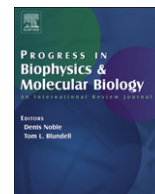




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Review

Electromagnetic cellular interactions

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ABSTRACT

Chemical and electrical interaction within and between cells is well established. Just the opposite is true about cellular interactions via other physical fields. The most probable candidate for an other form of cellular interaction is the electromagnetic field. We review theories and experiments on how cells can generate and detect electromagnetic fields generally, and if the cell-generated electromagnetic field can mediate cellular interactions. We do not limit here ourselves to specialized electro-excitabile cells. Rather we describe physical processes that are of a more general nature and probably present in almost every type of living cell. The spectral range included is broad; from kHz to the visible part of the electromagnetic spectrum. We show that there is a rather large number of theories on how cells can generate and detect electromagnetic fields and discuss experimental evidence on electromagnetic cellular interactions in the modern scientific literature. Although small, it is continuously accumulating.

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1. Introduction

Biological systems (e.g. cells) can communicate with each other and interact with the inanimate environment via many mechanisms and at many levels, depending on the type and complexity of the biological system and the nature of the information being communicated. Most known mechanisms of cell-to-cell communication in the current literature involve chemical or electrical signaling. In contrast, our understanding of non-chemical, non-electrical forms of communication is at a rudimentary level at best. In this article we review evidence for other forms of communication, with an emphasis on intercellular interactions via electromagnetic fields. These alternative forms of intercellular signaling may help explain phenomena that are hard to attribute to other forms of signaling (e.g. phenomena that require synchronous behavior among physically independent biological units).

Interest in such alternative forms of communication can be traced back at least to the second decade of the 20th century when a Russian scientist, Alexander Gurwitsch, showed an increased number of mitoses in a set of chemically isolated onion root cells that were in the vicinity of a group of actively dividing cells (Gurwitsch, 1923, 1924; Gurwitsch and Gurwitsch, 1924). Later, scientists discovered other pieces of the puzzle and gave more shape to this new form of intercellular communication. Before entertaining the idea of electromagnetic field (EMF) based cell-to-cell interaction, however, we should review the evidence that cells are indeed capable of generating and detecting EMFs (Fig. 1).

2. Evidence that cells generate electromagnetic fields

2.1. Type of EMF

Since Burr published his report on stable voltage gradients in various biological systems in 1935 (Burr and Northrop, 1935), many

scientists have found that these stable voltage gradients can be altered when the whole organism goes through any of a variety of biological processes including growth, local injury and other drastic changes. In his “electrodynamic theory of life”, Burr stated that “the pattern or organization of any biological system is established by a complex electrodynamic field, which is in part determined by its atomic physicochemical components, and which in part determines the behavior and orientation of those components.” “Electrodynamic” in Burr’s experiments refers to alterations that take place in a matter of hours to days. However, based on Maxwell’s theory of electromagnetic fields, we can safely consider such slowly varying electrical fields as “static” or “quasi-static”. Although, these quasi static electric fields may be involved in some significant physiological cell properties such as changes in distribution of ions (McCaig et al., 2005; Levin, 2003), they can not be considered as a mechanism for long-distance intercellular communication.

2.2. Measurement of EMF

2.2.1. History of detection of cellular EMF

In 1912, Gurwitsch borrowed the term “field” from physics and applied it to biology in his published theory of embryonic development (Gurwitsch, 1912, 1922). However, detection of a cellular EMF was first described in the visible spectrum by Scheminzký in 1916. He detected “emanations” of light from biochemical processes of yeast cells using photographic plates in light-tight chambers (Scheminzký, 1916). In 1918, Ludwig described the same phenomenon (“Hefenstrahlung”) (Ludwig, 1918). Independently, Alexander G. Gurwitsch did experiments with onion roots (Gurwitsch, 1923). He monitored the number of mitoses in a set of chemically isolated onion root cells that were in the vicinity of a group of actively dividing cells (Gurwitsch, 1923, 1924; Gurwitsch and Gurwitsch, 1924). He observed a significant rise in the number of mitoses if detector roots were separated from actively dividing

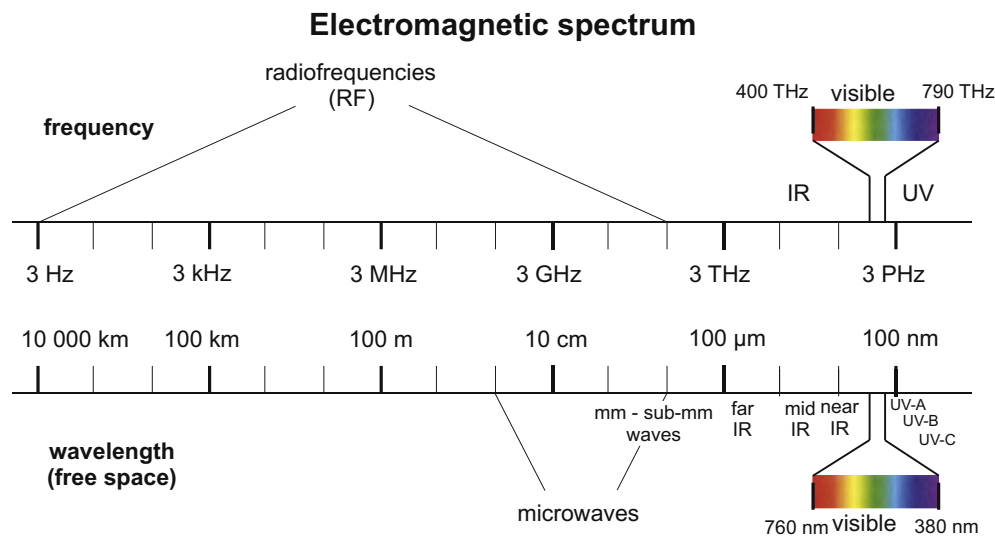


Fig. 1. The range of the electromagnetic (EM) spectrum covers different frequencies including visible light. The radiofrequency (3 Hz–300 GHz) wavelengths are named originally based on their use in technology for radio communication and broadcasting. However, radiofrequencies are used for many other applications nowadays. The visible part of the EM spectrum of light is sandwiched between the infra-red (IR) and ultraviolet (UV) regions and is sometimes denoted as the optical part of the EM spectrum.

roots by quartz glass but not by normal glass. The fact that UV light can pass through quartz but not regular glass suggested the existence of a form of cellular radiation of an electromagnetic nature which he named “mitogenetic radiation”. This study was the first to suggest that the emanation of light is not an incidental property of cells but one that might have relevance to signaling.

At that time, Gurwitsch and his colleagues used prisms for spectrum analysis and biological detectors for measurement of mitogenetic radiation. They found “finger-print” spectra for several enzymatic reactions and noticed spectral changes in light emission from cells following physiological changes (summarized in Gurwitsch and Gurwitsch, 1959). Inspired by Gurwitsch’s work, several independent laboratories in the Europe and USA gave rise to hundreds of articles, several books and dozen of reviews about mitogenetic radiation between 1920 and 1930 (see e.g. Rahn, 1936). Gurwitsch’s discoveries are a major topic of a recent review papers by Belousov (Voeikov and Belousov, 2007; Belousov, 1997).

In the 1930s, evidence for the existence of mitogenetic radiation took another leap forward when a group of scientists detected this radiation using modified Geiger–Müller detectors (Audubert, 1938; Rajewsky, 1931; Siebert and Seffert, 1933). Initially, it was believed that mitogenetic radiation only emanated from cells during their replication phase, and that the spectrum of this radiation belonged to the portion of the UV light spectrum that is invisible to human eyes. The invention of photomultiplier tubes allowed direct measurement of very small quantities of emitted light in the visible spectrum from germinating seeds as reported by Colli and Facchini (1954), Colli et al. (1955). Later on, other researchers applied a similar technique that was capable of detecting single photon emissions and showed that there is light emission from other biological substrates. Most of these experiments were done by groups in the International Institute of Biophysics, which was established by Popp who coined the term “biophoton”.

2.2.2. Experimental detection of EMF of biologic origin in infra-red, visible and ultraviolet ranges

There are only a few reports in the literature regarding direct measurement of infra-red (IR) light in biology independent of the thermal blackbody radiation that is related to the thermal level of the system and can be detected in living objects as well as inanimate materials. One of these reports was from Fraser and Frey who measured infra-red activity from electrically stimulated crab nerve (Fraser and Frey, 1968). Non-thermal millimeter wavelength radiation from nerves was detected by Gebbie and Miller (1997) from electrically stimulated frog gastrocnemius muscle. Coherent non-thermal infra-red emissions between 1000 and 1500 cm^{-1} were detected from myoglobin crystals after Argon laser photostimulation (Groot et al., 2002).

There is an abundance of published work regarding spontaneous emissions of light from cells. This is usually referred to as ultra weak photon emission (UPE), biological luminescence or biophotons. Cells usually emit one to thousands of photons/s cm^2 over the technical noise level (inherent noise activity of the instrument sensor). Generally, cells show increased UPE intensity when they undergo physiological changes. This is particularly true when they are exposed to chemical or physical stressors (Multi-author review, 1992; Slawinski, 1990, 2003). Rapidly and transiently increased UPE or a “death flash” can be observed when cells are damaged irreversibly (Slawinski, 2005).

The non-linear relationship of the object temperature and the UPE intensity suggests that UPE from living cells does not originate directly from simple chemiluminescent reactions for which kinetics are governed purely by the law of Arrhenius. For example the UPE measured from fresh whole blood samples had a linear correlation with temperature in the range of 35–38 °C. However, UPE intensity

deviated from this linear correlation and decreased regardless of the direction of temperature changes when the temperature was dropped down to 32 °C or raised up to 43 °C (Voeikov et al., 2003). The same phenomenon was also observed in green peas (*Pisum Sativum*) and barley (*Hordeum vulgare*) (Slawinski and Popp, 1987). In these examples, the bio-objects created a hysteresis-like dependence of UPE on the cycling temperature, i.e. the intensity of UPE at a given temperature depended on whether the cells were experiencing rising or falling temperatures.

Other evidence came from observations that there are unique patterns of UPE from cells during specific phases of the cell cycle during cell replication (Konev et al., 1966; Quickenden and Que Hee, 1974, 1976; Quickenden and Tilbury, 1983, 1991; Multi-author review, 1988). For example, Mei found a broad peak of UPE intensity in synchronized *Saccharomyces cerevisiae* cells in late S phase, which coincides with increased DNA synthesis, and G2 phase (chapter in Popp et al., 1992).

Another mode of active optical behavior of living cells is “delayed luminescence”. This term denotes the emission of photons from a biosystem after previous excitation e.g. by external light. This optical activity is similar to autofluorescence, but exhibits a decay time that is larger in magnitude by several orders. In fact, biological organisms may require up to days (depending on the sample type) until the level of spontaneous optical activity is restored. In this technique, tissues or cell cultures are excited using a light source with a defined spectral composition and intensity. The excitation phase is followed by a phase of early luminescence, followed by delayed luminescence. Although inducing luminescence may seem to be an artificial phenomenon that only belongs to experimental conditions, most living systems function under conditions of constant sunlight or are stimulated by natural EMF over a wide spectral range, which can, technically speaking create a natural condition similar to stimulated luminescence. Thus, it would not be surprising if this form of EMF emission was also involved in the process of cellular communication. However, the details and practical use of these techniques are beyond the scope of this manuscript.

2.2.3. Indirect cellular EMF detection by dielectrophoresis

An EMF can also be detected indirectly using a technique called dielectrophoresis (DEP) (Pohl, 1978). In this technique, EMFs are detected by the effect of a non-uniform electric field on a neutral particle via a polarization force. One of the pioneers in measuring cellular EMFs using the DEP method is Herbert A. Pohl (Pohl, 1980b, 1983a, 1979, 1980a, 1981; Roy et al., 1981; Pohl et al., 1981; Pohl, 1982, 1983b; Rivera et al., 1985; Phillips et al., 1987; Pohl, 1984, 1985; Pohl and Pollock, 1986; Pollock and Pohl, 1988). In the DEP method, the electric field induces a dipole moment in sample particles and the resulting force acting on them is the force of the electric field on a dipole. Since Pohl used small particles of a few micrometers in size to probe cellular EMFs, he often used the term “micro-DEP” (μ -DEP). In this method, particles were either repelled from or attracted to the surface of cells depending on whether they had a lower dielectric constant (BaSO_4 , SiO_2 , Al_2O_3) or higher dielectric constant (BaTiO_3 , SrTiO_3 , NaNbO_3) than the suspending medium, which was usually water-based. Based on these experiments, Pohl estimated that the frequencies of cellular electrical oscillations were in the radiofrequency range (5 kHz–9 MHz) (Pohl, 1980b; Pollock and Pohl, 1988). These experiments included tests of several types of cells including bacteria, fungi, algae, nematodes and mammalian cells, all of which showed, under suitable conditions, a dielectrophoretic effect caused by a cellular EMF (Pollock and Pohl, 1988). Other investigators reported similar findings for diverse cell types including human leukocytes (Pohl and Lamprecht, 1985; Hölzel, 1990, 2001; Pokorný, 1990; Jandová et al., 1987).

2.2.4. Direct electronic detection of cellular EMFs

One of the first pieces of direct evidence for the generation of EMFs in the spectral region of kHz–GHz by cells was obtained in a series of experiments that used direct electronic detection of EMF from a single cell or a suspension of cells. Using a spectrum analyzer, Jafary-Asl and Smith found EMF signals in the range of 7–80 MHz that were emitted from *S. cerevisiae* (Jafary-Asl and Smith, 1983; Del Giudice et al., 1989). Later on Rivera and Pohl (Pohl and Pollock, 1986) detected a spectrum of signals from the alga *Netrium Digitus* with peaks around 7 and 33 kHz followed by Hölzel who extensively analyzed the frequencies of a different group of cells in the MHz region (Hölzel, 1990; Hölzel and Lamprecht, 1995, 1994; Hölzel, 2001). Disagreeing with Jafary-Asl and Smith, Hölzel claimed that the frequencies reported by them were mainly artifact, probably due to positive feedback coupling in the amplifier. With improvement in detection techniques other researchers aimed to detect cellular EMF during the process of mitosis in cells they were partially successful in the MHz region (Jelínek et al., 1999, 1996; Pokorný et al., 2001) but had limited success in the range of 30–300 GHz (Jelínek et al., 2002, 2005, 2007; Kučera, 2006).

It is worth mentioning that currently available direct measurements of cellular EMF techniques can only detect radiative cellular EMF components. However, measurement of the nonradiative (near field) cellular EMF component, which is bound to cell structures and only measurable in the vicinity of cell boundaries, awaits further developments of special sensors using nanotechnology (Kučera et al., 2010) (Table 1).

2.2.5. Detection of cellular vibration states by spectroscopic techniques

EMFs, especially up to the region of a few THz, are generated by vibrations of electrically polar structures (see Section 2.4.2). The frequency of the vibrations of polar structures is equal to the frequency of the generated EMF.

Lower frequencies of mechanical oscillations (down to a few hundreds of Hz) can be optically detected (Piga et al., 2007; Popescu et al., 2007; Tuvia et al., 1998) and/or image analyzed (Koniar et al., 2009; Hargaš et al., 2008).

In Raman or Brillouin spectroscopy, one can use the “R” ratio (the ratio of anti Stokes and Stokes line intensities) to measure excitation of these vibrations above thermal levels. If the vibrations were excited above thermal levels, they could generate EMFs above the thermal noise level. Although detection of non-thermal vibrations in the THz region using Raman shifts has been reported for biological systems (Webb et al., 1977; Webb, 1980; Drissler and Santo, 1983; Drissler and MacFarlane, 1978; Del Giudice et al., 1985), it was criticized by others as being an artifact (Layne and Bigio,

1986; Layne et al., 1985; Furia and Gandhi, 1984, 1985; Cooper and Amer, 1983). Nevertheless, a model for interpretation of Raman spectra of metabolically active cells based on tight interplay between coherent electric vibrations and solitons has been proposed (Del Giudice et al., 1982). Similarly, as was argued by Kučera et al. (2010) about measurement of local cellular electrical oscillations, successful measurement of non-thermally excited vibrations in cells is possible only with near field modifications of inelastic scattering spectroscopy techniques (Kneipp, 2007) (such as Raman or Brillouin scattering) since the nanometer spatial resolution and measurement in close vicinity of the sample (under micrometer) is necessary.

2.3. Correlation of cellular EMF activity with cellular metabolism, vitality and replication

Some studies show a clear correlation of cellular function and cellular EMF activity. Pohl et al. (1981) observed significant cellular EMFs variations in yeast cells using the dielectrophoretic technique while exposing them to various chemical toxins, physical injury or during cell mitosis. There are also reports correlating the peak of cellular electromagnetic emission during cellular replication to the process of microtubule reassembly to form mitotic spindles, binding of chromatids to form kinetochore microtubules, and elongation of mitotic spindles during anaphase A and B (Pokorný et al., 2001). This suggests a crucial role of microtubules in generation of cellular EMFs (Pokorný, et al. 2001).

2.4. How cells can generate EMFs

2.4.1. Basis of EMF generation

The classical description of the generation and behavior of EMFs is given by Maxwell's equations. A static electric field is generated by a static charged particle. Both an electric field and a magnetic field are generated if a charged particle moves at a constant velocity. The term electromagnetic field is reserved for a condition where a charged particle is accelerated. In most cases the acceleration of a charged particle takes place in the form of an oscillation. Therefore, the electric and magnetic fields also oscillate. Any temporal change (nonzero time derivative) of the electric field gives rise to a magnetic field and vice versa. This creates an interconnection between oscillating magnetic and electric fields. The higher the frequency of the oscillation, the more electric and magnetic fields are mutually coupled. An oscillating charge may radiate an electromagnetic field. A radiated electromagnetic field, usually called an electromagnetic wave or, simply, radiation, carries away

Table 1

Direct electronic detection of EMF cellular signals. Indirect detection of cellular EMF, for instance by its dielectrophoretic effect, is not included.

Organism	Frequency or wavelength	References
alga <i>Netrium Digitus</i>	7 kHz, 33 kHz	Pohl and Pollock (1986)
yeast <i>Saccharomyces cerevisiae</i>	0.4–1.6 kHz	Jelínek et al. (2009), Cifra (2009)
	1, 7, 50 (60)–80 MHz	Jafary-Asl and Smith (1983), Del Giudice et al. (1989)
	8–9, 8.2 MHz	Jelínek et al. (1999, 1996), Pokorný et al. (2001)
	1.5, 2.6, 5.7, 18, 52 MHz	Hölzel (1990), Hölzel and Lamprecht (1995, 1994), Hölzel (2001)
	42 GHz (attempt only, not considered significant)	Jelínek et al. (2002, 2005, 2007), Kučera (2006)
yeast <i>Schizosaccharomyces pombe</i>	3.1, 4.8 MHz	Hölzel (1990), Hölzel and Lamprecht (1995, 1994), Hölzel (2001)
frog gastrocnemius muscle (electrically stimulated)	0.2–2 mm	Gebbie and Miller (1997)
crab nerve (electrically stimulated)	3–10 μ m	Fraser and Frey (1968)
various types of bacteria, plant, fungi, mammalian cells	visible, UV	a great abundance of data, e.g. books: Popp et al. (1992), Popp and Belousov (1996), Belousov et al. (2000), Popp and Belousov (2003), Van Wijk and Shen (2005), Belousov et al. (2007), Musumeci et al. (2003), Burr (1985), Multi-author review (1988, 1992), Multiauthor (2003, 2008)

energy. If the frequency of an oscillating charge is high and approaches the optical part of the EMF spectrum, the generated electromagnetic waves start to manifest their particle-like properties in their interactions with matter. This is when we can speak about particles of light or photons rather than electromagnetic waves. The generation of photons is usually interpreted as a process where a charged particle “drops” from a higher energy (excited) state to a lower energy (ground) state ($hf = E_2 - E_1$), where h is Planck’s constant, f is the frequency of the photon, E_2 is the energy of the excited state and E_1 the energy of the ground state. Although they may sound different, these two processes, charge oscillation and change of charge energy state, are intrinsically the same.

All objects, whether living or nonliving, are continuously generating EMFs due to the thermal agitation of their particles that possess charge. The EMF spectrum that is generated is described by Planck’s law for the ideal case of a blackbody in thermal equilibrium. EMFs generated thermally have a random, non-coherent character. However, the question remains whether the EMF that is generated by a biological entity is a simple EMF generated by an object or is part of a biological property of a living system. Based on the evidence presented later in the manuscript, it seems that such EMFs are an integral part of biological systems and are thus part of purposeful processes.

Physically, living biological systems are non-equilibrium (they have different energy levels than their surroundings) and open (they can exchange energy and matter with the surroundings) thermodynamic systems. Such systems may locally decrease entropy (increase order). Since living systems are not in thermal equilibrium, their electromagnetic (or generally, vibrational) spectrum may also deviate from the thermal spectra given by Planck’s law. Furthermore, the important question is whether the generated biological EMF can have a coherent component, since coherence enables very efficient energy and information transfer via the spatial and dynamic formation of interference patterns. Answer to this question may be at least partially elucidated when we describe the structures and processes responsible for cellular EMF generation.

2.4.2. Basis for cellular EMF generation

Various cell functions are associated with moving charges in cellular compartments and may generate EMF. For example, membrane depolarization (a neuron firing at several hundred Hz (Buzsaki et al., 1992)) generates oscillations of electric charges with higher harmonics, creating an EMF with a frequency up to 10 kHz (Collins et al., 2001). However, this phenomenon is limited to a group of specialized cells in higher organisms and not all cells in an organism are involved in the process of membrane depolarization. The question arises whether non-specialized cells that are not involved in cell membrane depolarization are also capable of generating coherent EMFs, and if so how. In 1968, Herbert Fröhlich postulated that biological systems exhibit coherent longitudinal vibrations of electrically polar structures (Fröhlich 1968a,b, 1969). Electrically polar structures by definition contain electric charges and can, under special conditions, generate EMFs when they vibrate. Not surprisingly, a majority of protein molecules are electrically polar structures. So, where in the cell could these coherent vibrations exist? The original Fröhlich model was general and did not limit the process to any particular cellular structure. In his model, when the energy supply exceeds a critical level, the polar structure enters a condition in which a steady state of non-linear vibration is reached and energy is stored in a highly ordered fashion. This order expresses itself in a long range phase correlation, which is physically similar to superconductivity and superfluidity, where the behavior of particles is communal and inseparable. The energy source in this model is metabolic energy, and the non-linearity of

the vibrating system is caused by a strong static electric field. The existence of very strong static electric fields in the cell membrane led Fröhlich to consider cellular membranes to be the source of the postulated vibrations. He also proposed the existence of a selective resonant interaction of similar frequencies of biomolecular EMFs between two systems (Fröhlich, 1972, 1975, 1970). In an elaborate and extensive series of publications, Fröhlich proposed a role for EMFs in cell growth regulation and deregulation (Fröhlich, 1980). In particular, he and some other workers thought that these processes could be involved in the development of neoplasias (Fröhlich, 1977, Fröhlich, 1978; Cooper, 1981; Fröhlich and Kremer, 1983; Fröhlich, 1988; Pokorný et al., 2008; Pokorný, 2009).

Fröhlich’s conjecture stimulated much enthusiasm in the scientific community and based on his theory, it was predicted that biomolecular EMFs would appear in the range of 100–1000 GHz. In fact, a group of researchers used Raman spectroscopy to probe the predicted vibrations (see Section 2.2.5). Fröhlich’s model also inspired several other studies and models that tried to address the same topic (Wu and Austin, 1979, 1978, 1977, 1981; Mills, 1983, 1994, 1995; Turcu, 1997; Paul et al., 1983; Tuszyński, 1985, 1988; Chatterjee and Fritz, 1987; Chatterjee et al., 1983; Paul et al., 1984; Tuszyński, 1985b; Bolterauer, 1999; Kouba, 1998; Pokorný, 1987, 1982a; Pokorný et al., 1984; Pokorný and Fiala, 1994; Pokorný et al., 1991; Pokorný and Wu, 1998; Pokorný, 1982b; Pokorný et al., 1986a,b; Mesquita et al., 1998; Šrobár and Pokorný, 1996; Šrobár, 2009a,b; Amoroso, 1996; Bolterauer and Tuszyński, 1989; Tuszyński, 1985a; Tuszyński et al., 1992; Chatterjee and Fritz, 1987; Bolterauer et al., 1991; Tuszyński, 1987; Tuszyński et al., 1984; Tuszyński and Paul, 1991; Ristovski et al., 1992; Kocić et al., 2000, 2001; Mesquita et al., 1998, 1996, 2005, 2004; Jaggard and Lords, 1980; Illinger, 1982; Reimers et al., 2009; McKemmish et al., 2009; Hyland and Rowlands, 2006).

After the discovery of the cellular cytoskeleton, microtubules (MTs) became a serious candidate as the source of cellular EMFs. This was due to the fact that MTs fulfill requirements for the Fröhlich system and for generation of electromagnetic fields. They are composed of tubulin heterodimer subunits that electrically are highly polar. MTs resemble hollow tubes whose growth (driven by tubulin polymerization) is nucleated by centrosomes near the cell nucleus. MTs are characterized by their perpetual alternation between growth (tubulin polymerization) and shrinking (MT depolymerization). This dynamic instability provides a constant influx of energy via assembly and disassembly of GTP rich tubulin heterodimer subunits (Caplow et al., 1994; Caplow, 1995; Caplow and Shanks, 1996). The other energy supply for MT vibration could come from the movement of MTs aligned with motor proteins or the energy that is dissipated from mitochondria (Pokorný et al., 2008; Cifra et al., 2010). Mitochondrial ATP production by the citric acid cycle has an efficiency of ca. 40%. The remainder of the energy usually dissipates as infra-red vibrations as well as infra-red and optical (Hideg et al., 1991) radiation. Efflux of energy from mitochondria represents the most significant source of energy for excitation of vibrations. Mitochondria are also a source of strong static electric fields – in the range of 10^6 V/m – due to the creation of a hydrogen ion gradient. This static electric field of mitochondria penetrates up to a few micrometers into the cytosol (Tyner et al., 2007). The presence of mitochondria along the length of MTs leads to non-linearity caused by their strong static electric fields; it also provides the required energy for EMF generation through dissipated energy. Furthermore, microtubules are capable of vibrations in kHz to GHz regions (Sirenko et al., 1996; Wang et al., 2006; Wang and Zhang, 2008; Gu et al., 2009; Wang et al., 2009; Portet et al., 2005). The excitation and vibrations of MTs were the mainstay of the model that was proposed by Pokorný (Pokorný et al., 1997; Pokorný, 1999; Pokorný et al., 1998). Some scientists raised doubts

about his theory because they assumed that a viscous cytosol should dampen any vibrating cytosolic organelles (Foster and Baish, 2000; Adair, 2002). The cytosol could have a dampening effect on organelle vibrations if there was a “no-slip” boundary condition between cellular structure and the surrounding cytosol. However, as argued by Pokorný (2003, 2005), a lowered mobility of ions in the cytosol creates a “slip” between the microtubule, the adjacent ionic layers and the cytosol, which makes vibrations of microtubules in the cytosol physically plausible.

Other cellular structures were also considered as potential sources of EMF generation. Technically, any cellular structure or substructure can oscillate at its resonance frequency (Eigenfrequency) when excited by energy, unless strongly damped. For example, Smith calculated that a spherical cellular membrane has a mechanic resonance frequency of 10^{10} Hz (perpendicular to the membrane surface) and a circumferential frequency of 10^8 Hz (parallel to the membrane surface); electromagnetic resonance of the cell membrane (parallel to the membrane surface) occurs at a frequency of 10^{13} Hz (Jafary-Asl and Smith, 1983). Weak resonances in the region around 36–38 GHz have also been detected on erythrocyte ghosts in suspension (Blinowska et al., 1985).

Another cell membrane related theory of EMF generation stems from Russian scientists, especially from Devyatkov and his coworkers (Devyatkov et al., 1991; Betskii et al., 1988). They considered deformations and asymmetries of polar cellular membrane as a mechanism for generation of acousto-electrical waves whose EM radiative component depends on deviation from the healthy state; in the case of healthy nondividing cells, both the radiation from and the EM sensitivity of the cells is lowest. Vacuum wavelengths of the generated EM waves fall into the region of millimeter waves due to the geometric and mechanical properties of cellular membrane.

The other theory of EMF generation relates to the electrosoliton. The electrosoliton is the electrical counterpart of soliton. The soliton is a self-reinforcing solitary wave (a wave packet or pulse) that maintains its shape while it propagates. Electrosolitons can be viewed as moving charges that provide transport of charge in biological systems and can be considered as an important contender of EMF generation in the microwave frequency region (Brizhik and Eremko, 2003; Brizhik, 2003; Brizhik and Eremko, 2001; Musumeci et al., 2003).

While the previous model is more or less physical and based on non-linear wave theory, an electrochemical model was proposed by Pohl. He suggested that cellular EMFs can be generated by the coupling of oscillating chemical reactions to physically mobile ions within the regions of the cell to produce charge waves (Pohl et al., 1981; Pohl, 1982). In this model, oscillations of ions can be induced by chemical reactions, and the direction of oscillations will be steered by laminar and filamentous cellular structures. Indeed, oscillating chemical reactions such as the Belousov–Zhabotinski reaction are typical in non-equilibrium systems such as biological systems (Voeikov et al., 2001a,b; Epstein and Showalter, 1996; Epstein et al., 1983). Nevertheless, Pohl’s model of cellular EMF generation has not been developed further.

Authors of all of the above mentioned models created them directly to describe how cellular EMFs can be generated. To the contrary, there is also a recent model by Gov which analyzes mechanical vibrations based on the membrane-bound cytoskeleton (Gov et al., 2003; Gov and Safran, 2004, 2005a,b; Gov and Gopinathan, 2006; Lin et al., 2006), not mentioning any connection to electric oscillations. However, we assume that these mechanical vibrations can drive oscillations of electrically polar transmembrane proteins, thus generating electric oscillations. Nevertheless, in his model, the frequency limit of the oscillations only reaches 30 Hz, which is the upper frequency of membrane mechanical oscillations

that have been detected experimentally by some authors (Brochard and Lennon, 1975; Korenstein and Levin, 1990; Tuvia et al., 1998, 1992; Bitler and Korenstein, 2004; Tuvia et al., 1997, 1999; Krol et al., 1990; Popescu et al., 2007, 2006).

Parallel to these studies trying to explore the origin of cellular EMFs in the frequency range under the THz range, other researchers focused on the optical region of the EMF and cellular optical properties. These studies started with the work of Gurwitsch in the 1920s on mitogenetic radiation, which was believed to be in the UV region of the EMF spectrum (Voeikov and Belousov, 2007; Gurwitsch and Gurwitsch, 1959; Rahn, 1936). After an extensive review of this field, we now know that electronically excited biological systems can generate UPE in both UV and visible regions, which is the basis for many chemiluminescence assays. These assays are usually designed to detect reactive oxygen species (ROS) (Boveris et al., 1980; Cadenas, 1984; Van Wijk et al., 2008). Thus, it is not surprising that the mitochondria, which are an important cellular source of ROS production, could be the major cellular organelles that emit UPE (Hideg et al., 1991; Cadenas et al., 1980; Creath, 2008). Mitochondria are also a very likely source of near infra-red EMFs as has been proposed in other studies (Albrecht-Buehler (2000); Tuszyński and Dixon, 2001).

In his study, Slawinski reviewed physicochemical luminescence processes that could generate UPE in cells (chapter in Multi-author review, 1988). He proposed that there is a possibility of photo-emission from collective molecular interactions such as relaxation of superhelical DNA, or movement of other biomolecules, or collective excitations in the electric field of biomembranes and perturbed cytosolic water.

Voeikov is another scientist that believes that there is a nontrivial role of cellular water in generation of electronically excited EMF (Voeikov, 2005). It was experimentally shown that various low energy physical processes such as ultrasound agitation, passing water through capillary tubes and external microwave EMFs can influence UPE generation in water (Vaks et al., 1994). Preparata and del Giudice (Preparata, 1995) have theoretically shown that liquid water is effectively composed of two phases: a gas phase and a “coherent domain” phase. Coherent domains behave as reservoirs of quasi free electrons which can be released just by modest excitation (Del Giudice et al., 2009). Separation of these two phases of water is likely to occur near interfaces, such as membranes, hydrophilic surfaces, polar molecular backbones, etc. Indeed, it was experimentally shown by many authors (e.g. Zheng and Pollack, 2003; Zheng et al., 2006; Chai et al., 2008, 2009; Pollack et al., 2009) that water near interfaces exhibits quite different properties than bulk water. These include solvent exclusion, higher viscosity, lowered thermal motion of molecules, separation of charge, different spectroscopic properties etc.

Even though it is plausible to consider UPE from biosystems, how can we explain the alleged coherence in UPE phenomena (Popp and Yan, 2002; Popp et al., 1992, 2005; Bajpai, 1999, 2003; Popp and Belousov, 1996)? Popp proposed that DNA in the nucleus is the main source for stimulated or spontaneous coherent radiation. This is mainly due to the luminescent properties of the nucleotides, due to their cooperative behavior while in DNA and due to continuous supply of energy through metabolic processes (Popp et al., 1992, chapter 5) and (Popp and Li, 1990). Popp and his coworkers suggested, based on experimental data of other workers (Vigny and Duquesne, 1976), that stacking of bases in DNA provides suitable conditions for stimulated radiation (Popp et al., 1989, chapter by Popp). He also suggested that DNA is suitable as a photon storage system due to its spatial conformation. Following his theory, unwinding of DNA during replication causes greater UPE. In fact, he showed that unwinding of DNA induced by ethidium bromide resulted in increased UPE (Rattemeyer et al., 1981, 1984). To explain

how there could be coherence in spontaneous EMF radiation, Popp also considered Dicke's theory (Dicke, 1954; Scully and Svidzinsky, 2009). This theory explains how two radiating molecules that are within the coherence length of their radiation of each other can give rise to a correlated radiation. In fact, from an informational point of view, the coherence properties of UPE in photocount statistics (PCS) are of greater importance than the intensity of UPE. Other scientists studied the special states of coherent cellular UPE (squeezed states) in super- and sub-Poissonian models (Bajpai et al., 1998; Bajpai, 2004, 2005; Popp et al., 2002).

It is generally accepted that the main sources of cellular UPE are excited molecules such as reactive oxygen species (ROS), which can release their energy in rather random chemical reactions as photons. However, ROS can also provide energy to macromolecular structures which can store the energy and release it in a more coherent fashion. We are of the opinion that macromolecules can modify the statistical properties of light due to their ordered structure and store or emit UPE. The effect of structure and order in biosystems on emitted light was explored by a few researchers. (Yan et al. (2005)) found that homogenized solutions of cytoplasm have different photocount distributions than intact cellular structures, despite the fact that the chemical composition is the same. In this study, they observed that a whole leaf exhibits oscillations in emitted delayed luminescence intensity after illumination, while leaf homogenates or isolated chloroplasts in buffered solutions do not show such oscillations. Budagovsky et al. (2002) and Borodin et al. (2008) used degree of spatial coherence of laser light scattered from leaves of various plants to determine the plant health state. Works of Yan and Budagovsky showed that the order in biosystems influences statistical and coherence properties of the light re-emitted after excitation. Thus, it is rather safe to assume that the coherence (more generally stated, statistical properties) of endogenous biological light – UPE – are influenced by the order in biosystems. The order in biosystems that influences the coherence properties of UPE could be related both to spatial morphological order and temporal (dynamic) order.

The question that remained to be answered is “can we reconcile the models that explain cellular EMF generation in the kHz–GHz range with the models that explain UPE (EMFs in the optical region)?” At first glance, models describing the generation of lower frequency cellular EMF use vibrations of electrically polar structures while generation of optical EMF is rooted in electronic excitation of molecules. However, some scientists such as Swain and Popp proposed that there is a connection between the different spectral ranges of cellular EMFs. They proposed that if certain non-linear conditions are fulfilled, upconversion (summing the energy of photons to give higher energy photons) of photons even in the range of GHz frequencies could give rise to optical photons (Swain, 2006; Popp, 2006; Swain, 2008). It also remains to be seen whether a down conversion could be a mechanism for generation of lower EMF frequencies from energy in the optical EMF spectrum.

3. Evidence for cells being affected by electromagnetic fields

3.1. The effect of EMFs on biological systems

There is a huge body of evidence in the literature that sheds light on our understanding of how biological tissues are being affected by external EMF (Research Center for Bioelectromagnetic Interaction, 2010). However, those acquainted with this research field know that there has been an unsettled discussion for several decades regarding the safety and degree of influence of these fields on living organisms. With advances in technology, we are now exposed to increasing numbers of EMFs including extra low-frequency EMFs from electric power lines, EMFs from cell phones and microwaves,

and many EMFs at mid range frequencies such as house appliances and remote controls. From a biophysical standpoint, static and low-frequency electric fields are attenuated rather rapidly by mobile ions in solution or in the cell cytosol within a very short distance, which allows only minimal electric field penetration into the tissue. Nevertheless, static or low-frequency fields may have biological effects due to redistribution of ions and weak currents they induce (Marino and Becker, 1977; Robinson, 1985; Portier and Wolfe, 1998; McCaig et al., 2005; Funk and Monsees, 2006; Teissie, 2007; Funk et al., 2009). The biological effects of low-frequency magnetic fields have been the subject of more extensive studies since they can penetrate deeper into tissues (Marino and Becker, 1977; Adey, 1980; Robinson, 1985; Frey, 1993; Hong, 1995; Portier and Wolfe, 1998; Volpe, 2003; Funk and Monsees, 2006; Funk et al., 2009).

For instance, it has been shown that low-frequency EMFs can act at the cellular level and affect various cell functions, including cellular proliferation and differentiation (Foletti et al., 2009; Lisi et al., 2006, 2008a,b; Ventura et al., 2005; Ross, 2005), apoptosis (Tian et al., 2002; Tofani et al., 2001; Santini et al., 2005), DNA synthesis (Takahashi et al., 1986; Litovitz et al., 1994), RNA transcription (Goodman et al., 1983), protein expression (Goodman and Henderson, 1988), protein phosphorylation (Sun et al., 2001), redox-mediated rises in NFκB and cell damage (Wolf et al., 2005; Regoli et al., 2005), microvesicle motility (Gölfert et al., 2001), ATP synthesis (Zrimec et al., 2002), hormone production (Paksy et al., 2000), antioxidant enzyme activity (Kula et al., 2000), metabolic activity (Milani et al., 2001), and inhibition of adherence (Jandová et al., 2001).

EMFs of intermediate frequencies (kHz–MHz) under suitable geometry have been shown to be effective in arresting the growth of cells (Giladi et al., 2008) – with direct applications to cancer treatment (Kirson et al., 2004, 2009).

The effect of higher frequency EMFs on biological systems can be divided into thermal effects and non-thermal effects. The extent of the thermogenic effect depends mainly on EMF intensity, which is associated with the specific absorption ratio (SAR). Obviously, increased temperature or a thermal effect can result in changes in cellular function and, under extreme conditions, can result in cellular damage. However, in this section, our focus is on non-thermal, direct effects of EMFs on biological systems. There are numerous published papers that show different modifications of biological systems after exposure to weak EMFs (Belyaev, 2005a,b; Belyaev et al., 2000; Phillips et al., 2009; Banik et al., 2003; Kumar, 2003; Sienkiewicz, 1998; Berg, 2004; Repacholi et al., 1998). There are also many published studies that show the lack of any significant effect of weak EMFs on biological systems (see list of reviews above). In general, many earlier studies in this field of bioelectromagnetics suffer mainly from poor design, lack of appropriate control groups, and an abundance of confounding factors, which in turn results in reduced reliability and reproducibility. One of the major problems lies in the fact that there could be uncontrolled microthermal effects at subcellular levels even in the setting of weak EMFs. This is particularly true in non-uniform media such as living cell samples. Furthermore, Morrissey et al. have shown in an *in vitro* experimental model that even a small temperature rise (0.2 °C) can cause biological effects (Morrissey, 2008). Thus, meticulous dosimetry is a crucial aspect of designing experiments for studying the biological effects of EMFs. Besides the difficulties in controlling EMF microthermal effects, there are several other non-thermal parameters of EMF which are very important and could act as confounders in research if they are not meticulously considered in the study design. A few examples of these parameters include frequency of the carrier wave, modulation frequency, near field/far field effect, polarization, duration of the exposure, continuous or pulse wave exposure, shape of the pulse and, finally, presence or absence of a static magnetic or electric field. To complicate the

matter even more, there are several biological factors that also play a major role in the response of a biological system to EMFs. One of the major parameters is the stage of cell differentiation and replication. This factor plays a major role in the way cells respond to the same EMF, and can create a significant problem in reproducing research results (Belyaev et al., 1996; Osepchuk and Petersen, 1997; Belyaev et al., 1998, 2000; Osepchuk et al., 2002). Regardless, it is hard to interpret the effects observed in biological systems that are exposed to weak EMFs solely based on minute thermal effect of these fields (Belyaev, 2005a,b; Belyaev et al., 2000). For example, in extensive studies performed by Grundler and Keilmann on yeast cells, there was either suppression or stimulation of cell growth when cells were exposed to certain frequencies of electromagnetic radiation around 42 GHz in a resonant-like manner – the effect on growth rate was positive at certain frequencies and negative at other frequencies. The distance between adjacent frequencies was about 8 MHz (Grundler et al., 1977; Grundler and Keilmann, 1978; Grundler, 1983; Grundler and Keilmann, 1983, 1989; Grundler et al., 1982, 1988; Grundler and Kaiser, 1992; Grundler, 1995). This phenomenon does not seem to be related to biological system geometry, but more likely to frequency of cellular vibration states and their higher harmonics. Another example represents the effects that occur in certain intervals of EMF power (Shcheglov et al., 1997; Belyaev et al., 1996; Adey, 1980) or for certain EMF polarizations (Belyaev et al., 2000). It has been found that there are similarities in the effects of certain low frequency EMF and high frequency (RF) EMF amplitude-modulated by the same low frequency (Blackman, 1985; Schwartz and Mealing, 1993; Shcheglov et al., 2002; Lass et al., 2002; Ayrapetyan et al., 2009).

Bioeffects of low intensity 30–300 GHz EMFs (millimeter wavelength EMFs) have been studied extensively in cell and animal models, but not generally accepted by western scientists. Nevertheless, stemming especially from workers of eastern origin, there are even human studies in the form of double-blind placebo-controlled trials that evaluated the efficacy of millimeter wavelength EMFs in the therapy of peptic ulcers, postoperative wound infection, pulmonary tuberculosis and rehabilitation of patients after myocardial infarction (Pakhomov et al., 1998; Pakhomov and Murphy, 2000; Rojavin and Ziskin, 1998; Betskii and Lebedeva, 2004; Fedorov et al., 2003).

There are also studies that explored the effect of low level light on biological systems particularly at cellular and subcellular level (Smith, 1991; Hamblin and Demidova, 2006; Hawkins and Abrahamse, 2007). Since the bioeffects of low level optical EMF (light) are more accepted than the bioeffects weak the microwave and lower frequency EMFs, we will not go into such details. In a series of experiments, researchers showed that low intensity light can influence physiological processes, particularly the activity of Na^+/K^+ -ATPase (Kilanczyk et al., 2002), intracellular Ca^{2+} concentrations (Volotovskii et al., 1993; Alexandratou et al., 2002; Greco et al., 2001), mitochondrial ATP synthesis (Passarella et al., 1984), production of reactive oxygen species (Lubart et al., 2005) and cell proliferation (Khadra et al., 2005; Tuby et al., 2007).

3.2. How EMFs affect biosystems

3.2.1. How cells react to non-visible EMFs

Numerous models have been proposed to explain how weak EMF fields interact with biological systems (Beneduci, 2008; Foster, 2000; Karimov and Shcheglov, 2000; Berg, 2004; Sonnier and Marino, 2001; Chiabrera et al., 1985; Challis, 2005; Ho et al., 1994; Kanokov et al., 2009). In most of these models, a subcellular organelle or even a single biomolecule was the target of a cellular EMF. Regardless, one of the major problems in explaining how an EMF affects a biosystem lies in the fact that the quantum of energy

of an EMF with a frequency lower than a few THz in most cases is less than the average energy of thermal noise (kT constraint) and if, the EMF has a quantum of energy smaller than the average energy of thermal noise, then EMF absorption cannot significantly influence biosystems by mechanisms other than thermal effects (Adair, 2003). Thermal noise has an average energy of kT (at a room temperature of 20 °C, kT corresponds to 26 meV, which is the energy of a single EMF quantum with a frequency of 6.2 THz and a wavelength of 48 μm in a vacuum).

However, (Binhi and Rubin, 2007) explain that the notion of a “ kT constraint” originates from a statistical physics model and is only applicable to systems near thermal equilibrium. In such systems, low intensity, low-frequency EMFs cannot change the mean energy of cellular structures or more precisely, the vibrational energy of their molecules stored in their degrees of freedom. Degrees of freedom is the expression for the ways a molecule or a structure can move (vibrate, rotate, etc.) and the energy absorbed by a degree of freedom of a molecule from an EMF at a certain frequency can not be “stored” or accumulated at that certain frequency if that degree of freedom is coupled to other degrees strongly. In that case, the energy at that certain frequency will be redistributed to other degrees of freedom (frequencies) and will be dissipated very rapidly. However, biological systems, in general, are not in thermal equilibrium. In fact, they are thermodynamically far from equilibrium and some of their degrees of freedom are weakly coupled to others or to the surrounding heat bath. Thus thermalization time (time needed to redistribute energy into other degrees of freedom) may be significantly greater than in systems in thermal equilibrium (simple inanimate or dead systems). Indeed, in some systems, this number may even be greater than the characteristic lifetime of the system. Thus, it is not surprising that EMFs can induce a significant change in energy in some degrees of freedom before dissipation or redistribution of the energy. In particular, Binhi showed that the magnetic component of the EMF can interact with cellular magnetic nanoparticles. Natural magnetic nanoparticles are indeed found in many organisms from bacteria to humans (Posfai and Dunin-Borkowski, 2009; Kirschvink et al., 1992a,b). Long-lived rotational states of some molecules within protein structures, spinning magnetic moments in radical pairs, and even the magnetic moments of protons in liquid water are other possible targets of magnetic field interactions (Binhi and Rubin, 2007; Binhi, 2001).

Pilla et al. (1992) also calculated that the kT limit can be decreased significantly for cell arrays in tissues where cells are conductively connected through gap junctions. This decrease could be on the order of ten to one hundred fold, and depends on the frequency of the EMF as well as cellular dimensions.

The other problem with the effect of EMFs on biosystems has to do with the coupling of energy. From an engineering standpoint, the absorption of EMF energy is inefficient when the receiving antenna, which in this case is a group of molecular dipoles, is very small compared to the radiating EMF wavelength. However, coupling and energy transfer of an EMF to cellular structures may become greater if there is a resonant interaction of the EMF with vibrational modes of the cellular structures (Adair, 2002). But is it possible to have a long-lived vibrational movement of cellular structures in a viscous medium such as the cytosol? In his book on quantum electrodynamics in matter, Preparata et al. predicted that liquid water is composed of two phases, distinct coherent domains of water which have different physical properties such as viscosity and density compared to “bulk” gas-like water which is present between the coherent domains. Based on this theory, other scientists proposed that the microvolumes of water that surround biomolecules may exhibit drastically lower damping (Preparata, 1995; Del Giudice et al., 2002; Zhadin and Giuliani, 2006). This is supported by several studies, in which extremely sharp resonances in low-frequency magnetic fields have been observed (Foletti et al.,

2009; Lisi et al., 2006; Zhadin, 2000; Zhadin et al., 1999; Pazur, 2004; Grundler and Kaiser, 1992; Belyaev, 2005a; Smith et al., 1987). A low damping rate (collisionless movement) is a necessary assumption in a model proposed by Liboff which describes a resonance of moving ions in an external magnetic field by a cyclotron resonance mechanism (Liboff and Jenrow, 2000; and chapter in Chiabrera, et al. 1985). In his model, an alternating magnetic field acts resonantly on a moving ion, and the frequency of the resonance depends on the magnitude of the static magnetic field component that is parallel to the alternating magnetic vector.

Coupling of EMFs in the microwave and millimeter wave regions to vibrations of cellular structures is also consistent with a part of Fröhlich's theory (Fröhlich, 1980; Betskii and Lebedeva, 2004; Pakhomov et al., 1998) which predicts resonant interaction of biomolecular structures with external electromagnetic fields. With a similar assumption, Sinitsyn et al. (2000) proposed generation of intrinsic resonance frequencies by water clusters in biosystems exposed to weak EMF in the range of 50–70 GHz. At these frequencies, water-molecule oscillators lock on to the external signal frequency and amplify the signal by means of synchronized oscillations or regenerative amplification. Then, waves at these frequencies can pass through aqueous media almost without significant resistance and penetrate deep into the biosystem. Betskii and Lebedeva (2004) also pointed to stochastic resonance as a possible process involved in EMF non-linear interactions with biosystems (Anishchenko et al., 1999; Neiman et al., 1998). In stochastic resonance the sensitivity of a system to weak periodic signals is actually increased when the optimal level of random noise is added (Hänggi, 2002).

In three engineering models, Barnes (1992) explains the mechanisms through which EMFs can be extracted from a noisy background by biological systems. The first model is based on oscillation injection-locking processes in which the signal-to-noise ratio can be less than one. The second model is based on parametric amplification and assumes that the external signal and the biological process can have different frequencies and that the necessary requirement for functioning of this model is phase stability of the EMF frequency. When the phase of the signal is unstable, no net amplification can take place. In the third model, Barnes uses a computer model to simulate a neural network, which can be trained to identify a field of definite frequencies at signal-to-noise ratios much less than one. The prerequisite for all of these models is the existence of long lasting coherence of the external EMF.

The other theory in this field was proposed by Tsong et al. (Westerhoff et al., 1986; Tsong and Astumian, 1986; Markin and Tsong, 1991a,b; Tsong, 1992). This group proposed electro-conformational coupling (ECC) for cellular enzymatic systems. In this theory, enzymatic systems, particularly those in membrane structures, are able to receive, process and transmit high and medium level intensity periodic potentials, e.g. electric potentials. ECC theory describes four-state enzyme systems that convert electric field energy into chemical potential energy if the frequency and the strength of the applied field properly match the characteristics of the system. They also proposed a theory of oscillatory activation barriers (OAB) (Markin et al., 1992; Tsong, 1992), one which explains the processing of low level periodic electric potentials. Unlike the ECC model, in the OAB model the electric field does not influence the chemical equilibrium of a reaction. Instead, it alters the rate of a reaction by interacting with the activation barrier. In the OAB model the energy barrier between the states of an enzyme can be set oscillating by the applied oscillating electric field. The rate of reactions involving the enzyme can be influenced by a certain frequency of the electric field. Tsong, in his molecular recognition theory of electric fields (including both ECC and OAB) in 1992 suggested that “a molecule which is immobilized, oriented or tumbling more slowly

than the frequency of the periodic field, may interact with the field to produce chemical effects that are uncommon in homogeneous solution” (Tsong, 1992). These conditions are fulfilled when the molecule is part of a complex structure. This strongly suggests that for detection of a weak field, molecules need to be in a structured or anisotropic medium (e.g. in a membrane where the field is amplified, in macromolecular filaments of the cytoskeleton, or in cellular bound water) to rectify and accumulate the energy. It has been shown experimentally (Tsong, 1992; Knox and Tsong, 1984; Serpersu and Tsong, 1984; Liu et al., 1990), as predicted by the ECC and OAB theories, that there are optimal frequencies and amplitude intervals of oscillating electric fields and optimal ligand concentrations for efficient coupling of the electric field to enzyme systems. To test the ECC theory, they did a series of experiments in which the electric field could induce cation pumping activities of Na^+/K^+ -ATPase and Ca^{2+} -ATPase in human erythrocytes, and ATPase activity in beef heart mitochondria. Supporting the OAB theory, they successfully stimulated ATP hydrolysis by weak periodic electric fields. Optimal frequencies depended on the system under investigation and varied from 10 Hz to 1 MHz. Applied electric field intensities in the experiments of Tsong et al. were about a few V/cm for low level fields and a few kV/cm for high level fields. It is known that externally applied electric fields with frequencies less than 1 MHz, can be amplified by cellular membranes (Foster and Schwan, 1989; Tsong and Astumian, 1986). The basis for this phenomenon resides in the properties of the membrane that allow it to act as an insulator and effectively blocks the passage of the electric field across the membrane. This results in a drop of voltage across the membrane (V_m) given by the equation

$$V_m = 1.5E_{ex}R \quad (1)$$

where E_{ex} is the intensity of the uniform externally applied electric field, and R is the radius of the cell. Intensity of the field in the membrane E_m is given by the drop in voltage (potential difference) across the membrane divided by d – the thickness of the cell membrane

$$E_m = \frac{V_m}{d} = 1.5E_{ex}\frac{R}{d} \quad (2)$$

Thus the field in the cellular membrane E_m is effectively amplified compared to the intensity of the external applied field E_{ex} by the ratio $1.5 R/d$. Taking this into consideration along with the ECC and OAB theories, Tsong proposed that under suitable conditions even detection of a weak field at the level of a few nV/cm by membrane macromolecules is feasible. The ECC theory can also be used to explain how a molecular system can serve as a generator of specific EMFs if the inverse mechanism of the ECC model (conversion of chemical energy to electrical energy) is being considered (Tsong, 1989; Tsong and Gross, 1994).

Stoykov et al. (2004) used a computational model to examine a non-thermal electromagnetic interaction mechanism in living cells with a focus on non-linear behavior of the cellular sodium ion channel when exposed to microwave EMFs. Their results imply that an amplitude-modulated microwave electric field can induce low-frequency ion currents in cellular sodium channels. This may be due to a non-linearity that is inherent in the ion-flow process.

In addition to membrane-bound enzymes and pumps, other membrane macromolecules were also considered as the potential unit of cellular EMF reception. Weaver and Astumian (1990) presented a physical model in which membrane macromolecules can directly respond to very weak external electrical fields as low as (10^{-6} V/cm) if there is a narrow band of frequencies to which the molecule responds, or if signal averaging occurs. If the applied signal is periodic and there are means of time integration of the

signal in the biosystem, then the noise is averaged out over subsequent periods of integration and thus, the signal-to-noise ratio is increased.

Balzano examined EMF interactions with biosystems from a non-linear thermodynamics perspective (Balzano and Sheppard, 2003, 2007; Balzano, 2003). He showed that a non-linear interaction could be due to scattering of EMFs from (molecular) oscillators located in strong electric field gradients in regions such as membrane surfaces. He also proposed that non-linear mechanisms could appear if stress (elastic) waves were induced due to EMF heat generating properties. A sudden, pulse-like absorption of a large amount of EMF energy can cause heat stress and expansion of the exposed biosample. Elastic waves induced in this manner could interact with incoming photons and give rise to modulation products of frequencies different than the incoming EMF, which could induce bioeffects.

Several other theories consider simple or complex molecules as the unit of cellular EMF reception. Van Zandt (1978) proposed that resonant interactions between biological molecules could assist several types of interactions. He showed that forces between localized, oscillating vibrating molecules could be modified based on the relative frequency difference between the two oscillators. The proposed model resembles that of the Fröhlich theory proposed in the 1960s–1970s, but there are slight differences which cause the interactive force of oscillators in Van Zandt's model to be much more selective. Later on, Karimov et al. (1999) proposed a model for the mechanism of interactions of weak EMFs with biomolecules. The focus of this model was based on the dependence of the natural resonances of biomolecules on the shape of the EMF signal and features of biomolecular structures and active centers containing metal atoms.

In 2002, Goodman and Blanck proposed the existence of electron transport (charge flow) in DNA. They suggested that low-frequency EMFs can interact directly with electrons in DNA molecules which eventually translate into biosynthesis of proteins (Goodman and Blank, 2002). Edwards et al. (1984, 1985) also provided evidence of direct resonant DNA absorption of microwaves. His findings were later challenged by some scientists (Foster et al., 1987; Gabriel et al., 1989), while others supported them (Tao et al., 1987b, 1987a; Mei et al., 1981; Lindsay and Powell, 1983; Van Zandt, 1986; Van Zandt and Saxena, 1989).

Kaiser considered coupling of EMFs to intracellular Ca^{2+} oscillations as a possible mechanism (Grundler and Kaiser, 1992; Kaiser, 1995, and chapter in Chiabrera et al., 1985). In his general model, he explained how non-linear oscillators could manifest specific phenomena including synchronization, sub- and superharmonic resonances, and frequency and intensity sensitivity. This implies that biosystems can be very sensitive to changes in the parameters of external EMFs. Several other scientists proposed similar models that focus on calcium as the probable candidate for reception of EMFs in biosystems (Gapeyev and Chemeris, 2000; Liboff and Jenrow, 2000).

Another interesting theory in this field deals with free radical chemistry. Vaks et al. (1994) showed that low energy physical factors such as microwave EMFs may induce homolytic cleavage of water and produce free radicals. Free radicals are also produced constantly in cells by mitochondria, chloroplasts or during other cellular metabolic reactions and serve as a mediator in several cellular metabolic pathways.

Keilmann proposed another theory based on molecular spin. Spin is a kind of intrinsic degree of freedom of molecules and particles, which, in simple words, could be imagined as a rotational movement of particles around their own axis. Spin is weakly coupled to other degrees of freedom and thus can be non-thermally populated. EMFs can influence the population of spin states. Triplet and radical molecules are the special target of this theory since they

have nonzero spin. Hence, if these molecules belong to a reaction chain where the reactivity is dependent on spin, EMFs can influence the reaction rate (Grundler and Kaiser, 1992; Adey, 1993) and triplet mechanisms (resonance dependency on EMF frequency) (Keilmann, 1986; Grundler and Kaiser, 1992).

In the other model, Brizhik et al. discussed the effect of EMFs on biosystems through influences of solitons, which can provide dissipationless energy and information transfer (Brizhik and Eremko, 2003; Brizhik, 2003; Brizhik and Eremko, 2001). In their theoretical analysis, they showed that the spectrum of biological effects of EMFs can be divided into two major bands. The lower frequency band is connected with an intense form of EMF energy absorption and consequent emission of sound waves by solitons. In contrast, the higher frequency bands can induce soliton transitions to delocalized states and thus are able to destroy the soliton and disrupt the transfer of energy and information.

Another important point in understanding the nature of the effect of EMF on biosystems can be inferred from Levin's conclusion (Levin, 2003). In his view, when it comes to affecting a biosystem, the intensity of the EMF is of much lesser importance compared to the amount of information it carries. He concluded that the effect of an EMF can be in general much stronger on the whole biosystem compared to its effects on cells, cell organelles and macromolecules (original citation (Barnothy, 1964)).

3.2.2. How cells react to EMF in the visible range

Interactions of biosystems with EMFs in the visible range have a wider acceptance among scientists since quanta of such EMFs have higher energy levels than the average energy of thermal noise. Most of the models in this field closely link EMF reception to EMF emission. This is done because in most cases both processes share a common particulate matter and mechanism. It seems that a major problem for current molecular biologists is to understand how very weak EMFs in the range of a few to tens of photons in the visible range could couple to molecular signaling pathways. Is there any amplification mechanism in the cell? In fact, in his original theory, Gurwitsch proposed that there is an amplification mechanism in cells that lies in the initiation of branching chain reactions (Voeikov and Belousov, 2007). Gurwitsch also stressed that the sensitivity to small numbers of energy quanta is possible only in systems in a "non-equilibrium state" (originally in German "unausgeglichene Konstellation") (Gurwitsch and Gurwitsch, 1959). This closely agrees with the physical properties of living systems which are in a non-equilibrium thermodynamic state.

In his review article, Karu explains how cytochrome c-oxidase can act as a photoreceptor in eukaryotic cells exposed to monochromatic red to near-IR radiation (Karu, 1999; and chapter in Musumeci et al., 2003). Albrecht-Buehler also suggested that cells are capable of detecting EMF in the near infra-red range. However, he proposed that centrioles serve as the "eyes" of the cell (Albrecht-Buehler, 2010, 1992). This is based on their specific structure and perpendicular conformation, which enable these organelles to detect the source and the angle of the irradiated IR EMF.

Gao et al. (2009) used a molecular approach to explain the effects of light on living cells. According to his theory, low level laser light affects cell proliferation mainly through the activation of the mitochondrial respiratory chain and its cellular signaling.

Hamblin and Demidova (2006) proposed similar mechanisms to explain how light can affect cells. In their theory the molecular targets of low level light are mainly cytochrome c-oxidase and photoactive porphyrins and the mitochondria is the major site of for this reaction.

Amat et al. (2006) proposed a mechanism for visible and IR light bioeffects based on oscillatory electric fields induced in biomolecules. This hypothesis traces the effect of the light to the effects of

EMFs of lower frequencies, which have been reviewed earlier in this paper. Amat supports his hypothesis by pointing to the similar bio-effects of light and lower frequency EMFs known from the literature.

Cosic (1997), in her model, proposed that there is a resonant interaction between macromolecules that plays an essential role in their bioactivity. In fact, she coined the term Resonant Recognition Model (RRM). She showed that there are certain periodicities within the distribution of energies of delocalized electrons along a protein molecule that are critical for protein functions such as interactions with their targets. A charge moving through a protein backbone can produce EMF radiation or absorption with spectral characteristics corresponding to the potential energy profile of the protein. The RRM enables the calculation of these spectral characteristics which lie in the infra-red and visible spectral EMF regions. In simple words, the key point of Cosic's finding is the assignment of specific spectral characteristic of proteins to their specific biological function (Pirogova et al., 2008). This theory could also be the base for designing new peptides which would have desired biological activity based on their precalculated spectral characteristics (Cosic, 1994; Cosic and Pirogova, 2007).

4. Evidence for cells interacting through electromagnetic fields

4.1. Are biological systems really interacting through EMFs?

Data from the literature provide evidence that EMFs can be generated and received by biological systems in a broad range of frequencies; from static fields to a wide range of optical and non-optical EMF spectra. However, whether the biosystems that are capable of emitting and receiving EMFs are really using them for intercellular or intracellular interaction is a matter of fundamental debate.

Most of the experiments that try to explore the hypothesis of biosystem interactions through EMFs are centered around experiments that explore correlated behavior of chemically separated biosystems. As mentioned earlier, the first scientific report on distant non-chemical interaction between two distantly placed biosystems was described by Gurwitsch in the 1920s who dubbed the mediating phenomenon as "mitogenetic radiation" due to its mitosis stimulating effect (Gurwitsch, 1923, 1924). Following his report, many scientists did thousands of experiments in which they used a group of cells or biological tissues (inducer) undergoing a certain process using a stimulator and measured the response in another group of cells or biological tissues (detector) that are not

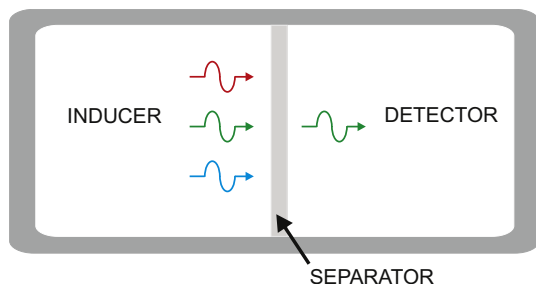


Fig. 2. Typical setup used in experiments on cellular electromagnetic interactions. INDUCER: a source of EMF – a cell culture undergoing or stimulated to undergo some process. DETECTOR: a cell culture detecting and reacting to the EMF from the inducer. SEPARATOR: ensures a chemical separation of the cell cultures and determines transmitted spectrum to find out which part of the EMF spectrum is mediating the interactions.

exposed to any stimuli and are chemically separated from the inducer by a separator (see Fig. 2). The main purpose of the separator is to chemically separate the inducer from the detector while permitting transmission of certain specific EMF frequencies (wavelengths). In these experiments, the inducer and detector may or may not be from the same type or species.

Examples of the selected inducer and detector include ex-vivo cultured cells, cell suspensions and cell lysates. In a recent review on distant cell communication by Trushin (Trushin, 2004b), one can find several examples of experiments from Russian language papers that can not be easily accessed in other languages or obtained by internet searches. Those works are mentioned in this paragraph. In an elaborate set of experiments, Kaznacheev observed that cellular cytopathic effects in a cell culture plate can be mimicked in a distinct cell culture plate that was not exposed to the cytopathic stimulus and was separated by quartz glass from the cells that were exposed to the cytopathic stimulus. He called this phenomenon a "mirror cytopathic effect" (Trushin, 2004b; Kaznacheev and Mikhailova, 1981, 1985). In these experiments, Kaznacheev used various types of human or animal cell cultures as inducer and detector and Coxsackie A-13 virus or mercury chloride as cytopathic agents (stimulus). He used regular glass as well as quartz glass for physically partitioning the inducer and the detector cells in a parallel set of experiments. He observed that the biochemical effects in the detector cells were only observed in those cells that were separated from the inducer cells by quartz glass and not by regular glass. Therefore, he suggested that the mirror cytopathic effect is most likely due to ultraviolet or infra-red EMFs emanating from inducer. Trushin's review also shed light on a series of similar experiments by Kirkin in 1981 who observed changes in growth in distant detector cells and Mostovnikov and Kholkhov found a significant increase in chromosomal aberrations in distant detector cells while the inducer cells were treated with cytopathic agents such as fast neutrons and laser beams.

In other sets of experiments, Grasso et al., (1991) used yeast cell cultures plates and positioned two culture palates (one inducer and one detector) in front of each other at a 6 mm distance. The exposure time was 30 s at room temperature and the cells were partitioned only by air. Control samples were maintained far from inducer cells. After keeping the samples and controls for 8 min at 26 °C, biological processes were stopped by adding a formaldehyde solution and samples were checked for their growth and gemmae formation. They found that detector yeast cells had their growth and gemmae formation significantly increased compared to control cells.

In another set of experiments on yeast, Musumeci et al. (1999) tested the possibility of an intercellular optical EMF interaction in physically separated yeast cell cultures. They used a temperature sensitive strain of yeast cells in order to control the growth phase of the cell culture. The growth rate of the detector cells that were optically connected with inducer cells in the active growth phase was significantly different compared to the growth of detector cells that were optically connected with inducer cells that were not dividing.

Budagovskii et al. (2001) (cited also in Trushin, 2004b) published an observation that pollen grains manifest increased germination rates in detector cells but not in control cells. His observation indicates that volatile molecules are not a strong candidate for transferring information from inducer to detector since the control was placed in the same gas environment.

Bacteria can also use distant signaling for spore germination and other physiological functions. In his experiment, Nikolaev reported that through distant interactions *Pseudomonas* bacteria can significantly reduce their adhesive capacity (Nikolaev, 2000a; and chapter 20 in Belousov et al., 2000). He also found a distant effect of *Pseudomonas putida* on *Bacillus subtilis* spore germination (chapter 12 in Belousov et al., 2007). He compiled his and others'

findings on distant interactions of bacteria in his recent review (Nikolaev, 2000b). Also, Trushin prepared critical reviews on distant interactions of bacteria (Trushin, 2004a, 2003d).

Both Trushin and Laager in their two independent experimental works observed a link between UPE and growth parameters of optically connected cultures of *Escherichia coli* (Trushin, 2003a,b,c; Laager, 2008).

Plant seeds also seem to use UPE in their interactions. Kuzin and Surkenova (chapters in Popp and Belousov, 1996; Belousov et al., 2000) found that low dose gamma ray irradiation of *Raphanus sativus* seeds (inducer) can result in similar changes in detector seeds not exposed to gamma rays and separated from the inducer seeds by a quartz barrier. This effect was abolished when the experiment was repeated with a glass barrier, which blocks the passage of ultraviolet UPE.

Optical interactions have also been reported in human cells. Voeikov and Novikov reported optical interactions of neutrophils via UPE during their respiratory burst (Voeikov and Novikov, 1997; and chapter in Popp and Belousov, 1996). Shen et al. (1994), and chapters in Popp and Belousov (1996), Belousov et al. (2000) found that PMA (phorbol myristate acetate) induced respiratory bursts in inducer neutrophils can result in a similar reaction in optically-coupled detector neutrophils.

Rowlands et al. observed that rolexaux formation of erythrocytes does not follow simple Brownian laws of motion and proposed cellular EMFs generated as described by Fröhlich theory as a plausible explanation for this complex group of cellular interactions (Rowlands, 1983; Rowlands et al., 1981, 1982; Sewchand and Rowlands, 1983).

Other scientists also showed distant interactions between mammalian cells through EMF coupling. Zhang and Zhang (2007) found that osteoblasts emitted UPE after being stimulated by weak, low-frequency, pulsed, electromagnetic fields. Far more interestingly, the emitted UPE promoted the proliferation of other osteoblasts. In our own laboratory, we showed that H₂O₂ treated colon cancer inducer cells (CaCo-2 cells) increased NFκB activation, caused cytoskeletal structural damage and reduced total cell protein content in optically-coupled detector cells (Farhadi et al., 2007).

Albrecht-Buehler pioneered a series of experiments in which he used various cell culture models and investigated the effect of both weak artificial and cellular infra-red EMFs on cell functions and properties. He observed interactions of baby hamster kidney (BHK) cells separated by a thin glass film (Albrecht-Buehler, 1992). Cells on one side tended to orient (traverse) themselves based on the orientation of the cells on the other side of a separator made of glass. To determine the interaction wavelength, various types of separators with different thickness and material were used. He determined that the wavelength of interaction radiation is in the red to near infra-red range. The number of traversing cells decreased inversely with the thickness of the separator plate. He also observed that 3T3 (Albrecht-Buehler, 1991, 2005) and CV1 (Albrecht-Buehler, 1995) extend their pseudopodia towards distant near infra-red sources, e.g. latex particles which scattered light. For 3T3 cells (Albrecht-Buehler, 1991), about 25% of cells extended lamellipodia towards a single near infra-red scatterer. If two such scatterers are provided, 47% of the cells extended towards them. The strongest response was achieved for electromagnetic fields at wavelengths of 800 nm, intermittently modulated (60 periods per min with rectangular or sinusoidal variations in amplitude). Albrecht-Buehler proposed that motile cells have rudimentary near infra-red vision, where the detectors can be perpendicularly oriented centrioles in cell centrosomes. Infrared EMFs were found to play a role in mutual cellular interactions (Albrecht-Buehler, 1992, 2005, 2010, 1995).

The other potential contender responsible for cellular optical interaction is the mitochondrion. Batyanov (1984) found in a series

of experiments that there is an optical interaction between rat liver mitochondria in a suspension. He showed that changes in the rate of oxygen consumption of optically connected samples are different from controls. Mitochondria are well known sources of reactive oxygen species, so it is easily conceivable that they are the source of UPE. Furthermore, Batyanov's experiments show that mitochondria can even respond to UPE from other mitochondria.

Whether distant cell signaling process is species-specific or not is another important question. Budagovsky et al. found that human whole blood has a stimulatory effect on germination of radish seeds when those are optically coupled. They proposed that biosystems are able to convert random photon emission to coherent photon emission depending on their physiological state (chapter in Belousov et al., 2007). To verify that the coherence of the UPE is the major factor in distant interactions of biosystems, investigators performed experiments using standard photometric four-sided quartz cuvettes with two opposite transparent sides and two matte sides. The matte sides (random phase screen) scatters the light, reduces the spatial coherence, and the transparent sides (ordered phase screen) preserves the spatial coherence, while both sides transmit the same intensity of light. Researchers observed significant reduction in the ability of UPE from human blood to influence germination of radish seeds when separated by random phase screens as compared with ordered phase screens. We consider this study of extraordinary importance since it experimentally showed that statistical ordering (coherence) of UPE plays an important role in UPE-based communication between biological systems. Temporal statistical ordering imposed by intermitting the UPE flux by rotating disc (which contained regularly distributed apertures) between two cell cultures was used by Gurwitsch and Gurwitsch (1934) to increase the efficiency of UV mediated mitogenetic effect.

Is the distant (non-chemical) interaction in biosystems limited to interactions at the cellular level? It seems that biosystem interaction has been reported at the level of plants, and primitive biosystems such as insects and other biosystems. In the study conducted by Burlakov, he used loach *Misgurnus fossilis* L. embryos of different ages that were kept in quartz cuvettes for 20–24 h. In his experimental setting, embryos could only communicate through optical means. He observed some morphological changes in embryo development when these embryos were optically connected. He did not observe these changes in control embryos that were not exposed to the other group (Burlakov et al., 2000).

Popp and Chang (chapter in Ho et al., 1994) also reported synchronization of flashes of fireflies and dinoflagellates when cultures were connected optically. From these experiments, the authors suggested that electromagnetic bio-communication plays an important role in the interactions of whole organisms. Also Belousov et al. demonstrated optical interactions of fish eggs and embryos via UPE (Belousov et al., 2002b, 2003, and chapter in Belousov et al., 2000).

Jaffe discovered that marine plants polarize the growth of distant fucus eggs by means of UPE up to distances of at least 10 mm (Jaffe, 2005, 2004). Marine plants used in the experiment were *Egregia menziesii*, *Fucus furcatus* and *Zostera marina*. Fels (2009) showed that *Paramecium caudatum* can interact via UPE in both the UV and visible parts (>340 nm) of the EMF spectrum. Experiments on interactions of cell populations took place in darkness. Cells affected cell division and energy uptake in neighboring cell populations. These effects were positive (stimulating) or negative (suppressing) depending on whether the quartz or normal glass cuvette was used and on the number of cells in the inducer and detector populations.

Galle also suggested that insects use UPE as a way to interact. In his study, adolescent *Daphnia magna* create UPE and the intensity

of UPE had a non-linear dependence on the population density with distinct maxima and minima (Galle et al., 1991; Galle, 1993, chapter 14 in Popp et al., 1992).

Callahan has performed extensive experimental and theoretical work on electromagnetic aspects of insect olfaction (Callahan, 1977a,b, 1985a, 1977c, 1985b; Callahan et al., 1985; Callahan, 1981). His main motivation was to explain long range attraction of various insects to each other and to sources of light. One of the proposed, modeled and tested mechanisms was based on the hypothesis that certain insect exoskeleton structures serve as antenna for infra-red radiation emitted by excited attractant molecules. A critical review of Callahan's theory and experiments was recently published in Traill (2005, 2008).

Becker was another scientist that worked on the role of EMF in insect biology. He showed that the behavior of insects such as termites is dependent on mutual EMF interactions during gallery building. In his numerous experiments, termites acted differently when colonies were separated by an aluminum plate compared to non-conductive plates. These behavioral changes were observed across different species of termites, suggestive of trans-species communication among different species of termites (chapter in Popp et al., 1989 and numerous references therein) (Table 2).

4.2. Caveats and obstacles in experiments on cellular EMF interactions

Scientists were skeptical (Bateman, 1935; Hollaender and Claus, 1935, 1937) after Gurwitsch discussed his findings and proposed his theory of "mitogenic radiation". Moreover, Gurwitsch's experimental protocol was arduous and interpretation of the data was rather complex. For example when he used yeast cells, they should be used as detectors in log phase and as inducers in the phase of exponential growth. Then, the effect on detector cells should be evaluated (number of cells counted) in the beginning of the exponential phase. Other peculiarities were also involved (Voeikov and Belousov, 2007; Gurwitsch and Gurwitsch, 1943(1999); Volodyaev et al., 2009). Thus, it is not surprising that some groups of scientists who tried to reproduce Gurwitsch's results were not able to do so due to lack of adhering to his stringent methods (Gurwitsch and Gurwitsch, 1943(1999), p.286–293 Gurwitsch and Gurwitsch, 1959) giving rise to intense debates and controversies. Using detector devices that lacked adequate sensitivity to detect weak UPE, such as modified Geiger–Müller counters or certain photographic plates, was another reason why scientists could not easily reproduce the experimental data in their lab, undermining trust in Gurwitsch's findings on mitogenetic effects. This mistrust happened because of a few dozen negative reports in spite of several hundred positive reports that confirmed the existence of "mitogenetic radiation" (Gurwitsch and Gurwitsch, 1943(1999)). Wainwright (1998) in his review evaluated some of the problems with reproducibility of mitogenetic radiation. He himself had trouble in reproducing his own data but in several cases reproduced the experiments and provided evidence for UPE cellular interactions in double-blind experiments with bacteria (Wainwright et al., 1997). After the Second World War, research in this area ceased almost completely in Western countries until the 1970s. Over several recent decades the existence of UPE from cells has been confirmed repeatedly by many scientists all around the world (Multi-author review, 1988, 1992; Popp et al., 1992; Popp and Belousov, 1996, 2003; Van Wijk and Shen, 2005; Belousov et al., 2007). Although various effects resulting from EMF cellular interactions have been reported, the mitogenetic effect of UV UPE is not widely known. In order to rehabilitate Gurwitsch's original findings, computer based image evaluation methods are being developed by Russian scientists (Volodyaev et al., 2009).

An other interesting obstacle in reproducing results from experiments on EMF cellular interactions was reported by Kaznacheev and Mikhailova (1981, 1985). They observed that the "mirror cytopathic effect" (mediated by UV UPE) varied with the month of the year. The effect was at its nadir between December and January and at its peak from June to August, see Fig. 3.

Especially for the perspective of experimental workers in this field, it may be informative to show the typical reproducibility of the experiments during the most and least favorable parts of the year (Figs. 4 and 5). Please note that this depends also on the geographical latitude (Kaznacheev's experiments have been carried out mostly in Novosibirsk, 55° 02' N, 82° 55' E).

It is well known that there are periodic changes of many biological processes at the level of single cells to whole organisms with diurnal, monthly, annual and other periods. Brown et al. found that there are annual fluctuations of perspiration activity in various species. He found that O₂ consumption (potato samples), even when using a respirometric system keeping conditions constant, including pressure, fluctuates almost sinusoidally over the year, with a minimum during October–November and a maximum (rate doubled) in April–May (Brown, 1958). Furthermore, it has been discovered that seed water uptake (Brown and Chow, 1973), small animal motility (Webb and Brown, 1965) and tree stem diameters (Zürcher et al., 1998) fluctuate periodically. Candidate external pacemakers for these phenomena include periodic gravitational changes due to the moon (the cause of regular tides), periodic changes in magnetic fields due to solar activity and the relative position of the earth to sun.

Kaznacheev (Kaznacheev and Mikhailova, 1981, 1985) and other scientists (Volpe, 2003) studied the effect of changes in the natural magnetic field on mitotic activity and on the viability of cells. Positive results suggested that if basic cellular processes are influenced to periodically change their magnitude, the same should be the case for EMF cellular interactions.

Based on the analysis of more than 12 000 experiments over a period of more than 10 years on 12 cell lines (Kaznacheev and Mikhailova, 1981, 1985), it has been discovered that results in the field of distant cellular interactions can be affected by several variables such as heliogeomagnetic conditions: solar activity, geomagnetic disturbances and geographic latitude (Trushin, 2004b; Kaznacheev and Mikhailova, 1981, 1985).

Results of analysis similar to cross correlation analysis of observed magnitudes of the "mirror cytopathic effect" and geomagnetic factors (Ap index, polarity of interplanetary magnetic field, F index) gave the following results (p. 97 in Kaznacheev and Mikhailova, 1981): the Ap index of geomagnetic activity¹ was increased when the cytopathic effect was not present and vice versa. A negative orientation² of interplanetary magnetic field was present on days when there was a very high occurrence of the effect and a positive interplanetary magnetic field orientation was present when the effect did not appear. A higher solar flare index (SFI) has accompanied unsuccessful experiments. To summarize the effects of geomagnetic fields on cellular interactions, based on extensive experimental work we can state that disturbances of the natural electromagnetic background, including its amplitude and spectra, lowers the ability of cells to interact via their own electromagnetic fields. Additionally, to study the effect of natural magnetic fields on the mirror cytopathic effect, better controlled influences have been studied. It has been found, for example, that increased temperatures (to 38.5 °C instead of the standard 37°)

¹ Ap index is a measure of the general level of geomagnetic activity over the globe for a given (universal time) day.

² Magnetic field lines of interplanetary magnetic field pointing to the sun.

Table 2
Electromagnetic distant interactions of biosystems. Quartz is transparent basically for wavelengths of EMFs longer than 170 nm, normal glass is transparent usually for wavelengths longer than 350 nm. The upper limit of transparency is in near the infra-red at wavelengths of about 2000 nm. Exact transmission depends on the composition of the glass and its thickness.

Organism, cell type or structure	EMF wavelength or frequency, separator material	Effects	References
fibroblasts (human & chicken), monkey kidney tissue	UV or IR (<i>various separator materials tested: RF, sound excluded</i>)	Transfer of the effect of HgCl ₂ and viruses	Kaznacheev et al. (1980), Kaznacheev and Mikhailova (1981, 1985)
human embryonic fibroblasts, Hep-2 cells	UV excluded (<i>glass separator</i>)	Transfer of effect of high dose UV irradiation	Kaznacheev and Mikhailova (1981), cited in Trushin (2004b)
rat sarcoma cells	UV excluded (<i>glass separator</i>)	growth rate	Kirkin, cited in Trushin (2004b)
diploid cells of musclicutaneous tissue from human embryo	EMF 500–700 nm (authors' claim)	transfer of effects of neutron bombarding: chromosomal aberrations	Mostovnikov and Kholkhov, cited in Trushin (2004b)
yeast cells	UV-IR (authors' claim) <i>air separation</i>	growth rate	Grasso et al. (1991)
yeast cells	<i>quartz (UV transparent cuvette)</i>	growth rate	Musumeci et al. (1999)
cherry pollen grain	<i>common gaseous environment, effect present only if cultures in direct optical contact</i>	germination rate	Budagovskii et al. (2001), cited in Trushin (2004b)
<i>Pseudomonas</i> bacteria	not UV (<i>glass separator</i>)	adhesive capacity	Nikolaev (2000a), also in chapter 20 in Belousov et al. (2000)
<i>Pseudomonas putida</i> → <i>Bacillus subtilis</i> spores	not UV (<i>acrylic plastic separator used</i>)	germination rate	Nikolaev (chapter 12. in Belousov et al. 2007)
<i>Escherichia coli</i>	<i>vis-IR (effect present with vis-IR transparent glass, but not with opaque one)</i>	growth rate	Trushin (2003a,b,d), Laager (2008)
<i>Raphanus savitus</i> seeds	<i>UV (effect present with quartz (UV transparent), but not with glass separator)</i>	seed germination and development	Kuzin and Surkenova (chapter in Popp and Belousov, 1996 and chapter 18 in Belousov et al., 2000)
neutrophils	<i>quartz separated (UV transparent)</i>	effect of inducer on PMA or zymosan induced respiratory burst in detector	Voievkov and Novikov (1997), Novikov et al. (chapter in Popp and Belousov, 1996), Shen et al. (chapter 26 in Belousov et al., 2000)
neutrophils	<i>quartz separated (UV transparent)</i>	PMA stimulated inducer increases UPE and O ²⁻ in detector	Shen et al. (1994), Shen et al. (chapter in Popp and Belousov, 1996)
whole blood	<i>quartz vials (UV transparent)</i>	effect of back-reflected (by aluminum foil) photons on blood UPE	Voievkov et al. (2003)
erythrocytes	<i>cells in blood plasma and other liquids</i>	rolleau formation kinetics	Rowlands (1983), Rowlands et al. (1981, 1982), Sewchand and Rowlands (1983)
osteoblasts	<i>UV-IR (effect present through copper net, but not through black glass)</i>	proliferation promotion in detector, when inducer stimulated to proliferate	Zhang and Zhang (2007)
CaCo-2 (colon cancer) cells	UV-IR expected, but not verified – <i>glass separators</i>	increased NFκB activation, structural damage and reduced total protein in detector, when inducer treated by H ₂ O ₂	Farhadi et al. (2007)
mammary gland explants	UV visible (<i>effect through the quartz, not through the opaque separator</i>)	changed protein secretion, lipid peroxidation and UPE in detectors when inducers treated by oxytocin, acetylcholine, epinephrine, nor-epinephrine	Galantsev et al. (1993), Moltchanov and Galantsev (chapter in Popp and Belousov, 1996)
BHK, CV1, 3T3 cells	IR (<i>various separator materials tested</i>)	mutual orientation, motility	Albrecht-Buehler (1992, 1991, 2005, 2010, 1995)
human blood → radish seeds	(<i>quartz separator used</i>)	germination rate	Budagovsky (chapter 4 in Belousov et al., 2007)
loach <i>Misgurnus fossilis</i> L. embryos	(<i>quartz separator used</i>)	morphological changes in embryo development	Belousov et al. (2002a,b, 2003), chapters 23 and 24 in Belousov et al. (2000), Medvedeva (2008)
frog <i>Xenopus laevis</i> eggs and embryos	(<i>quartz separator used</i>)	decreased UPE intensity	Volodyaev (2007), Volodyaev and Belousov (2007)
fireflies, dinoflagellates	UV-IR expected, but not verified (<i>effect present with quartz glass separator, but not with a copper one</i>)	synchronization of UPE	Chang et al. (1995), chapter 12 in Ho et al. (1994), Chang et al. – chapter in Popp and Belousov (1996), chapter 21 in Belousov et al. (2000)
marine plants <i>Egregia menziesii</i> , <i>Fucus furcatus</i> , <i>Zostera marina</i> → <i>Fucus</i> eggs	UV-IR expected, but not verified	polarization of growth	Jaffe (2005, 2004)
ciliate <i>Paramecium caudatum</i>	some effects through UV, some through visible-IR <i>glass or quartz cuvettes used</i>	cell division and energy uptake	Fels (2009)
<i>Daphnia Magna</i>	UV–visible expected, but not verified (<i>organisms in water environment</i>)	non-linear dependence of UPE on population density - presumably by EMF interaction	Galle et al. (1991), Galle (1993), chapter 14 in Popp et al. (1992)
fungus <i>Gaeumannomyces graminis</i> → bacteria <i>Pseudomonas corrugata lux</i>	UV (<i>effect present through the quartz, not through the glass separator</i>)	increased luminescence	Wainwright et al. (1997)
termites	(<i>aluminum separator dismissed the effect</i>)	different gallery building structure	Becker, chapter in Popp et al. (1989) and number of references therein (in German)
moth <i>Trichoplusia Ni Huebner</i> , lovebug <i>Plecia Nearthica Hardy</i>	IR?	long range attraction/interaction	Callahan (1977a,b, 1981, 1985a, 1977c, 1985b), Callahan et al. (1985), Traill (2005, 2008)
mitochondria	UV	oxygen consumption rate	Batyanov (1984) and chapter in Popp and Belousov (1996)
Other partial reviews on EMF (distant) cellular interaction			Trushin (2004a,b, 2003c), Nikolaev (2000b), Wainwright (1998)

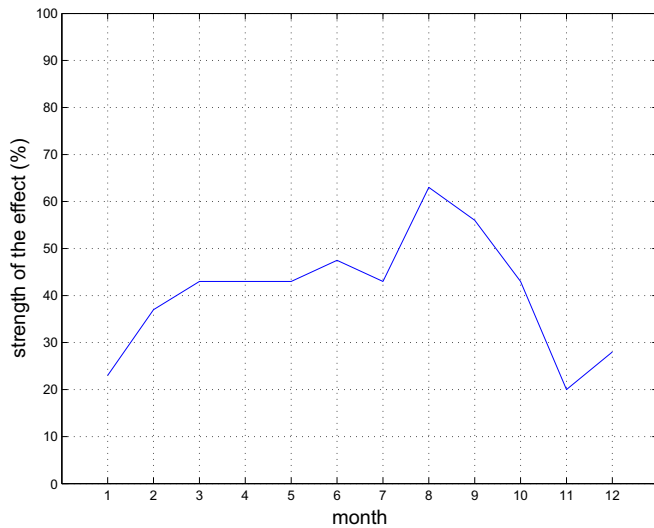


Fig. 3. Strength of the mirror cytopathic effect averaged from more than 12 000 experiments over the period of 10 years on 12 cell lines. Adapted from Kaznacheev and Mikhailova (1981).

(p. 104 in Kaznacheev and Mikhailova, 1981) and UV pre-irradiation (human embryonic fibroblasts and Hep-2 cells) (Kaznacheev et al., 1979) of detector cultures increased mirror cytopathic effect compared to control.

The problem of reproducibility has also been discussed by Rahn in his book in 1936: “Professor Gurwitsch has told that in his experience such a condition usually remained for several days, or even for a number of weeks, and it was impossible to produce even the simplest mitogenetic effect.” Rahn further mentions that there were times that cell cultures did not produce the test result for unknown reasons: “...discussing this point with the various investigators in this field, practically all seem to have had the same experience.” (p. 93–95 in Rahn, 1936). Nevertheless, the observed effect, when present, was well outside experimental error. These statements exactly fit

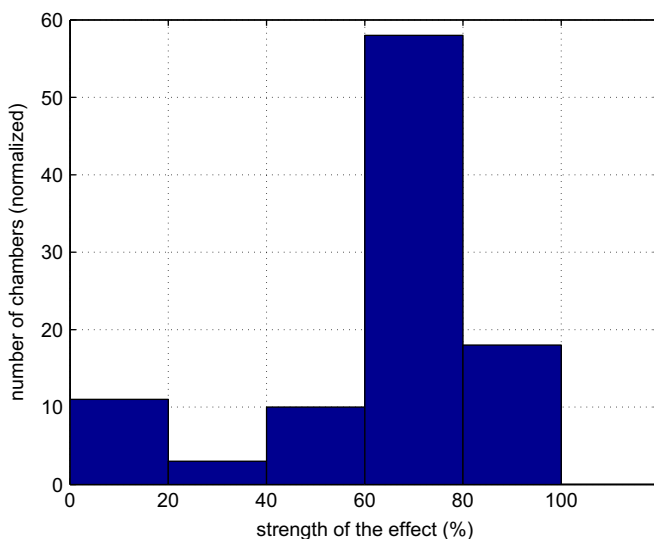


Fig. 4. Histogram of the strength of the mirror cytopathic effect (dependent on UV EMF cellular interaction) in August, the statistically most favorable month. The abscissa shows the strength of the effect in individual experiments, the ordinate is the number of experiments with a given strength. An average of more than 12 000 experiments over a period of 10 years on 12 cell lines. Adapted from Kaznacheev and Mikhailova (1981).

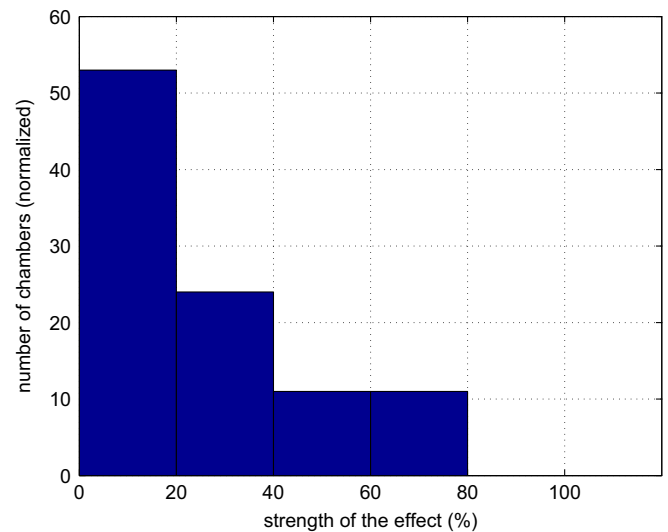


Fig. 5. Histogram of the strength of the mirror cytopathic effect (dependent on UV EMF cellular interactions) in November, statistically the least favorable month. The abscissa shows the strength of the effect in individual experiments, ordinate is the number of experiments with a given strength. An average of more than 12 000 experiments over a period of 10 years on 12 cell lines. Adapted from Kaznacheev and Mikhailova (1981).

findings of Kaznacheev, who had the same experience and found correlations of unsuccessful experiments with both sudden and periodic disturbance in the natural electromagnetic background as explained earlier. Thus, state of the electromagnetic background has to be taken into account in order to minimize negative results (such as by Quickenden and Tilbury, 1985), if it is the goal of the experiment.

Despite an abundance of research on production of cellular EMFs or on the effects of effect of EMFs on biosystems in the literature, there is very little recent data published in English on direct cell-to-cell interactions as was observed by Albrecht-Buehler, or Farhadi et al. who used direct cell-to-cell radiation passing through mechanical or optical barriers.

4.3. How cells interact via EMFs

Popp with his theories was one of the pioneers in proposing physically oriented models for UPE interactions based on the coherence properties of delocalized cellular EMFs and their interference (Popp et al., 1989). Based on one of his theories applied by Galle (Galle et al., 1991; Galle, 1993), every biological system displays a complex wave pattern that is probably species-specific. Interference of the complex wave pattern, can occur if the pattern are similar and the origins are from the same species (chapter in Ho et al., 1994). Popp's early work included studies of the optical properties of carcinogenic substances in which he noted that some carcinogenic substances have almost the same molecular structure as their harmless counterparts, except that they differed in their optical absorbance spectrum (Popp, 1976; chapter in Belousov et al., 2000). He suggested that carcinogens cause harm by interfering with normal inter- and intracellular optical communication. This notion was later supported by the work of Sung (chapter in Popp et al., 1989).

If cells are communicating with each other using EMFs, then how can cells avoid environmental EMFs that are much stronger than the weak EMFs generated by other biosystems? A well known, but in this context very interesting fact was pointed out by Letokhov: the wide spectrum of UV light – 230–280 nm (UV-C) – is almost completely absorbed by the ozone layer. In fact, the intensity of photons in this spectrum of extraterrestrial origin drops from

10^{13} to ca. 10^{-27} photon/($\text{cm}^2 \text{ s}$) in passage through the atmosphere (chapter in Musumeci et al., 2003). In his report he argues that this range is an excellent candidate for cellular communication on Earth. His theory is consistent with findings of several experiments mentioned before (Section 4.1).

In his master's thesis, Laager (2008) proposed a metabolic photo-communication model. In his model the detected optical EMF can be attenuated by light-absorbing cell components. The part of the spectrum that is filtered out depends on the state of the cell. Thus, we can consider that EMF signals can be modulated by cell components based on cell conditions.

In an other interesting theory, Mayburov based his observation on information theory and stated that UPE signal rates emitted from cells are reminiscent of a binary data exchange via optical channels (Mayburov and Volodyaev, 2009). He further proposed that photon energy can reversibly convert into an exciton (the bound state of an electron-hole) which can propagate along molecules in biosystems. He thought that the pulses of photons enable synchronization among cells.

Tsong et al. based their theory on the study of electric field effects of transmembrane proteins. They proposed that oscillatory electric fields (kHz–MHz range) generated at the level of cell membranes can serve as signals for cellular communication (Tsong, 1989, chapter in Ho et al., 1994). The oscillatory electric field can be transduced by electroconformational changes in enzymes and vice versa. Tsong refers to the oscillatory electric field of cells as the “language of cells” (Tsong, 1989).

Endogenous biological fields are not limited to electromagnetic fields. For instance, Matsushashi et al. proposed that cellular interactions may involve an endogenous cellular acoustic field and radiation. They detected sound signals from cultures of *E. Coli* (Matsushashi et al., 1998). Indeed, the possibility of acoustic field interactions between cell cultures has not been seriously excluded in several experiments mentioned earlier in our review.

However, if cells can communicate directly with each other using a form of EMF, is it possible to record these signals using a device and replay the recorded signal to see if we still can generate the desired effect? Benveniste's group was one of the first who used electronic methods, not only for transfer of biological signals (Thomas et al., 2000), but also, for recording and replaying the signals³. In fact, their experiments formed the bases for the term “digital biology”. There is, however, much controversy about Benveniste's experiments, including inability of some workers to reproduce them (Jonas et al., 2006), while others could (Endler et al., 1995; Bellavite et al., 2006). Regarding the possible EMF nature of biological signals, we refer the reader to recent experimental papers by Montagnier et al. (2009a, 2009b), which describe the capacity of some bacterial DNA sequences to induce electromagnetic waves at high aqueous dilutions. Here we must remark that the physical method used in the work of Montagnier for the detection of EMF signals (unscreened solenoid coil) is highly susceptible to artifacts due to technogenous EM noise.

In addition to a possible EMF-mediated cellular information transfer (Van Wijk, 2001), there can be a pure EM force interaction between cells without transfer of any specific information. Cells create oscillatory electric fields in their surroundings, which can act via dielectrophoretic forces on other particles (Pohl et al., 1981 and Section 2.2.3). The oscillatory field will thus affect adherence of cells to substrates and to other cells (Pokorný et al., 1983; Pokorný and Wu, 1998). The adherence of cells plays an important role in cancer.

³ We need to emphasize here that the spectral composition of the cellular EMF can be theoretically recorded, but it is not the only aspect of the cellular EMF. Another aspect, which is probably impossible to substitute technically, is the complex spatial and temporal distribution of the field in the cell and organism.

Disintegration of the cytoskeleton is connected with metastatic growth (Suresh, 2007). The cytoskeleton, especially microtubules, is expected to be a source of vibrations that generate cellular EMFs at least in the kHz–GHz range. Theoretical calculations showed that disturbed electromagnetic fields of a cell lead to reduced adhesive forces between cells, which may contribute to metastatic growth (Pokorný, 2006; Pokorný et al., 2008; Pokorný, 2009).

5. Conclusion and future prospects

There is no doubt that biosystems can be affected by EMFs at several levels. There is also little doubt that biosystems can be the source of EMFs. The main question at hand is whether biosystems use EMF for a purposeful interaction (communication) and if so at what level of the bio-organism will it happen? The amount of data that support the latter notion is rapidly mounting at the same speed as the increasing number of questions that need to be addressed. One of the major problems in this line of research originates in the lack of the usual reproducibility that scientists are used to observe in their experiments. Several scientists blame these discrepancies on known confounding factors such as geophysical properties of the experiment place, timing and season of the experiment, lighting or conditions of the laboratory and cosmophysical factors. Not to mention that the scientist as a biosystem is also a source of measurable UPE (Van Wijk and Van Wijk, 2005; Van Wijk et al., 2006; Creath and Schwartz, 2006; Cifra et al., 2007; Kobayashi et al., 2009), which can unintentionally influence the experimental process during handling of the samples. The prime question is “Why we should care if cells interact via EMF?”. If the existence of distant cell communication proves to be true, there would be a substantial impact on our understanding of biology and biological research. Mastering and influencing the distant signaling system in biosystems can open a whole new horizon in our approach to biology. Then, the applications in biology and medicine could be astonishing.

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