Spectra-H lux devise recently is applied in several Departments of Sheba Medical Center as pain relief device in patients suffering from painful disorders.

BIOLOGICAL BASIS OF PHOTOTHERAPY

REVIEW OF SCINTIFIC PUBLICATIONS

ACTION OF LIGHT ON ANIMAL AND HUMAN CELLS

Light absorbing biomolecules

In order to affect the living cell, light has to be absorbed by some molecules -chromophores. Energy of light photon can elevate electron in a chromophore molecule to the excited state changing the physical-chemical properties of the molecule. This can lead to measurable biological effect, which may be realized at the cellular, tissue and organism levels.

The main biologically active chromophores of living tissue absorbing light in a visible region of the spectrum (400-700 nm) are *melanin, porphyrins and hemoproteins, flavins and flavoproteins*. These molecules have various important functions and are involved in biological processes of metabolism, homeostasis, immune response, differentiation and proliferation.

Melanin a dark biological pigment (bio chrome) found in skin, hair, eyes, and some internal organs. It is a main absorber of light in the skin. It absorbs light energy in the broad range of the spectrum from ultraviolet to infrared wavelengths. It converts and dissipates absorbed light energy in the form of heat. It can act not only as a sunscreen but also as a scavenger of active chemical species and as a part of immune defense system. For example, it was found that skin melanin and neuromelanin (melanin of brain neurons) can act as natural antioxidant [**Krol and Liebler, 1998; Wilczok et al., 1999]**. One of the functions of melanin and melanin containing organelles (melanosomes) and cells (melanocytes) in skin is to inhibit the proliferation of bacterial, fungal and other parasitic infections of the dermis and epidermis [**Mackintosh, 2001].**

Porphyrins and hemoproteins. Porphyrins are tetrapyrrolic pigments that occur widely in nature, and play very important roles in various biological processes. They play important roles in electron transport systems and function as prosthetic groups (non-amino

acid part of a conjugated protein) of many enzymes - biochemical catalysts increasing the rate of reaction. Due to their importance to all living systems and their intense coloring, they have been nicknamed the "pigments of life". They absorb light in blue, green and red regions of the spectrum. Heme is one of the porphyrins that are essential for the many biological functions. Heme serves as the prosthetic group of numerous hemoproteins. Hemoproteins play such vitally important roles as transport (hemoglobin, myoglobin, neuroglobin, cytoglobin and leghemoglobin), catalysis (peroxidases), hormone synthesis and regulation (thyroperoxidase, cytochromes P-450), active membrane transport and electron transfer (cytochromes a,b,c). Some enzymes of hemoprotein nature are involved in control of protein synthesis, and cell differentiation.

Hemoglobin is the hemoprotein molecule in red blood cells which carries oxygen from the lungs to the body's tissues and returns carbon dioxide from the tissues to the lungs. It was shown that hemoglobin may serve as a carrier protein for hormone melatonin in the blood and discharge it in the target organs [**Gilad and Zisapel, 1995]**. Subsequently, the efficacy of melatonin's action as a hormone or antioxidant in target tissues may be enhanced.

Peroxidases are enzymes with heme prosthetic groups that have the ability to catalyze the oxidation of a wide variety of substrates by hydrogen peroxide. Human cells contain at least four peroxidases: myeloperoxidase, eosinophil peroxidase, lactoperoxidase and thyroid peroxidase. Peroxidases were fond in blood cells of immune system (neutrophils, leucocytes, monocytes and macrophages). Upon activation these cells kill bacteria. These enzymes play various functions and one of them is an improvement of immune response. The thyroid peroxidase participates in formation of the thyroid hormones.

Cytochromes are generally membrane-bound proteins that contain heme groups and carry out electron transport. They are found in the mitochondrial membrane. Several kinds of cytochrome exist, e.g. cytochrome *a*, cytochrome a_3, cytochrome *b*, cytochrome *c*, cytochrome c_1, cytochome *f* , cytochrome(s) P450. Cytochromes act as carriers of electrons (or hydride ions) in the series of complex enzymes known as the electron transport chain. An important class of monoxygenases is called cytochromes P450. They represent a class of similar enzymes that each contains a heme. This family of enzymes is involved in the metabolism of many drugs and dietary substances, and in the synthesis of steroid hormones and other extracellular lipid signaling molecules. It was shown that a cytochrome P450 plays an important role in regulation of melatonin metabolism [**Facciola et al., 2001**].

Flavins and flavoproteins. In tissues there is a broad distribution of flavins. The majority is found in flavocoenzymes [mainly flavin adenine dinucleotide (FAD)] and lesser amounts are in a mononucleotide [riboflavin-5'-phosphate (FMN)]. Flavoproteins are proteins containing a flavine prosthetic group. Flavoproteins are often an integral part of important electron transport pathways in living systems including such important processes as respiration, photosynthesis, nitrogen fixation, signal transduction, detoxification, DNA repair, and metabolism. Many of these processes represent an important means by which chemical and photochemical energy is transformed and transported within the cell as electro-chemical potential. Flavins absorb light in a blue-green region of the spectrum.

There are over 40 known flavoproteins, all playing important roles in the oxidation processes in the body that help create energy. Based upon their function these proteins are divided into several classes, including vitamins (e.g. riboflavin, vitamin B2) and variety of enzymes, e.g. electron transferases, dehydrogenases, disulfide oxidoreductases, oxidases and monooxygenases. Respiratory flavoprotein enzymes (e.g. NADH-dehydrogenase) catalyze (speed) oxidation-reduction reactions.

Flavoproteins have been implicated as playing signal transduction roles in programmed cell death [**Miramar et al., 2001**] and regulation of biological clock [**Sancar, 2000**].

Energy production and electron transport (respiratory) chain.

In animal cells, the energy for cellular growth and functions is generated by oxidation of substrates (sugar, protein and fat). Substrate oxidation consists of cascade of chemical reactions and includes three steps: glycolysis, citric acid cycle and electron transport (Fig.1).

Overall metabolic process contains three kinds of reaction sequences: 1) reactions in which the substrates themselves are oxidized; 2) reactions in which the electrons are transported through the electron transport chain, and 3) reactions involved in the coupling of the electron transport to synthesis of ATP (adenosine triphosphate, adenosine plus three phosphate groups).

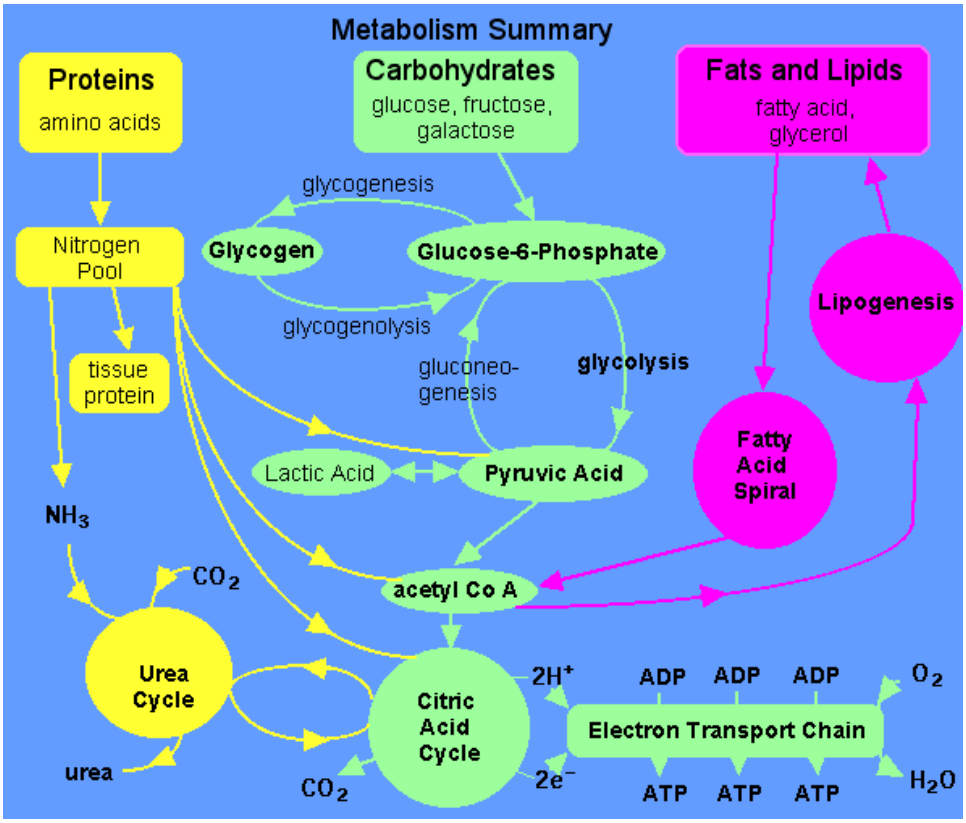

Fig.1. Metabolic process scheme.

Mitochondrial respiratory chain (Fig.2) is located in inner mitochondrial membrane and includes more than 80 peptides organized in 5 enzymatic complexes (I-V) that transfer electrons (received from donors in intermediary metabolism) from one complex to another to final acceptor, oxygen, eventually resulting in the formation of ATP from ADP (adenosine diphosphate, adenosine bonded to two phosphate groups).

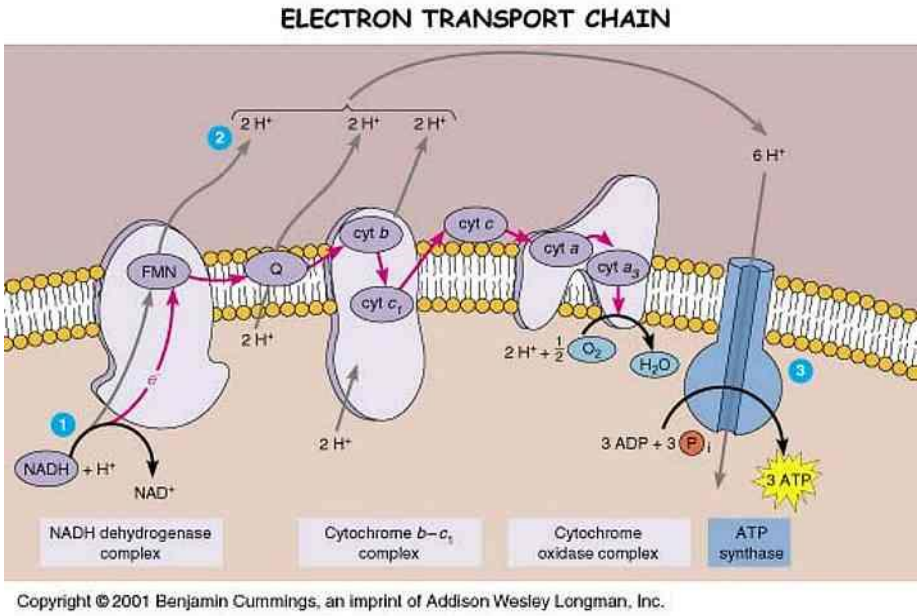

Fig.2. Electron transport chain.

Enzymatic complexes of mitochondrial electron transport chain include flavoproteins (FP, NADH-dehydrogenase), hemoproteins (cytochromes) and coenzymes (e.g. FAD, FMN, NAD - nicotinamide adenine dinucleotide).

Electron transport in electron transport chain:

NAD → FP → Cytochromes → O_2 + ATP (energy)

The ATP/ADP cycle provides energy for cellular activity. When energy is necessary the third phosphate group breaks off from ATP. This forms ADP and releases energy. When a phosphate group is freed up, it may move on to another molecule in a process called phosphorylation. In the energy production cycle in the mitochondria, energy is stored when ATP is produced from ADP and a phosphate group "P".

The discrete steps in oxidation of substrates, called oxidative phosphorylation, may be regarded as the passage of electrons through the electron transport chain until finally oxygen itself accepts the electrons and water is formed.

Light effect on cellular respiration

Depending on its wavelength, electromagnetic radiation in the form of light can excite macromolecules by transferring energy to electrons and can initiate conformation changes in proteins. This action of light may cause cascade-like processes in cellular organelles (e.g. outer membrane, mitochondria, endoplasmic reticulum, nucleus**)** leading to modification or/and stimulation of the cell growth and function.

Mitochondria are the power centers of the cell - organelles which provide the energy to cellular growth and function e.g. moving, dividing and production of secretory products. They have a double membrane. The outer membrane is fairly smooth. But the inner membrane is highly convoluted, forming folds called cristae. The cristae greatly increase the inner membrane's surface area. Cellular respiration occurs on these cristae and food (sugar) is combined with oxygen to produce ATP (adenosine-tri-phosphate) - the primary energy source for the cell.

It is generally assumed that the stimulating action of visible and near infra-red light is due to excitation of chromophores such as flavins and cytochromes in the respiratory chain (RC) of mitochondria. There is a direct correlation between the action spectrum (light wavelength of biostimulation) and the absorption spectrum of the pigments of the RC. Low level laser from the red and the near infrared region corresponds well with the characteristic energy and absorption levels of the relevant components of the mitochondrial RC. This laser stimulation vitalizes the cell by increasing the mitochondrial ATP production. **[Wilden and Karthein, 1998]**. Numerous studies support this concept by observation of various stimulating effects on mitochondria.

Light irradiation at 632 nm with energy dose 2 J/cm^2 and light power of 10 mW lead to an increased activity of cytochrome c oxidase [**Pastore et al., 1994]**. This increase is observed in both the electron transfer and proton pumping.

Photostimulatory effect on a model of rat liver mitochondria was also obtained using 660 nm light at a power density of 10 mW/cm^2 [**Yu et al., 1997**]. Photoirradiation at this wavelength and light doses ranged between 0.6 and 2.4 J/cm^2 significantly increased mitochondrial oxidative metabolism (oxygen consumption), phosphate potential, energy charge and enhanced the activities of NADH and some electron chain enzymes (e.g. ubiquinone oxidoreductase, ferrocytochrome C and ferricytochrome C oxidoreductase).

Radiation of isolated liver mitochondria with a He-Ne laser (632 nm) brought about enhanced ATP-ADP metabolism, an increased content of ATP, a growth of the electric

potential across inner membranes and pH in matrix, as well as small changes in the matrix configuration [**Passarella et al, 1988**].

Several studies demonstrated that He-Ne laser at 632 nm causes activation of mitochondrial DNA replication and enhances cytosolic and mitochondrial protein synthesis [**Greco et al., 1989; Vacca et al., 1993, 1994, 1996**]. In the studies of **Greco** et al. **[1989]** and **Vacca** et al. **[1993]** stimulation in the synthesis of all mitochondrial transcription (RNA) and translation (protein) products was observed at an energy dose of 5 J/cm^2 (power density 12 mW/cm^2). In the more recent paper, similar effect was observed using fluence rates 7 and 12 mW/cm^2 at a much lower energy dose of 0.24 J/cm^2 [**Vacca et al., 1996**].

Red light irradiation (632 nm, 0.24 J/cm^2) triggers an increase of the mitochondrial membrane potential and, in a cascade-like manner, increase in intracellular Ca2+, leading to cell stimulation by both direct and indirect action mechanisms **[Greco et al., 2001].**

This concept is supported by recent study of **Bortoletto** et al. [**2004**], where similar phenomena of changes in mitochondrial membrane potential after irradiation at 635 nm with energy density of 0.1 J/cm^2 were found.

Light effect on cellular growth (proliferation) and functions

It has been demonstrated on various kinds of cells *in vitro* that low level photoradiation can stimulate cellular proliferation. Cultures of skin cells keratinocytes and fibroblasts are often used as a very suitable model to study biological action of light irradiation *in vitro*.

Boulton and Marchall [**1986**] showed that irradiation of two types of fibroblasts with a He-Ne laser for 15 min (1 W/m^2) accelerated the growth of the population in the exponential phase and attachment of cells to substrate.

Irradiation of keratinocytes with He-Ne laser (1.5 J/cm^2) induces an increase in the rate of cell proliferation [**Yu et al**., **1996**].

A stimulating effect on the long-term proliferation of skin fibroblasts was shown after laser irradiation at 670 nm, (power density 40 mW/cm^2) and the energy densities 2 J/cm^2 and 12 J/cm^2 [**Bednarska et al., 1998].** Similarly, **Almeida-Lopes** et al. [**2001**] showed an improvement in fibroblast proliferation after laser irradiation in visible (670 and 692 nm) and near-infrared (780 and 786 nm) regions of spectrum at light dose of 2 J/cm^2. Analogous results with significant stimulatory effect on proliferation of fibroblasts were revealed in response to laser irradiation with 665 and 675 nm [**Moor et al**., **2005**].

Low power laser irradiation (632 nm, 0.10-6.3 J/cm^2) of human vascular and cardiac cells in vitro results in a statistically significant increase of secretion of vascular endothelial growth factor and rate of cell proliferation [**Kipshidze et al., 2001]**.

In study of **Sroka** et al. [**1999**] a significant increase in the cellular mitotic rate (indicating proliferative activity) in both normal and malignant cells was seen 24 hours after light illumination with 410 nm, 635 nm and 805 nm.

Several studies demonstrate positive effect of laser radiation on blood cells. **Iijima** et al showed that low-power green (543 nm) and red (632 nm) laser light have a protective and stabilization effect on membranes of red blood cells **[Iijima et al., 1991**]. Blood lymphocytes can be activated by low-level laser light irradiation at both 632 and 660 nm [**Smol'yaninova et al., 1991; Stadler et al., 2000**]. In the study of **Stadler** et al [**2000**] lymphocyte proliferation after irradiation using 660 nm light (40 mW/cm^2, 1.5-3 J/cm^2) was higher in the presence of hemoglobin. Authors suggest that hemoglobin can absorb light energy and amplify light-induced biological effects. This concept is supported by study of **Mi** et al. [**2004**]. They showed a positive effect in the modulation in rheological properties of blood cells after irradiation with green (532 nm) and red (632 nm) light. In this study green light was more efficient than red light.

Low-power laser irradiation has been found to produce photobiological effects with evidence of interference with immunological functions. **Funk** et al. [**1992; 1993**] have found that irradiation with He-Ne laser (632 nm, 18.9 J/cm^2) increases levels of cellular cytokines (e.g. interleukin-1 alpha, interleukin-2, tumor necrosis factor-alpha and interferon-gamma), which play an important role in immune response. Interleukins are specific mediators of cellular functions that stimulate the growth and activities of certain kinds of blood cells (e.g. leucocytes, immune cells) and promote hematopoiesis (blood cell formation). Similar results with a significant enhancement in production of interleukins (interleukin-1 alpha and interleukin-8) was also observed after irradiation of skin cells using He-Ne laser at 1.5 J/cm^2 [**Yu et al**., **1996**].

Photoirradiation using light emitting diode (LED) light sources show very similar to laser effects. An increase in rate of fibroblast proliferation in vitro was demonstrated after illumination using green (570 nm), red (660 nm) and infrared (950 nm) LEDs [**Vinck et al., 2003**]. It was found that green light yielded a significantly higher effect than red and infrared light. In the more recent study **Vinck** et al. [**2005**] demonstrated the efficacy of green LED irradiation (570 nm, power output of 10 mW, 3 min, radiation exposure 0.1 J/cm^2) for enhancement of fibroblast proliferation and viability.

ACTION OF LIGHT ON HUMAN BODY

Light therapy techniques and applications

From the facts that there are many positive effects of light on human health, light therapy is one of the oldest therapeutic methods used by humans. Currently light therapy or phototherapy is considered part of physiotherapy.

The potential therapeutic effect of light on tissue or organism level depends upon the wavelength transmitted to the target tissue. Ultraviolet radiation (UV, 100-400 nm) contains shorter wavelengths than visible light (400-700nm). The shorter the wavelength, the greater the photon energy but the less the penetration through biological tissues; conversely, the longer the wavelength, the less the photon energy but the deeper the penetration through the skin. The greater the photon energy, the greater the potential for harm: even low exposures to UV may cause sunburn and skin cancers, while visible and infrared light at the same doses is not dangerous. Moreover, visible and infrared light has been shown to be therapeutic.

A wide variety of light therapies e.g. phototherapy of neonatal jaundice, phototherapy of psoriasis, bright light therapy for Seasonal Affective Disorder (SAD), photodynamic therapy of cancer, high power laser therapy for surgery and cosmetics, low power laser therapy for bio-stimulation have evolved over the past 100 years. All light treatment modalities have different mechanisms of biological action. Some of these mechanisms (e.g. neonatal jaundice, phototherapy of psoriasis, bright light therapy of SAD, photodynamic therapy of cancer, high power laser therapy) are well studied and established, while others (photobiostimulation) are not understood yet.

Technically, all light treatments may be divided into two categories – light illumination of eyes and light illumination of skin.

Light illumination of eyes by full spectrum light screens or boxes is used in the treatment of Seasonal Affective Disorder (SAD). Some people may be more sensitive to decreasing exposure to light during the shorter winter days, leading to neurochemical imbalances (e.g. decreased serotonin level) that promote endogenous depression. Exposure to full spectrum light for a short period of time each morning appears to be successful in alleviating the symptoms of the winter blues and may be effective in treating other types of depression as well [**Roberts**, **1995, 2005**].

Skin illumination by coherent (lasers) or noncoherent (lamps, LEDs) light sources is used for other popular forms of light therapy. Psoriasis is treated by UV lamps. Red light lamps, lasers and LEDs are used for photodynamic therapy of cancer. LEDs and lasers [e.g.

helium-neon laser (He-Ne) , radiation at 632.8 nm; the gallium-aluminum laser (Ga-Al) , 630-685 nm; the helium-neon-arsenate laser (He-Ne-As), 780-870 nm, and the gallium-arsenate laser (Ga-As), (904 nm)] are the most commonly used sources for low-level (power, intensity) laser therapy. The use of LEDs (light-emitting diodes) as light sources is the next step in the technological development of light therapy. The emission band of LEDs lies in a wide region of the spectrum and they can be used instead of lasers when low power irradiation is needed.

For low power photo-biostimulation or photo-biomodulation procedure, lasers and LEDs are applied directly to the respective areas (e.g., wounds, sites of injuries) or to various points on the body (acupuncture points, muscle-trigger points). When low level lasers or LEDs are used to stimulate acupuncture points throughout the body, healing effects are similar to that seen with acupuncture needle stimulation of acupoints. This technique is sometimes referred to as laser-puncture (although no actual puncturing of the skin occurs). A number of different small hand held devices utilize LEDs to stimulate acupoints for relaxation, pain relief, and other health problems. Some researchers suggest that acupuncture meridians actually conduct light, somewhat like the fiberoptic guides in communication and electronic systems. Illumination of large skin areas (e.g. wound and injury sites) with low power red or infrared light has been shown to cause analogous anti-inflammatory and pain relief effects [**Schindl et al., 2000].**

<u>Skin structure and functions</u>

The skin is the largest organ of the body. It has 4 major functions: endogenous homeostasis (e.g. regulation of body temperature and fluid balance), metabolism (e.g. vitamin D synthesis,), sensory input, and to serve as a barrier to external threats (e.g. infection, mechanical injury, ultraviolet light). The skin organization includes structural cells (fibroblasts, keratinocytes and melanocytes), glands (sweat and sebaceous), and specialized nerve receptors for stimuli (changes in internal or external environment) such as touch, cold, heat, pain, and pressure. The skin is the main physical barrier between the environment and internal homeostasis (regulation of internal environment in order to maintain a stable condition by means of multiple control mechanisms). The skin has basically three layers that are affected by light: the epidermis (outer layer), the basal cell membrane (median layer) and the dermis (inner layer).

The epidermis has several layers that contain different cell types. Keratinocytes produce keratin, a protein that gives skin its strength, flexibility and waterproofs.

Melanocytes produce melanin, the dark pigment that gives skin its color. Merkel's cells are probably involved with touch reception. Langerhans' cells are messenger cells that report to the immune system about the injury or change in the skin.

The dermis is much thicker than the epidermis and is comprised of fibroblasts (cells having a major role in the synthesis and reorganization of extracellular matrix), blood and lymph vessels, collagen and elastin fibers and the bases of hair follicles and sweat glands.

Numerous studies show that skin functionality is highly complex and expresses endocrine (hormonal) activities with self-regulatory properties. The skin neuroendocrine system comprises locally produced hormones and neuro-endocrine mediators that interact with corresponding specific receptors in the body [**O'Sullivan et al., 1998; Slominski and Wortsman, 2003]**. Skin neuropeptides are a group of molecules functioning as neurotransmitters, neuromodulators, potential growth factors, and hormones. The skin neuroendocrine system acts to preserve cutaneous structural and functional integrity and to maintain systemic homeostasis [**Spellberg**, **2000**]. Some cutaneously synthesized neuropeptides are light-dependent and have pronounced systemic effects. For example, light-induced vitamin D3 and parathyroid hormone-related protein are involved in vital metabolic processes, including calcium and phosphorus metabolism, mineralization of the skeleton, regulation of parathyroid hormone production and significant effect on the immune system [**Holick, 1981; DeLuca, 2004]**. Recent investigations demonstrate that human skin can produce serotonin and transform it into melatonin [**Slominski** et al., **2002; 2003; 2005**]. These biomolecules have various important functions including antioxidant activity, anticancer and immunomodulation properties as well as signals of photoperiod to the body [**Maestroni 1993; Young and Matthews, 1995; Macchi et al., 2004]**.

Possible biological mechanisms of phototherapy on tissue and organism level

Despite of a large number of publications in the field of low-power light therapy, the exact biological mechanism(s) of this treatment is not known.

Photochemical reactions are discussed to be a possible basis of low power (1-100 mW/cm^2) phototherapy [**Karu**, **1999**]. When light enters the skin it is refracted, scattered and absorbed by various biomolecules (e.g. proteins, DNA, RNA, amino acids, hemoglobin and melanin) in the skin cells and in the blood. Absorption of light quanta can lead to photoexcitation (promotion of electronically excited states) of photoacceptors (flavins, cytochromes and porphyrins) in cellular mitochondria. The next step is the transfer of energy from the excited biological molecule to intracellular biochemical reactions. It is supposed

that cellular enzymes are the main targets of direct or indirect action of light. Direct action includes local heating or change in redox properties of electron chain enzymes in mitochondria. Indirect action can involve intermediate electron acceptors, such as oxygen or nitrogen, follow by generation of reactive oxygen species (e.g. singlet oxygen, superoxide anion) or nitric oxide (NO) [**Karu et al., 2004; 2005**]. Activation of enzymes initiates the secondary reactions (cellular signaling cascade) e.g. increasing of ATP synthesis, production of second messengers (e.g. Ca^{2+} ions, cyclic AMP), stimulation the synthesis of DNA and RNA. Activation of an enzyme or the induction of the synthesis of an enzyme needs only a few photons. Once an enzyme is activated, it can catalyze thousands of chemical reactions. There is a large amplification factor involved; a few photons can produce a huge biological effect [**Smith**, **2005**]. Thus, low power light irradiation can lead to pronounced biological responses such as increased cell proliferation and functionality.

Visible and infrared irradiation has different effects on molecules. Visible light can produce chemical changes while infrared radiation can only produce physical changes in molecules. In spite of this, both result in similar biological effects.

On tissue and organism levels therapeutic action of light is demonstrated as anti-inflammatory and immuno-modulatory effects, accelerated regeneration of damaged tissues and the improvement of blood circulation in organs. This may be associated with 1) growth of the activity of certain cells such as leukocytes and phagocytes, as well as an increased content of calcium ions in the cytoplasm of these cells; 2) enhancement of cell division and cell growth; 3) activation of the synthesis of proteins and cytokines, and 4) improvement of blood circulation in the bloodstream due to the relaxation of vessel walls (vasodilatation) [**Vladimirov et al., 2004**]. All these reactions may be connected with light -induced stimulation of neuroendocrine system. Neuroendocrine system is a universal system of response, control and organism protection. It is known that local treatment of skin by light may initiate systemic responses on organism level by involvement of some hormones and neurotransmitters (e.g. vitamin D3). Melatonin and serotonin have been also shown to be mediators of light-induced immunomodulation [**Korf et al., 1998]**.

J. Walker in a double blind study demonstrated that repeated irradiation with a low-power (1 mW) helium-neon laser produced relief in patients with chronic pain and this effect was associated with a large increase in the urinary excretion of 5-hydroxyindoleacetic acid, the degradation product of serotonin. **[Walker, 1983].**

In 1998 **Campbell and Murphy** published in Science journal pioneering and very expressive results demonstrating shifting human circadian clock and changing melatonin

level by light exposure of popliteal region (behind the knee) **[Campbell and Murphy, 1998].** They suggested that extraocular light may be transduced into a signal that is received and processed by the human central nervous system **[Murphy and Campbell, 2001].** It has been hypothesized that extraocular illumination of body sites with visible light may lead to systemic effect similar to ocular illumination with the mechanism of eye-brain regulation in serotonin/melatonin balance**.** However, further clinical studies did not show change in circadian phase, circadian-related sleep disorders or melatonin level at extraocular illumination of popliteal region in humans [**Lockley et al., 1998; Hebert et al., 1999**; **Lindblom et al., 2000; Lushington et al., 2002**; **Ruger et al., 2003**]. Despite this fact, there are a number of scientific publications indirectly supporting the hypothesis of **Campbell and Murphy**. These studies demonstrate the existence in human skin some elements of neuroendocrine and immune systems that can be activated by low power light stimuli.

One such element of diffuse neuroendocrine system is hormone melatonin. Melatonin is a regulator of biological rhythms. It is synthesized in the rhythmic fashion, primarily by the pineal gland. However, melatonin production have been found also in extrapineal tissues e.g. retina, harderian gland, gastrointestinal tract, lung, liver, kidney, adrenals, thymus, thyroid, pancreas, ovary, carotid body, placenta and endometrium as well as in non-neuroendocrine cells like mast cells of skin, natural killer cells, eosinophilic leukocytes, platelets and endothelial cells [**Zawilska, 1996; Kvetnoy, 2002]. Bjarnason** et al. have found that the human clock genes are expressed in human oral mucosa and skin, with a circadian profile consistent with that found in the suprachiasmatic nuclei of the brain [**Bjarnason et al, 2001**]. **Slominski** et al. published results with evidence that serotoninergic and melatoninergic systems are fully expressed in human skin [**Slominski** et al., 2002]. Several publications show involvement of blood cells in serotonin/melatonin homeostasis [Finocchiaro et al., 1991; **Carrillo-Vico et al., 2004**; **Cedeno et al., 2005]. Finocchiaro** et al. demonstrated in vitro stimulatory effect of visible light on human peripheral blood mononuclear leukocytes with modulation of cellular serotonin/melatonin metabolism **[Finocchiaro et al., 1995].** In vivo, visible light can penetrate epidermal and dermal layers of the skin to a depth of 2-3 mm and directly interact with circulating lymphocytes to modulate immune function. It has been found immuno-modulatory effect in peripheral blood of patients after illumination of skin areas (400 cm^2) with visible light (12-40 J/ cm^2) [**Samoilova et al., 2000; 2004; Zhevago et al., 2004; Zhevago and Samoilova, 2004**]. The treatment caused enhanced phagocytic activity of monocytes and granulocytes, enhanced cytotoxic

activity of natural killers, and induced secretion of tumor necrosis factor from mononuclear leucocytes.

CONCLUSION

Therapeutic effect of low-power light treatment has been demonstrated in a lot of scientific publications. The exact mechanism(s) of this treatment is not known yet. The most popular theories suggest that light-induced photochemical reactions can activate metabolic and functional properties of cells. Systemic effect of phototherapy may be associated with activation of cellular components of neuroendocrine and immune systems existing in skin and circulating blood. An occurrence of such a mechanism is discussed in literature. **Roberts** proposes that skin may be an alternative (to eye-brain system) pathway for light-induced immune modulation of the body [**Roberts, 2000**].

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