

**Measurement of spontaneous photon emission from
the human body: technical aspects, parameters, time
and temperature dependent fluctuations of photon
emission**

DIPLOMA WORK

MICHAL CIFRA

UNIVERSITY OF ŽILINA
FACULTY OF ELECTRICAL ENGINEERING

Department of Electromagnetic and Biomedical
Engineering

Specialization: BIOMEDICAL ENGINEERING

**Supervisor of the diploma work:
prof. Dr. Roeland Van Wijk, Ph.D.**

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Abstract

This work deals with the issue of ultraweak spontaneous photon emission from the human body and time and temperature dependent fluctuation of the photon emission from human body

First part deals with historical and theoretical background of the photon and biophoton concept, theories of the biophoton source, the measurement systems, the overview of human biophoton research and the properties of the photon emission from the human body.

Second, experimental part of this work brings information about the dynamics of the time dependent human photon emission fluctuations in the interval of 24 and more hours. Temperature dependent fluctuations of the human photon emission caused by induced as well as spontaneous cooling have also been measured and evaluated.

List of the used abbreviations

ATP - adenosine triphosphate

BPE – biophoton emission

BIOPHOTON – a photon(s) of visible or UV range which emanates in ultra low intensity from biological systems

CCD – coupled charge device

EB – ethidium bromid

EES – electron excited states

IR – infrared (light)

LDL - low density lipoproteins

MGR – mitogenetic radiation

PE – photon emission

PMT – photomultiplier

QE - quantum efficiency

ROS – reactive oxygen species

SE – spontaneous emission

SOD – superoxid dismutase

UV – ultraviolet, light in ultraviolet spectrum

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1. Introduction [1,2]

The fact that all living biological systems, including humans, constantly and spontaneously emit small amount of photons in visible and UV part of electromagnetic spectrum is in generality not very well known among scientists in the fields of medicine, biomedicine and biophysics. Physicists know very well, that electromagnetic radiation from every body depends primarily on his temperature, as described by Planck's law of black body radiation. However, organisms don't reach such a high temperature to be able of emitting visible light as, for example, Sun or incandescent tungsten (wolfram) filament in light bulb does. Biologists describe chemiluminescence visible by naked human eye from variety of insect or marine species. Special chemical-enzymatic processes that cause this luminescence are not present in all living systems and therefore cannot explain the phenomenon of spontaneous photon emission in all living systems generally. Terms such as ultraweak photon emission and spontaneous ultraweak, low-level, or dark bio-/chemi-luminescence, etc. are now in general use to describe these phenomena, in order to distinguish them from general bioluminescence.

Impossibility to see this ultraweak photon emission is due its ultra low intensity, order of $10^{-16} - 10^{-18} \text{ W/cm}^2$, that is 1-100 photons/ s.cm^2 , while the threshold sensitivity of human eye can individually differ from $10^{-12} - 10^{-14} \text{ W/cm}^2$, or $10^6 - 10^4$ photons/ s.cm^2 .

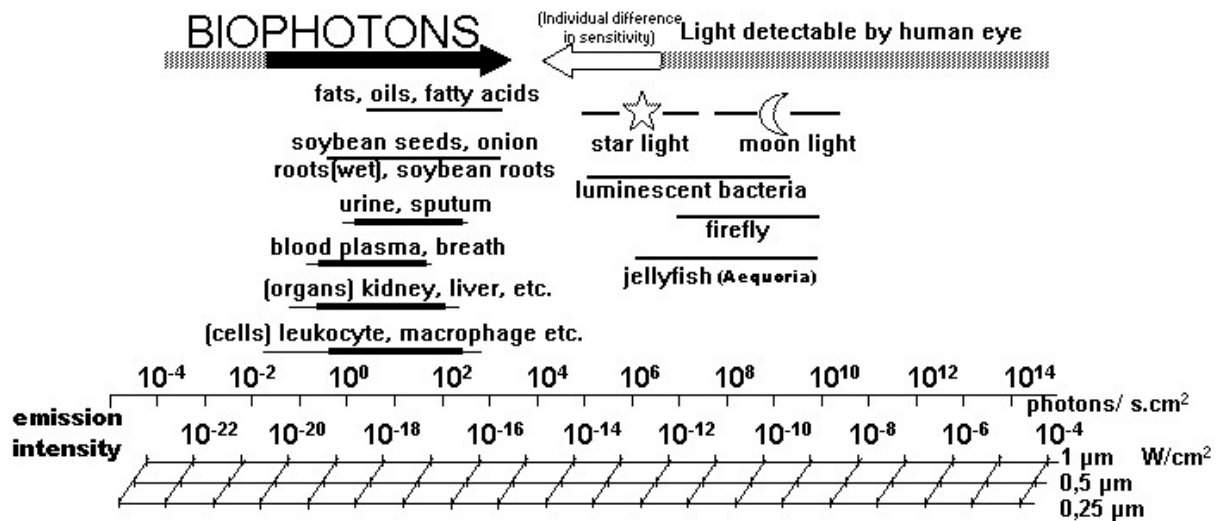


Figure 1.1. – Comparison of photon emission intensity from various biological samples and human eye sensitivity [1]

Despite low intensity of photon emission from organisms, many papers of various research groups suggest the possible biocommunication and bioregulatory effect from this ultraweak radiation field [3,11,61,76,82-84,86,94-98], leading to new view in biology and medicine.

There are several theories of origin of this ultra weak spontaneous photon emission. However, general consensus about its origin was not reached yet.

The coherence of photon emission from various organisms is described [31-35], however not in terms of classical definition of coherence, but in terms of quantum coherence.

Another phenomenon tightly bound with spontaneous emission is delayed luminescence. It can be defined as long term decay of higher photon intensity due to previous excitation of the system. This effect takes much longer than normal fluorescence (microseconds), typically minutes to hours. Delayed (super delayed, induced, etc.) luminescence, as well as spontaneous emission, can be observed in all living systems. This work is oriented on theoretical and practical evaluating of spontaneous emission.

2. Historical background [3]

Russian biologist Alexander Gawrilowich Gurwitsch, born in 1874 in Ukraine, studied at the Medical faculty of Munich University in Germany, worked in leading biological centers in Switzerland and Germany, was interested in one of the most enigmatic phenomena in biology – morphogenesis. He desired to understand the mechanism of emerging of complex tissues and organs, of organisms with unique architecture from rather primitively structured embryo cells.

Many observations and considerations, emerging in the publications of his theory of morphogenetic fields (1912), led him in 1923 to crucial experiments with two onion roots. A tip of an onion root - the inducer - was directed at the wall of another onion root - the detector. After they had been kept for some time in this configuration the number of mitoses at the detector side facing the tip of the inducer root significantly exceeded that at the opposite one. If a glass plate was introduced between the inducer and the detector, there was no stimulation of mitotic activity. A quartz plate shielding the tip of the inducer root did not interfere with its action. If the tip of the inducer was aimed at a metal mirror in such a way that its reflection fall onto the wall of the detector, stimulatory action again was observed.

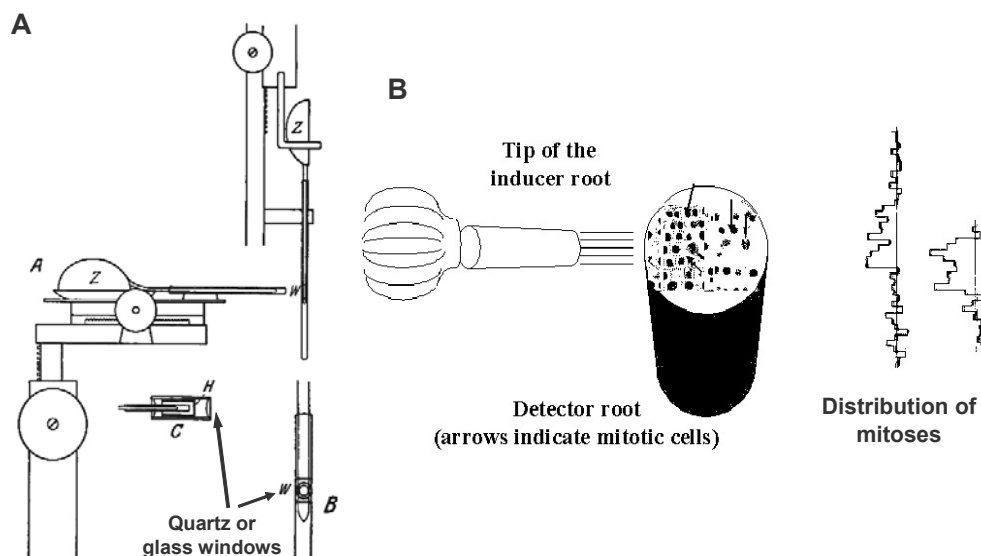


Figure 2.1. - Schematic presentation of “onion” experiment of Gurwitsch. **A.** Installation of an inducer root (horizontal) and a detector root (vertical) on moving tables of microscopes. Z – onion bulbs, C – tip of the inducer root fixed in an air-tight chamber, H- and W – quartz or glass windows. **B.** Sketch of experimental results evaluation. A detector root was sliced below and above the “irradiated” zone and an excess of mitotic cells on the left (irradiated) or right (non-irradiated) sides of the root on each slice was calculated. Two indented lines at the left illustrate the results of two representative experiments. Significant excess of mitotic cells on the left side over the average distribution was observed in the irradiated region. [3]

These results could be explained neither by chemical nor by mechanical action of one root on another. The most plausible explanation of the effect was the following: a living organism can emit photons which stimulate cells to divide. These photons belong to the ultraviolet region of the spectrum since quartz but not glass is transparent for them. Gurwitsch named this photonic flux “mitogenetic radiation” – MGR.

It is well known that UV-light is hazardous for living cells. However, when the light beam of an UV-lamp was attenuated several thousands times, the number of mitoses increased. Thus, the conclusion that UV-photons induce the performance by a living cell of its major function - reproduction - had been proved. It also turned out that the effect of light on living systems strongly depends on its intensity and duration of action: excessive illumination resulted in suppression rather than stimulation of cell divisions.

In 1924 Professor Gurwitsch was elected the head of Histology Department at the Medical Faculty of Moscow State University, and the investigation of the new phenomenon had shown that MGR is produced by various animal and plant tissues, by microorganisms. Ordinary yeast culture turned out to be most convenient test-system. By all experiments understanding of physiology of both biological detectors and emitters of MGR is needed. For example irradiation needs to be done when cells are in certain phases of the cell cycle, otherwise no effect will be observable. The lack of understanding of necessary physiology leads to failing in experiments that were aimed to reproduce the effect. This was probably the main problem of studies that purportedly refuted existence of MGR.

A.G. Gurwitsch was nominated for Nobel price, first time in 1927 [4] and failed in getting it missing only one vote [5] (Nobel committee responsible for voting of winner of Nobel price for medicine and physiology has nowadays 50 members). Many laboratories and researches in the USSR, Germany, France, Italy and Japan began to experiment with MGR. From 1923 and up to 1939 hundreds of papers, a dozen of comprehensive reviews on MGR appeared. In the thirties, MGR (or at least radiation) was successfully registered in several laboratories by modified Geiger-Muller counters to have sensitivity in UV wavelength band.

During 2nd World War many research centers were destroyed. Upcoming biochemistry and the fact that cell growth can be generally stimulated by radiation and, even more effectively, by hormones, evoked slowly scepticism about Gurwitsch's

discovery which got then more and more subject of discrimination and MGR fell into oblivion. A.G. Gurwitsch passed away in 1954.

In the beginning of the 1960s in the Department of Biophysics on Moscow State University emission of photons from organisms in visible range was detected by the first photoelectric detectors. However, sensitivity of these early detectors was very low. The research group of prof. Tarusov indicated that this ultra weak photon emission was a by-product of random lipid peroxidative reactions and recombinations of active forms of oxygen. This point of view is still dominant in biophysics, biochemistry, and physiology.

In the 1970s the German physicist Fritz-Albert Popp refreshed Gurwitsch ideas and works. He was inspired by Herbert Fröhlich, father of superconductivity theory, who was theoretical physicist in the field of solid state physics, who later applied theoretical physics on biological systems stating that biological systems are:

- highly nonlinear, far away from thermal equilibrium and must be treated as thermodynamically open systems that constantly carry out work to keep this non-equilibrium
- macroscopic quantum systems that are able to produce coherent oscillations.

Professor Popp connected ideas of A.G. Gurwitsch, H. Fröhlich, I. Prigogine and other outstanding scientists with his quantum physical theories and experimental research with highly sensitive photomultiplier systems. Popp revived the idea of a regulative und bicommunicative role of the ultra weak photon emission of biological systems emanating from all living systems on the principles of coherence and introduced the name “biophoton” to distinguish it from classical bioluminescence.

3. Theory of photon

The nature of photon, as we understand it, and processes that give rise to a photon are very peculiar, especially in biological systems, which are highly dynamic and complex. That radically increases effort needed to explain these processes.

Modern theory of light is indivisibly connected with quantum theory of light, which says that light “consists” of light “particles”, called photons.

3.1. Brief history of photon concept [6,7,9,10]

Already in 17. century Francis Bacon and Isaac Newton, inspired by renaissance thinkers, had been persuaded that light is particle-like, although in an other meaning as we are told by nowadays quantum physics. This opinion was refuted for almost a whole century by the experiment of Thomas Young in 1803. He showed diffraction and interference of light, suggesting its wave-like nature.

Modern concept of electromagnetic energy quanta was introduced by Max Planck in 1900 to explain spectral distribution of blackbody radiation. In 1905, apart from his article about Special Relativity Theory Albert Einstein published an article about the photoelectric effect. For its discovering, and other contributions, he received the Nobel Price in 1921.

Photoelectric effect is based on the following observations:

1. The number of electrons emitted by the metal depends on the **intensity** of the light beam applied on the metal; the more intense the beam, the higher the number of electrons emitted will be.
2. The emitted electrons move with greater **speed** if the applied light has a higher **frequency**.
3. No electron is emitted until the light has a **threshold** frequency, **no matter how intense the light is**.

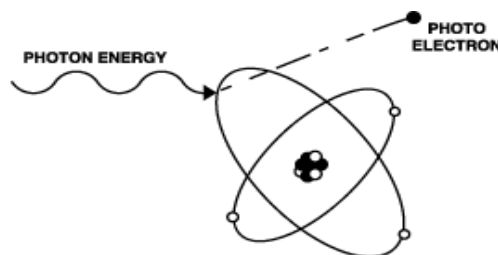


Figure. 3.1. - Einstein's Explanation of Photoelectric Effect[9]

The photoelectric effect is not explainable by wave-like nature of light.

3.2. Description of photon [6, 8, 12, 13, 16, 17, 18]

Photon is quantum of electromagnetic radiation. The term photon (from Greek $\phi\omega\varsigma$, "phōs", meaning light) was first used by American physical chemist Gilbert Lewis in 1926. It is usually connected with meaning of “particle” or quantum of light, which is only narrow band of broad electromagnetic spectrum, but *it can be generally used for naming a quantum of electromagnetic radiation of any wavelength*. In the next chapters (3.1.2., 3.1.3.) the word “photon” will refer, unless closer specified, to quantum of electromagnetic radiation of any wavelength.

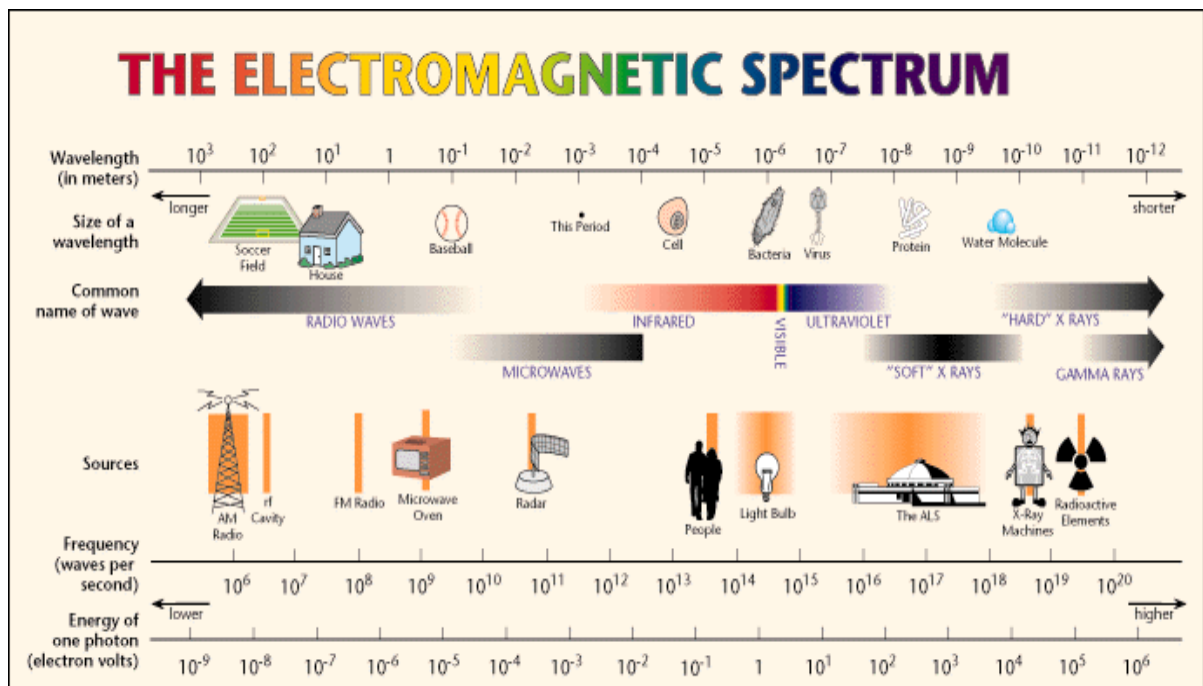


Figure. 3.2. - Electromagnetic spectrum [26]

In particle physics, a photon is one of the elementary particles and its sign is γ (gamma), although in nuclear physics this symbol refers to high energy radiation.

Paraphrasing physicist Fritz-Albert Popp: “The reasonable imagination of a photon may be obtained by looking at it as a process rather than as a particle. This process provides the interaction of an electromagnetic field with an ideal detector which is able to absorb the energy of this field completely at a definite space-point without thermal dissipation.”[12]

On a question “What is a photon ?” we can also answer in the words of Roy Glauber, who was awarded in 2005 by Nobel Price in physics for his contribution to Quantum Optics: “A photon is what a photodetector detects.”

The fact that we consider a photon as a particle in quantum physics cannot be interpreted/seen in terms of a discrete position in space-time but in terms of a discrete amount of energy one photon can have. This amount of energy can be expressed in multiple ways:

$$E = h.f = \frac{h.c}{\lambda} = p.c$$

E – energy of photon (eV or J)

f – frequency (Hz)

λ – wavelength (m)

p – momentum (in quantum mechanics or relativity physics often expressed in eV/c)

c = 299 792 458 m/s – speed of the light

h = 6.6260755. 10⁻³⁴ J.s = 4,135 667. 10⁻¹⁵ eV.s - Planck's constant

Photons are in permanent movement; they always propagate with the speed of light. In any medium with higher density than vacuum, electromagnetic radiation consisting of photons interact with atoms, more precisely said, with electrons, depending on photon frequency. Basically, photons are absorbed by electrons and reemitted after small delay, what causes global slowing of propagation of electromagnetic radiation. These interactions are described by Quantum Electrodynamics [14]. Classically we describe changing of speed of light propagation by index of refraction. Physical constant “Speed of light” always remains the same.

Photons have neither mass nor electric charge. However, they carry energy and momentum and have polarization (related to spin). Location in space and in time is no longer a means for theoretically distinguishing photons as elementary particles. Photons belong to group of particles called bosons. Bosons are particles with integer spin and they do not obey Pauli exclusion principle. For example electrons (belong to the group of particles called fermions - they have non-integer spin), do obey Pauli exclusion principle.

The Pauli exclusion principle is a quantum mechanical principle formulated by Wolfgang Pauli in 1925, which states that no two identical fermions may occupy the same quantum state simultaneously. It is one of the most important principles in

physics, primarily because the three types of particles from which ordinary matter is made—electrons, protons, and neutrons—are all subject to it. The Pauli exclusion principle underlies many of the characteristic properties of matter, from the large-scale stability of matter to the existence of the periodic table of the elements. [15]

Fact that photons are bosons and do not obey Pauli exclusion principle is at the foundation of laser theory because laser operation requires many photons to occupy a single mode of the radiation field.

As spin one particles, photons obey Bose-Einstein statistics. The repercussions of this fact are very significant both for conception of the photon and for technology. In fact Planck's law for the distribution of blackbody radiation makes use of Bose-Einstein statistics.

Bose-Einstein distribution – energy distribution for bosons:

$$f(E) = \frac{1}{Ae^{\frac{E}{kT}} - 1}$$

The quantum difference which arises from the fact that the particles are indistinguishable

$f(E)$ – the probability that particle will have energy E

k – $1.380658 \cdot 10^{-23}$ J/K – Boltzmann's constant

T – absolute temperature [°K]

E – energy of the particle [J]

3.3. Generation of photon [8, 13, 17-19]

The creation of a photon is connected with the law of energy conservation. There are basically few ways how the photon can be generated, in some sense they overlap.

Generally, the wavelength that is most likely to be absorbed is most likely to be emitted by any object, as absorption and emission are mutually interconnected processes.

3.3.1. Line emission [19, 20, 21]

Electron in atom descends from higher to lower energy level. The energy difference between higher(E_h) and lower(E_l) energy level is radiated in form of photon:

$$E_h - E_l = E_p = h \cdot f$$

Absorption and emission of photons by electrons are just two sides of one coin. Electron must first gain energy for example by photon absorption to reemit the absorbed energy in the form of a photon. Electrons can “jump” only between, or exist on, discrete energy levels, which are specific for each atom. Therefore atoms have specific absorption and emission spectra, a fact that is deeply exploited in astronomy, spectroscopy, etc. to identify substances. These spectra aren't continuous but they are consisting of lines, or narrow bands, therefore the term *line emission* is commonly used.

Assume an electron in a given atomic orbital. The energy of its state is mainly determined by the electrostatic interaction of the (negative) electron with the (positive) nucleus, calculable using the principal quantum number.

$$E_n = -chR_\infty \frac{(Q/e)^2}{n^2}$$

E_n – energy level of electron [eV or J]

Q – charge of nucleus [C]

n – number of energy level [dimensionless]

$e = 1,60217733 \cdot 10^{-19}$ C - charge of electron – elementary charge

$c = 299\,792\,458$ m/s – speed of the light

$R_\infty = 1.0973731568525 \cdot 10^7 \cdot \text{m}^{-1}$ – Rydberg's constant.

$h = 6.6260755 \cdot 10^{-34}$ J.s = $4,135\,667 \cdot 10^{-15}$ eV.s - Planck's constant

The typical magnitude is in order from 1 eV to 10^3 eV. The Rydberg levels depend only on the principal quantum number. Energy has negative sign, because it is bound energy.

There are several influences that cause the line emission to contain not single spectral lines (gross structure), what would be expected from discrete energy levels, but narrow bands around these lines (fine and hyperfine structure).

Intrinsic influences on electron energy levels:

Fine structure

Fine structure of line emission arises from relativistic kinetic energy corrections, spin-orbit coupling (magnetic dipole interaction of the electron spin with the magnetic field due to the movement in the electric field of the nucleus) and the Darwin term (contact term interaction of s-shell electrons inside the nucleus). Typical magnitude 10^{-3} eV (or cca 0,001nm shifts around main spectral line).

Hyperfine structure

In atomic physics, hyperfine structure is a small perturbation in the energy levels (or spectrum) of atoms or molecules due to the magnetic dipole-dipole interaction, arising from the interaction of the nuclear magnetic dipole with the magnetic field of the electron. Typical magnitude is 10^{-4} eV (or ca 0,0001 nm shifts around main spectral line).

Here belongs 21 cm line (1420.4058 MHz), which is created by transition of hydrogen electron from parallel (electron and proton spinning in the same direction) to anti-parallel (electron and proton are spinning in opposite directions) spinning configuration. It is observed from hydrogen clouds in interstellar medium.

Electrostatic interaction of an electron with other electrons

If there is more than one electron around the atom, electron-electron-interactions raise the energy level. These interactions are often neglected if the spatial overlap of the electron wave functions is low.

External influences on electron energy levels: [19, 23, 24]

Zeeman effect[23, 24]

The Zeeman effect is the splitting of a spectral line into several components in the presence of a magnetic field.

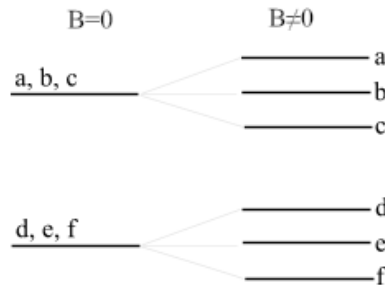


Figure 3.2. – Splitting of spectral lines in the presence of magnetic field[]

Paschen-Back effect

For strong magnetic fields Zeeman splitting does not give a correct description of the energy levels, since the splitting effect becomes nonlinear. This is known as the Paschen-Back effect.

Stark effect[25]

Interaction of electrons with an external electric field. It is analogous to the Zeeman effect, the splitting of a spectral line into several components in the presence of an magnetic field.

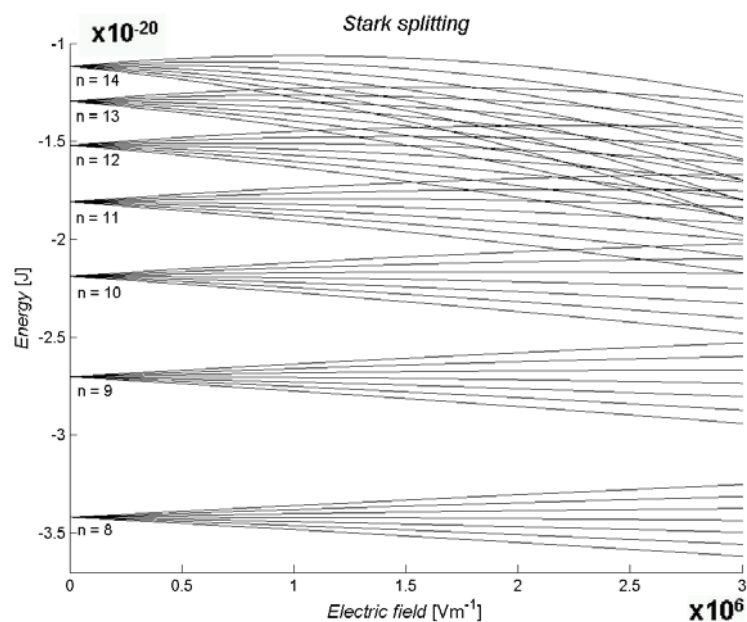


Figure 3.4. Stark splitting of hydrogen emission lines[25]

Molecules, since they contain more atoms, have in addition to single atomic spectra even more complex spectra lines implying from their spatial structure and electronic, vibrational, rotational transitions.

3.3.2. Acceleration of a charged particle [27-34]

Acceleration of a charged particle, whether electrons, protons or ions, produces electromagnetic radiation because of the conservation of energy and momentum. Since a photon carries off energy and momentum, conservation means that the energy and momentum of the emitting particle change due to the emission of a photon. Photons produced by acceleration of charged particles can be of wide range of wavelengths, ranging from low energy radio photons in terrestrial antennas to high energy photons from human-built particle accelerators and cosmic processes around black holes.

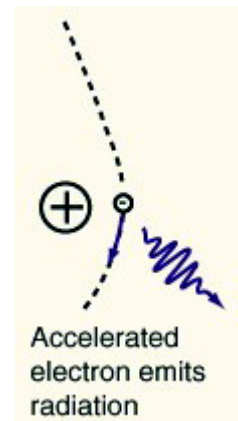
Means of generating photons by particle acceleration can be divided into few groups by the processes that give rise to acceleration.

Bremsstrahlung – (from German: *braking radiation*) Radiation emitted or absorbed when a free electron is accelerated in the field of an atomic nucleus but remains in a hyperbolic orbit without being captured. Since Bremsstrahlung is not quantized, photons of any wavelength can be emitted or absorbed. Therefore the spectrum of Bremsstrahlung itself is continuous.

The *bombarding of electrons*, if it has enough energy, can also eject electrons from the inner shells of the atoms of the target, and the quick filling of those vacancies by electrons dropping down from higher levels gives rise to sharply defined peaks in a spectrum (For example characteristic x-rays in X-ray machines).

Secondary radiation - Bremsstrahlung is a type of "secondary radiation", in that it is produced as a result of stopping the primary radiation (beta particles).

Plasma - in a plasma the free electrons are constantly producing Bremsstrahlung in collisions with the ions.



Antennas – alternating electric current, what is in fact stream of there and back vibrating electrons, is most common source of low energy electromagnetic radiations, low energy photons. Radiation frequencies are equivalent to frequencies of current that feeds the antenna, however efficiency of this emission depends on antenna shape and size.

Cyclotron, synchrotron, linear accelerators – all of them accelerate charged particles, in various ways and product is usually highly energetic electromagnetic radiation - photons. In most cases, generation of electromagnetic radiation is only by product of particle acceleration.

Charged particles are accelerated by electric field, and/or rotated by means of Lorentz force in magnetic field.

Particle acceleration processes occur quite common in universe and the high energy electromagnetic radiation that is produced hereby allows astronomers to observe and identify sources of such radiation (black holes, etc.), which are not observable in visible spectrum.

3.3.3. Blackbody radiation [35-38, 41]

Any object, whether inanimate or living, with temperature higher than 0 °K is source of electromagnetic radiation – photons.

The “Blackbody” is an idealized model of object that is “black” for all wavelengths i.e. it has an emissivity of exactly one at all wavelengths, what means that it absorbs all the energy that reaches it. The amount of total emitted energy by the ideal blackbody depends only on the temperature and is given by Stefan-Boltzmann law:

$$I = \varepsilon \cdot \sigma \cdot T^4$$

I – irradiance or power density [W/m²]

T – absolute temperature [K]

ε – emissivity (for ideal blackbody ε = 1, for real objects (“grey bodies”) ε < 1)

σ - 5.670400 · 10⁻⁸ W·m⁻²·K⁻⁴ - Stefan-Boltzmann constant

The spectrum of emitted radiation depends on the temperature of the object. According to quantum statistics principles, the spectral volume density of radiation

energy can be determined by calculating the equilibrium distributions of photons, for which radiation field entropy is maximum and taking into consideration that the photon energy with frequency f is equal to hf , where h is Planck's constant. Max Karl Ernst Ludwig Planck considered the radiation of ideal gas that obeys Bose-Einstein statistics and derived the famous formula that has been named after him. Planck's law describing electromagnetic radiation of any object by its temperature:

$$I(\lambda, T) = \frac{2\pi hc^2}{\lambda^5} \cdot \frac{1}{e^{\frac{hc}{\lambda kT}} - 1}$$

I – spectral radiance, flux [power unit/(surface unit x wavelength unit)] e.g. [W/(cm².μm)]

T – absolute temperature [K]

λ – wavelength [wavelength unit] e.g. [μm]

h - 6.6260755. 10⁻³⁴ J.s – Planck's constant

c - 299 792 458 m/s – speed of the light

k - 1.380658. 10⁻²³ J/K – Boltzman

Both Stefan-Boltzmann law and Planck's law of blackbody radiation can be found in several variations, as spectral radiance or energy density dependent on frequency or wavelength with various dimension prefixes. Actually integration of Planck's law of blackbody radiation through all wavelengths leads to Stefan-Boltzmann law and derivation of Planck's law leads to Wien's displacement law which describes the peak wavelength for a blackbody of given temperature:

$$\lambda_{MAX} = \frac{2,897 \cdot 10^{-3}}{T}$$

λ_{MAX} – peak wavelength [m]

T – absolute temperature [K]

2,897. 10⁻³ K. m - Wien displacement law constant

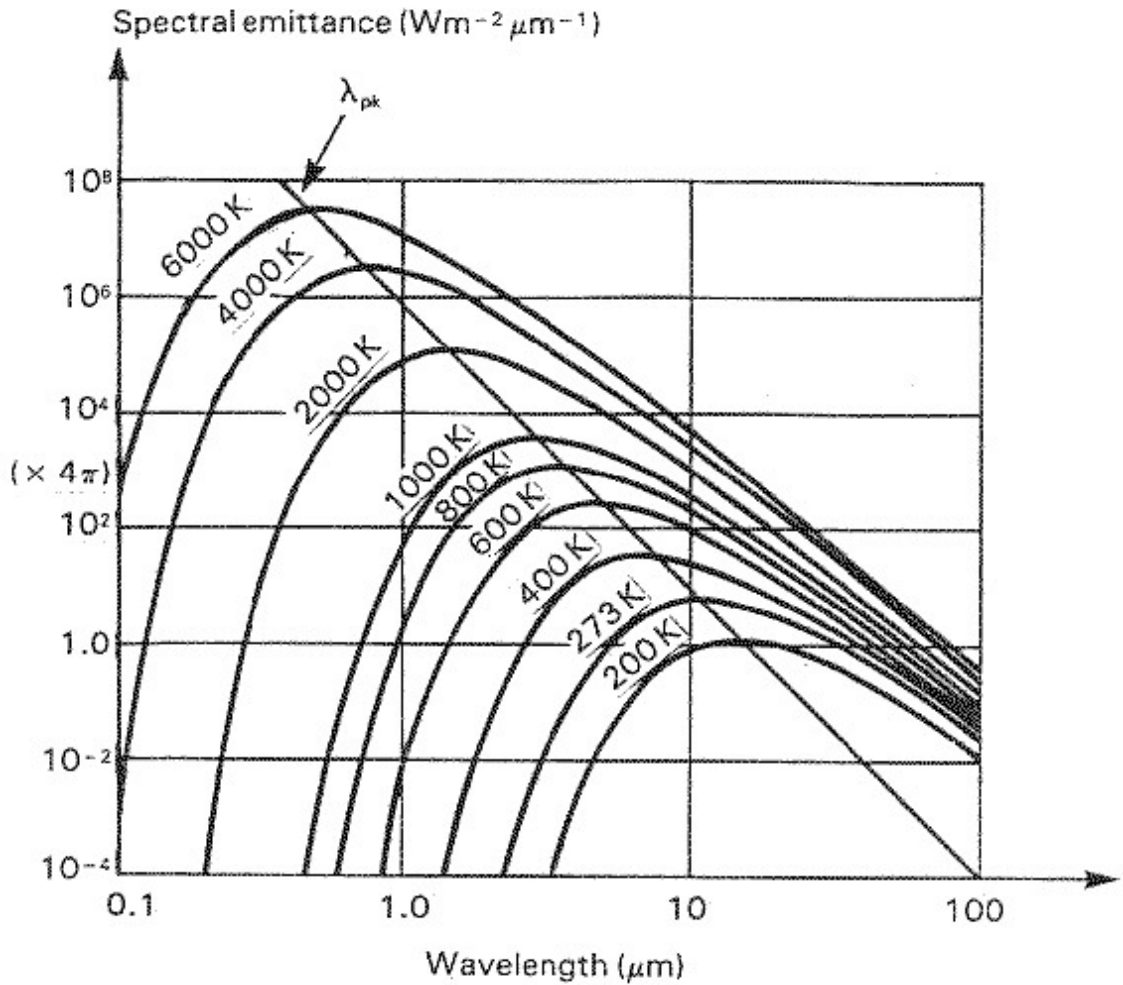
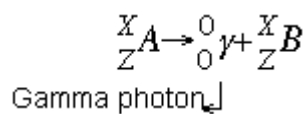


Figure 3.5. – Blackbody spectrum of objects with various temperatures, line showing the peak wavelength [39]

3.3.4. Gamma decay [40, 42, 43]

High energy photons are generated in certain type of atomic nucleus decay – gamma decay. After alpha or beta decay, some daughter nuclei have excess energy (they are in excited state), and they become stable (transition to ground state) after emission of gamma photons. Thus, gamma photons are emitted almost at the same time as beta or alpha rays are emitted.

Gamma decay follows the form:



In gamma decay, neither the atomic number nor the mass number is changed.

3.3.5. Annihilation of matter and antimatter

When particle and antiparticle collide and are annihilated, energy is released, depending on the particle type. In case of annihilation of an electron and its antiparticle positron (underlying process in the Positron Emission Tomography) it gives a rise to two gamma photons, each with energy of 0,511 MeV and opposite direction to conserve energy and momentum:

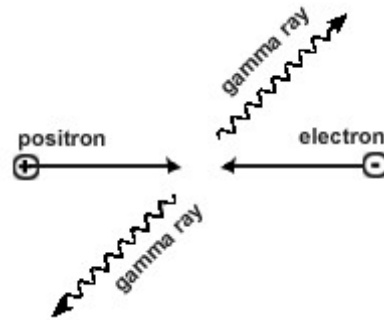


Figure 3.6. – Annihilation of positron and electron[44]

4. Theories of biophoton origin

After brief overview of possibilities how electromagnetic radiation can be generated, we can discuss each possibility and try to find corresponding processes that could occur in organisms and give rise to a continuous ultra-weak flux of photons with wavelength of ca. 200-750 nm, i.e. 6,5 – 1,5 eV, respectively. Closer elucidation of these processes can lead to an understanding of biophoton nature. It is worthwhile to note that general consensus about biophoton origin has not been reached yet in scientific community.

In the following chapters the term “photon” will refer specially, unless otherwise specified, to quantum of electromagnetic radiation of visible and UV band, what is the range of biophoton phenomenon.

4.0.1. Line emission

Line emission, i.e. excitations of electrons to higher energy states and their consequent relaxation to lower energy state accompanied by photon emission can easily occur in any system that undergoes excitation by such an amount of energy that is sufficient to provide electron “jump” to higher levels. However, one must not forget non-linearity of biological systems and possible coupling of lower energy modes to higher energy mode.

There are common physiological processes in organism that provide electron excitation in the course of free radical, reactive oxygen species reactions, that are capable of giving off an amount of energy equivalent to visible and UV range photons. This turns out to be one of the theories of biophoton origin

4.0.2. Acceleration of a charged particle

Processes of acceleration of charged particles certainly do take a place in the microcosmos of the organisms. There are plenty of ion and electron transfers in any living system, which are influenced by weak but omnipresent static geomagnetic field as well as electric fields and magnetic fields generated by innumerable electro-processes on microscopic scale. Excitations of neural and other excitable cells are ubiquitous. However to produce electromagnetic quanta of energy reaching that of visible range (1,6 – 3,1 eV), acceleration would require electric or magnetic fields of such a high power that are not present in biological systems or their normal surroundings. Therefore

continuous generation of biophotons in biological systems by these means can be excluded.

4.0.3. Blackbody radiation

Although the ideal blackbody (emissivity equals one for all incident wavelengths and spectrum of radiation depends only on body temperature) does not exist, we can make good approximation with model of the so-called “grey” body, where the emissivity is lower than one. Stefan-Boltzmann law then implies that “grey” has always lower radiation output than ideal blackbody because of the lower emissivity.

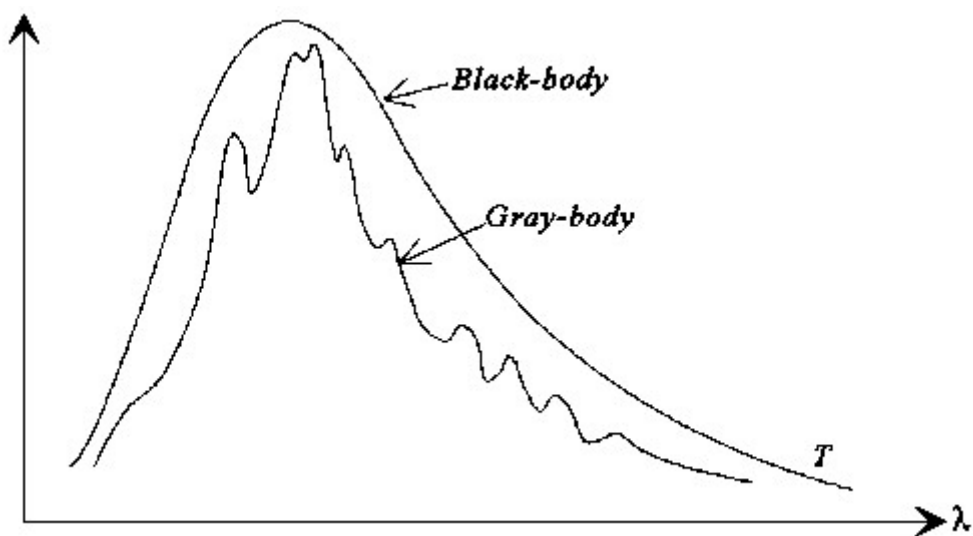


Figure 3.7. – Ideal black-body and more real grey-body of the same temperature – comparison of spectral characteristics [45]

When we consider blackbody of temperature of 310 K (ca. 37 °C),

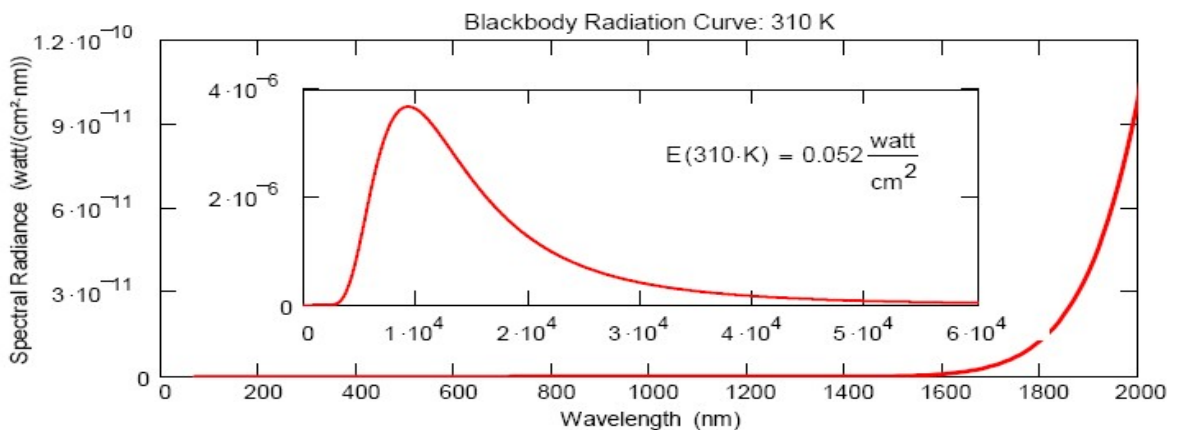


Figure 3.8. – Radiation of blackbody by temperature of 310 K [46]

its peak wavelength of spectral characteristic is approximately at 9,3 μm . This can be seen from the *Figure 3.8.* , as well as counted from Wien's displacement law. The fact that the human body and other, inanimate bodies of temperature ranges that are common to be found on Earth (~ 300 K), has biggest fraction of electromagnetic radiation in far to mid infrared spectrum has its use in thermography.

As the human body is not an ideal blackbody and doesn't absorb (and therefore does not emit all, but only fraction) energy of all incident radiation, its characteristic spectral curve will always lay under the ideal blackbody spectral curve. The most important fact, when searching for biophoton origin, is that blackbody of 310 K has no quantum modes in the visible range of electromagnetic radiation. It can be clearly seen that spectral radiance practically fades out to zero W/cm^2 already by the wavelength of 1,6 μm . According to Planck's law of blackbody radiation, to reach the intensity of biophoton flux ($10^{-18} - 10^{-20} \text{W}/\text{cm}^2$) in the red beginning of visible region a blackbody of the temperature more than 600 K is needed and to reach the equivalent flux in the UV region, temperature of more than 1000 K would be necessary. As this certainly does not take place under physiological conditions, we can exclude blackbody radiation as the cause of the biophoton emission. However, there are other models of quantum cavities, that can be found in human body and fulfill conditions to give rise to such a flux of photons.

4.0.4. Gamma decay

As gamma decay produces only high energy gamma photons, it is not probable source of biophotons, because of the wavelength range. Even when the gamma photon could scatter by means of Compton scattering and photoelectric effect to produce ions and radicals (that could give a rise to photon in visible range), as the gamma decay is a step of nuclear decay, it is very improbable to take place in biological systems under normal conditions.

4.0.5. Annihilation of matter and antimatter

As this phenomenon is connected with pair production by means of high energy processes like pair production by gamma rays and the results is again high energy photon, it is very unlikely to take place in organism under physiological conditions.

4.1. ROS [47, 53]

Reactive oxygen species (ROS) are highly reactive molecules involving oxygen. They are often associated with free radicals as their reactions often lead to creation of free radicals and some ROS are even radicals themselves. A free radical is a cluster of atoms one of which contains an unpaired electron in its outermost shell of electrons. This is an extremely unstable configuration, and radicals quickly react with other molecules or radicals to achieve the stable configuration of 4 pairs of electrons in their outermost shell (one pair for hydrogen).

Reactive oxygen species are:

- molecules like hydrogen peroxide
- ions like the hypochlorite ion
- radicals like the hydroxyl radical - it is the most reactive from all of them
- superoxide anion which is both ion and radical

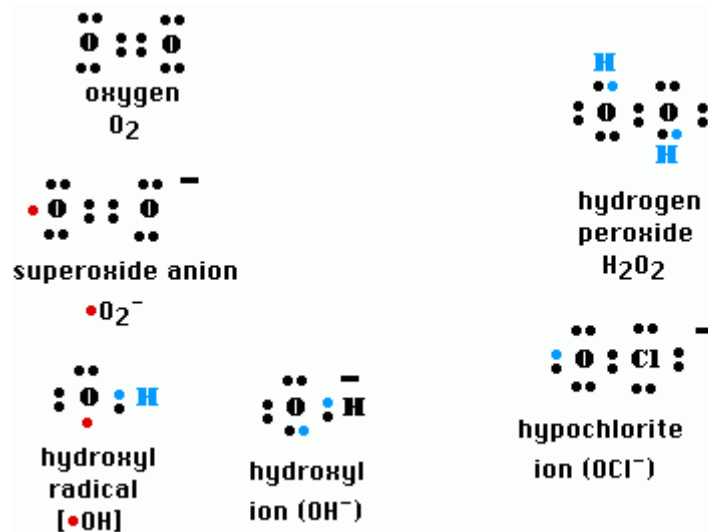


Figure 4.1. [47] – Oxygen molecule and some ROS (Reactive Oxygen species) with their electron configuration. Unpaired electrons are in red. Blue shows an additional electron creating an electron pair

ROS occurs naturally in organisms. Humans possess mechanisms to produce them that can be found in organisms has an endogenous source. They are crucial for normal physiology but an excessive amount of them is considered to be the source of many pathologies and degenerative diseases.

Reactions and recombinations of free radicals, ROS are highly exergonic due to processes that lead to generation of electron excited states and therefore reaction energy output can reach >1 eV, thus reaching visible part of electromagnetic spectrum.

4.1.1 ROS Formation

Reactive oxygen species are formed by several different mechanisms:

- the interaction of ionizing radiation with biological molecules

- a product of cellular respiration (in mitochondria). Some electrons passing "down" the respiratory chain leak away from the main path and go directly to reduce oxygen molecules to the superoxide anion. The major product of mitochondrial respiration is ATP but under energy demand and stress conditions ratio of ROS/ATP production increases

- synthesis by dedicated enzymes in cells e.g. phagocytic cells like neutrophils and macrophages, where the ROS are basis for protection from pathogens like viruses, bacteria and fungi, i.e.

- NADPH oxidase (in both type of phagocytes)

- myeloperoxidase (in neutrophils only)

Oxygen supply is necessary condition for ROS production in organism. Either part of mitochondrial respiration or enzyme catalyzed reactions provide activation of oxygen leading to ROS generation. Activation can be understood as increasing its reactive capability, what can be done in several ways. One of them is by adding an electron and creating superoxide anion($O_2^{\bullet-}$, “•” being a symbol of an unpaired electron), an other is turning of oxygen to excited singlet state.

Oxygen, like all other molecules, has an even number of electrons, but unlike others in which electrons with opposite spins are paired (this may be written as $\uparrow\downarrow$), two of them are unpaired. Oxygen is symbolized as $O_2\downarrow\downarrow$ or $O_2\uparrow\uparrow$. Such a constitution of an outer electron shell is termed a *triplet state*. Particles in a triplet state possess an excess of energy over the state in which all their electrons are paired and which is called a ground

singlet state. Unlike all other molecules, oxygen never possesses a ground singlet state. If it acquires an energy quantum sufficient enough (~ 1 eV) to reverse a spin of one of its unpaired electrons, ($O_2\downarrow\downarrow + h\nu \rightarrow *O_2\downarrow\uparrow$) it jumps into excited singlet state.

Natural ROS production by activated neutrophils was considered long time to be only case where ROS are produced. However, during the last decade NADPH-oxidase which utilize ROS production were detected in a variety of cells – lymphocytes, endothelial cells of blood vessels[50, 56], ocular lens[51], colonic epithelial cells[52], cells of connective tissues, cardiomyocytes, smooth muscle cells from human arteries[49], they are present in spermatozoa and eggs where they are grossly activated at the moment of fertilization[48]. Similar enzymes were found in plants and microbes[48]. It was also reported that eosinophils rather actively consume oxygen even in a resting state and they do this exceptionally through the one-electron way of oxygen reduction, producing superoxide[48].

ROS are also produced by enzymes that previously were not regarded as ROS generators, for example, by gamma-glutamyl-transpeptidase. One of the recent findings is that even immunoglobulins are oxygenases: they catalyze water oxygenation to H_2O_2 with singlet oxygen. Certain type of oxidases(Cyanide-resistant) were found in mitochondria of animals, plants, fungi, protists[53]. One may conclude that the omnipresence of ROS producing enzymes in organisms can give rise to flux of photons as is observed in biophoton phenomenon.

4.1.2. Significance of ROS

ROS, more specifically their radicals, are generally ill-famous because of thousands of studies and papers that observe their malign effects *in vitro*. They are considered to be the cause of degenerative diseases and aging. Since radicals possess one unpaired electron they are highly reactive. In desire to get the pair for their electron they easily react with almost any biomolecule and may cause damage to cellular structures. But their real damaging capability is caused by the fact that the oxidized biomolecule (whose electron is taken by radical) becomes a radical itself which can oxidate another molecule etc. These chain reactions are biggest threat of their activity.

Most common target of free radicals are lipid membranes of cells, especially hydrogen on double bonded carbon of unsaturated fatty acids.

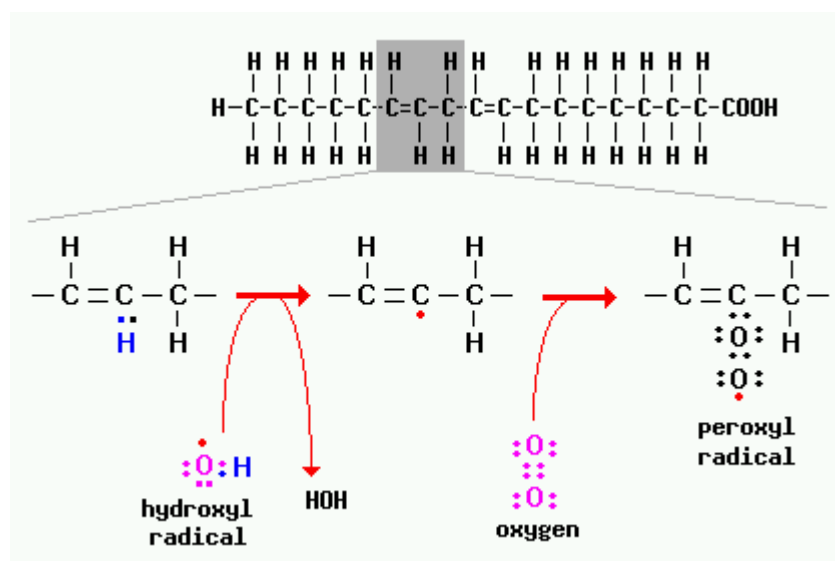


Figure 4.2. – Example of typical radical chain reaction with unsaturated fatty acid initiated by lipid peroxidation[54]

However, as was already mentioned above, ROS are of great importance in normal physiology. Phagocytes and neutrophils synthesize them in great amounts in organisms to destroy pathogens like viruses, bacteria and fungi[55]. So-called “respiratory burst” occurring in neutrophils is accompanied by multiplied non-mitochondrial consumption of oxygen, which leads to production of ROS.

There is rapidly growing number of studies dealing with the issue of redox signaling (the concept saying that electron-transfer processes play a key messenger role in biological systems), where another crucial role of ubiquitous ROS is showing up[60]. One could speculate if it isn’t actually photon signaling or photon communication what takes place in these processes as ROS reactions may lead to generation of excited electronic states and consequently to photon generation.

As energy demand in organism increases, the production of ROS grows too. It is worthwhile to remind that human brain although being just 2 % of human weight it consumes 20 % of human oxygen intake. Since brain tissue doesn’t contain more mitochondria than other metabolically active tissues [58], it may be suggested that oxygen is consumed in non-mitochondrial, enzymatic reactions where ROS are generated.

Some studies indicate that active oxygen (superoxide anion radical) may even serve as necessary “ignition” of oxidative combustion [57]. It has been found that ordinary air that is completely ridded of negative air ions (actually superoxide anion

radicals $O_2^{\cdot-}$) accelerates death of rats and mice, with symptoms showing disturbances in neurohormonal regulation and pituitary insufficiency [59]. A minute amount of air ions is enough to restore health status from the pathological conditions to normal.

4.1.3. Regulation of ROS activity

Since an excessive amount of ROS and radicals can be harmful, an “antioxidative system” of ROS activity regulation was evolved in organisms. Organisms are able to produce strong macromolecular antioxidant enzymes. For example ubiquitous superoxide dismutase(SOD) catalyses the recombinations of two $O_2^{\cdot-}$ radicals producing H_2O_2 and oxygen, and catalase reduces H_2O_2 to water and oxygen.

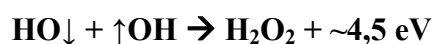
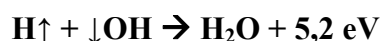
There are natural antioxidants as ascorbic acid(vitamin C), tocoferol(vitamin E) or beta-carotens, but their functionality in regulation of ROS activity is less effective than by endogenous antioxidant enzymes.

Greater amount of oxygen intake is used to produce ROS under physiological conditions in aerobic organism than was previously thought[61] based only on immediate reaction by catalyzing enzymes that degrades and recombine ROS to lower energetical state accompanied by emission of photon of visible range. Why would organism do such an, on the first look, useless and energy wasting activity? If we consider probable essential communicative and triggering effect of the redox signaling and photon emission, this activity turns out from being irrational to highly substantial if not crucial.

4.1.4. Energy generation in ROS reactions

As already mentioned above, ROS and radical reactions and their catalysed recombinations are exergonic enough to give off the quantum of energy in the form of photon in visible or UV spectrum due to electron excited states that are created when unpaired electrons of radicals recombine.

Here are some examples of the reactions that can occur in water milieu of biological systems[62, 63]:





Complexity of biomolecules and their reactions with ROS, radicals does not lead to single spectral lines of biophoton emission but broad spectrum centered around these and other values can be observed. There are as many vibrational sublevels as there are molecules that interact together. When there are enough molecules influencing each other, there are so many sublevels of macromolecular orbitals that we no longer perceive their discrete nature: they form a continuum. We no longer consider energy levels, but energy bands.

4.1.5. Some studies and facts suggesting the plausibility of radical and ROS theory of biophoton origin

It is common knowledge in physiology[55] that production of ROS by neutrophils in respiratory burst during immune reactions is accompanied by luminescence. NADPH oxidase systems and subsystems that are responsible for this non-mitochondrial one-electron reduction of oxygen is however found not only in neutrophils and other cells of the mammalian immune system but omnipresent in almost all living organism, as shown above. Equally omnipresent are endogenous antioxidant systems as superoxide dismutase and catalase are not only present in higher organism but even in anaerobic bacteria, indicating presence of ROS and radicals in these organisms.

The author of this work has observed that biophoton emission from body surface is generally higher in the state of classical influenza and under inflammation processes when the immune system is highly activated and immune cells are actively producing ROS.

It has been found that biophoton emission from human hand increased in oxidative environment and decreased in nitrogen environment[64], what may indicate significance of oxidative processes in biophoton origin.

More indications are to be found in chapter 6. Overview of human biophoton research

4.2. DNA

While ROS and radical theory of biophoton origin is relatively simple and easily understandable due to more or less common biochemical approach, DNA theory of biophoton origin is much more complex. Deep understanding of quantum mechanics, biophysics, biology, and physiology is needed. This combination is rarely found in one individual, therefore not many are able to judge the plausibility of this theory. Lack of comprehension may lead to neglecting or denying of this theory. However we cannot neglect nor deny it as it offers explanation to phenomena that are hardly explainable by other theories. Although ROS and DNA theory may overlap in explaining some phenomena and represent different points of view, they do not necessarily exclude each other. We can view both as complementary to each other.

In this theory the DNA helix in cell nucleus is considered to be quantum electrodynamic cavity that is constantly excited by metabolic activity of cell.

4.2.1. DNA structure [65, 66, 69]

Described for the first time by Watson and Crick in 1953, DNA is right handed double helix found in the nucleus and the mitochondria of eucariotic cells. It is carrier of genetic information. DNA stands for deoxyribonucleic acid. If we break down the name into the parts, we get the basic structure of DNA. DNA is a long string of nucleotide units attached to one another, where thre basic components are present:

- 1) a sugar molecule
- 2) a phosphate group
- 3) a nitrogenous base

The sugar molecule in DNA is called deoxyribose, hence the name deoxyribonucleic acid.

The Nucleotides of DNA

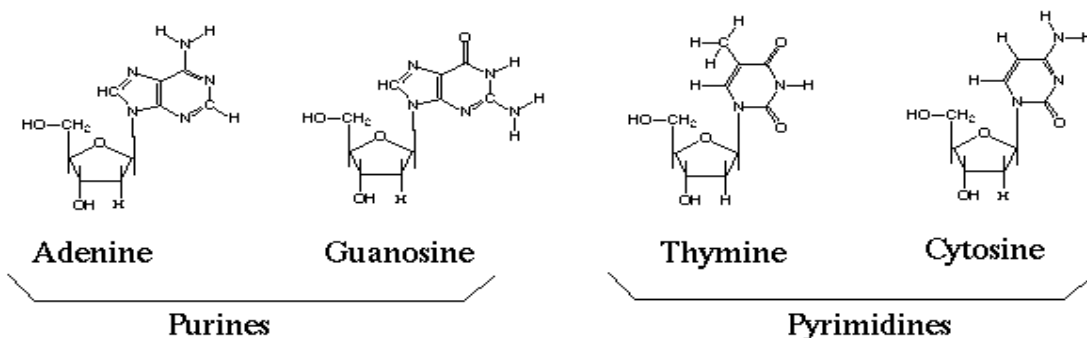


Figure 4.3. Nucleotides of DNA[66]

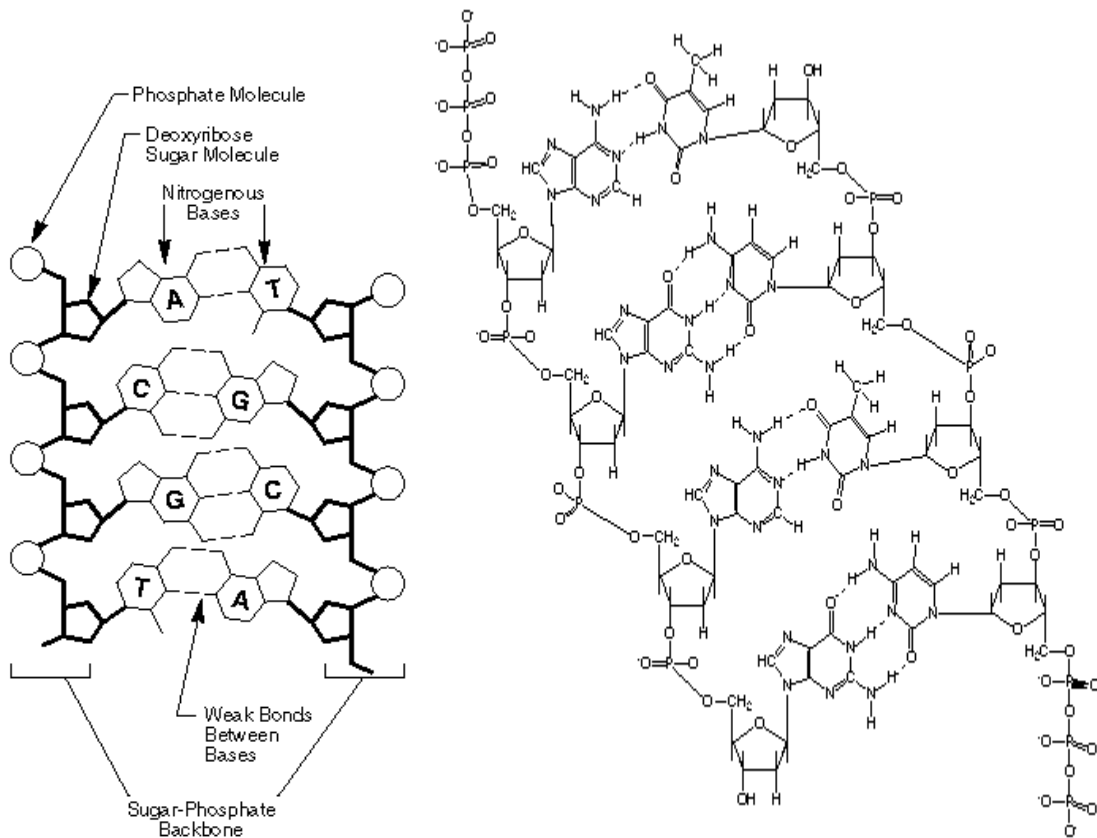


Figure 4.4. A – left: schematic planar structure of DNA double strand[65]

Figure 4.4. B – right: more detailed structure of DNA double strand[65]

There are 4 different types of bases in DNA: adenine, guanine, thymine, and cytosine. They are commonly abbreviated as the letters A, G, T, and C, respectively. DNA strands are connected by hydrogen bonds between nitrogenous bases. There are only A – T bonds and G – C bonds created.

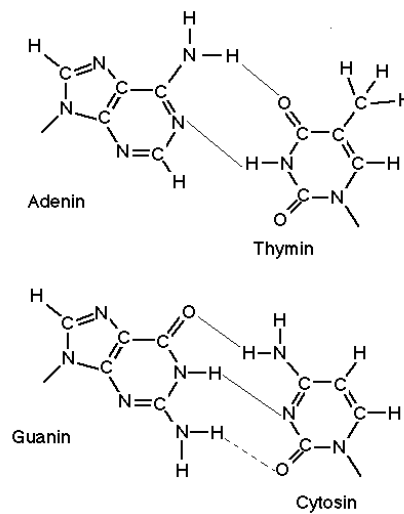


Figure 4.5. –structure of DNA bases [67]

The bases are oriented perpendicular to the helix axis. They are hydrophobic in the direction perpendicular to the plane of the bases (cannot form hydrogen bonds with water). The interaction energy between two bases in a double-helical structure is therefore a combination of hydrogen-bonding between complementary bases, and hydrophobic interactions between the neighboring stacks of base-pairs[75].

DNA spatial structure is what makes it unique among other cell structures. In eukaryotic cells, DNA double helix is wrapped around a protein core of 8 histone molecules creating *nucleosome* unit. DNA with its proteins is also called *chromatine*. Nucleosomes with DNA are ordered in “beads on the string” fashion and this string (~10 nm fiber) is coiled to form 30 nm wide chromatine fiber (“supercoil”). Higher order folding varies with cell cycle.

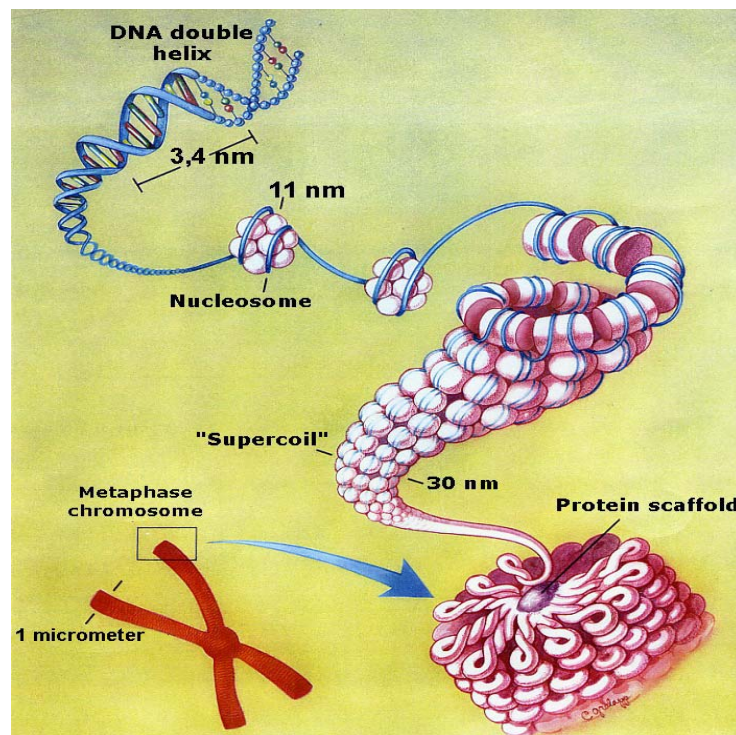


Figure 4.6. – Artist’s realistic representation of DNA spatial conformation in larger units[68]

4.2.2. Excited states in DNA [70]

Although major approach to DNA research takes into consideration its static structure, we have to consider DNA as dynamic structure, which is rich in movements, vibrations and undergoes excitations and relaxation to ground states.

It has been known for a long time that DNA is luminescent materials[71]. It is primarily due to the excitability of DNA bases. The fluorescence spectra of native and denatured calf thymus DNA are distributed around the wavelength of 350 nm. These

spectra have been identified as excimers and exciplexes built by base-molecules due to the base stacking[70].

An excimer (originally short for excited dimer) is a short-lived molecule that bonds two molecules in an electronic excited state, if there is enough excitation energy present and molecules are close enough. While an excimer is a molecule that forms a dimer(refers to a molecule composed of two similar subunits or monomers linked together) from the same molecule in the excited state, an exciplex is a molecule that forms a dimer from different molecules in the excited state. An excimer forms a dimer only in the excited state; excimers do not form dimers in the ground state [72]. We may point out an interesting fact namely that the excimer (exciplex) formed by the base molecules of DNA must be classified as a so-called intramolecular excimer. Significance of this fact will be revealed later.

4.2.3. Dicke's theory [70]

American physicist Robert H. Dicke stated, based on his observation, as first in 1954[73], that two oscillators cannot be considered independent if they lay not further apart as the coherence length of their radiation. Therefore, if they fulfill certain conditions, their radiation is mutually coherent.

Dicke has shown that "in the usual treatment of spontaneous radiation by a gas, the radiation process is calculated as though the separate molecules radiate independently of each other. This simplified picture overlooks the fact that all the molecules are interacting with a common radiation field and hence cannot be treated as independent." He first considered the problem under the assumption that the gas molecule distances have quite small dimensions compared to a radiation wavelength. The result shows that the radiation rate of a coherent spontaneous radiation process is proportional to the square of the molecular concentration rather than linearly to the concentration as the usual radiation theory has predicted. He called this new strange emission "superradiance". The opposite case, of the states having abnormally low radiation rates, when in the ideal case for an even number of molecules the radiation rate equals zero, be named "subradiance".

4.2.4. DNA as source of coherent radiation

There are more structures in a cell (protein complexes, DNA) that can be considered potential source of coherent radiation due to their small mutual distance. Intramolecular exciplexes and excimers are most probable sources as the distances of polymer substructures (nitrogenous basis, in case of DNA) are very small (0,34 nm B-DNA, 0,26 nm A-DNA). However, the DNA due to its highly complex and condensed spatial conformation seems to be the best candidate for the quantum cavity source of biophoton emission.

It was found that exciplexes are formed in DNA even in room temperature [77], however to form exciplex or excimer the distance of forming monomers has to be smaller than 0,2 nm [78]. Therefore monomers that form the exciplexes / excimers must be in dynamic movement or oscillation to reach this distance.

In the DNA, stacking of bases provides conditions for stimulated radiation [76]. Bases can be like working medium, the long double helix can be like an optical cavity and pumping of energy can come from ATP. Above mentioned fact that intramolecular exciplexes / excimers are present is important because the threshold for generating stimulated coherent emission from intramolecular excimers is much lower than that of intermolecular excimers. The technique for producing stimulated coherent emission from intramolecular excimers was built for the first time in 1973 already [74].

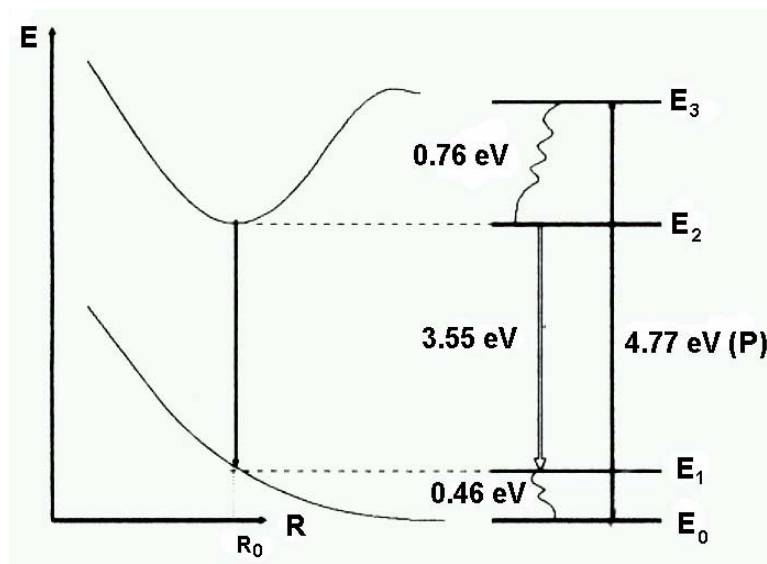


Figure 4.7.5. – Four level model of excimer formed by two interplanar neighbour DNA base molecules distanced 0.2 nm. $E_3 - E_0 = 4.77 \text{ eV}$ (P – pumping energy), $E_3 - E_2 = 0.76 \text{ eV}$ (binding energy), $E_2 - E_1 = 3.55 \text{ eV}$, 320-350nm (excimer emission), $E_1 - E_0 = 0.46 \text{ eV}$ (repulsive energy) [70]

The term coherence is usually connected with monochromatic or narrow band light. When we speak about the broad band phenomena of biophoton emission and its coherence, it is important to keep in mind that neither a narrow bandwidth (single quantum modes), nor high intensities are necessary conditions for quantum coherence [76, 79].

As we are dealing with quantum phenomenon, we can describe it by its quantum statistical properties. However, the intensity measured from some biophoton sources (for example the intact human body) is too low to use the standard approach of determining the degree of coherence with correlation functions as it is common in laser experiments.

The conditions for the quantum coherence are observed in properties and behaviour of biophoton emission [80-85]:

Poissonian and even sub- Poissonian or super- Poissonian distribution of photo count statistics (sub- and super- Poissonian distribution indicate presence of “squeezed light”) in $\Delta t < \tau$

(Δt length of time interval, τ – coherence time of chaotic field) [86, 87]

The Poissonian probability function:

$$p(n) = \frac{\langle n \rangle^n e^{-\langle n \rangle}}{n!}$$

describes the probability $p(n)$ of detecting of n photons in a given time interval Δt . $\langle n \rangle$ is the mean value of the number of detected photons in the time interval Δt from all measurement. However chaotic light shows thermal distribution:

$$p(n) = \frac{\langle n \rangle^n}{(1 + \langle n \rangle)^{n+1}}$$

One of the easily checkable properties of Poissonian distribution is that its mean value ($\langle n \rangle$) that has to be equal to the variance ($\langle \Delta n^2 \rangle$) (a square of standard deviation):

$$\langle n \rangle = \langle \Delta n^2 \rangle$$

whereas

$$\langle \Delta n^2 \rangle = \langle n \rangle (1 + \langle n \rangle / M)$$

is valid for completely random field with M modes[76]. However, it is worthwhile to be noted that Poissonian distribution can be reached not only by coherent source but even by random source if $\Delta t > \tau$ and the random source is of many modes ($M \gg 1$) [76, 86].

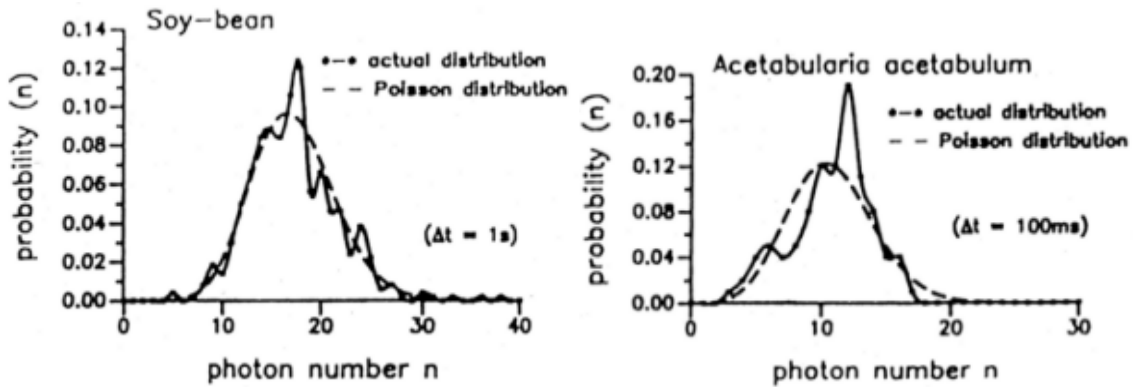


Figure 4.8. [88]– Poissonian distribution of photon counts found in simple biological samples is considered to be present in higher organisms where is hardly measurable because of low photon intensities

Hyperbolic decay after excitation of organisms [89, 84]

If we consider relaxation from a higher energy state in a random system, radiation intensity $n(t)$ is proportional to the number of radiators:

$$n'(t) = -\lambda.n(t)$$

which solution is:

$$n(t) = n(0). e^{-\lambda t}$$

which is the exponential relaxation typical to random, non-coherent decay, e.g. radioactive decay. However in coherent systems all radiators are coupled to each other and radiation intensity after excitation is proportional to the square of the number of radiators:

$$n'(t) = -\mu \cdot n^2(t)$$

with the solution:

$$n(t) = \frac{1}{\mu \cdot (t + t_0)}$$

if $t_0 = 1 / (n(0) \cdot \mu)$. The solution is a hyperbolic function typical for the relaxation of cooperative systems as biological systems are.

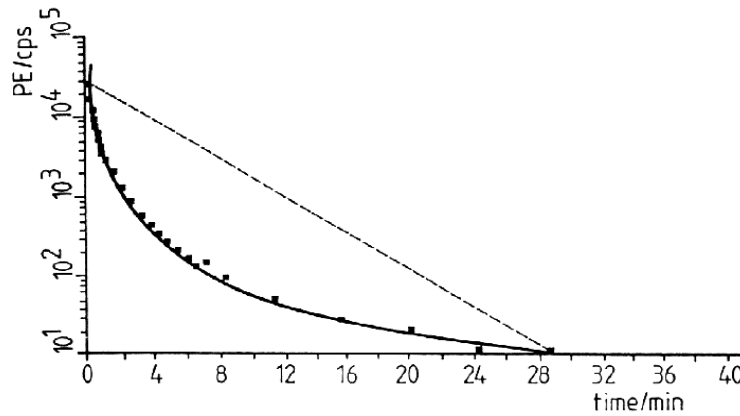


Figure 4.9. [89]– Delayed luminescence of a leaf of the plant *Bryophyllum daigremontanum* after illumination with red light of a wavelength of 676 ± 1 nm. The approximation of decay with exponential function (dashed line) does not fit, hyperbolic function (full line) fits perfectly

In the coherent system apart from superradiance (the maximum intensity of the emitted light scales with the square of the number of atoms [90]) even phenomenon of subradiance can take place, depending on the parameters of the coherent system. The subradiance state can also be called the photon trapping state. The trapping mechanism is a coherent migration of photon (excitation energy) within the system of cooperative molecules. The trapping time can be several orders of magnitude larger than the natural decay time of a single molecule and is mainly a function of the density of the molecules. Subradiant trapping mechanism behind the theorem of photon storage in large and dense molecules as DNA, can be of great significance in biological systems [76]. It is conceivable that this trapping can influence metabolic and cellular events by triggering amplification mechanisms and promote photochemical processes even in the dark.

4.2.5. Possible role of non-coding DNA

Human haploid cell contains total DNA about $3 \cdot 10^9$ base pairs, but from this number only 1-1.5% serves for encoding proteins. The huge majority of DNA does not code neither proteins nor RNA. Such a kind of DNA is called as non-coding (or “junk”) DNA. It contains lot of repetitive sequences. Up to now the functions of this repetitive DNA is unknown. Non-coding DNA may be necessary for biophoton emission as working medium and as optical cavity.

4.2.6. Some studies and facts suggesting that the DNA or chromatin is a source of biophoton emission

- separated nuclei show higher re-emission of photons after excitations than whole cells, indicating absorption or trapping mechanisms in an intact cell[91]
- intercalation of ethidium bromide(EB) between DNA base pairs changes DNA conformation by means of uncoiling the DNA helix[92]. Depending on the EB concentration the DNA helix can be uncoiled and with the rising concentration even be coiled to opposite helicity(from right handed to left handed). If DNA is considered a photon storage due to its spatial conformation, its unwinding should cause higher emission of photons. It is indeed so as can be seen on Figure 4.11.

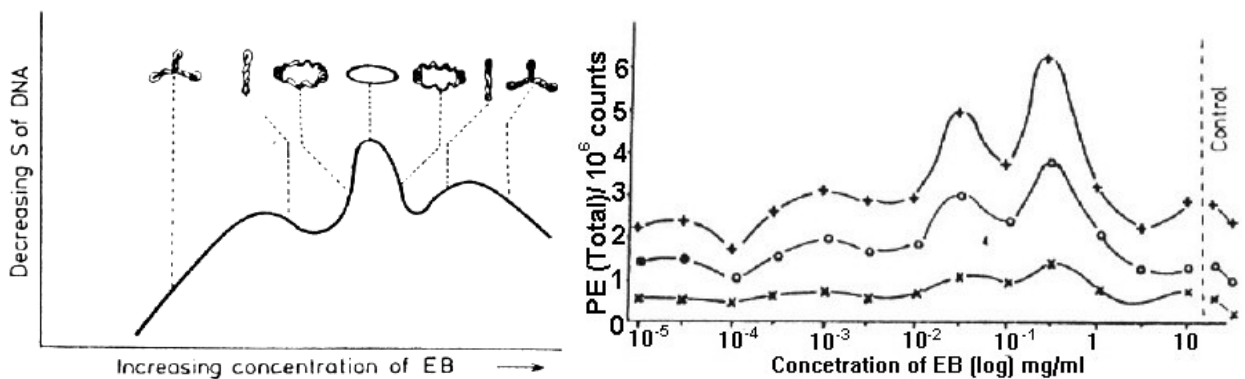


Figure 4.11. A[92] – On the left: decreasing of sedimentation of DNA indicates unwinding of DNA after treatment with ethidium bromide. In higher concentration DNA is even rewinded in opposite rotation.

Figure 4.11. B[92] – On the right: photon emission (PE) from cucumber seedlings with increasing concentration of EB shows similar dependence as unwinding and rewinding of DNA. Three curves show the PE after 1,2,5 hours of adding of EB

- nonlinear dependencies of photon emission on cell density(cress[95], daphnia[94], drosophila[96]) may suggest communication effects, subradiance or superradiance in the terms of Dicke on macroscopic scale

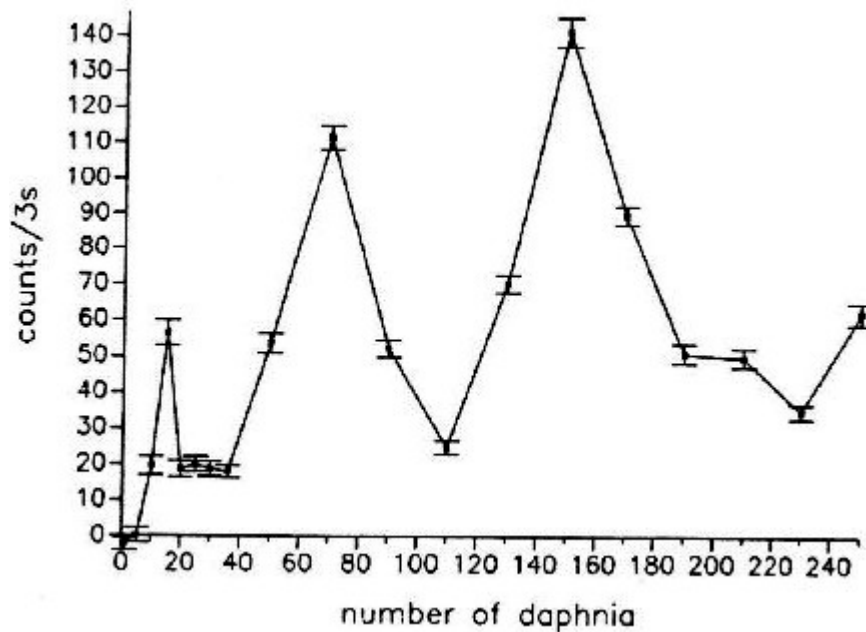


Figure 4.12[94] - The intensity of photon emission as a function of the number of individuals of *Daphnia magna* in 15 ml cuvette. The diagram shows the mean values and the 2-fold standard error of 150 readings.

- different dependencies of photon emission on cell density discriminate between healthy and cancer cells[97]. It is noteworthy that some cancerogenic substances have almost the same molecular structure as their harmless counterparts, however differ in absorbance spectrum. [98] Further, carcinogenic polycyclic hydrocarbons have in common almost the same level of optical transitions. That suggest some intercellular optical communication perturbation in neoplastic processes
- DNA alone, without its protein complexes compared to whole chromatin(DNA + its proteins) reemits much less photons after illumination

[93]. It can be interpreted that its spatial conformation is necessary for its function of photon storage.

	Total photon counts in 0.5 s corresponding with 10⁶ cells	transmission 500 nm %
Whole cells	4051 + 770	18
Cytoplasmic fraction	1736 + 581	87
Nuclear fraction	5670 + 1463	36
Chromatin fraction	13 230 + 3206	44
DNA (phenol-extracted)	388 + 72	96

Table 4.1. [93] – shows photon reemission from the cell fractions

4.3. Other possible sources of biophoton emission [130]

Apart from the two main of means of the photon generation in biological systems, which are subject in majority of the literature that is trying to identify the source of the biophoton emission, few more are worthy of mentioning:

- Collective excitations in perturbed water and cytosol
- Molecular interactions in the electric field of biomembranes
- Microtubules

These sources and mechanisms may be partially connected or be a functioning parts of the mechanisms described above. It seems to be plausible that photon emission may have multiple sources, which are functionally interconnected.

5. Technical prerequisite for measurement of biophoton emission

Measurement devices used to quantify the biophoton emission must be capable of detecting minute amounts of photons (10^{-18} W/cm², few photons of visible range), which requires high sensitivity as well as low noise to reach high signal/noise ratio.

5.1. Photomultipliers [99, 100, 101]

Photomultipliers (PMT) are the most common devices used for detecting low amounts of light. They have a wide use in spectroscopy, gamma cameras, in-vitro assay, scintillation counting, biotechnology, high-energy physics experiments, aerospace, low light detection, etc. For the purpose of measuring of small amounts of light quanta they operate in single photon counting mode.

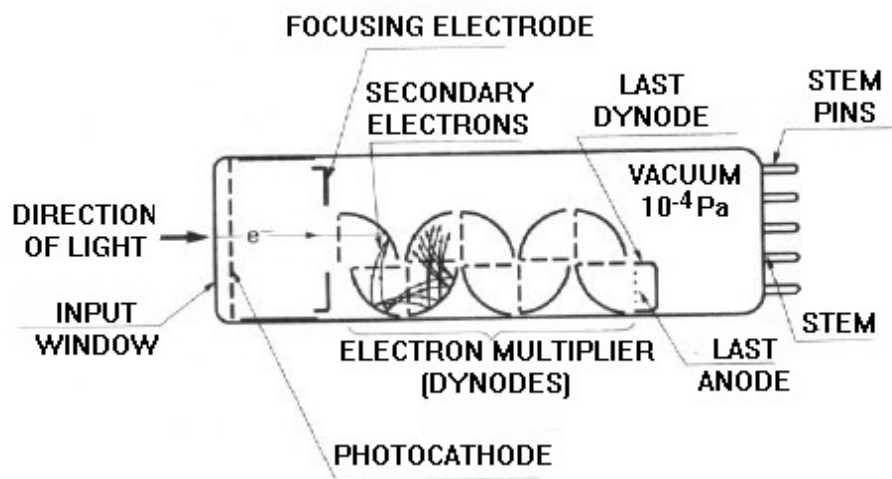


Figure 5.1 – Scheme of the photomultiplier [99]

The principle of PMT functioning lies in photoelectric effect and acceleration of charged particles (here: electrons) in an electric field.

Light, i.e. photons, enter through an *input window*. They fall on a *photocathode* of suitable material to knock the electrons out to the vacuum of the tube. Electrons are focused and accelerated in an electric field to the first *dynode*. Since the incident electron gained the energy by acceleration, it knocks out more electrons from the dynode. Those are called *secondary electrons*. This process continues cascade-like to next dynodes, each with higher positive voltage. After multiplying the flow of electrons falls on the last dynode – anode. In the next steps is this already detectable electric current amplified in a classical way and processed by hardware and software.

5.1.1. Characteristics of photomultipliers

Quantum Efficiency

Quantum efficiency (QE) is the number of photoelectrons emitted from the photocathode divided by the number of incident photons and is generally expressed in percent:

$$QE = \frac{\text{Number of photoelectrons produced}}{\text{Number of incident photons}} \times 100\%$$

QE as function of wavelength is spectral response characteristic of PMT. QE usually reaches just a few tens of percent even for good PMT's.

Another measure of QE is *radiant sensitivity* S :

$$S = \frac{\text{Photoelectron current [A]}}{\text{Incident power[W]}}$$

Radiant sensitivity is easier to measure directly.

Spectral response range

Spectral response range is defined by two border wavelengths, short and long. Region between these two wavelengths is called *spectral response range*.

To enumerate all of characteristics and parameters of PMT's that imply from their structure and describe their behavior is beyond the scope of this work. We can at least mention several of them: *gain, dark current, various time characteristics, linearity of response, stability, voltage and light hysteresis, polarized light dependence, etc.*

5.1.2. The Tube

A photomultiplier tube that includes dynodes must be evacuated to 10^{-5} Pa to work properly. Otherwise molecules of gas can cause collisions with accelerated electrons and stopping them from reaching the dynodes or cause additional noise themselves.

5.1.3. Input window of photomultiplier

The material of the input window determinates the short wavelength of *spectral response range*. It is important to keep in mind that input windows can generate loss in *quantum efficiency* due to reflection and absorption of incident light. There are many possible materials, however most common choices are:

MgF₂ crystal - a magnesium fluoride (MgF₂) crystal allows transmission of vacuum ultraviolet radiation down to 115 nm.

Sapphire (Al₂O₃) - sapphire shows an intermediate transmittance between the UV-transmitting glass and synthetic silica in the ultraviolet region. The short cutoff wavelength is 150 nm

Synthetic silica - synthetic silica transmits ultraviolet radiation down to 160 nm.

UV glass (UV-transmitting glass) - most suitable for transmission of UV light. The short cutoff wavelength is 185 nm

Borosilicate glass - this is the most commonly used window material. It is often called "Kovar glass" because it has a thermal expansion coefficient very close to that of the Kovar alloy which is used for the leads of photomultiplier tubes. The short cutoff wavelength is 300 nm

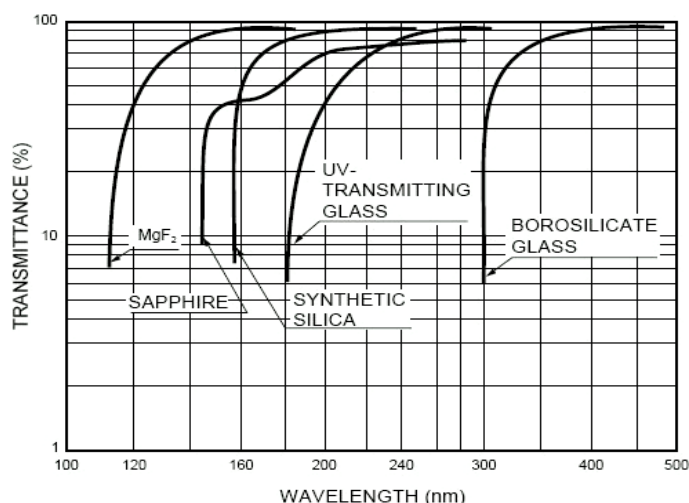


Figure 5.2. [99] – spectral transmittance of various PMT input window materials

5.1.4. Photocathode

The material of photocathode determinates the long cutoff wavelength of *spectral response range*. Most photocathodes are made of a compound semiconductor mostly consisting of alkali metals with a low work function. Each photocathode is available as transmission (semitransparent) type or a reflection (opaque) type. Relatively low (not exceeding 30%) *quantum efficiencies* that are common for PMT and generally for all devices with detection based on photocathode are understandable when we briefly look on the basic problem of photocathode thickness. If the photocathode would be too thick, it may easily happen that photons get absorbed in the material and knocked electrons do not manage to get out. If the photocathode would be too thin, photon could pass the photocathode without collision with an electron. There are several kinds of photocathodes which are currently in practical use:

Cs-I: solar blind, good for vacuum ultraviolet, $\lambda < 200$ nm

Cs-Te: solar blind, good for vacuum ultraviolet, $\lambda < 300$ nm

Sb-Cs: from UV to visible, used often for reflection type photocathodes

Bialkali (Sb-Rb-Cs, Sb-K-Cs): high sensitivity and lower dark current than Sb-Cs

High temperature, low noise bialkali (Sb-Na-K): possible to use in temperatures up to 175°C (usual photocathodes are only good up to 50°C). Low dark current in room temperatures

Multialkali(Sb-Ba-K-Cs): from UV to infrared (IR)

Ag-O-Cs: from UV to IR, however higher sensitivity in longer wavelengths

GaAs (Cs): 300-850 nm

InGaAs (Cs): similar to previous but reaches to 1000 nm

InP/InGaAsP(Cs), InP/InGaAs(Cs): utilizing PN junction, providing new possibility of counting photons even to 1,7 μ m, when cooled down to -80°C

5.1.5. Types of dynodes

Photomultiplier dynodes are usually made of alkali antimonide, BeO, MgO, GaAs or GaAsP and usually give out 5-10 secondary electrons per incident electron. Dynodes can be found in various geometries [99]:

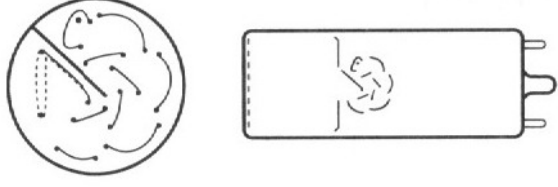
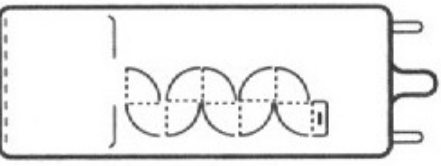
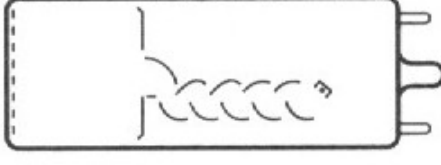
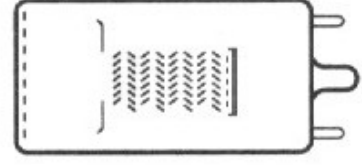
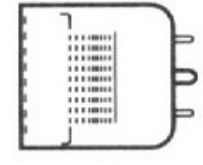

Types of electron multipliers	Scheme of dynode geometry
<p>Circular-cage type</p> <ul style="list-style-type: none"> - compactness - fast time response 	
<p>Box-and-grid type</p> <ul style="list-style-type: none"> - widely used - superior in photoelectron collection efficiency. - high detection efficiency and good uniformity 	
<p>Linear-focused type</p> <ul style="list-style-type: none"> - fast time response - good time resolution - excellent pulse linearity 	
<p>Venetian blind type</p> <ul style="list-style-type: none"> - suitable for large photocathode diameter 	
<p>Fine-mesh type</p> <ul style="list-style-type: none"> - possible to operate in highly magnetic environments - position-sensitive capability when used with a special anode 	
<p>MCP (MicroChannel plate)</p> <ul style="list-style-type: none"> - position-sensitive capability when used with a special anode - improved time resolution as compared to other discrete dynodes 	

Table 5.1. [99]

Voltage on dynodes is usually distributed by voltage divider. Typical voltages used in operating of PMT's are in range from hundreds to thousands Volts.

The term “Biasing” means providing each successive dynode with more positive voltage than the previous one in order to make sure that electrons move from dynode to dynode and amplification takes place.

5.1.6. Associated circuits

The design of the associated circuits depends on the nature of the signal. In the high and medium light levels, where the current output is more or less continual, AC and DC methods of measuring current output are used. In low light level measurements, single quanta of light, photons, are counted.

In the single photon counting method shown in Fig. 5.3., the output pulses from the photomultiplier tube are amplified and only the pulses with an amplitude higher than the preset discrimination pulse height are counted. This method allows observation of discrete output pulses from the photomultiplier tube, and is the most effective technique in detecting very low light levels.

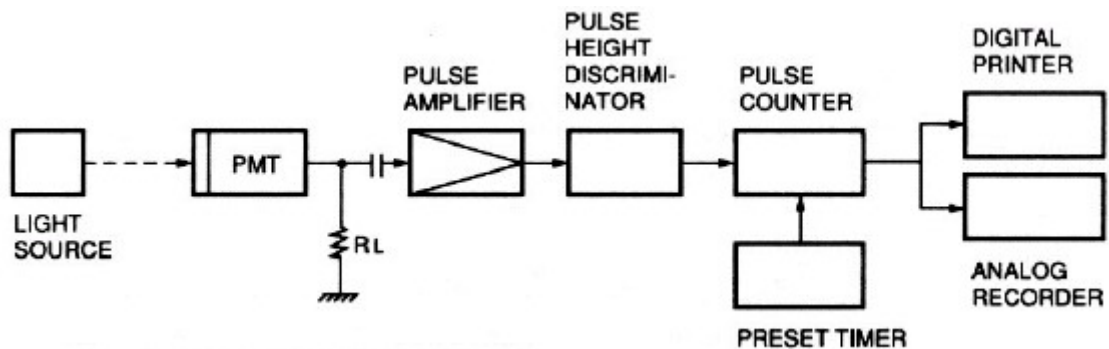


Figure 5.3.[99] – scheme of single photon counting method circuit

5.1.7. Noise in PMT, its sources, reduction and shielding

There are numerous reasons for a signal to appear even though there was no incident photon:

Causes of noise

1. Thermionic emission current from the photocathode and dynodes – can be reduced by cooling the PMT, what may however reduce sensitivity
2. Ionization of residual gas molecules by photoelectrons – may cause “afterpulsing”
3. Glass scintillation when electron hit the glass PMT envelope may be lowered by operating on lower voltage

4. Ohmic leakage – can cause constant dark current and worsened by dirt and humidity
5. Field emission current – can occur when electrons are pushed out of the photocathode, if the electric field is too high
6. Cosmic rays, radiation from radioisotopes contained in glass envelope or environmental gamma rays
7. Electronic noises

We can reduce the influence of strong magnetic fields by soft ferromagnetic materials in shielding(μ -metal) and using magnetic field resistant designs of PMT's, electric fields by grounding the coating of PMT.

5.1.8. Special photomultiplier types

Usual types of PMT's can detect single photons with time resolution in order from μ s to ns, while it was reported that experimental setups with fast read-out electronics can reach even hundreds of ps[102].

However, typical designs of photomultipliers don't allow position detection i.e. we can get information about the incident photon with high temporal resolution as mentioned above, but we do not know *where* the photon has hit the photocathode. To get this spatial information, special *Fine-mesh* and *MCP* (Micro Channel Plate) dynodes have been developed.

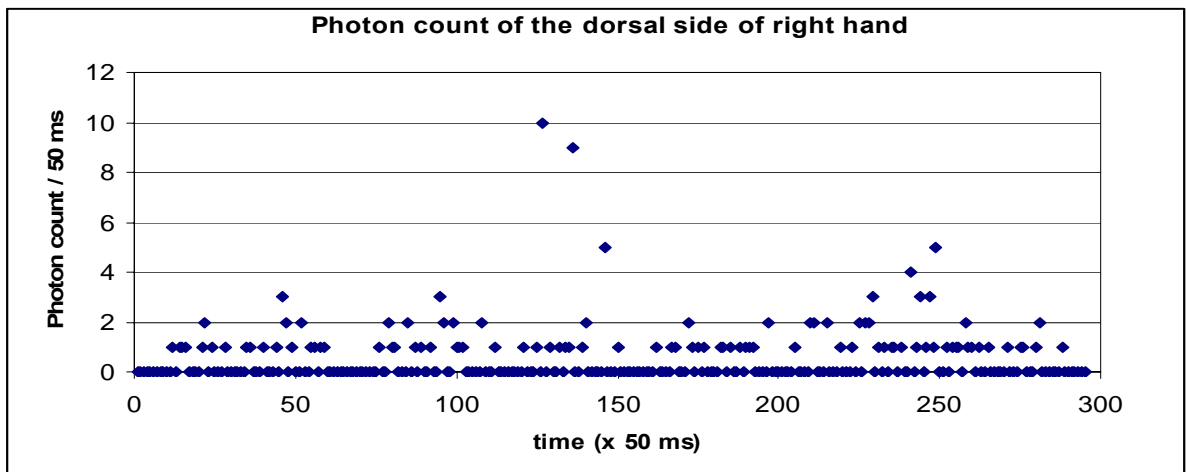


Figure 5.4. -Example output from photomultiplier measurement, single photon counting procedure. Notice the discrete values and length of the time interval (dwell time, gate time). Photomultiplier brings information about photon signal with high time resolution, however most PMT types lack possibility of giving the spatial information. Raw outputs are just long set of numbers that needs to be processed further.

5.2. CCD systems [100, 103, 104]

A charge-coupled device (CCD) is an array of metal-oxide-semiconductor (MOS) capacitors which can accumulate and store charge due to their capacitance. It is the core of ubiquitous digital cameras.

Photons knock out electrons which are stored in a potential well resulting by applied voltage. These charges can be shifted from one pixel to another pixel by digital pulses applied to the top plates (gates). In this way the charges can be transferred row by row to a serial output register.

Cameras where the light penetrates through the gate structure to reach the region where electrons are collected, are called front-illuminated. More sophisticated in the production, but with a higher sensitivity are cameras where the CCD chip is exposed from the opposite side. These cameras are called back-illuminated.

Advantages of CCD detectors are relatively high quantum efficiency reaching 70% to 90 %, and the fact that output brings spatial information about photon intensity in form of a picture.

The “shift-register style” of data read-out takes some time, usually tens of milliseconds or more, depending on CCD architecture. Therefore the main CCD disadvantage is their time resolution.

For measurements of low light intensity, what has been originally developed for astronomical observation, highly sensitive CCD camera system with a thinned back-illuminated CCD detector, cryogenically cooled to below -100 °C is suitable. The readout rates are restricted in order to reduce the readout noise, occurring in the buffer amplifier built into the CCD chip. Thus this type of camera is referred to as a slow-scan CCD camera. Although the slow-scan limits the time resolution of a measurement to the order of tens of minutes, this is not insignificant for biophoton imaging, considering the integration time.



Figure 5.5 .- output from CCD system:

on the left: hands under weak illumination

on the right: emission of photons from human hands integrated for 15 minutes

5.3. Other measurement systems [100]

Beside the commonly used photomultipliers and cryogenically cooled CCD cameras, there are other systems that can be used for measurement of ultra low light levels. Hybrid photodiodes, Avalanche photodiodes, Visible Light Photon Counters, Solid State Photomultipliers, Quantum Dots and their combinations are few of them.

However, most promising are Superconducting Tunnel Junctions. The types suitable for measurement of ultraweak light are not commonly used due to problematic cooling to extreme cryogenic temperatures (around 1 K). They bring all features that an ideal detector should have:

- photon counting/timing
- wavelength measurement
- position information

Superconducting Tunnel Junction is made of two tiny films of superconducting metal separated by a thin insulating layer. When operated below superconductor's critical temperature (below 1 K), equilibrium is easily perturbed by visible photon that carry well enough energy to break Cooper pairs that make up superconducting state. The greater the energy of absorbed photon the more of these pairs are broken up. By measuring the total charge, we could get the information about photon wavelength, what with connection with high quantum efficiency and spatial information could make breakthrough in all fields where ultra low level of light needs to be detected.

6. Overview of human biophoton research[2]

Although photon emission from human samples has been studied since the 1930s, when Gurwitsch's discovery of mitogenetic radiation became better known, first studies dealing with human photon emission from intact human body were published in the middle of the seventies of 20th century. Since then large number of studies were carried out, to enlighten the behavior of this phenomenon, to find correlation with internal and external physical and physiological parameters and to develop possible application of measurements of human photon emission. A brief overview of photon emission research of human body samples as well as intact human body is given.

6.1. Samples from human body

6.1.1. Non-diluted blood [105]

Photon emission from non-diluted blood stabilized by heparin or sodium citrate was studied. Photon emission was amplified by standard probes for superoxide radical (lucigenin) or less specific probe luminal for a variety of reactive oxygen species (H_2O_2 , ClO^- , OH^\bullet , etc.) It was found that:

- oxygen supply to blood increases the photon emission
- addition of substances that stimulate immune reaction(zymosan), induced respiratory burst by neutrophils and significantly increased the photon emission
- addition of 0,5 % of free hemoglobin to undiluted blood quenches the photon emission significantly, while the concentration of hemoglobin packed in erythrocytes reaches 35-40% and doesn't quench the photon emission
- dependency of photon emission on temperature is highly non-linear

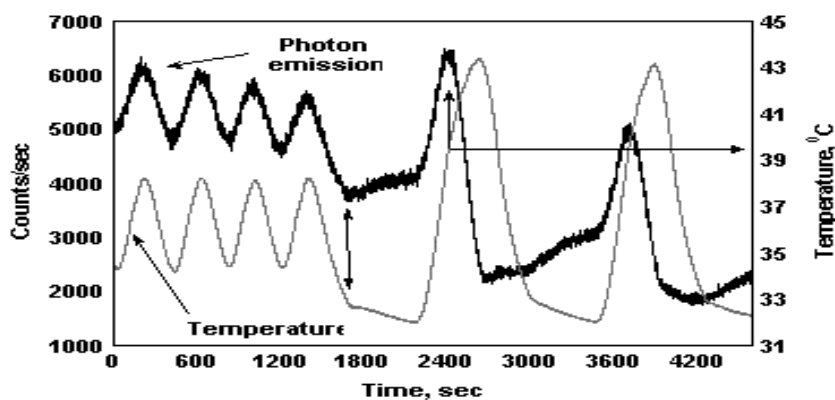


Figure 6.1. [105]– Nonlinear dependence of blood PE on temperature is obvious from this graph. It is worthwhile to note, that in the above displayed temperature interval no blood denaturation took place.

- back reflection of the photons changes the pattern of photon emission of blood sample

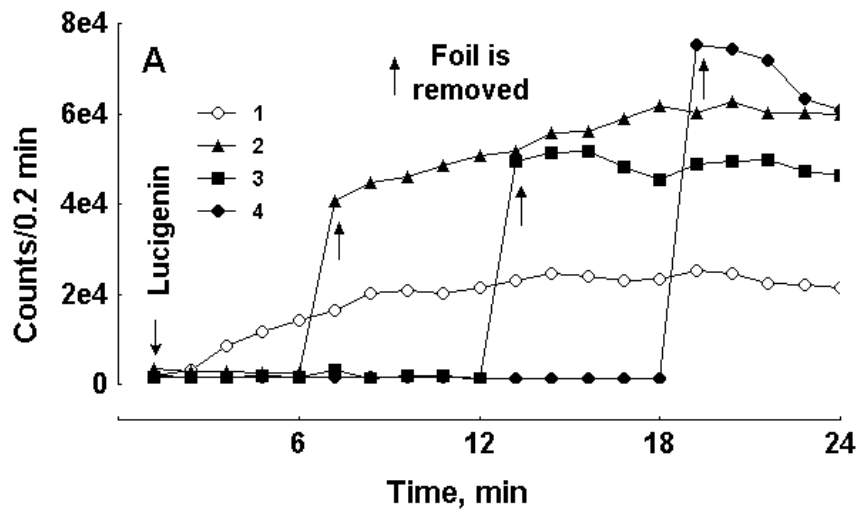


Figure 6.2.[105] - Effect of aluminum foil screen on lucigenin photon emission development in preparations of blood. Lucigenin is standard probe for presence of superoxid radical. Curve 1 – control test tube, curves 2-4 -- test tubes were wrapped with foil before lucigenin addition. Foil was removed from respective test tubes at moments indicated by arrows. In each experiment a preparation of blood was distributed in equal portions among 4 samples, which were counted in the mode of rotation. Back reflected photons seem to inhibit activity of neutrophils in production of superoxid radical

- self-irradiation of blood may reactivate respiratory burst in it especially at the stage of PE decline

6.1.2. Blood plasma

- [108, 110] increased photon emission was found from blood plasma of:
 - o smokers
 - o persons with certain diseases connected with increased LDL (low density lipoproteins) blood levels
 - o persons who had previously eaten meal/substantial meal
 - o elder persons
- comprehensive review in [108] mentions among other higher photon emission from plasma of bone cancer patients, alcohol liver injuries patients
- [112] different spectral distribution has been observed in photon emission of plasma of hemodialysed patients and photon emission intensity was higher [111] compared to the spectrum of healthy individuals, after luminescence induction by hydrogen peroxide and hydrogen radicals

- [111] blood plasma samples from patients with malignant tumor in liver or biliary tract, as well as show generally higher intensity than in healthy cases
- [113] increased photon emission from heated blood plasma samples of cancer patients and pregnant women has been found, compared to healthy patients as control subjects.

6.1.3. Cell suspensions

- under certain conditions which include lack of agitation of a suspension, an optimal buffered medium containing nutrients and access to air, neutrophils are able to develop oscillating behavior of photon emission initiated by immune reaction stimulating substance [105]
- leukocyte suspension irradiated by $0,03 \text{ mW/cm}^2$ electromagnetic radiation of certain, depending on the species, extra high frequency(42,19; 46,84; 53,53 GHz) for 30 min showed increased photon emission compared to the control sample[106]
- tumor Wish cells photon re-emission after illumination was found to be decreasing with cell density whereas normal amnion cells showed increasing photon re-emission[114]

6.1.4. Tissues

- samples of cancer, cancer-edge and healthy tissue from 23 patients have NOT shown any significant difference in photon emission. Always the comparison between the PE from healthy and from cancer tissue within one patient (the PE from his tumour tissue has been compared to PE of his healthy tissue) has been carried out. Only spontaneous emission was measured [107].
- samples of cancer tissue showed generally higher photon emission, or induced photon emission than the control samples from healthy tissue(tumour tissue PE of one patient has been compared with healthy tissue from other non-case). However intensities for some cases from control and cancer tissue were overlapping. Spontaneous emission and delayed luminescence were measured [109]

6.1.5. Breath

- intensity of breath sample photon emission increased immediately after the start of physical exercise and dropped down swiftly after end of the exercise[111]

6.1.6. Urine

- photon emission(PE) from urine samples from patients with inflammatory or necrotic diseases tends to be higher compared to the urine samples PE from healthy individuals[111]

It is generally valid that contact of biological samples with oxidative substances (even the amount that is naturally present in fresh air) increases the intensity of their photon emission. Inert atmosphere (argon, nitrogen, lack of air supply) inhibits photon emission.

6.2. Intact human body [2]

Research on photon emission from the intact human body could only be carried out when the measurements systems had evolved enough to detect very low intensity of photon emission. Technical prerequisites for this appeared thirty years ago. First research groups were very enthusiastic to find slightly higher photon counts than the intrinsic noise(dark count, background) of the measurement system when they pointed photomultiplier on the part of the human body.

To measure photon emission from any part of human body, a special dark room or dark chamber must be constructed, to avoid any ambient light from masking photon emission, since we can not distinguish which photon comes from the body, which is from ambient light or which are actually false impulses from photomultiplier noise (described in the previous chapter, *5.1.7. Noise in PMT, its sources, reduction and shielding*). Noise fluctuations of measurement devices combined with low signal/noise ratio as well as impossibility to filter out the noise from the signal represent the biggest obstacles of human biophoton research.

Most of the research of human biophoton emission is oriented on finding possible diagnostic application of these measurements.

6.2.1. Basic parameters of human photon emission

Photon emission from intact human body could be named photon emission from the skin, since we do not have probes small enough to observe photon emission from internal structures of human body. However one should not be confused with this name: the origin of human photon emission may not be only in the top layers of the skin.

Human photon emission is of ultra-low intensity in the order of single to tens of photons per centimeter squared per second. Since quantum efficiencies of state-of-art photomultiplier systems and cooled CCD systems reach on their peak spectral sensitivity 30% and 70%, respectively, we usually detect net photon counts of few to some tens of photons per second per whole sensitive area of device, what is commonly few tens of squared centimeters. The experiment in [115] shows that matching the refractive index of transmission media(usually air) from hand to PMT input window refractive index by using mineral oil or water leads to as much as two fold increase in detected photon counts by lowering the number of reflected photons.

Due to the ultra-low human photon emission intensity its spectral distribution can be practically measured only by sharp cut-off optical filters.

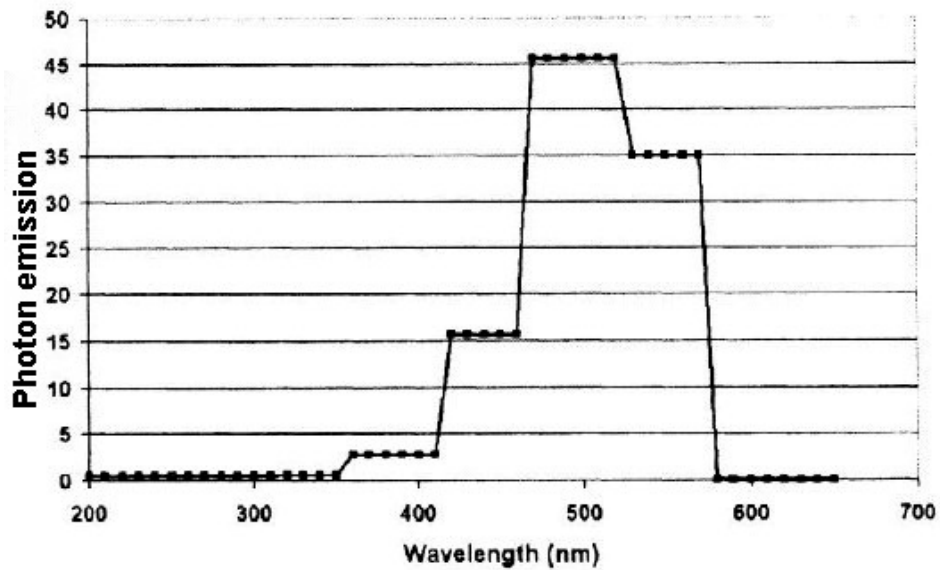


Figure 6.3. [80] – example spectral distribution of spontaneous emission of the hand was achieved by computing the average counts for a standardized wavelength range, followed by a mathematical correction for the average sensitivity of recording in each wavelength range. Photomultiplier tube EM9235QB was used

Like in all organisms, both spontaneous photon emission as well as the delayed luminescence is present in human body. Delayed luminescence can be best approximated by hyperbolic decay, what points to cooperative behavior of radiating system.

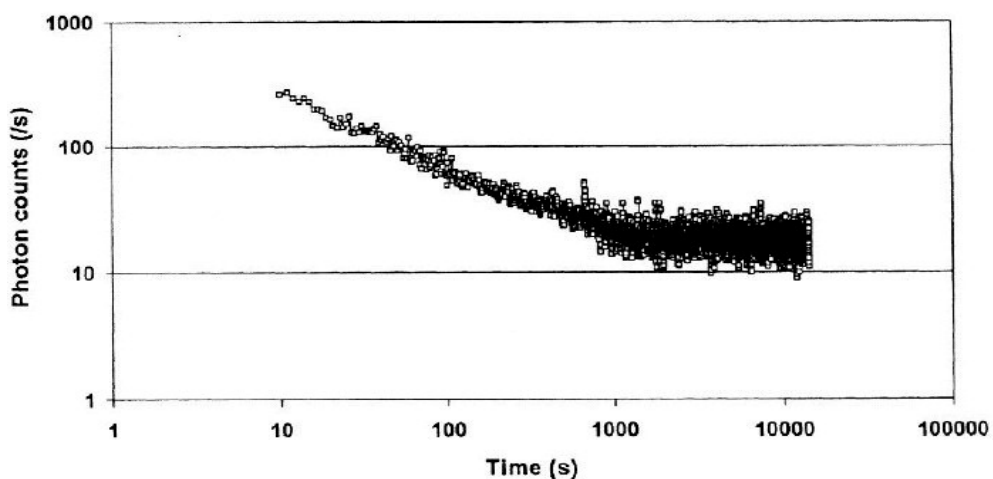


Figure 6.4. [80] – example delayed luminescence of the hand induced by strong artificial sunlight. This fact implies a need to avoid UV rich sunlight two hours and one hour of dark accommodation prior to measurement of spontaneous emission (SE) in order to avoid any delayed luminescence in SE measurement. Hyperbolic decay in $\log x - \log y$ scale is visible as linear decrease

6.2.2 Temperature and oxygen dependency

Temperature and oxygen dependence of photon emission from intact human body has been studied in several groups. Parameters of the environment (in most cases air) as temperature, oxygen content and moisture can be relatively easily and independently controlled and changed. However, although we can influence parameters as skin temperature and oxygen supply in intact human body, we cannot precisely and independently control them. This is caused by the fact that temperature as well as oxygen content in skin is indivisibly connected to blood perfusion.

- it has been found that changes of skin temperature of hand positively correlate with changes of photon emission intensity of hand after externally induced changes of skin temperature [115]
- higher photon emission intensity of hands has been observed when surrounded by oxygen gas, and a ca. by 40% lower emission intensity could be observed when surrounded by nitrogen gas compared to normal air [115]
- [116] conclusion from measurement of 5 subjects - the cuff applied on the upper part of the arm and inflated subsequently to higher and higher pressure (under diastolic > over the diastolic > over the systolic) caused restriction of blood supply. With restricted blood supply, the arm's temperature as well as its photon emission decreased; when a certain level was reached, photon emission stabilized whereas temperature was further decreasing
- no positive correlation between distribution of surface temperature and biophoton emission was found [117]

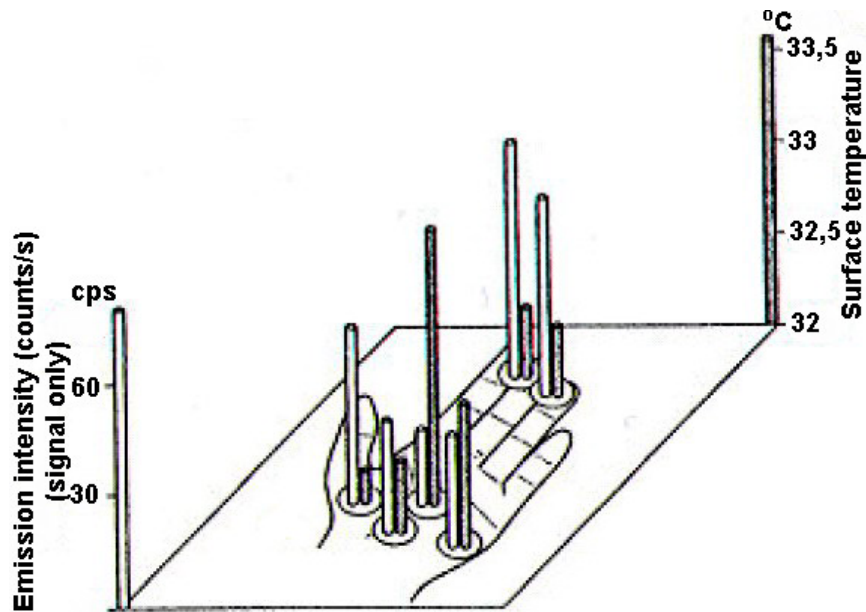


Figure 6.5. – distribution of biophoton emission intensity and surface temperature on the left palm of a healthy male 34 years old subject [1]

- lowering of the PE with the increase of temperature of the same point was reported in [117] in reproduced trials by treatment of moxibustion(traditional Chinese medicine healing technique) compared to the control where photon emission was increasing with increasing temperature
- in one subject however reproducible rise of photon emission while decrease of temperature has been observed while actively performing Qi-Gong (traditional Chinese medicine healing and health maintaining technique)[118]

6.2.3. Spatial distribution of photon emission

There is no clear spatial correlation with temperature of surface skin temperature and photon emission, as can be seen on *Figure 6.5.*, but there is more or less typical pattern of photon intensity of human body.

- it has been found and is known in human biophoton research that generally more structured parts(i.e. hands compared to other parts of arm[118]) emit more photons than less structured, flat parts of body, i.e. abdomen

- interesting paper [119] shows and quantifies anatomic characterization of human ultra-weak photon emission from 12 body locations of 20 healthy subjects with special cryogenically cooled CCD system and movable photomultiplier. Cheeks, palms

and the neck has been found the most contributive to the total emission of the 12 observed locations

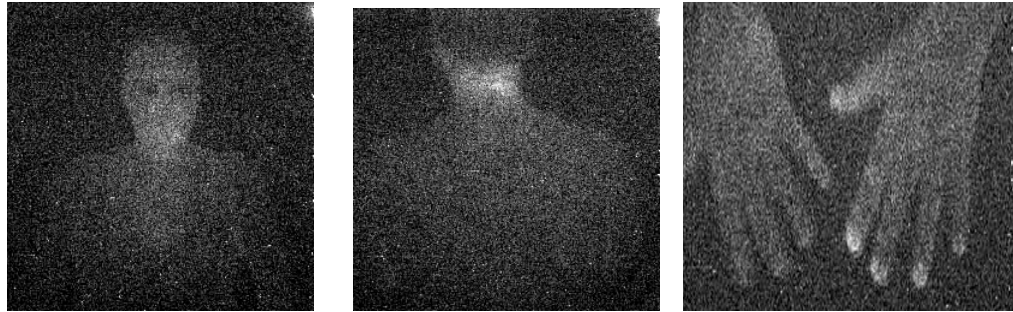


Figure 6.6. – shows human photon emission intensity distribution as detected with special cryogenically cooled CCD camera. From left to right: front of the upper torso and face, back of the upper torso and neck, palm and back of the hand

- right-left symmetry of photon emission intensity is observed in healthy cases[120]
- there is a tendency for higher PE from dominant hand than non-dominant hand
- significantly higher photon intensity is observed from nails, but not from fingertips [128]. There are speculations about this considering refractive index of nails as the cause

6.2.4. Health and disease

There are several papers that describe deviation of photon emission parameters of subjects in the state of disease or injury. The potential usefulness of biophoton emission measurements for non-invasive diagnostic purposes is actively being investigated.

- fresh scratch on the skin as well as internal bleeding under the nail are manifested by higher emission intensity, compared to the healthy sites[117]

- the two-dimensional pattern from the index and middle fingers was used to differentiate hypothyroidism, a lower state of metabolic activity than normal. Biophoton emission in patients with hypothyroidism or patients after thyroidectomy was always less intense than control cases [123]

- it has been reported in [124] that patient in progressive stage of multiple sclerosis shows high right-left asymmetry as well as 10-20 higher photon emission from hands than is usual in healthy subjects

- higher asymmetry in the patients suffering by cold is observed in [125], and symmetry tends to restore with recovery

- [120] - large right-left asymmetries have been found in patients with hemiparesis, compared to the healthy cases. After acupuncture treatment symmetry recovered to the level of healthy cases

6.2.4. Biological rhythms

Intensity of biophoton emission is specific for individual, may vary even 5-fold between the healthy subjects. However, photon emission varies even in the same subject spatially as described in 6.2.2. *Spatial distribution of photon emission* as well as in time. Until now, mainly only long term rhythms and changes have been observed.

- in [118] is, apart from other facts, that human photon emission is higher in summer than in winter
- authors of [126, 127] which observed biophoton emission and delayed luminescence from hands and forehead of one subject for several months and found weekly, monthly and several other periodicities by Fourier analysis. Apart from that, correlation of left-right hand emission has been observed, while the forehead showed anticorrelation to hands
- in Korean paper [122] has been shown that photon emission from palms and dorsal sides of the hands of 3 subject varies during the year(measured once a week), with lowest emission during September. Greater fluctuations has been found on dorsal side of the hand than on the palm
- photon emission intensity from 29 body sites was found to be slightly higher in afternoon than in the morning [121]

7. Temperature and time dependent fluctuations of spontaneous human photon emission

It can be seen in the brief overview of human biophoton research that photon emission varies not only spatially (different emission in different anatomic location) but also temporally (emission changes in time). It has been found that under no direct external influence photon emission doesn't vary rapidly in time scale of minutes and already few minutes long measurement gives reasonable value of photon emission intensity. But what basic factors, like temperature for example, influence fluctuations of photon emission? How fast can these changes occur? If we want to find the influence of more complex factors on biophoton emission, we need to know the influence of standard physiological parameters on photon emission and its dynamics at first. We need to have the knowledge of dynamics, especially to explain more complex effects that may be either linear or non-linear composition of simple changes of basic parameters, and to exclude the possibility of false direct association of changes in photon emission to some complex effect, when the actual cause may be just a change of basic parameter. An example of a direct association of photon emission to a complex effect can be the association of increased photon emission intensity from hands to increased physical activity. Because it is generally considered to be valid, that photon emission rises with physical activity (complex effect), one should be aware of the enhanced blood perfusion in hand if one observes increased emission.

Intensity of spontaneous photon emission is not the only parameter that can be utilized to characterize photon emission. One can utilize, additionally, photon count statistics parameters or even delayed luminescence and its decay kinetics, to detect changes in the dynamics and nature of the photon emission.

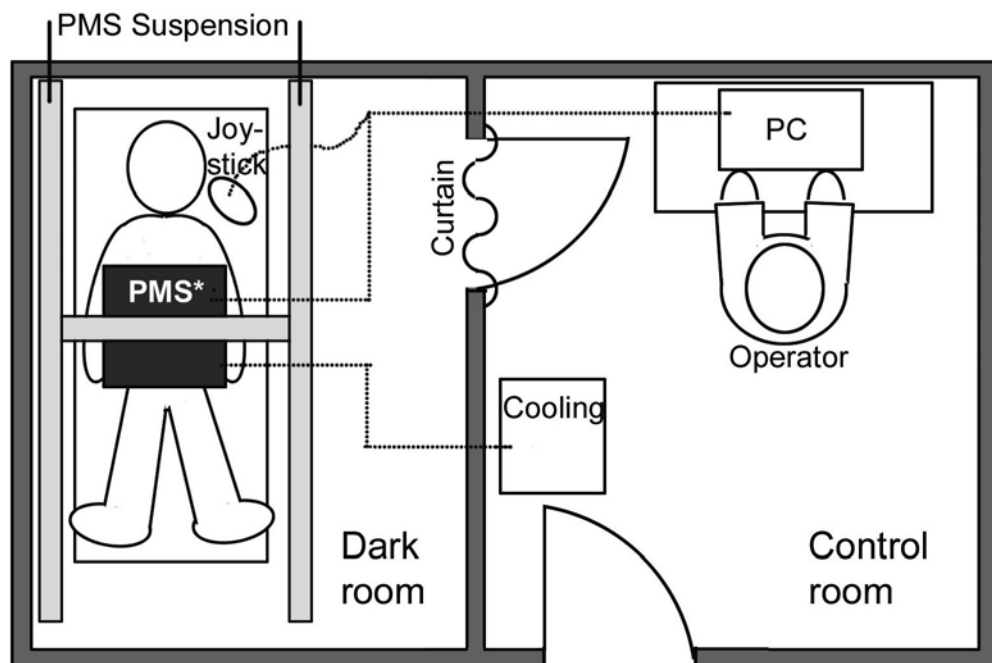
The main aim of the experimental part of this work is:

- to study the fluctuations in spontaneous photon emission from human subjects in the order of hours, recording the photon emission in regular time intervals during day and night
- to correlate these fluctuations with skin surface temperature of the measured locations
- to study the kinetics of temperature induced changes on photon emission
- to record photon emission and skin surface temperature of various locations of the human body

7.1. Measurement system

A special darkroom (2m x 2m x 1m) and photon counting device are used to record photon emission of the subjects. Walls and ceiling of that darkroom are covered with black matt paint. The temperature of the dark room was registered before and after every set of measurement. During longer stays (more than 1-2 hours) of the subject in the darkroom air temperature increased slightly (max. 2 degrees °C). The temperature of the dark room was commonly 20 °C. The dark room could be vented. The darkroom had a bed inside and subjects could be easily measured in lying or sitting position. The darkroom was built inside a control room without windows, and separated by a light tight door and black matt curtain to ensure that there is no light leakage to dark room. The sufficiency of these measures is easily verifiable by the control measurements with the PMT shutter closed and opened, when there is subject in the room. It can be seen that

The control room contains the electrical equipment (high voltage supply for PMT, PMT shutter and steering system), active air moisture filter, cooling unit for PMT and a remote computer system for the photomultiplier as well as. Control room is illuminated by red light.



* Photomultiplier System

Figure 7.1. –schematic depiction of measurement rooms (courtesy of Heike Koch)

The photomultiplier (EMI 9235 QB, selected type) used could be manipulated in 3 directions. It is a 52-mm diameter, especially selected, low noise, end window photomultiplier that was mounted in a sealed housing under vacuum with a quartz window. An additional ring at the front of the photomultiplier tube allowed measuring a 9-cm-diameter area at a fixed distance from the body (70 mm). The photomultiplier has a spectral sensitivity range of 200–650 nm. It is maintained at a low temperature of -25°C in order to reduce the dark current (electronic noise). Typical dark current (electronic noise) under these conditions is ca. 5 counts per second (cps). The photomultiplier is protected from the light by the automatic shutter that opens and closes on the beginning and on the end of the measurement, respectively. It even closes by opening the door during the measurement to prevent damage to photomultiplier tube from bright light.

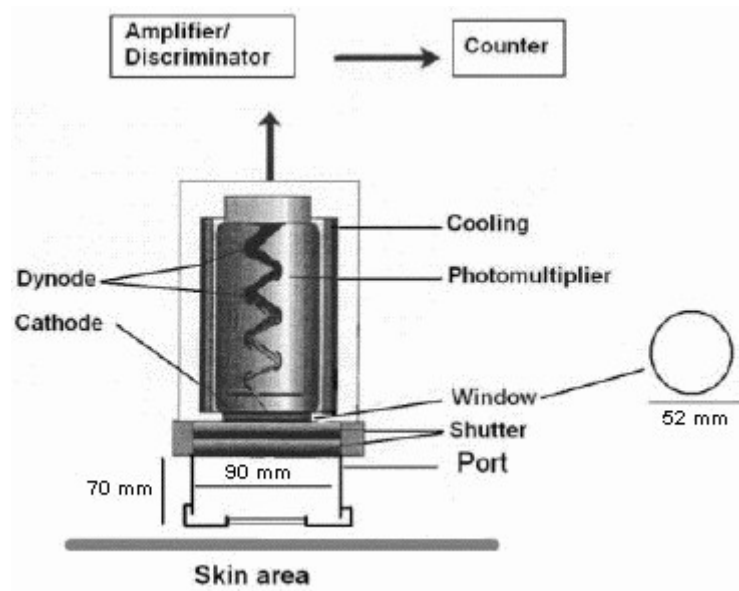


Figure 7.2. – dimensions of the PMT

Temperature measurements were carried out with a thermocouple thermometer with red LCD display and sensitivity of $0,1^{\circ}\text{C}$. The sensitive thermocouple end was gently pushed to skin by small, flat, smooth polystyrene cube to ensure reproducibility of the measurement.

7.2. Measurement software and basic measurement settings

The PC software PMS5 runs under DOS environment and has simple graphic interface. It is functional enough to efficiently manipulate with photomultiplier measurement system. Apart from the advanced settings, which description is beyond the scope of this work, it offers the setting of these parameters:

- file name, file number of the stored data file
- number of the shot, length of the shot. Shot is basic unit of the measurement. Depending on the user choice, 1-99 shots can be in one file. Length of the shot is basically how long will be the column of the one shot.
- gate time (bin size, dwell time in other literature) determines the time interval of photon counting, actually the time resolution, i.e. length of the shot: 3600, gate time: 50 ms will result in $3600 \times 50 \text{ ms} = 180 \text{ s}$ (3 min) long measurement. The output will be column of 3600 rows, each containing number of the photons counted in 50 ms intervals
- program for either spontaneous emission or delayed luminescence measurement. Under delayed luminescence setting, length of the light stimulation, delay time of starting the measurement after the light excitation, etc.

There are many more settings concerning the moving of photomultiplier, automatic settings, etc.

The raw data are saved to computer hard disk as “*.CND” files, the heading of the data containing all settings and time information about the stored data are in the corresponding “*.CNP” files. The “*.CND” files contain always three columns. Lengths of these columns depend on the length of the stored shots and number of the shots in the file. First column denotes the number of the shot, second the number of the time interval the shot and third shows actual number of photons detected in this time interval.

7.3. Measurement protocols

There were several protocols defined, one for each type of the measurements. They include rules, timing and settings for the measurement that had to be followed and kept to gain comparable set of data.

There are a few rules that are common to all of them:

- dark accommodation for at least 1 hour prior to measurement. It means to keep measured sites on body in low (preferably red) light condition to eliminate delayed luminescence. Practically it was achieved by wearing gloves on hands, long sleeves or staying in the dim red light of control room. Dark accommodation includes 10 minute long stay in a dark room in the same position as the measurement was carried out to avoid some abrupt changes in photon emission due to change of body position, i.e. from standing position to laying position.

- the “dark count” is controlled before and after every measurement to ensure the basic counting capability of PMT system and in order to have approximate dark count value to subtract it later from measured counts in data analysis to gain net value of photon emission. “Background” (measurement of empty darkroom) was performed occasionally in order to control if there isn’t any light leakage due to lowly probable but possible damage of door light tightening functionality. If it would be found that background is unusually higher than dark count of PMT, it would mean that there is either leakage of light or unwanted source of light in dark room.

7.3.1. „24 hours“ measurement protocol

Subject is measured every two hours in time span of 24 hours (even during the night). The protocol had few versions, but majority of measurements has been done on palms and dorsal (back) sides of both hands, few measurements on forehead and neck, too

When it was observed by sample temperature measurement that photon emission of hand seems to follow temperature changes of hand, systematic measurements of the body temperature had been started.

7.3.2. Induced cooling measurement protocol

Temperature changes have been induced by external cooling. In the first part, dark accommodated palm and dorsal side of the hand have been measured to obtain the baseline value of photon emission as well as temperature under the stable temperature conditions. In the second part, palm sized ice cube covered with thin plastic to avoid the skin moistening during contact was used to cool down the palm while the photon emission of dorsal side of the hand was measured. In the third part, the ice cube was removed to allow recovery of the temperature. The photon emission and temperature directly after the removal of the ice as well as during the recovery have been measured.

7.3.3. Whole body spontaneous cooling measurement protocol

In order to find more information about the dependence of the photon emission of various anatomic sites on skin surface temperature, the photon emissions as well as surface skin temperature of 12 spots of the body have been measured in two cycle measurement.

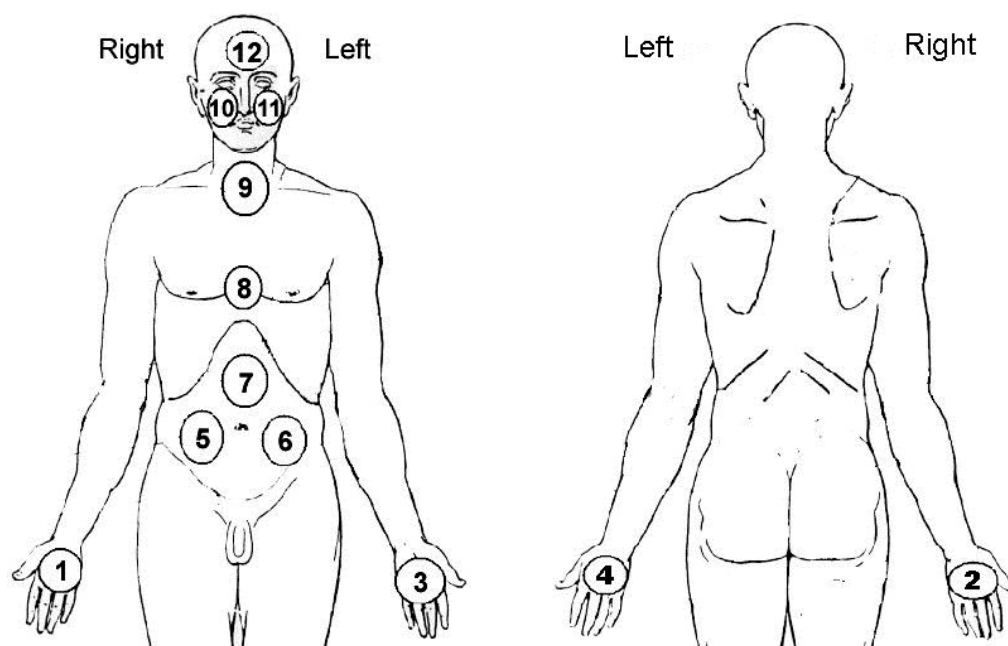


Figure 7.3. – depiction of the measured sites, the numbers are showing the order in which they are measured in every 12 spot measurement cycle

1	2	3	4	5	6	7	8	9	10	11	12
Hand (palm) right	Hand (dorsal) right	Hand (palm) left	Hand (dorsal) left	Abdomen right	Abdomen left	Solar plexus	Heart	Throat	Cheek right	Cheek left	Forehead

Table 7.1. – table showing number of anatomic location and its code name

In the first measurement cycle, dark accommodated subject wearing more layers of clothing to keep the stable body temperature was lying on the bed in the dark room and had the temperature as well as the photon emission of the all twelve depicted spots measured. After the subject removed the upper layers of clothing, keeping just the minimum the subject spontaneously cooled down in the following period 30 min, depending on room temperature. Temperature of the dark room air was measured before and after every 12 spots measurement cycle to ensure that it is low enough (varied from 19-21 °C) to cause the cooling and thermoregulation changes. After the 30 min of the spontaneous cooling, the second 12 spots measurement cycle of photon emission and temperature has been carried out.

7.4. Data analysis – Results

Since the number of the stored data in shots is usually reaching thousands of counts in one column, automatic preprocessing or so-called “cutting” of these columns into the single shot columns considerably reduces amount of manual work. Scripts written in Python programming language were used for this preprocessing and for basic analysis (counting of average values, joining single time intervals into bigger ones – to gain longer gate times from the shorter ones) as well as the fast fourier analysis, plotting and saving the figures. Variety of relatively simple processing scripts has been written for this purpose.

Microsoft Excel 2003 has been used for more advanced analysis of data sheets as well as creating the graphs and charts.

7.4.1. “24 hours” measurements

While there are several papers describing extra long term (weeks, months, year) changes of the human photon emission, there is a lack of published papers in English dealing with the systematic observation of changes of photon emission during the day with high and regular sampling interval because they are highly time consuming and connected with several practical difficulties. One paper [121] describes on 4 subjects that the PE from all of 29 measured sites tends to rise in the afternoon, actual increase vary between individuals. Do we know, however, if the emission only simply rises towards the evening and then decreases in the night or are there any greater fluctuations? Bringing more information on this issue was the main aim of the measurements of “24 hours” protocol.

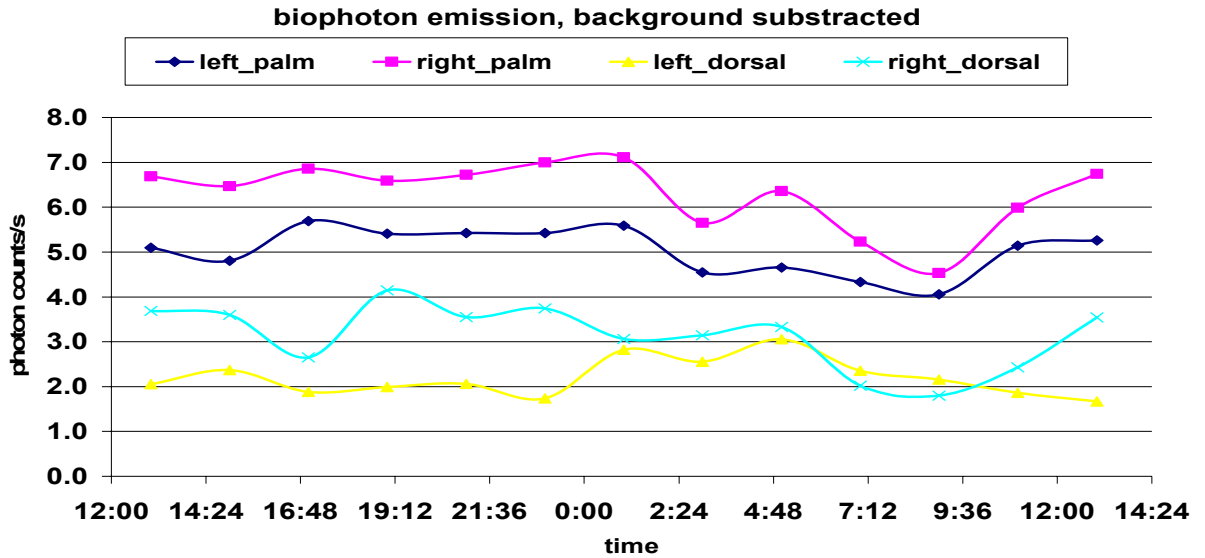


Figure 7.5. – example of 24 hour long measurement. Every two hours were left, right palm as well as the dorsal side (back) of the left and right hand measured. The subject has his right hand dominant, depicted measurement shows higher photon emission from the right hand, what confirms already known fact of tendency for higher human photon emission from dominant hand.

These “24 hours” measurements were carried out in winter period (January) and keeping the protocol as described in 7.3.1. “24 hours” measurement protocol.

To quantify the photon emission fluctuations, evaluation of all “24 hours” and whole day measurements that fulfilled given conditions has been done. For all measured spots (mostly palms, back of the hands, few times even forehead and neck), *difference between the highest and lowest measured value* (averaged from every two hours measured photon counts) within every “24 hours” measurement, as well as the relative difference((maximal value-minimal value)/average)), and the standard deviation has been calculated. The following table is output from large calculation sheet:

MAX	8.327			3.531			3.101			1.858		
MIN	1.636			1.389			1.171			0.777		
AVG	3.579			2.429			2.325			1.350		
MAX		1.110			1.149			0.644			0.393	
MIN		0.241			0.504			0.250			0.205	
AVG		0.628			0.793			0.458			0.310	
MAX			1.858			1.367			0.971			0.590
MIN			0.462			0.418			0.365			0.261
AVG			1.068			0.761			0.703			0.433
	1	2	3	1	2	3	1	2	3	1	2	3
	PALMS (20*)			BACK OF HANDS (10*)			FOREHEAD (3*)			NECK (3*)		

Table 7.2.

* - number of “24 hours” and whole day measurements performed on given anatomic location, i.e. PALMS(20*) means that left and right palm were measured in 10 “24 hours” or whole day measurements

1 – the *difference between maximal and minimal measured value* of photon emission of given anatomic location within one “24 hours” measurement, in photons/s

2 – *relative difference* – difference(described in 1) divided by average emission of given anatomic location within “24 hours” measurement

3 – *standard deviation* of “24 hours” measurement measurement

MAX – maximal value of given parameter (1, 2 or 3) from all measurements of given anatomic locations

MIN - minimal value of given parameter (1, 2 or 3) from all measurements of given anatomic locations

AVG – average value of given parameter (1, 2 or 3) from all measurements of given anatomic locations

Example for comprehension of meaning of values in *Table 7.2.*: Value **8.327** in the top left corner shows the *maximal (MAX) difference* in the PE intensity that ever occurred in one day(one „24 hours“ long measurement) on palm. It implies that in all other measurements it has never been observed that the PE from palm changed within 24 hours more than by 8,327 photons/s (averaged).

The next value **1.636** photons/s means, that in one low fluctuating „24 hour“ measurement, emission fluctuated only within this small interval, what is the *minimal(MIN)* observed PE intensity fluctuation in our measurements on palms ever. Finally the (*AVG*) means that the most PE intensity fluctuations on palms within measured interval (24 hours or whole day) were around the value **3.579**.

It is important to remind that the absolute values of the PE intensity fluctuations are intensity dependent, therefore is recommended to judge the fluctuability of PE intensity of particular anatomic location by *relative difference*, the values in the green color. What does this table actually tell us? It shows that although the highest fluctuations(at least from here measured points) in absolute values (photon counts) are usually on palms (due to their high PE intensity), relative fluctuability is highest on dorsal sides of the hands (back of the hand), whereas forehead and neck seems to be most stable in the PE intensity.

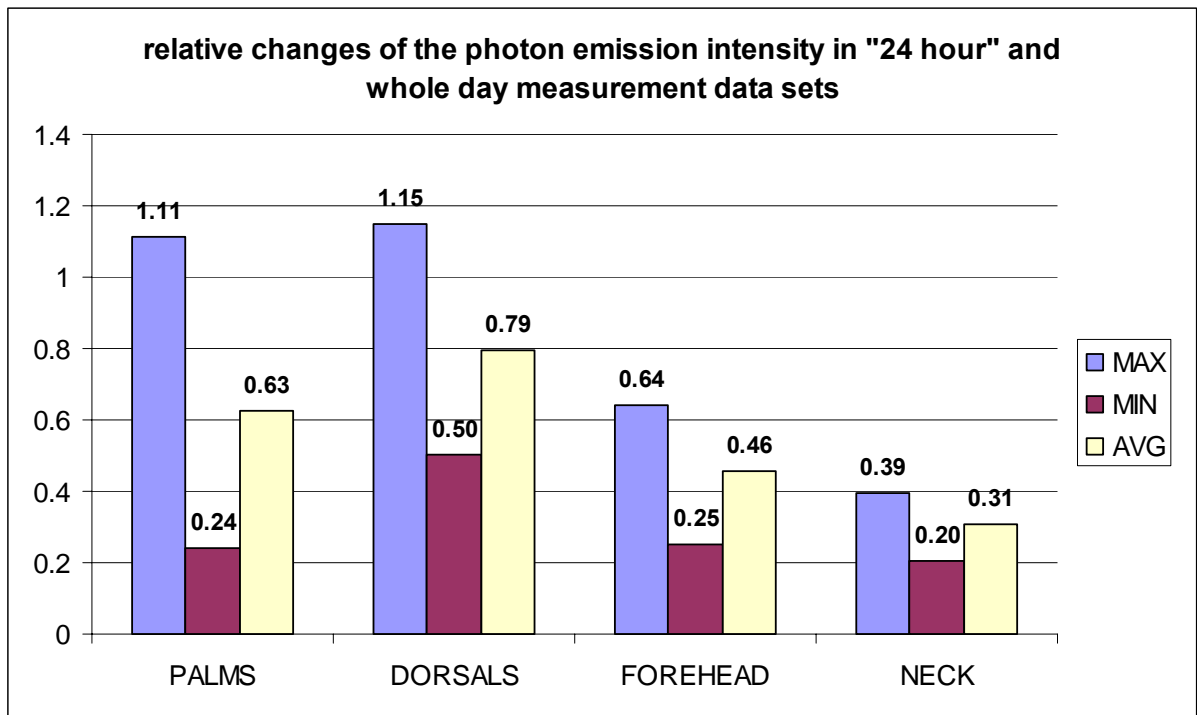


Figure 7.6. – depicts relative changes of the photon emission intensity in “24 hour” and whole day measurements. Columns in the figure represent green colored values from Table 7.2.

The amount of data from “24 hour” measurements provides possibility to calculate correlation between left and right hand PE intensity to rate the left-right symmetry of fluctuation changes:

Average correlation	Left palm	right hand - dorsal side
left hand – dorsal side	0.555±0.51 (high SD due to low S/N) (MAX=0.968,MIN=-0.341) *5	0.387±0.55 (high SD due to low S/N) (MAX=0.907,MIN= -0.294) *5
Right Palm	0.759±0.21 (MAX=0.951,MIN=0.195) *11	0.747±0.15 (MAX=0.899,MIN=0.716) *5

Table 7.3. – asterisk(*) informs about the number of measurements (24 hour or whole day measurements), where both location were measured. MAX and MIN indicate maximal and minimal correlation values in the set used to calculate average correlations which are in this table

It is important to note that values in the Table 7.3. are more or less illustrative, but give an idea about left-right as well as palm-dorsal hand side symmetry. Indeed, there is a trend of same course of changes of the left and the right side and of hand-dorsal hand side of PE intensity. However, as can be seen from MIN values in the Table 7.3., it doesn't necessary have to be so in all cases.

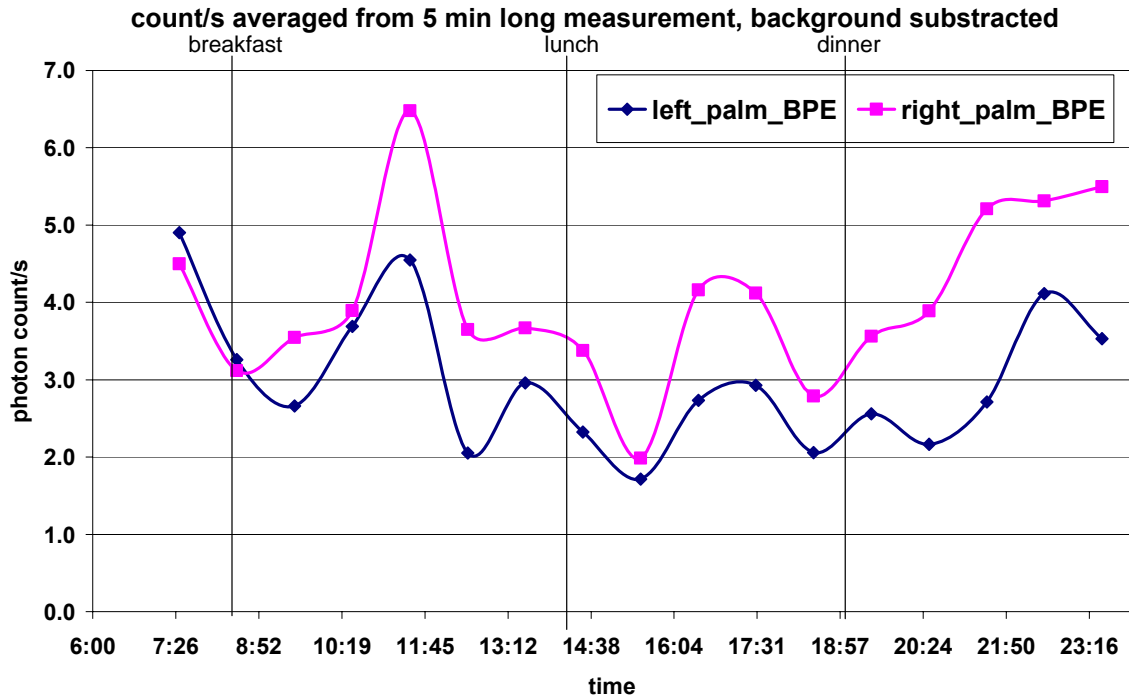


Figure 7.7. – one example of intensive whole day measurement (measurement interval 1 hour) showing the same trend of intensity changes for left and right palm. The correlation between left and right palm PE intensity is 0.711 in this case

The average pattern of daily PE intensity fluctuations of dorsal sides of the hands and the palms was calculated from five “24 hour” measurements of 3 subjects (2+2+1).

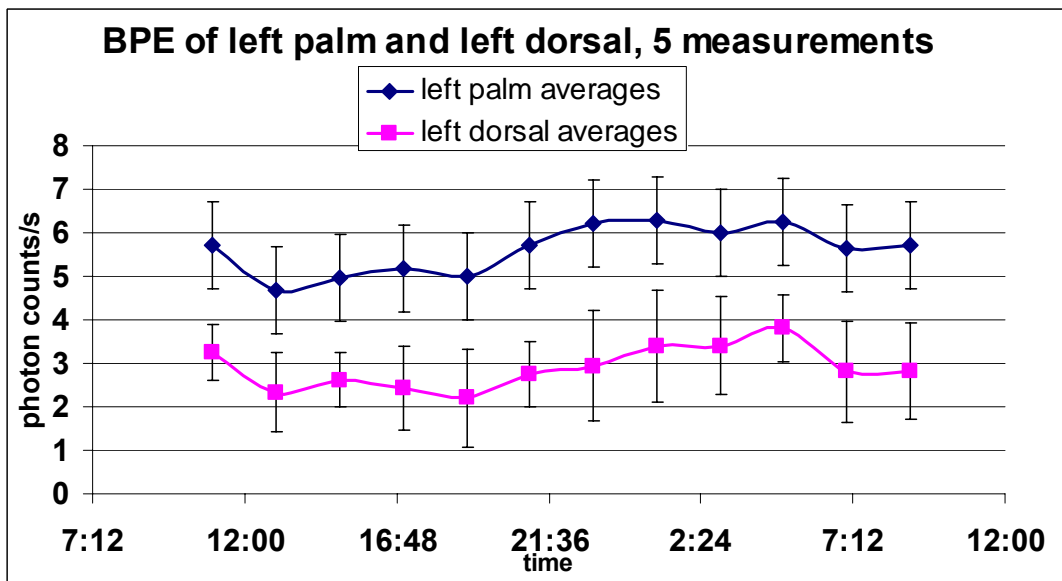


Figure 7.8. – The pattern of daily PE intensity fluctuations of the left palm and the dorsal side of the left hand calculated from 5 measurements of 3 subjects. Error bars depict standard deviation for each location and time point

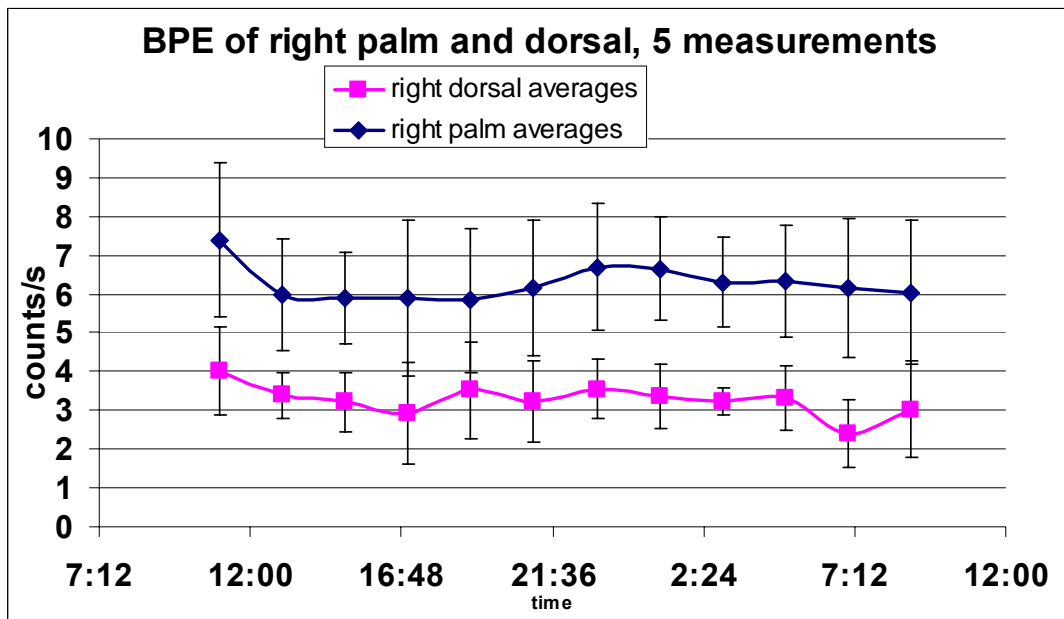


Figure 7.9. – The pattern of daily PE intensity fluctuations of the right palm and the dorsal side of the right hand calculated from 5 measurements of 3 subjects. Error bars depict standard deviation for each location and time point

In order to find out if there are any strong and regular rhythms of the PE in the order of hours, the Fourier analysis of all 24 hour or longer measurements have been performed. Commonly observed peaks were associated to circadian cycles (0,9-1,2 cycles/24 hours), another often appearing peak was assigned to rhythms in order of 6-8 hours (3-4 cycles/24 hours). However, the number of samples(12-26 samples/) used per analysis was relatively small and we cannot draw any deeper conclusions from this analysis.

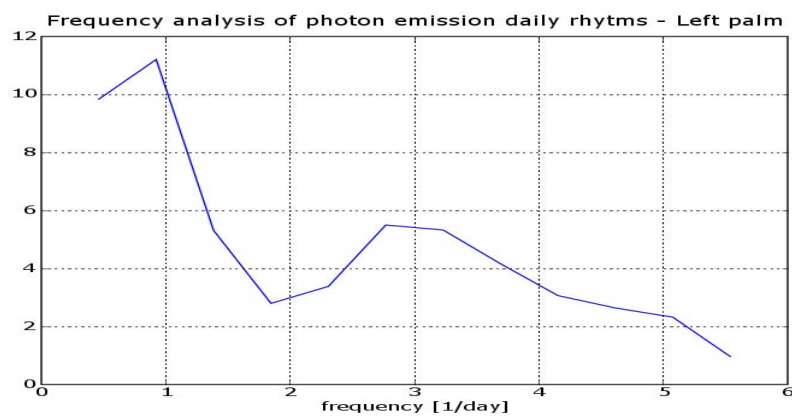


Figure 7.10. – shows Fourier analysis of photon emission of the left palm from one measurement. Number of samples was 26, sampling interval 2 hours(the measurement was spanning over 52 hours). X-axis shows frequency of cycles/day. Major peaks are visible near 1 cycle/day and 3 cycles/day

There have been several “24hour” or whole day long BPE measurements carried out, in parallel with temperature measurements. Temperature has been measured constantly before or/and after each PE measurement during the day. Most data about the PE and temperature come from the measurements of the palms.

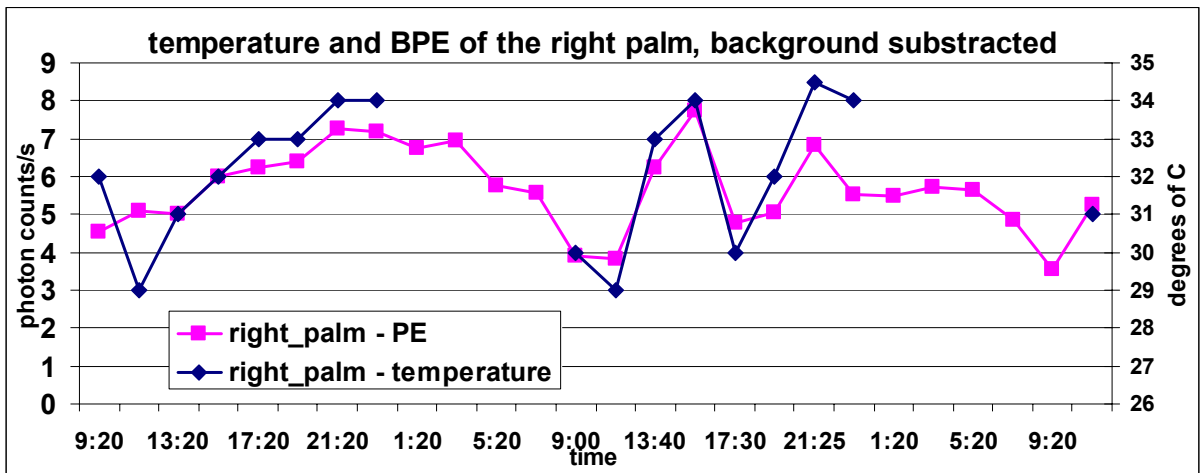


Figure 7.11. – example of correlating(0.834) temperature and the photon emission of the right palm over the interval of 50 hours long measurement. Temperature has not been measured during the night not to disturb the sleeping subject

The observation of the relationship of the PE intensity and the temperature of the palms is summarized in the following table. Since there is not enough “24 hour” or whole day measurements of the photon emission AND the temperature from other locations, they are not published here.

Average correlation	Left palm PE	Right palm PE
Temperature of given location	0.548±0.23 (MAX=0.843,MIN=0.213) *6	0.525±0.26 (MAX=0.834,MIN=0.198) *6

Table 7.4. – asterisk () informs about the number of measurements (24 hour or whole day measurements), where both PE and temperature of the given location were measured and used for correlation. MAX and MIN indicate maximal and minimal correlation values in the set used to calculate average correlations which are in this table*

Positive but not very strong average correlation of the PE and temperature of the palm has been found. It is necessary to note that correlation does not imply causation; however it may point to direct or indirect relationship.

7.4.2. Induced cooling measurements

In order to verify the influence of temperature and thermoregulation reactions of the body on changes of temperature, measurements of photon emission and skin surface temperature have been carried out according to the protocol described in 7.3.2. *Induced cooling measurement protocol.*

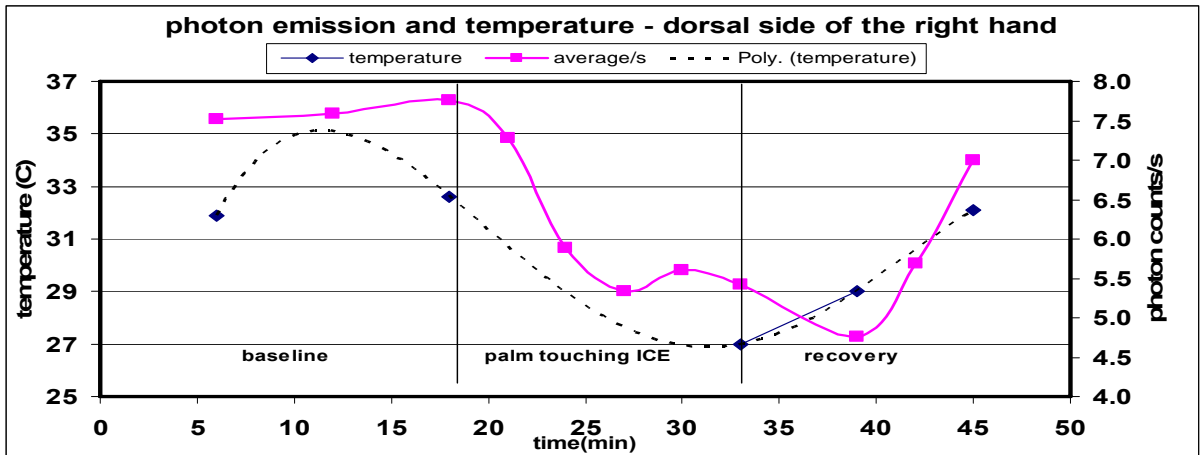


Figure 7.11. – Example of the dynamics of the photon emission and the temperature of dorsal side of the right hand while the right palm was touching the ice. Temperature curve is approximated by the 4th order polynomial

To evaluate the influence of the ice induced cooling on the photon emission and the temperature, three values for the temperature and the photon emission of palm and the dorsal side of the hand have been calculated for every measurement:

- baseline value – average from all baseline period values
- ice value – value on the end of the ice treatment period
- recovery value – last and most stable value, taken from the end of the recovery period

Two ratios have been additionally calculated for each of the ice induced cooling measurement (10 measurements where the dorsal side of the hand has been included; 7 measurements where the palm has been included):

- ice value / baseline value ratio shows to what extent dropped the given parameter during the ice treatment compared to the stable baseline value before the ice treatment
- recovery value / baseline value ratio shows how much differed given parameter from baseline value after the recovery

Average of these ratios is depicted on the *Figure 7.12.*

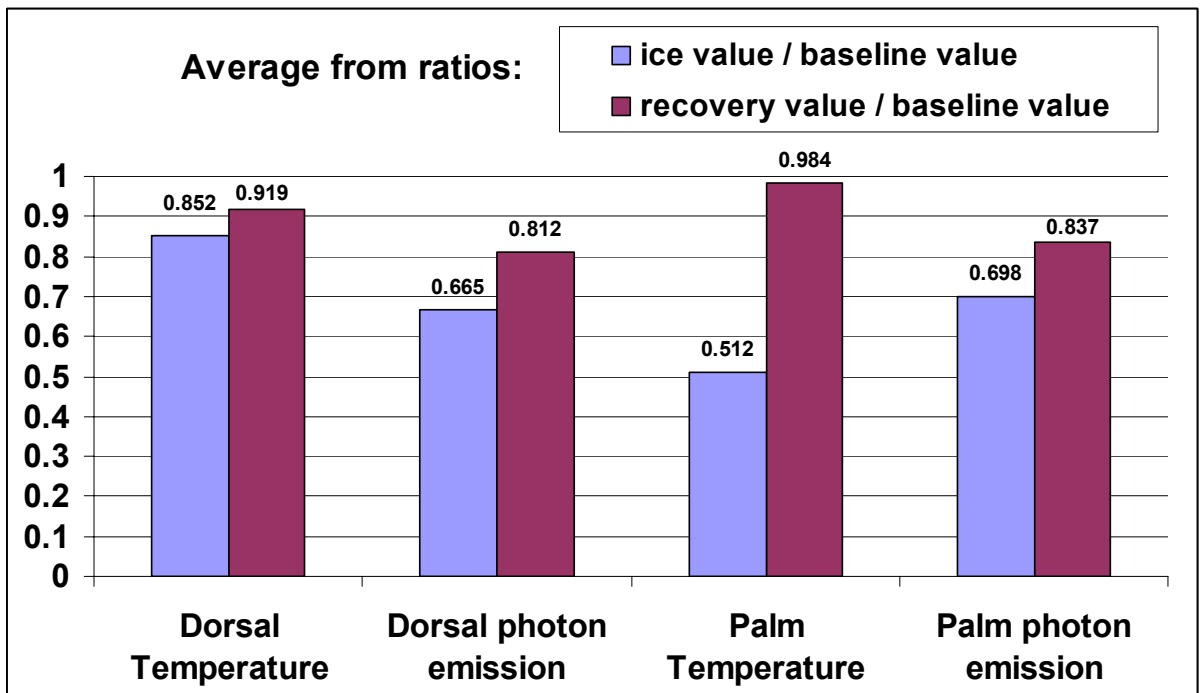


Figure 7.12. – depicts average of the ratios that describe relative changes of given parameters: the temperature of the dorsal side of the hand, the photon emission of the dorsal side of the hand, the temperature of the palm, the photon emission of the palm

One can see from the *Figure 7.12.* that there is positive correlation between the surface skin temperature changes induced by ice cooling, however it is not linear. For example, while the temperature of the controlled point surface skin temperature on the palm almost recovers (0,984) after 15 minutes, the photon emission of the palm is still lower (0,837) than the baseline value. Another interesting fact visible from this figure is that palm recovers(both the photon emission and temperature) faster from the ice cooled state than the dorsal side of the hand, although the palm was touching the ice directly. It indicates different thermoregulatory and compensation activity on the dorsal side of the hand than the palm, when the palm is cooled down directly.

Simple correlation of the temperature and the photon emission intensity values was calculated for every ice induced cooling measurement. The average value from all calculated correlations is 0.72 ± 0.192 for the palm photon intensity and temperature correlation and 0.71 ± 0.467 for the dorsal photon intensity and temperature correlation.

The hands as well as all extremities are subject of the considerable blood supply alterations what results in large temperature changes. However, is there comparable

degree of correlation of PE intensity and skin surface temperature of other anatomic sites?

7.4.3. Whole body spontaneous cooling measurements

In order to explore the behavior of other anatomic locations in the response to cooling, spontaneous body cooling by means of reduced clothing has been chosen. 12 anatomic locations have been measured for the temperature and the photon emission before and after the cooling according to the protocol described in 7.3.3. *Whole body spontaneous cooling measurement protocol.*

Two subjects have been measured; each has undergone six times the cooling process.

The correlation calculation between the photon emission and temperature of an anatomic location was the first approach for the examination of some relation between the skin surface temperature and the photon emission intensity.

Correlation PE-temperature	Hand (palm) right	Hand (dorsal) right	Hand (palm) left	Hand (dorsal) left	Abdomen right	Abdomen left	Solar plexus	Heart	Throat	Cheek right	Cheek left	Forehead
subject 1	0.76	0.81	0.79	0.85	0.48	0.70	0.54	-0.32	0.67	0.81	0.74	0.38
subject 2	0.63	0.67	0.76	0.73	0.40	0.52	0.58	0.61	-0.50	0.66	0.48	0.24

Table 7.5. – Correlation between the photon emission intensity and the temperature of the 12 anatomic locations

As seen from the *Table 7.5.*, and confirmed by many measurements, the highest correlation of the temperature and the photon emission occurs mostly on hands, however even here it hardly exceeds 0,8. Other anatomic points show only medium and low correlation between the temperature and the PE intensity, some are very subject-dependent.

Relative difference of the temperature and the PE intensity before and after the cooling has been calculated for all observed anatomic locations for both subjects for all cooling experiments. Average from these relative differences has been calculated to characterize subject's individual PE response to cooling.

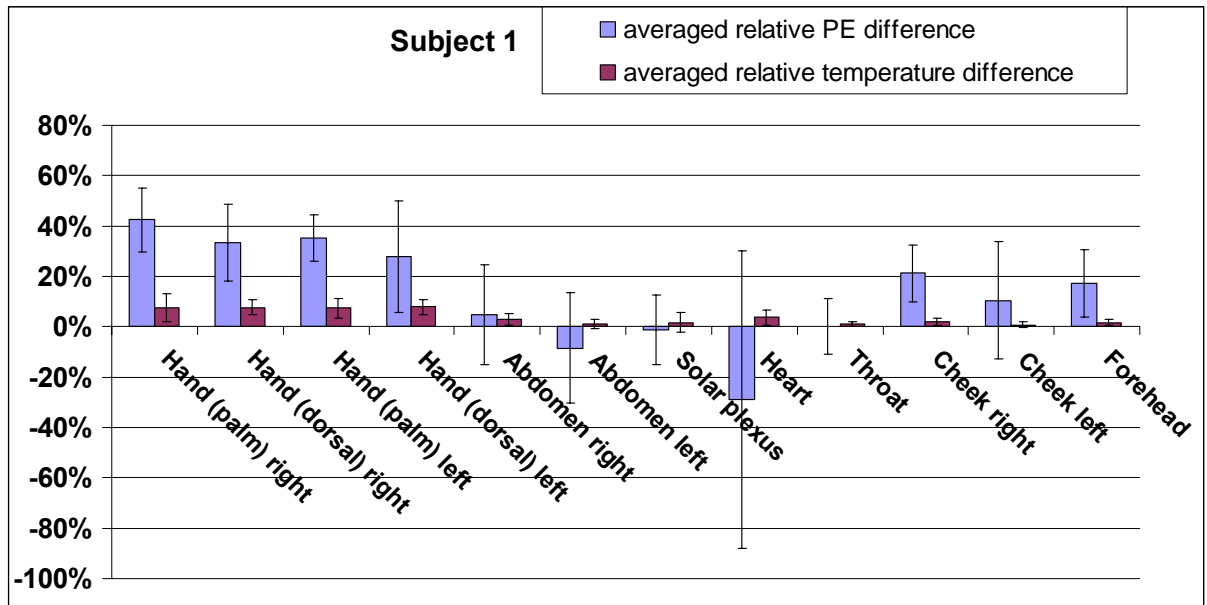


Figure 7.13. – average of relative differences before and after the cooling counted from six cooling experiments. Error bars show standard deviation of relative differences from this six measurements

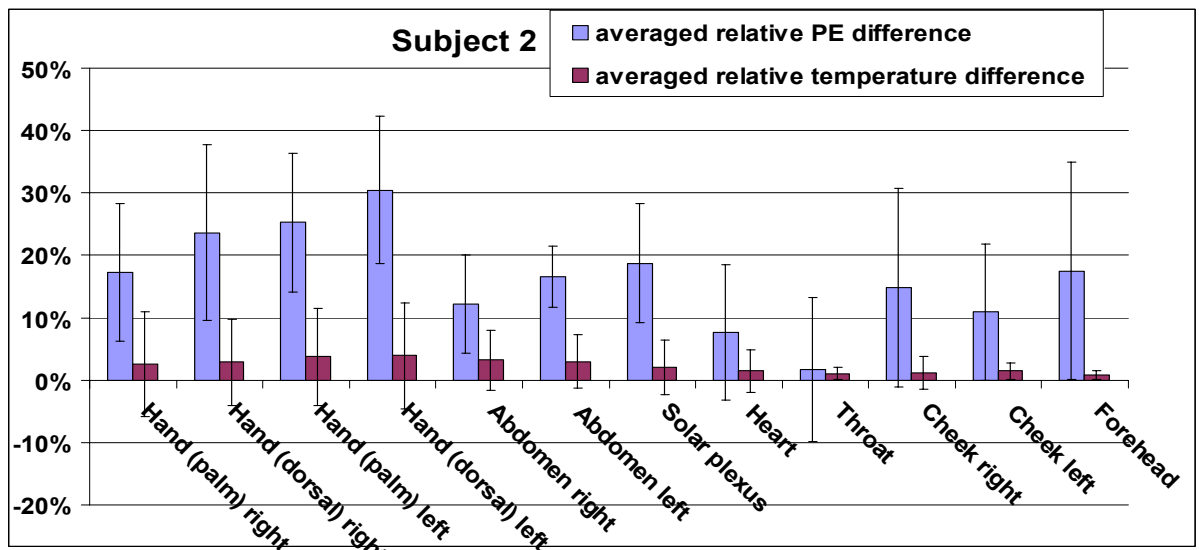


Figure 7.14. – average of relative differences before and after the cooling counted from six cooling experiments. Error bars show standard deviation of relative differences from this six measurements

When the correlation of the PE intensity between left-right symmetric anatomic locations has been calculated, it showed up that it is very high (~0,9), higher than temperature-PE intensity correlation of single location. Since the correlation of the temperature between left-right symmetric anatomic locations is very high as well (>0,9), it is sure that there is no linear relation between the skin surface temperature and photon emission intensity. It must be more than one factor that influences the PE intensity.

One has to keep in mind what are the body reactions to the coldness. Blood vessels in the extremities constrict to decrease the rate of the heat loss from the large surface of the limbs and to maintain the body core temperature on the stable level. Therefore one may suppose that the largest decrease and changes of the temperature as well as the photon emission intensity occurs on hands, as was observed in all previous types of measurements. Temperature of the body locations as forehead, throat, heart and solar plexus is relatively stable and one would expect only slight decrease in PE due to the slightly lower skin surface temperature, if the photon intensity is just simple function of the temperature or the bloody supply. However, there are sometimes occurring unexpected changes of the PE intensity, either negatively correlating or non-correlating with skin surface temperature at all, about which origin one can only speculate so far.

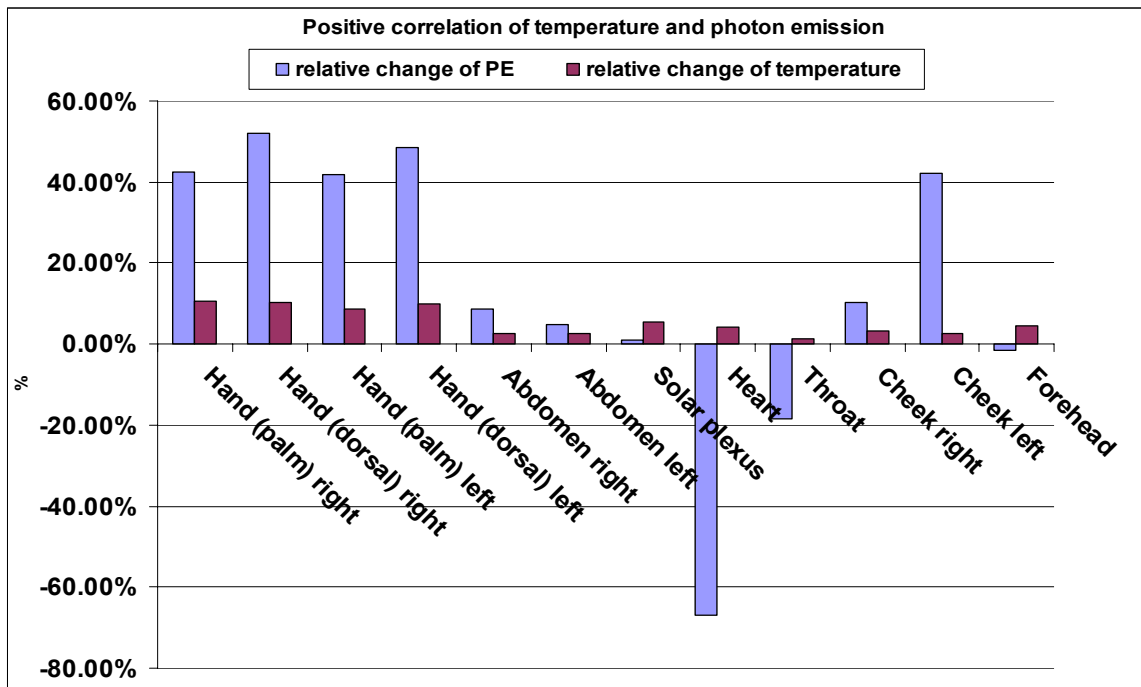


Figure 7.15. – relative differences before and after the cooling counted from one of the measurements of the Subject 1. Despite lowering the skin surface temperature of whole body by the spontaneous cooling due to the minimal clothing, the heart spot PE intensity increased. All possible artifacts that could cause such an increase in emission have been excluded. It is not the only case of strong anticorrelation of torso spots that increased their PE after decrease of the skin temperature

Since only two subjects were measured, although several times, and the PE intensity differences vary significantly between these two subjects in some anatomical locations, one cannot make any further conclusion about the temperature-PE correlation

for some anatomic spots that varied between two observed subjects. Research will continue with expanding the number of measured subjects under the same protocol.

It is worthy to note one more parameter that has been calculated not only for whole body measurements but also for the “24 hour” measurements. The parameter has sign “ δ ” or Q (both are to be found in literature) and it is way how to calculate deviation of measured photocount statistics from the Poissonian distribution. Poissonian distribution in $\Delta t < \tau$ (see 4.2.4. *DNA as source of coherent radiation*), the necessary condition of the coherent field, has its mean value ($\langle n \rangle$) equal to variance ($\langle \Delta n^2 \rangle$).

δ is calculated in this manner:

$$\delta = \frac{\langle \Delta n^2 \rangle - \langle n \rangle}{\langle n \rangle}$$

Therefore for the ideal Poissonian distribution, $\delta=0$. We can use δ as arbitrary and simplified tool of determining the fulfillment of necessary condition of coherence of measured photon statistics .

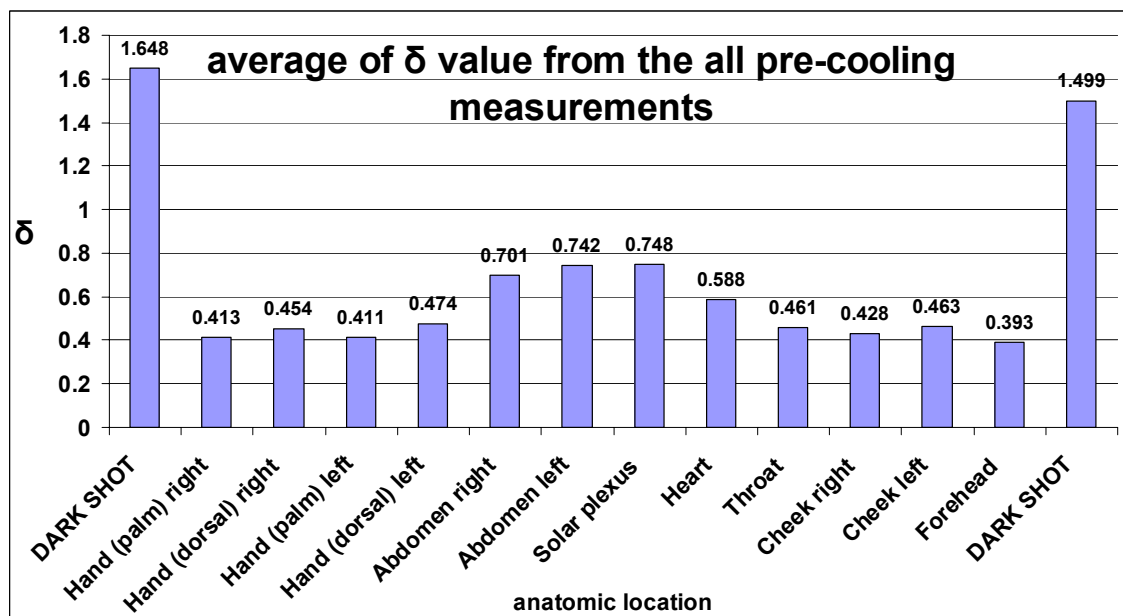


Figure 7.16. - δ of the PE of all measured spots is significantly lower than for the dark shot distribution (photomultiplier noise) that is of chaotic, thermal origin

If the intensity is higher, δ value is less vulnerable to random high counts that naturally occur as a part of noise. When the intensity is low (low S/N ratio), it is more

influenced by thermal PMT noise, therefore it is simple to understand the positive correlation of the PE intensity and the δ value, that is observed for the low number of photon counts.

7.5. Discussion of results

The analysis of the “24 hour” and longer measurements with sampling interval 2 hours yielded these basic results:

- relative intensity fluctuations of the photon emission(PE) from the back of the hand are slightly higher than those from the palms. In average, the photon emission of the dorsal sides of the hand fluctuated within 79,3% over or under the mean value within the one measurement cycle (24 hours), whereas the palms that have higher intensity of the photon emission show 62,8% relative intensity fluctuations.
- there is degree of left-right as well as palm-dorsal symmetry correlation in changes in ”24 hours” measurements. It is not particularly high (averaged correlation values 0,387-0,759) but is visually recognizable on the graphs
- average pattern of daily PE intensity fluctuations of dorsal sides of the hands and the palms was calculated from five “24 hour” measurements. There were no significant and robust difference found between the photon emission intensity during the day and night. The PE doesn’t necessary decrease in the night, as it was previously supposed
- question of some intrinsic regular PE oscillations period of hours is hard to answer since the photon emission is influenced by temperature, blood and oxygen supply, which has some irregularity, too. Fourier analysis of the photon emission intensity from 7 measurements longer or equal to 24 hours showed occurrence of the peaks of ca. 1 cycle/day and 3-4 cycles/day
- average correlation of the temperature and the photon emission of the palms calculated as average from the correlations of each of six “24hour” protocol measurement was 0.548 ± 0.23 for the left palm and 0.525 ± 0.262 for the right palm

The measurements of the photon emission and the temperature of the hand palm and the dorsal side of the hand were performed before, during and after the touching an ice cube with the palm in order to find more about changes of the photon emission when there is strong external factor that activates the thermoregulatory system. Following has been found:

- palm recovers(both the photon emission and temperature) to the baseline value faster from the cooled state than the dorsal side of the hand does, although the palm was touching the ice directly, what can be understood as stronger and faster thermoregulatory compensation due to the stronger cooling effect on the palm
- the recovery of the skin surface temperature to the baseline value from the cooled state was faster than recovery of the photon emission
- average value of the correlation the photon intensity and the temperature from all measurements is 0.72 ± 0.192 for the palm and 0.71 ± 0.467 for the dorsal side of the hand

After finding positive and relatively high correlation of the skin surface temperature and the photon emission on the hands, further questions about the correlation of the photon emission intensity and the temperature of the other anatomic locations have been asked. Measurements of photon emission and the temperature of 12 anatomic locations gave the first clue about the possible answer:

- relatively high correlation($\sim 0,75$) of the photon emission intensity and the temperature of both of the hands(palms, dorsal sides) have been confirmed
- temperature and the photon emission correlation of other anatomic locations vary between the subjects as well as within subject.
- observation of proportional contribution of single anatomic locations to total emission is in agreement of previously observed[119] photon emission intensity distribution on human body – more structured parts like hands and face emit more photons than that of smoother character(abdominal parts, stomach)

Under normal conditions, blood supply dependent temperature changes are essential in fluctuations of the photon emission from extremities. However, other

factors, which are not completely elucidated up to now, may also play important role in changes of the photon emission changes and in some cases even such strong role that they can overcome expected change of the photon emission due to the temperature change.

Although the correlation of the photon emission and skin surface temperature is not high in some cases, there is a particular positive correlation. Therefore it is strongly recommended to register the skin temperature by every experiment that relies on comparison between two values of the photon emission in different time, to avoid false conclusions.

The δ (or Q) value, the indicator of deviation from necessary condition of coherent field (the Poissonian distribution in $\Delta t < \tau$, $\delta=0$), shows for every measurement done, that photon emission of human body tends to fulfill this condition. The photomultiplier noise, which is of thermal origin, has significantly higher δ value than the human biophoton emission, what is confirming already known fact [129, 85].

8. Conclusion

In the first part, the historical and theoretical background of the photon and biophoton concept, theories of the biophoton source, the measurement systems, the overview of human biophoton research and the properties of the photon emission from the human body have been evaluated. In the second, experimental part, the measurement system and protocol have been described; measured data have been analyzed, quantified and depicted into the graphs, figures and tables. The last section summarizes the acquired data and points out the preliminary conclusions.

The aim of the experimental part of this work was to bring more information about the dynamics of the time dependent human photon emission fluctuations in the interval of 24 and more hours, because the data concerning dynamics of human biophoton emission found in the literature are rather scarce and oriented more on the long term observation in time intervals of weeks and months. Temperature dependent fluctuations of the human photon emission caused by induced as well as spontaneous cooling have also been evaluated. The knowledge of both time dependent and temperature dependent photon emission fluctuations and their dynamics are crucial for evaluation of any further experiments that relies on comparison between two values of the photon emission in different time, to avoid false conclusions.

Observed daily fluctuations of the biophoton emission intensity are not negligible, the photon emission intensity from the hands may fluctuate in the physiological conditions up to 2 times around the value of the daily average.

Significant relationship between the skin surface temperature (probably blood supply caused temperature changes) and the photon emission from hands has been found for all subjects. However, non-significant temperature and photon emission correlation or even negative correlation has been found on some anatomic locations, varying between as well as within the subjects.

Previous observations of rather high agreement of photon count statistics with the Poissonian statistics suggesting coherent nature of the biophoton emission has been confirmed.

Not only possible bioregulatory and biocommunicative effects of biophoton emission but also sometimes its peculiar behavior stimulates the need for the further research of this phenomenon. If we understand underlying processes of the human

biophoton emission in the state of health and disease, it may serve as powerful non-invasive diagnostic tool.

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Appendix

First page of the documentation of the PMT used for measurements – 9235QB

52 mm (2 ") photomultiplier 9235B series data sheet



1 description

The 9235B is a 52mm (2") diameter, end window photomultiplier with blue-green sensitive bialkali photocathode and 13 high gain, high stability, SbCs dynodes of linear focused design. The 9235WB and 9235QB are variants for applications requiring uv sensitivity.

2 applications

- low light level detection

3 features

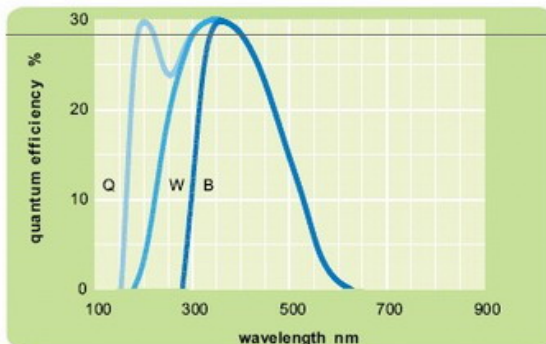
- high gain
- low operating voltage
- good SER

4 window characteristics

	9235B borosilicate	9235WB uv glass	9235QB* fused silica
spectral range**(nm)	290 - 630	185 - 630	160 - 630
refractive index (n _e)	1.49	1.48	1.46
K (ppm)	300	8500	<10
Th (ppb)	250	30	<10
U (ppb)	100	30	<10

* note that the sidewall of the envelope contains graded seals of high K content
** wavelength range over which quantum efficiency exceeds 1 % of peak

5 typical spectral response curves



6 characteristics

	unit	min	typ	max
photocathode: bialkali				
active diameter	mm		48	
quantum efficiency at peak	%		30	
luminous sensitivity	μA/lm		80	
with CB filter		9	12.5	
with CR filter			2	
dynodes: 13LFSbCs				
anode sensitivity in divider A:				
nominal anode sensitivity	A/lm		2000	
max. rated anode sensitivity	A/lm		5000	
overall V for nominal A/lm	V		1000	1400
overall V for max. rated A/lm	V		1100	
gain at nominal A/lm	x 10 ⁶		25	
dark current at 20 °C:				
dc at nominal A/lm	nA		6	50
dc at max. rated A/lm	nA		15	
dark count rate	s ⁻¹		300	
afterpulse rate:				
afterpulse time window	μs	0.1		6.4
pulsed linearity (-5% deviation):				
divider A	mA		30	
divider B	mA		100	
pulse height resolution:				
single electron peak to valley	ratio		2	
rate effect (I_a for Δg/g=1%):	μA		20	
magnetic field sensitivity:				
the field for which the output decreases by 50 %				
most sensitive direction	T x 10 ⁻⁴			
temperature coefficient:	% °C ⁻¹			± 0.5
timing:				
single electron rise time	ns		3	
single electron fwhm	ns		5	
single electron jitter (fwhm)	ns		6	
multi electron rise time	ns		4	
multi electron fwhm	ns		6.5	
transit time	ns		50	
weight:	g		100	
maximum ratings:				
anode current	μA			200
cathode current	nA			100
gain	x 10 ⁶		63	
sensitivity	A/lm		5000	
temperature	°C	-30		60
V (k-a) ⁽¹⁾	V			2300
V (k-d1)	V			300
V (d-d) ⁽²⁾	V			300
ambient pressure (absolute)	kPa			202

⁽¹⁾ subject to not exceeding max. rated sensitivity ⁽²⁾ subject to not exceeding max rated V(k-a)

7 typical voltage gain characteristics

