Light Guiding in Biological Tissue due to Scattering

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For a description of light propagation in biological tissue it is usually assumed that tissue is a random medium. We report a pronounced light guiding effect in cubes of human dentin that cannot be described by this standard model. Monte Carlo simulations which consider the microstructure of dentin are performed and successfully compared to experiments. Contrary to explanations so far, we show that light guiding is due to scattering by the tissue's microstructure. Exploiting this concept, light can be guided in arbitrary directions or locations without involving reflections or wave effects.

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Light propagation in biological tissue is usually described by the radiative transport equation assuming that biological tissue can be regarded as a (highly scattering) random medium [1,2]. In contrast to this assumption, biological tissues often exhibit an ordered microstructure, which has to be considered in the theoretical analysis of the light propagation. This is especially important for early diagnosis of diseases [3,4]. For example, most of the cancer types develop in the epithelium tissue layers, which are readily accessible for optical measurements.

Many tissues have a microstructure which consists of ordered elongated subunits, like the myofibrils in muscles or the collagen fibers in skin, tendons, or ligaments. These tissues show an anisotropic light propagation, which cannot be described by the normally used isotropic random media models [5–10].

In the case of dentin the anisotropic effects have been known for at least three decades. For example, it was shown that dental disks can magnify images [11] or that light is predominately conducted from the enamel-dentin-junction to the pulp [12]. In the literature, it was usually assumed that the anisotropic light propagation in dentin is due to wave guiding or fiber optic effects in the tubular microstructure of dentin [11]. Contrarily, in this Letter we show that the light guiding effect is caused by scattering on the tissue's microstructure.

We performed measurements on cubes of dentin which show a pronounced anisotropic light propagation that has not been reported in the literature to our knowledge. When a cube of dentin is illuminated perpendicularly onto a certain plane (the incident plane), almost all light is transmitted from one lateral plane (the effect plane) or remitted from the incident plane, see Fig. 1. Much less light is transmitted from the other planes.

In terms of light propagation, the microstructure of dentin is dominated by the tubules, which are cylindrical channels that run from the pulp to the enamel-dentin junction, see Fig. 2(a).

Figure 2(b) shows schematically the course of the tubules in the dentin. The cubes were cut from the periphery of the coronal dentin, where the tubules have a curved course. Thus, the tubules run from the incident plane of the cubes to the effect plane, see Fig. 2(b).

In order to show that the anisotropic light propagation is due to multiple scattering, we performed measurements of the transmitted and remitted light with a slow scan CCD camera and compared it to Monte Carlo simulations which consider the course of the tubules. The Monte Carlo method is a numerical solution of the radiative transport equation. It considers stochastically the transport of energy packages (here called photons) through the turbid medium.

The dental cubes were perpendicularly illuminated with a focused unpolarized HeNe laser (beam diameter: 10 μm, wavelength: $\lambda = 633$ nm) at the center of the incident plane, and the spatially resolved transmittance (or reflectance) from all six planes was measured with the CCD camera (Peltier cooled, 16 bit). We note that we also performed measurements with linearly polarized light and that we found very similar light guiding effects as for unpolarized light. Experiments on about 10 dental

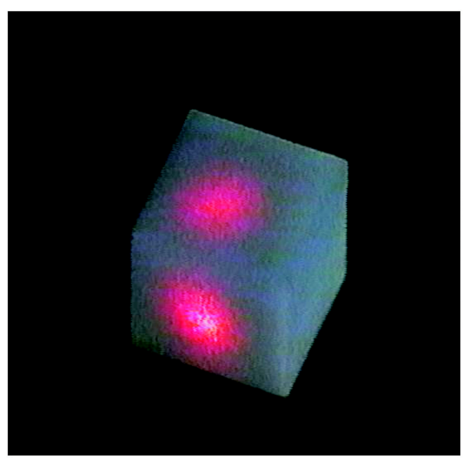

FIG. 1 (color online). Image of a cube of dentin (side length 2 mm) illuminated perpendicularly onto the center of one of the cube's planes (the front plane in the picture) by a HeNe-laser. Almost all light is transmitted from one of the lateral planes (the upper plane in the picture).

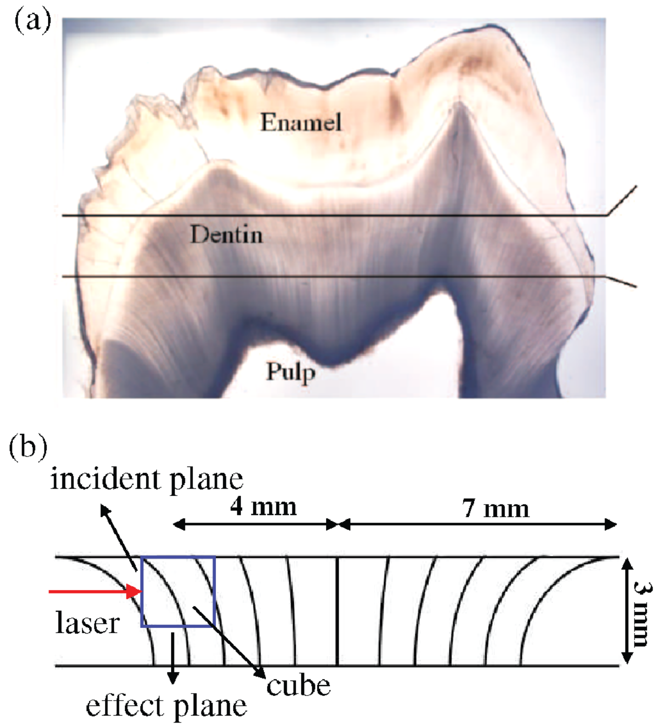

FIG. 2 (color online). (a) Microscopic image of a vertical slice of a human tooth showing the course of the tubules in the dentin; (b) model of the course of the tubules used in the simulations including the position of the cube and the incident laser beam.

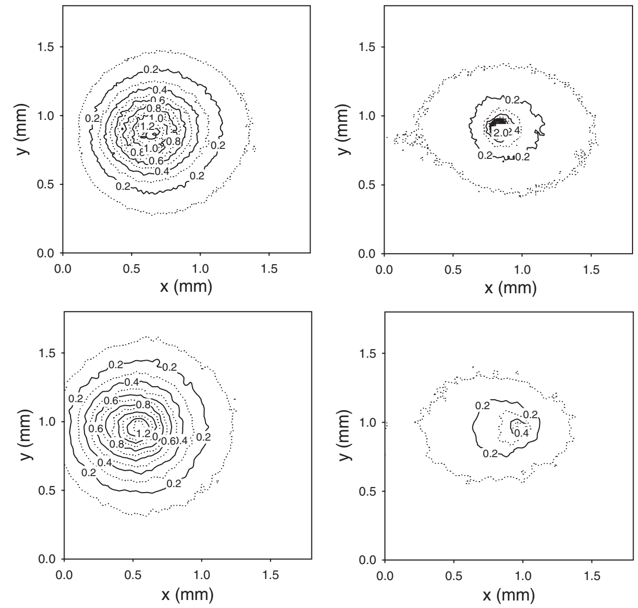

FIG. 3. (a) Spatially resolved transmittance from the effect plane, and (b) spatially resolved reflectance from the incident plane measured with the CCD camera. (c) Spatially resolved transmittance from the effect plane, and (d) spatially resolved reflectance from the incident plane calculated with the Monte Carlo method. The units for all figures are $1/\mathrm{mm}^2$.

cubes with side lengths between 1 and 3 mm were performed. During the measurements the cubes were placed in water to avoid alterations of the optical properties due to drying effects. We found for all cubes that most of the light is remitted from the incident plane or is transmitted from the effect plane. The transmittance from the other four planes was each less than 10% of that transmitted from the effect plane (data not shown). Figure 3(a) shows the spatially resolved transmittance from the effect plane and Fig. 3(b) the spatially resolved reflectance from the incident plane (right) of the dental cube shown in Fig. 1. Both figures were scaled by the same multiplicative factor to enable comparison with the theoretical data.

The intensity of the light transmitted from the effect plane is higher than that remitted from the incident plane. Solely, at the area where the laser beam is incident the reverse is true, because of the reflected light at the surface of the incident plane. We note that in Fig. 1 the intensity of the light transmitted from the effect plane is smaller than that remitted from the incident plane. This is because the measurement shown in Fig. 1 was performed in air. On air the scattering of dentin is increased due to drying effects, and, thus, the remitted light is increased and the transmitted light is decreased.

In order to explain these experimental data, we performed Monte Carlo simulations which consider the microstructure of dentin. As in the experiment, the cube's side length was 2 mm. The beam having a diameter of 10 μm was incident perpendicular to the center of the incident plane. The tubules in dentin have a cylindrical shape with a diameter of about 2 μm and a length of several mm. Thus, for calculation of the scattering functions of the tubules we solved the Maxwell equations for the scattering by an infinitely long cylinder for arbitrary incident angles [13] assuming a refractive index of 1.33 for the cylinder and of 1.52 for the surrounding material. These scattering functions were implemented in the Monte Carlo method [6]. The concentration of the tubules was set to $3 \times 10^4\ \mathrm{mm}^{-2}$, a typical value found in dentin. We assumed independent scattering by the tubules. For the course of the cylinders we used a simple model which approximates the microstructure of dentin. In the center of the dentin the tubules are straight vertical lines and at a distance of 7 mm from the center the course is a quarter circle, see Fig. 2(b). In between, the angles of the tubules are steadily changed from a quarter circle to a straight line. The course of the tubules at the six surfaces of the dental cube shown in Fig. 1 was observed with a microscope using an external oblique illumination. On the basis of these investigations the course of the tubules agrees with that of our model, if the center of the cube is positioned approximately 4 mm away from vertical course of the tubules, compare Fig. 2(b).

Besides the tubules, the collagen fibers have to be considered for the calculation of the light propagation in dentin. We assumed that the directions of the collagen fibers in dentin are isotropically distributed. Therefore, their optical properties are independent on the photons' incident directions. The scattering coefficient was set to $\mu_s' = 0.4\ \mathrm{mm}^{-1}$ considering the concentration and the refractive index of collagen fibers in dentin. As a scattering function, a Henyey-Greenstein function with an anisotropy factor of $g = 0.8$ was used. The absorption coefficient was assumed to be $\mu_a = 0.01\ \mathrm{mm}^{-1}$, a typical value for low-absorbing tissue in the red wavelength range. Since the absorption coefficient is low compared to the scattering coefficient of dentin ($\mu_s \approx 100\ \mathrm{mm}^{-1}$), the precise value of μ_a does not noticeably influence the results. For the refractive index of the cubes we used $n = 1.5$ (dentin) and of the surrounding medium $n = 1.33$ (water).

The spatially resolved transmittance from the effect plane and the spatially resolved reflectance from the incident plane obtained from the Monte Carlo simulations are shown in Fig. 3(c) and 3(d), respectively. All the main features of the experiments shown in Fig. 3(a) and 3(b) are reproduced by the simulations. As in the experiments, most of the light is transmitted from the effect plane, and also the peak intensity of the effect plane is shifted from the center to the left in direction to the incident plane. Also, a similar spread of the incident small laser beam can be seen at the effect plane. In addition, the transmittance from each of the other planes is smaller than 10% of that of the effect plane (data not shown). Solely, in the simulated reflectance data from the incident plane the intensity peak caused from the surface reflection found in the experiments cannot be seen in Fig. 3(d), because the reflection was not considered in the simulations due to the unknown roughness of the cube's surface.

In order to understand the anisotropic light propagation in the dental cube, the scattering of light by an infinitely long cylinder has to be considered. The scattered light is confined in a cone, which is oriented around the cylinder axis having an opening angle of ξ, where ξ is the angle between the incident beam and the cylinder, see Fig. 4(a). Figure 4(b) illustrates the propagation of a photon through the cube schematically taking multiscattering into account.

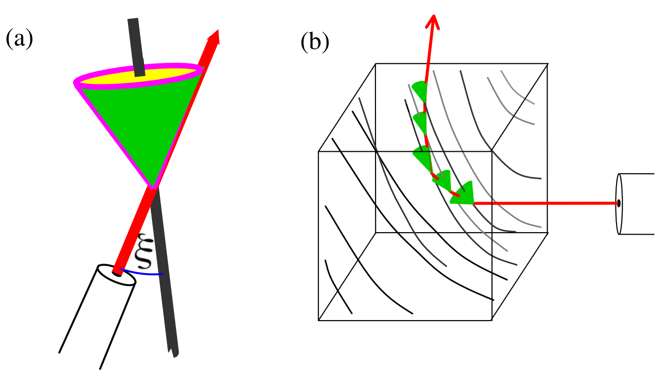

FIG. 4 (color online). (a) Light scattering by an infinitely long cylinder. (b) Schematic propagation of a photon through the dental cube.

First, the incident photon is scattered by a tubule. The scattering angle is small, because ξ is small. Thus, the photon will be scattered approximately in forward direction. We note that the smaller ξ the more isotropic is the scattering probability around the scattering cone [6]. Next, the photon is scattered by another tubule nearby, and again the scattering direction is close to that of the tubule's direction. Thus, as the tubules' direction is continuously changed, the photon's path follows approximately the overall directions of the tubules.

To demonstrate this light guiding effect more quantitatively, we present in Fig. 5(a) Monte Carlo simulations of photon paths trough a dental cube with side lengths of 2 mm. The paths of eight photons are shown. Each scattering interaction is indicated by a circle. The paths are projected onto a plane, which x axis and y axis lie in the incident plane and in the effect plane, respectively. In these

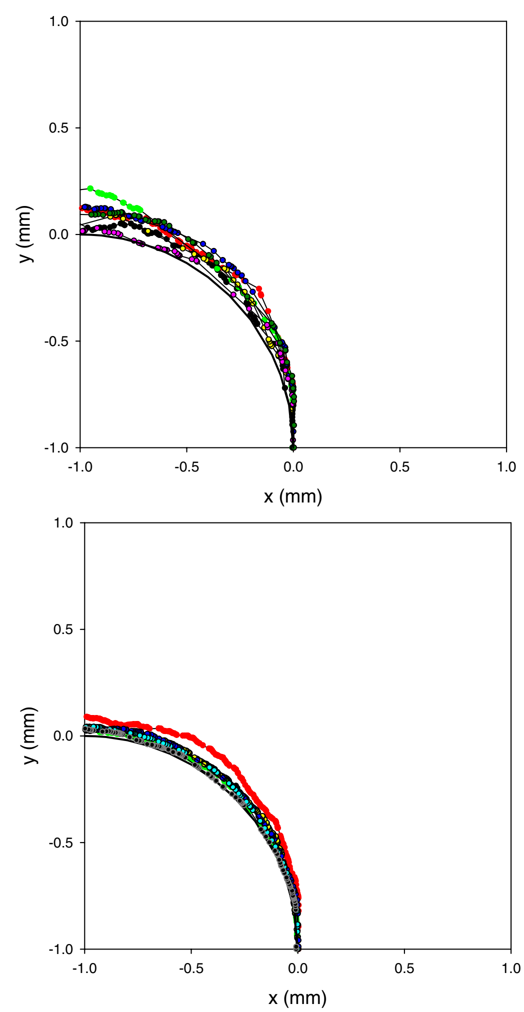

FIG. 5 (color online). (a) Path of eight photons through a dental cube with side length of 2 mm. Its microstructure exists of tubules that form a quarter circle with a radius of 1 mm; (b) same as (a) but the scattering coefficient is increased by a factor of 4.

calculations it is assumed that the course of the tubules has the form of quarter circles with a radius of 1 mm, and the centers of the circles lie on the line defined by the points ($x = -1$ mm, $y = -1$ mm) and ($x = 1$ mm, $y = 1$ mm). One of these quarter circles [that from ($x = 0$ mm, $y = -1$ mm) to ($x = -1$ mm, $y = 0$ mm)] is depicted in the graphs (line without symbols). It can be seen that the photons travel approximately along the course of the tubules. In average a photon is about 50 times scattered until it is transmitted. At the entrance of the light into the dentin ($x = 0$ mm, $y = -1$ mm) the photons' paths deviate somewhat from the course of the tubules, because here the directions of the tubules and of the incident photons are almost parallel and, thus, the scattering coefficient is low. Because the probability of a scattering interaction is low, the light propagates a longer distance along the incident direction. Thus, if the scattering coefficient or the diameter of the course of the tubules is increased, the light paths will be closer to the tubules' course.

For the simulations shown in Fig. 5(b) we increased the scattering coefficient by a factor of 4. As expected, the photons' paths are now closer to that of a quarter circle.

Additionally, we performed similar experiments with cubes cut from the center of the dentin where the tubules form almost straight lines, see Fig. 2. For perpendicular illumination in the enamel-to-pulp direction the beam is focused by the scattering of the tubules when transmitted from the opposite plane, whereas for illumination in the pulp-to-dental direction it is defocused. Again, we could reproduce the experimental results by Monte Carlo simulations.

In summary, we performed measurements on dental cubes, which were illuminated perpendicularly onto the center of a plane, and found that almost all transmitted light emerges from one lateral plane. We showed that this pronounced anisotropic light propagation can be described by multiple light scattering caused by the cylindrical microstructure of dentin. In order to exclude an explanation based on a fiber optic effect due to total reflection in the peritubular dentin, we investigated the transmittance from dental slabs having different thicknesses between 20 μm to 1 mm with an optical microscope. We could not observe any fiber optic effects either in the peritubular dentin nor in the tubules.

We note that many tissue types have a microstructure which also consists of aligned cylindrical scatterers, like the myofibrils in muscle, the collagen fibers in ligament or tendon, the prims in enamel, or the nerve fibers in nerve tissue. Thus, similar light guiding effects caused by scattering are expected in other biological tissues.

In addition, we showed that it is in principle possible to guide light in arbitrary directions and to arbitrary locations due to scattering on accordingly formed cylindrical structures. Regarding the multifarious possibilities nowadays to produce structured materials this effect could be used for a variety of technical light guiding applications.

In conclusion, we showed that the light propagation in macroscopical samples of dentin (dimensions >1 mm) can be quantitatively described with models based on its microstructure. The development of similar models for other biological tissues of the human body is an important task for future research projects, which will have a large impact for therapeutic and diagnostic applications.

Finally, in our opinion this new found effect "light guiding by scattering" might be an old principle well known by nature. In general, all light guiding effects found in biological media should be reviewed in order to verify if they are due to a fiber optic effect or due to scattering. We checked a few of them, and we found that the light guiding in wood is due to scattering and most probably this is also the case for seeds. In addition, the often discussed light guiding in the fur of polar bears might be caused by scattering by the hairs in a similar way as shown for the tubules in dentin. For these examples it has been discussed in literature that the observed light guiding is caused by fiber optic effects [14–16]. It seems reasonable that evolution used the presented effect to develop an "inexpensive" light guiding mechanism, for example, for low light conditions like in the depth of the sea.

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