LASER RADIATION PROPAGATION AND HEAT TRANSFER INTO CELLS AND TISSUES

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Abstract. In this paper we shall approach only the interaction of laser radiations with highly scattering and/or absorbing biological materials (i.e., opaque materials). The photon multiple scattering by cells and tissues is approached from the perspective of the general propagation theory (i.e., simple diffusion) of a particular physical magnitude, provoked by its conjugated gradient. In this way, one can obtain, in a particular case, the analytic expression of fluence rate for the isotropic tissues. The propagation theory is also conducting to the model of heat transfer (i.e., spatial-temporal evolution of the temperature) in the adjacent regions of target in which the laser absorbed energy was converted into heat.

Key words: laser radiation, tissue, absorption, multiple scattering, heat transfer, diffusion.

INTRODUCTION

Just from the very beginning of laser invention the physicians and biologists were interested in modalities in which this special light instrument could be efficiently used in order to replace the old conventional techniques and, at the same time, to overcome the inherent limits of classical medical and research techniques.

The expected future success of laser utilisation in medicine and biology should be based on the knowledge of the interaction between laser radiations and the living matter (i.e., biomolecules, cells, tissues, etc.) a very difficult task to be achieved.

In order to properly describe the interaction of laser radiations with the cells and tissues, one has to consider, in a first approximation, two classes of biological media [21]:

a) *opaque media* (strongly scattering and absorbing the light) like: skin, blood vessel walls, blood itself, lymph, brain and bones;

b) *transparent media* (weakly scattering and absorbing the light) like: cornea, aqueous humour, lens, and vitreous humour of the eye. Laser radiation interaction

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with opaque media could be analysed and properly described by a *model of multiple scattering of photon*s into anisotropic absorbing media as it is the case of biological media (e.g., cells and tissues).

In principle, near infrared radiation (NIR) and red radiations are scattered less than blue, green and yellow radiations. One can conclude that the NIR and red radiations penetrate more deeply into the skin potentially treating deeper cellular layers of the skin [10].

The penetration depth, defined as the tissue distance along which the laser radiation intensity is decreasing *e* times (i.e., approximately 40%) depends both on the radiation wavelength and the type of tissue. The greatest penetration depth is encountered in the radiations with wavelength in the spectral range (600–1,100) nm known, for this reason, as *optical window* of cells and tissues owing to the less efficiency of the endogenous absorbers [11].

INTERACTION OF LASER RADIATION WITH OPAQUE BIOLOGICAL MEDIA

Biological materials (e.g., cells and tissues) are, *par excellence*, anisotropic absorbing media with refractive indices, n_m, greater than the refractive index of air, n_a ($n_m > n_a$). This difference of the refractive indices is responsible for the *partial radiation reflection* (i.e., *Fresnel reflection*) taking place at the interface air-tissue (characterised by the *Fresnel reflection coefficient,* R_F) and for the *radiation refraction* into tissues.

Photon multiple and random scattering into tissue accompanied by photon absorption by the tissue components are responsible both for the collimated beam defocusing and attenuation as far as it penetrates the tissue.

The depth of radiation penetration into tissues consequently depends, on the one hand, on the multiple scattering process characterised by the *average scattering coefficient*, μ_s, of the tissue and, on the other hand, on the photon absorption process characterised by the *average absorption coefficient*, μ_a. Average values of the absorption coefficients for different and numerous normal and pathologic tissues are already published [13].

As a result of the two synergic effects, a collimated beam of laser radiation, with the incident intensity, I_0 (Wm^{-2}), will be attenuated by the tissue, along its direction, let say *Ox*, following the Lambert exponential law:

$$I(x) = (1 - R_F) I_0 e^{-\mu_t x} \tag{1}$$

where $\mu_t = \mu_s + \mu_a$ represents the *global attenuation coefficient* of the tissue.

The Fresnel reflection coefficient, R_F, is given by the formula [13]:

$$R_F = [(n-1)/(n+1)]^2 \tag{2}$$

where *n* represents the relative (against air) refraction index of the tissue.

The *natural attenuation length*, δ, that is, the length along which the incident flux intensity is decreasing *e* times, is given by the reciprocal of the global attenuation coefficient:

$$\delta = \frac{1}{\mu_t} = \frac{1}{\mu_s + \mu_a} \tag{3}$$

The natural attenuation length is decreasing with the blood content of the tissue and, in the case of VIS domain it is smaller for blue radiations and greater for the red ones.

LASER RADIATION SCATTERING BY THE CELLS AND TISSUES

From the morphological point of view, there are a large variety of cells with dimension in the 100 nm–20 μm range. It follows that the wavelengths of the available laser radiations are either greater or smaller than the cell dimensions. The smaller sub cellular organelles (e.g., mitochondria, Golgi apparatus, lysosomes, ribosomes, etc.) constitute the main scattering centres for laser radiations.

In the case of *elastic scattering*, the wavelength of the scattered radiation, λ_s, is equal to that of the incident radiation, λ_i ($\lambda_s = \lambda_i$).

An interesting type of elastic laser radiation scattering is that of the Rayleigh type which depends on: a) the wavelength of laser radiation, b) the dimension of the scattering centres (e.g., cellular organelles), and c) the difference between the refractive indices of the scattering centres and of cytosol.

One can demonstrate that the intensity, I_{SR}, of the Rayleigh scattered radiation depends on the reciprocal of the fourth power of the radiation wavelength, λ, and on the scattering angle, θ, according to the formula:

$$I_{SR} = \propto \frac{1 + \cos^2\theta}{\lambda^4} \tag{4}$$

which puts in evidence the fact that the short wavelength radiations are the most scattered and that the forward scattering (θ = 0°) is equal with the backward scattering (θ = 180°). It results that the radiations with a large wavelength, not being efficiently scattered, can penetrate deeper in tissue where they will have the chance to be absorbed or even to be partially transmitted. There is, however, a penetration limit due to their absorption by water or by chemical groups –CH and –OH.

Another type of elastic scattering is Mie scattering which becomes effective when the dimension of the scattering particles is comparable to the wavelength of the radiations. Unlike the Rayleigh scattering, in the case of Mie scattering, the

intensity of forward scattered light is greater than that of the backward scattered light. The intensity of the scattered light of the Mie type, I_{SM}, depends on the radiation wavelength according to the formula [3]:

$$I_{SM} \propto \frac{1}{\lambda^n} \tag{5}$$

where n varies in the range: 0.4–0.5.

In the case of *inelastic scattering*, the wavelength of the scattered radiation, λ_s, is different from that of the incident radiation, λ_i ($\lambda_s \neq \lambda_i$).

The inelastic scattering of the light by cells and tissues is weaker than that of the elastic scattering.

Two cases of inelastic scattering are encountered: a) *Raman scattering*, provoked by the excitation of molecular vibration energy levels and b) *Brillouin scattering,* due to the inhomogeneities induced by the shock waves of the generated ultrasounds.

The principal biological structures responsible for light scattering are presented in Table 1, the most important scattering centres being mitochondria and collagen fibres.

Table 1

The principal cellular structures responsible for the laser light scattering
(L = length, d = diameter, V = volume)

Cellular constituent (CC)	Refractive index	CC dimensions	Scattering theory	References
Cellular membrane	1.460	L = (8–10) nm	Mie	[18]
Collagen fibres	≅ 1.500	d > 1 μm	Rayleigh	[6]
Lysosoms		d = (0.5–2) μm	Mie	[1]
Mitochondria	1.428	L = (2–3) μm	Mie	[2]
		V = 1 μm^3		[12]
Nucleus	≅ 1.350	d = (2–10) μm	Mie	[4]
Cell	≅ 1.385–1.400	d = (18–25) μm	Mie	[19]

The light scattering by the tissue is characterised by the scattering coefficient, μ_s (cm^{-1}), whose typical values for the therapeutic windows (i.e., 600–1,500 nm) are embedded into the interval (150–550) cm^{-1}

PHOTON MULTIPLE SCATTERING MODEL

As we have already pointed out, an important modality of the interaction of laser radiation with biological material consists in photon scattering, conducing to

the propagation (transport) of photons from the place of impact with tissue to other adjacent zones.

In this model, we shall consider, for the sake of simplicity, an isotropic tissue, irradiated by a laser radiation which is not attenuated by absorption, but only by scattering, that is: $\mu_a << \mu_s$.

In this case, the tissue incident photons, produced by a short laser pulse, will be engaged in a stochastic propagation process through tissue of the «random walk» type which will determine their transfer from the initial place, where the photon concentration (i.e., intensity) is high, towards adjacent tissue regions where this concentration is lower or even zero.

These simplifying hypotheses permit to treat, by similitude, the stochastic photon propagation (as a result of multiple scattering) by the prism of *simple diffusion theory* of a scalar or vectorial magnitude along the gradient of a generalised force conjugated to its flux.

In this treatment, the flux of a physical magnitude, X, denoted by J_X, through a surface of area, S, is proportional to the gradient, ∇, of conjugated «force», Y_X:

$$J_{X,S} = - C_X S \nabla Y_X \tag{6}$$

where C_X represents a specific proportionality constant.

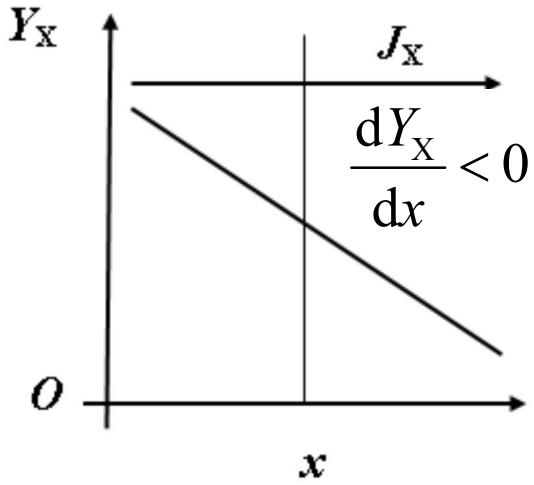

Fig. 1. Schematic representation of the flux of the magnitude, X, and of its conjugated force gradient, dY_X/dx, along the Ox axis.

To simplify the things, without affecting the general treatment, we shall follow the diffusion of the magnitude, X, along the direction, Ox (Fig. 1). Consequently, the equation (6) will have a simplified form:

$$J_{X,S} = -C_X S \frac{dY_X}{dx} \tag{7}$$

In many practical situations it is more concluding the *specific flux*, that is, the *flux density*, j_X (independent of the magnitude of the area, S) given by:

$$j_X = \frac{J_X}{S} = \frac{dY_X}{Sdt} \tag{8}$$

From the relations (7) and (8) it results the mathematical expression of the flux density of a magnitude, X, consequence of a conjugated force gradient, dY_X/dx:

$$j_X = -C_X \frac{dY_X}{dx} \tag{9}$$

For instance, in the particular case of substance diffusion, the flux density of mass (m, in kg) and substance quantity (ν, in mol) has the expressions, known as the *Ficks first law*:

$$j_m = \frac{dm}{Sdt} = -D\frac{d\rho}{dx}; \quad j_\nu = \frac{d\nu}{Sdt} = -D\frac{dC}{dx} \tag{10}$$

where D represents the *diffusion coefficient*, while $d\rho/dx$ and dC/dx represent the density and respectively, concentration gradients of the diffusible substance.

In the concrete case of stochastic photon scattering (i.e., photon diffusion) the Fick first law is expressing the density of the radiant flux, j_F, as function of the gradient of the *radiant flux* (i.e., *fluence rate*, F), according to the formula (9):

$$j_F = -D' \frac{dF}{dx} \tag{11}$$

where F represents the radiant flux of the photons ($[F]_{SI} = Jm^{-2}s^{-1} = Wm^{-2}$), and D' is the *diffusion constant* which, in the case of a tissue, is characterized by the anisotropy factor, g, according to the formula:

$$D' = \frac{1}{3\mu_s(1-g)} \equiv \frac{1}{3\mu_s'} \tag{12}$$

where μ_s' represents the so-called *reduced scattering coefficient.*

One can easily notice that, in the case of isotropic media (i.e., $g = 0$), the diffusion constant represents (see the formula (3)) a third of the *natural attenuation length,* δ.

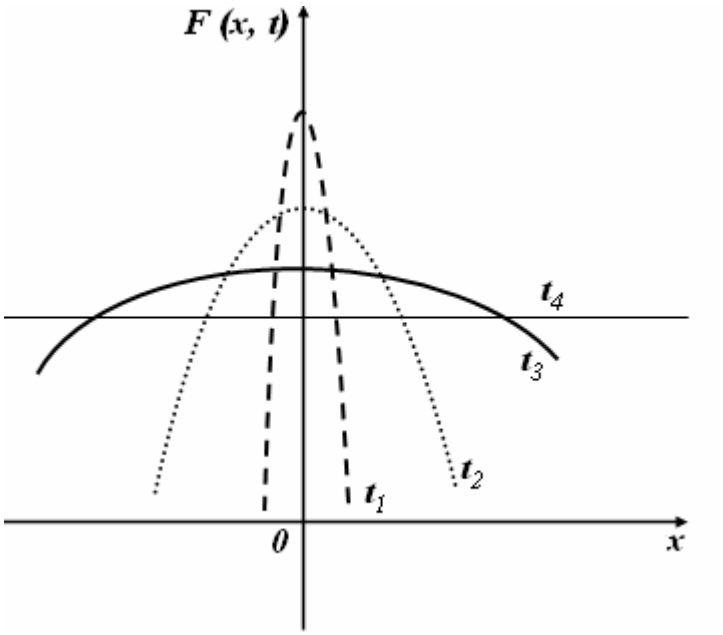

Fig. 2. Qualitative space-temporal dependence of radiant flux (i.e., the fluence rate): $t_1 < t_2 < t_3 < t_4 < \infty$.

Going further with the similitude between the photon scattering and substance diffusion, one can write the correspondent of the local *Fick second law*, in its one-dimensional variant:

$$\frac{\partial F(x,t)}{\partial t} = cD\frac{\partial^2 F(x,t)}{\partial x^2} \tag{13}$$

where c represents the photon speed in vacuum.

The solution, $F(x,t)$, of Eq. (13), in the case of a point source, which deposits, at a time, t, in the point, x, the energy, E_0, is given by [9]:

$$F(x,t) = c\,E_0\frac{e^{-\frac{x^2}{4cDt}}}{(4cDt)^{\frac{3}{2}}}, \tag{14}$$

a very useful relation in the study of photon interaction, by multiple scattering, with cells and tissues.

The solution (14) of Eq. (13) is an even function of spatial coordinate, that is, $F(x,t) = F(-x,t)$, which demonstrates (Fig. 2) that the *fluence rate* is symmetrical (in fact, in 3D, it has a radial symmetry) and, in a given point tissue location, it is decreasing exponentially in time.

LASER RADIATION ABSORPTION BY CELLS AND TISSUES

As a consequence of the absorption process of laser radiations by the enormous variety of molecules and macromolecules (components of the cells and tissues) a cascade of events are simultaneously taking place followed by cellular and tissular effects with positive or negative consequences on the whole body.

The post irradiation effects can be, *grosso modo*, classified in two categories: a) *radiative effects* (e.g., fluorescence and phosphorescence) not important for the present study, and b) *non radiative* (*thermal* and *non thermal*) important for this work.

The thermal effects, as a result of vibrational relaxation of the excited molecules and of the internal conversion (IC) are, in their turn, of two types [5]:

1) *Localized*, when a local heating of the biological medium is produced, followed by one of the phenomena: *coagulation* (when $t \sim 60$ °C), *vaporization* (when $t \sim 100$ °C), *carbonization* (when $t \sim 150$ °C) or *melting* (when $t >> 300$ °C, for instance, in the case of dental enamel);

2) *Delocalised*, when the radiation energy «deposited» as heat, is propagating from the place of the radiation-tissue impact: step by step (i.e., by *thermal diffusion*), by *convection* or by a *radiative process* (via infrared radiation) to regions more or less away, explaining thus the «distance effects » of the incident laser radiations.

The non thermal effects are mediated by *triplet* states of the excited molecules and consist in: photo additions (e.g., photo dimerisations) creating crosslinks between proteins and DNA, photo fragmentations, photo oxidations, photo hydrations, isomeriations, dimerisations, trimerisations, etc. [8, 16, 17].

HEAT TRANSPORT MODEL IN THE THERMAL INTERACTION

In the present model, we shall take into account the special case of the tissues into which, on the contrary to the above analysis, the photon multiple scattering is negligible as compared with their absorption ($\mu_a >> \mu_s$).

We shall make the additional assumptions: the energy deposition (due to the photon absorption) is not able to induce denaturation, vaporization, carbonization (combustion) or melting of the tissue, but only a local heating.

Heat generation, consecutive to the impact laser radiation-tissue, is determined, on the one hand, by the laser radiation parameters (i.e., wavelength, beam intensity, pulse duration) and, on the other hand, by the tissue parameters (i.e., scattering and absorption coefficients, anisotropy factor).

In exchange, the heat transfer by conductibility, convection and thermal radiation is solely characterised by tissue parameters like *specific heat*, *c*, and *thermal conductivity*, *k.*

For many tissues, one can find acceptable empiric approximations both for specific heat and thermal conductivity as functions of *tissue density*, ρ [20]:

$$c(\rho) = (1.55 + 2.8\ \rho_a/\rho);\ \ [c]_{SI} = \mathrm{J\,kg^{-1}K^{-1}} \tag{15}$$

$$k(\rho) = (0.06 + 0.57\ \rho_a/\rho);\ \ [k]_{SI} = \mathrm{W\,m^{-1}K^{-1}} \tag{16}$$

where ρ_a represents the liquid water density at the mammal body temperature (i.e., 37 °C).

The two above parameters can be incorporated into a single one, denoted by κ and known as *temperature conductivity coefficient* [15]:

$$\kappa = \frac{k}{\rho c} \tag{17}$$

The advantage of using the temperature conductivity coefficient, κ, resides on its very weak variation from tissue to tissue: the decrease of temperature conductivity coefficient, due to the decrease of the tissue water content, is compensated by the decrease of the tissue specific heat. Therefore, one can take a universal value for all tissues: $\kappa = 1.4 \times 10^{-7}$ m^2/s.

Heat propagation by conductibility represents the major mechanism by which heat is dissipated from the tissular impact point to the adjacent non irradiated regions.

Also, in this case, one can use the general transfer equation (9) in which will appear the heat flux density, j_Q, and its conjugated "force" (i.e., T), resulting a similar relation to Fick first law (in one-dimensional case):

$$j_Q = -k\frac{\partial T}{\partial x} \tag{18}$$

On the other hand, from the *continuity equation*, it results that the heat flux divergence, j_Q, depends on the heat variation of the unit volume, in a unit of time: $\partial q/\partial t$:

$$\nabla j_Q = -\frac{\partial q}{\partial t} \tag{19}$$

In one-dimension, Eq. (19) becomes:

$$\frac{\partial j_Q}{\partial x} = -\frac{\partial q}{\partial t} \tag{20}$$

One can also write the successive relations:

$$\frac{\partial q}{\partial t} = \frac{\partial Q}{V\partial t} = \frac{mc\partial T}{V\partial t} = \frac{\rho \ \ c\partial T}{\partial t} \tag{21}$$

Now, combining equations (20) and (21), one obtains:

$$\frac{\partial j_Q}{\partial x} = -\frac{\rho c\partial T}{\partial t} \tag{22}$$

Finally, from Eqns. (17), (18) and (22) it results the partial derivative equation which describes both the space and the time evolution of temperature, in the absence of a heat source:

$$\frac{\partial^2 T(x,t)}{\partial x^2} = \frac{1}{\kappa}\frac{\partial T(x,t)}{\partial t} \tag{23}$$

which is similar to the wave equation, but this time to a temperature wave equation.

In the presence of a heat source, h, the propagation equation (23) must be modified, in order to include the source contribution.

For the radiant flux, *I(x,t),* which is propagating in a non scattering medium, in *Ox* direction, the local heat deposition*, h(x,t)*, in unity of times and unity of volume, is given by:

$$h(x,t) = -\frac{\partial I(x,t)}{\partial x} \tag{24}$$

From the Lambert law (1) applied to a particular case of an absorbing but non reflecting (R_F = 0) and non scattering (μ_s = 0) medium, and from relation (24) it results:

$$h(x,t) = \mu_a I(x,t) \tag{25}$$

If one takes into account the heat source, *h(x,t),* then, the propagation equation (23), in the *Ox* direction (parallel to the incident beam), is of the form:

$$\frac{\partial^2 T(x,t)}{\partial x^2} = \frac{1}{\kappa}\left[\frac{\partial T(x,t)}{\partial t} - \frac{\mu_a I(x,t)}{\rho c}\right] \tag{26}$$

which can be adapted in the case of heat propagation into isotropic media.

The equation (26) is similar to other diffusion equations deduced under constant thermal properties and steady heat generation [7, 14].

In the above hypotheses on heat propagation, the solution of the partial derivative equation (26) has the form [15]:

$$T(x,t) = \frac{\mu_a}{\rho c \ \ (4\pi\kappa)^{\frac{1}{2}}}\int_0^t\int_{-\infty}^{+\infty}\frac{I(x',t')}{(t-t')^{\frac{1}{2}}}\exp\left[-\frac{(x-x')^2}{(t-t')}\right]dx'dt' \tag{27}$$

The solution (27) permits to follow heat propagation (i.e. temperature evolution) into a particular tissue, provided that the following parameters of the tissue are known: *absorption coefficient* (μ_a), *density* (ρ), *specific heat (c), temperature conductivity coefficient* (κ) and if the *intensity*, *I(x,t),* of a radiant flux is known, too.

The knowledge of modality of the heat propagation into tissues, from the "point" of laser radiation impact to the vicinal zones or the far ones, could explain the "distance action" of laser radiation.

For example, one can appreciate *the natural penetration depth*, x_p, as a function of time, that is, the distance on which the temperature is decreasing *e* times:

$$x_p(t) = (4 \quad \kappa \quad t)^{\frac{1}{2}} \tag{28}$$

It can be deduced from (28) that, if in t = 1 μs, the heat diffuses, in water, on a distance of 0.7 μm (as it is experimentally established). In the time interval, t' = 1 s = 10^6 t, the heat will migrate on a distance 10^3 times longer (i.e., 0.7 mm).

The last model predicts the spatial and temporal of temperature evolution into tissue if both the initial conditions and space limit conditions are well chosen, but the choice is not at all easy to be done. For this reason, it is necessary to emphasize that only an approximate temperature distribution can be estimated within this model.

Moreover, the effect of the laser radiation on the tissue depends on the initial temperature jump induced by the locally absorbed radiation. If this local temperature jump, ΔT, is minor (that is, $\Delta T < 5$ °C) the tissue is counterbalancing the thermal shock, by its thermoregulation mechanisms so that no notable effects will subsequently appear.

On the contrary, if the initial radiation impact induces a temperature jump, ΔT = (5–13) °C, this one is provoking a tissue *hyperthermia,* which will engender conformational changes of the proteins/enzymes, membrane alteration, or will provoke tissue necrosis (if hyperthermia lasts a few minutes).

Beyond the temperature of 50 °C, a severe attenuation of enzyme activity takes place, due to their denaturation, the enzymes preventing thus the cellular energetic transfer, blocking the defence natural processes and, implicitly, conducing to the death of an important cell fraction.

It results that, for an efficient laser therapy, the thermal effects induced by the laser radiations must be gentle, so that the local temperature jump does not exceed too much 42°C. Therefore, the irradiation parameters must be chosen so that the hyperthermia consecutive to laser irradiation is reversible (i.e., a recovering hyperthermia).

CONCLUSION

The photon multiple scattering by cells and tissues can be approached from the perspective of random propagation theory (i.e., simple diffusion) of a physical magnitude, provoked by its conjugated gradient. In this way, one can obtain, in a particular case of multiple photon scattering, the analytic expression of fluence rate for the isotropic tissues.

At the same time, the model of heat transfer, consecutive to thermal interaction of laser radiation with tissues, can describe the spatial-temporal evolution of the temperature in the adjacent regions of the target in which the energy is converted into heat.

The utilisation of laser radiation in the tissue treatment is expected to be efficient only if one succeeds to know the radiation scattering and absorption processes in order to adapt the laser radiation parameters to the particular treated tissue whose intrinsic parameters must be *a priori* known, too.

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