

Square law between spatial frequency of spatial correlation function of scattering potential of tissue and spectrum of scattered light

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Abstract. The significance of beam condition for scattered light from random tissue is analyzed for a practical optical imaging system with a finite numerical aperture. It is shown that in the transmitted illumination case, the information carrying part of the spectrum of the scattered light is proportional to the square of the spatial frequency of the spatial correlation function of the scattering potential of the medium. The result may be helpful in interpreting images obtained with microscopes in biological studies.

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One important way to obtain information about an object is to measure the light scattered or transmitted by the object. However, the basic question about these techniques is what information about the 3-D structure of the object can be, in principle, obtainable. Wolf answered this question in 1969.¹ He showed that within the first Born approximation, when the object is illuminated by a plane monochromatic light, incident in all possible directions, and the complex amplitude of the scattered light is measured in the far zone of the scatterer in all possible directions, only those 3-D Fourier components of the scattering potential, for which the spatial periods Δx , Δy , and Δz , satisfy the inequality

$$\frac{1}{(\Delta x)^2} + \frac{1}{(\Delta y)^2} + \frac{1}{(\Delta z)^2} \leq \left(\frac{2}{\lambda}\right)^2, \quad (1)$$

can be deduced from the measurements of the scattered light, where λ is the wavelength of the illuminating light.

In practice, such as in biological tissue imaging, the tissue under test is usually illuminated and the scattered (or reflected) light is detected within a limited range of incident angles, which is determined by the numerical aperture (NA) of the lens. When the NA of the lens is small, both the illuminated and reflected light can be approximated by a parallel beam. It is then desirable to evaluate at what level the 3-D

structure information of biological tissue is accessible in these cases, especially in high resolution microscopy. In a recent paper, a necessary and sufficient condition for a beam to retain its beamlike form after being scattered on a stochastic medium was reported.² In this work, we employ this result to analyze the possible Fourier components of the scattering potential of the object that can be obtained with a practical optical imaging system in biological study.

Tissue is a complex system in which light is scattered in propagation due to the spatial fluctuation of its refractive index, which can be written as a sum of its mean $\langle n(\vec{r}) \rangle$ and a varying part $\delta n(\vec{r})$, $n(\vec{r}) = \langle n(\vec{r}) \rangle + \delta n(\vec{r})$, where \vec{r} denotes the position within the tissue. It has been shown that within the first Born approximation, the spatial distribution of the scattering field is determined by the scattering potential of the tissue, which can be expressed as^{1,2}

$$F(\vec{r}, \omega) = \frac{1}{4\pi} \left(\frac{\omega}{c}\right)^2 [n^2(\vec{r}, \omega) - 1] = \frac{1}{4\pi} \left(\frac{\omega}{c}\right)^2 [\langle n(\vec{r}, \omega) \rangle^2 + 2\langle n(\vec{r}, \omega) \rangle \delta n(\vec{r}, \omega) - 1], \quad (2)$$

where c is the speed of light in vacuo. In Eq. (1), the second term $[\delta n(\vec{r}, \omega)]^2$ has been neglected due to the fact that $[\delta n(\vec{r}, \omega)]$ is very small compared with $\langle n(\vec{r}) \rangle$.³ The spatial correlation function of the scattering potential can then be expressed as⁴

$$C_F(\vec{r}_1, \vec{r}_2, \omega) = \langle F^*(\vec{r}_1, \omega) F(\vec{r}_2, \omega) \rangle = \frac{1}{16\pi^2} \left(\frac{\omega}{c}\right)^4 [\langle n \rangle^4 - 2\langle n \rangle^2 + 1 + 4\langle n \rangle^2 \langle \delta n(\vec{r}_1, \omega) \delta n(\vec{r}_2, \omega) \rangle], \quad (3)$$

where the angle brackets represent the statistical average, taken over the ensemble of the scatterer, $\langle \delta n(\vec{r}, \omega) \delta n(\vec{r}_2, \omega) \rangle$ is the refractive index correlation function, and from experimental data its spectrum can be expressed in the form of^{3,5}

$$\Phi(K) = \frac{\langle \delta n^2 \rangle L_0^3 \Gamma(m)}{\pi^{3/2} [\Gamma[m - (3/2)]] (1 + K^2 L_0^2)^m}, \quad (4)$$

where L_0 is the outer scale of tissue index inhomogeneities and describes the refractive index correlation length, m is a parameter ($1 < m < 2$),^{3,5} and $\langle \delta n^2 \rangle$ is the variance of the tissue refractive index fluctuation. Hence the Fourier transform of Eq. (3) is

$$\tilde{C}_F(K, \omega) = \left(\frac{\omega}{c}\right)^4 \frac{\langle n \rangle^2 \langle \delta n^2 \rangle L_0^3 \Gamma(m)}{4\pi^{7/2} [\Gamma[m - (3/2)]] (1 + K^2 L_0^2)^m}. \quad (5)$$

Here the constant terms in Eq. (3) that correspond to $\delta(K)$ in the spectral domain have been neglected, where $\vec{K} = k(\vec{s} - \vec{s}_0)$. Equation (5) can be rewritten as

$$\tilde{C}_F(K, \omega) = N(\omega) (1 + K^2 L_0^2)^{-m}, \quad (6)$$

where

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$$N(\omega) = \left(\frac{\omega}{c}\right)^4 \frac{\langle n \rangle^2 \langle \delta n^2 \rangle L_0^3 \Gamma(m)}{4\pi^{7/2} |\Gamma[m - (3/2)]|}. \quad (7)$$

In order that the incident light beam is still of the beam-like form after scattering, $\tilde{C}_F(K, \omega)$ must satisfy the condition²

$$\tilde{C}_F(K, \omega) \approx 0, \quad \text{unless } |\vec{K}|^2 \ll k^2, \quad (8)$$

where $k = \omega/c$. For tissue, L_0 is in the order of 4×10^{-6} m, and k is in the order of $7 \times 10^6 \text{ m}^{-1}$ ($\lambda = 0.83 \text{ }\mu\text{m}$). The relation $|\vec{K}|^2 \ll k^2$ means $K^2 L_0^2 < 1$. By applying the binominal expansion to Eq. (6) and retaining the first two terms of the expansion, we have

$$\tilde{C}_F(K, \omega) = N(\omega)(1 - mL_0^2 K^2). \quad (9)$$

Now consider a random polychromatic plane wave incident on a tissue surface in a direction specified by a real unit vector \vec{s}_0 . The incident field may be represented by an ensemble $\{U^{(i)}(\vec{r}, \omega)\}$ that is statistically stationary. Each realization of $\{U^{(i)}(\vec{r}, \omega)\}$ can be regarded as the time-independent part of a monochromatic wave function, and can be expressed as $U^{(i)}(\vec{r}, \omega) = a(\omega)\exp(ik\vec{s}_0 \cdot \vec{r})$, where the (generally complex) amplitude factor $a(\omega)$ is a frequency-dependent random variable, and $k = \omega/c$ is the wave number. The spectrum of the light is an important statistical characteristic of light and is given by $S(\vec{r}, \omega) = \langle U^*(\vec{r}, \omega)U(\vec{r}, \omega) \rangle$. Under the first Born approximation, the spectrum of the scattered light in the far zone of the scatterer can be expressed as⁶

$$S^\infty(l\vec{s}, \omega) = V l^{-2} \tilde{C}_F(K, \omega) S^i(\omega), \quad (10)$$

where V is the volume of the scattering object, l is the distance from the observation point to the reference point, \vec{s} is the unit vector in the direction of scattering, and $S^i(\omega)$ is the spectrum of the incident field. On substituting Eq. (9) into Eq. (10), we have

$$S^\infty(l\vec{s}, \omega) = N'(\omega, l)(1 - mK^2 L_0^2) S^i(\omega), \quad (11)$$

where $N'(\omega, l) = N(\omega)V/l^2$. Because $\vec{K} = k(\vec{s} - \vec{s}_0)$, we have

$$K^2 = 4(\omega/c)^2 \sin^2(\theta/2) = 4k^2 \sin^2(\theta/2), \quad (12)$$

where θ is the scattering angle ($\vec{s} \cdot \vec{s}_0 = \cos \theta$). Equation (11) shows that at frequency ω , the spectrum of the scattered light in the far zone is proportional to $(1 - mK^2 L_0^2)$. Because of the condition in Eq. (8), in Eq. (13) it is required that $\sin^2 \theta/2 \ll 1$, and we have the approximate relation $\sin \theta/2 \sim \theta/2$. Equation (12) may then be simplified to

$$K \approx k\theta. \quad (13)$$

Equations (11) and (13) show that when scattered light behaves in a beam-like form, it can be regarded as composing plane waves traveling along directions with small angles with z axes. The larger spatial frequency components of the scattering potential propagate in larger angles with respect to the incident direction \vec{s}_0 (chosen to be the positive z axis.). The light corresponding to the zero frequency component [the constant term in Eq. (11)] travels along the z axis. For a lens with an angular semiaperture α , the highest spatial frequency of the spatial correlation function of the scattering potential of the object that can be collected is $K \approx k\alpha$. The constant term

in Eq. (11) may be regarded as representing the background. Hence it is concluded that the information carrying part of the spectrum of the scattered light is proportional to the square of the spatial frequency of the scattering potential of the medium.

A similar result has been obtained in the scalar diffraction theory of monochromatic light in paraxial approximation.⁷ It can be shown that the light field distribution $U(x, y, z)$ in a plane $Z = z$ can be expressed as a superposition of plane waves $\exp[j2\pi(\alpha x/\lambda + \beta y/\lambda)]$ with direction cosines $\alpha = \lambda f_x$, $\beta = \lambda f_y$, and $\gamma = [1 - (\lambda f_x)^2 - (\lambda f_y)^2]^{1/2}$, where f_x and f_y are the spatial frequencies of the Fourier component $\exp[j2\pi(f_x x + f_y y)]$. Hence when $|\lambda f_x| \ll 1$ and $|\lambda f_y| \ll 1$, the wave vectors of the corresponding plane waves have small angles with respect to the z axis. This is known as the paraxial approximation.

However, there exist two differences between the diffracted monochromatic light field distribution around the propagation direction, and the spatial distribution of the scattered light around the incident direction. First of all, in monochromatic light illumination, the corresponding plane waves carry the spatial information of the field distribution. For scattered light from a random illuminated by a random polychromatic plane wave, the plane waves provide the structural information about the scattering potential of the medium. Second, for scattered light there exists a simple relationship between the spectrum of the scattered light in the far zone and the spatial frequencies K . For the diffraction of monochromatic light there is no such simple relation.

In conclusion, it is found that in the transmitted illumination case, such as in inverted biological microscopes, there exists a very simple relationship between the spectrum of the scattered light and the spatial frequency component of the spatial correlation function of the scattering potential of the medium under investigation. This result can be useful in interpreting images obtained with microscopes in biological study or evaluating the performance of a microscopic imaging system.

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