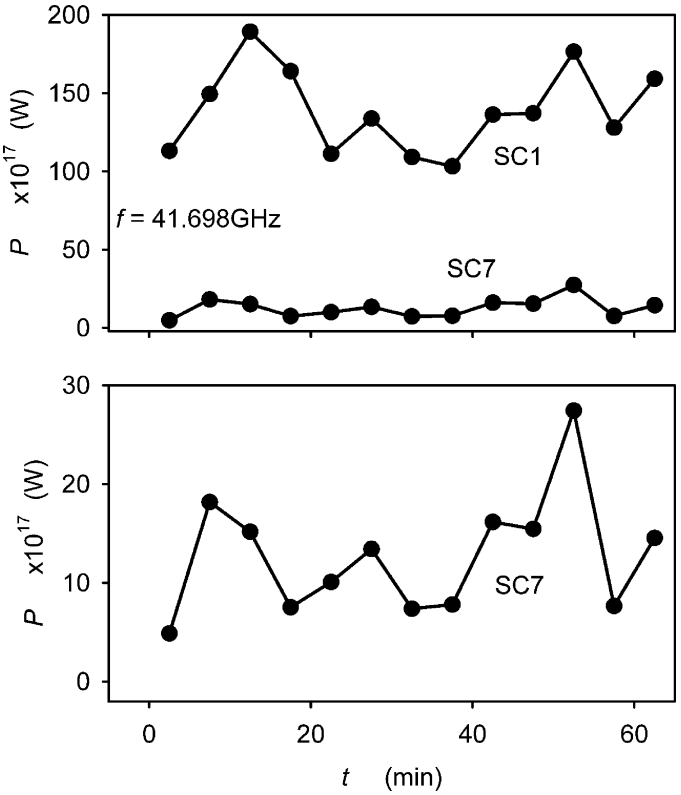


Figure 7. Evaluated signal measured on synchronous cells with the waveguide resonator sensor. a) Measurement SC1 was done one month prior to the measurement SC7. b) Data of measurement SC7 with enlarged scale at the vertical axis.

mitotic spindle, with binding of chromatids to kinetochore microtubules, and with elongation of mitotic spindles during anaphase A and B.

But the statistical evaluation revealed no significant differences among average measured values of the 3 media: synchronous cells, nonsynchronous cells, and sucrose. Figure 7 shows evaluated signal measured on synchronous cells with the waveguide resonator sensor. The data for the line SC1 were measured one month prior to the data for SC7. In both measurements, the same mutant *tub2-401* of cells was used. We are not able to explain the great difference of the levels, but the enlarged scale of Fig. 7b reveals the very similar shape of the lines SC1 and SC7.

Conclusion

Regardless of published results of other authors, oscillations in yeast cells at millimeter waves, if any, are very weak and the sensitivity of our system is not sufficient enough to detect them. The signal may be immersed in noise and the reproducibility of measured signals is not good. There are large differences among individual measurements.

As a first step in order to decrease the noise level we shall decrease the resolution bandwidth of the spectrum analyzer to 200 Hz by changing the computer

program. We also intend to improve the quality of the resonator as a sensor, or, possibly, to design a new type of sensor.

Acknowledgments

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