A method for measuring the complex refractive index of a turbid medium

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ABSTRACT

Based on the heterodyne interferometry and Fresnel equations, an alternative method for measuring the complex refractive index of a turbid medium. A light beam is incident on the boundary between a right-angle prism and a turbid medium. The phase difference between s- and p- polarizations of the reflected light occurs. The phase difference depends on then incident angle and the complex refractive index of a turbid medium; their relation can be derived from Fresnel equations. The phase difference can be measured accurately with the heterodyne interferometry. Because there are two unknown parameters to be estimated, at least the phase differences under two different conditions should be measured. Then, these measured data are substituted into the derived relation, and a set simultaneous equation is obtained. If the simultaneous equation is solved, the complex refractive index can be estimated. Because the reflected light from the boundary is measured, the scattering noises coming from the turbidity of the tested medium can be greatly reduced. In addition, this method has some merits such as simple optical setup, high sensitivity, high stability, and suitability for a little amount of the tested medium in its native state (without dilution).

Keyword: heterodyne interferometry, complex refractive index, turbid medium.

1. INTRODUCTION

A turbid medium is an important biochemical medium. Its complex refractive index is not only related with concentration, temperature, pressure, and wavelength, but also depends strongly on its quality. For example, there is an obvious complex refractive index difference between normal and abnormal tissues, which might be caused by the diseases[1,2]. Hence the measurement of complex refractive index can be used as an alternative method to judge the quality of a turbid medium. There are several methods for measuring the complex refractive index such as R-versus-θ method (reflectance versus incident angle method)[3], critical angle method[4,5], and ellipsometry[6]. In those methods, the reflectances at several incident angles are measured, they are substituted into Fresnel equations[7] to calculate the complex refractive index. Because the scattering property[8] of a turbid medium, those methods are difficult to measure a turbid medium accurately.

To overcome the drawback, an alternative method for measuring the complex refractive index of a turbid medium is presented in this paper based on the heterodyne interferometric technique and Fresnel equations. As a light beam is incident on the boundary between a high refractive index prism and the tested turbid medium, the phase difference between the s- and p- polarization components of the reflected light occurs. It can be measured accurately with the heterodyne interferometric technique. Because there are two unknown parameters, two phase differences at two different incident angles should be measured. If these data are substituted into the special equation derived from Fresnel equations, then the complex refractive index can be obtained. Compared with the general biological assay[9], this method needs not to add any extra reagent to dilute the tested turbid medium for testing. So this method can avoid damaging the property of the sample. In addition, this method has several merits, including a simple optical setup, easy operation, high stability, high measurement accuracy, and rapid measurement. We demonstrate its feasibility.

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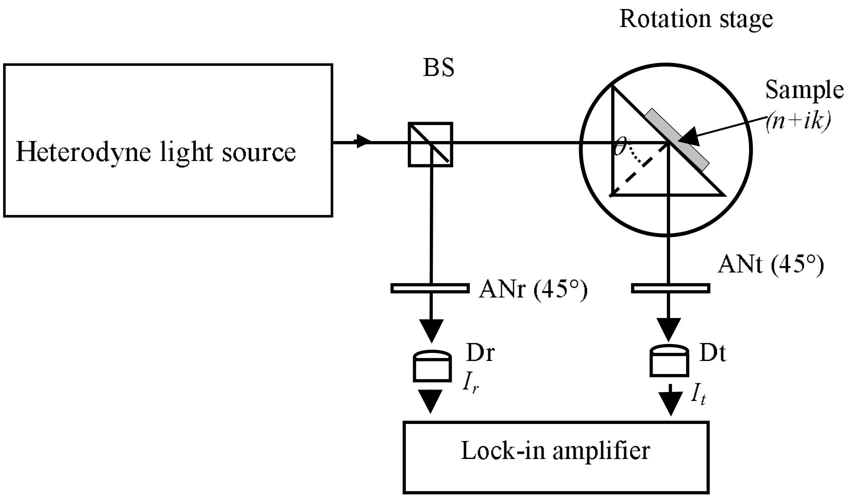
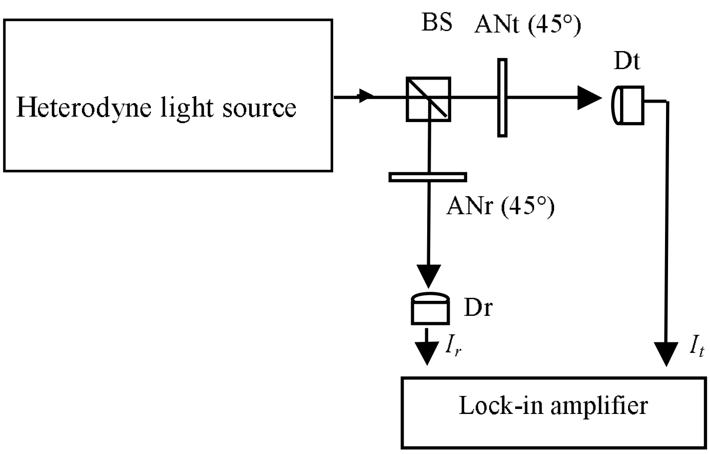
2. PRINCIPLE



(a)



(b)

Fig. 1 Schematic diagrams for measuring the phase differences owing to the reflections at (a) the boundary between a prism and a turbid medium, and (b) BS. BS: beam-splitter; AN: analyzer; D: photo detector.

The schematic diagram of this method is shown in Fig. 1(a). A light beam coming from a heterodyne light source is incident on a beam-splitter BS and divided two parts: the transmitted light and the reflected light. The transmitted light is incident at θ on the boundary between a high refractive index prism and the tested turbid medium. Both of them are located on a rotation stage. The light beam reflected from the boundary passes through an analyzer ANt and enters a photo detector Dt. If the amplitude of the light detected by Dt is E_t, then the intensity measured by Dt is $I_t = |E_t|^2$. Here, I_t acts as a test signal. On the other hand, the reflected light at BS passes an analyzer ANr and enters a photo detector Dr. If the amplitude of the light detected by Dr is Er, then the intensity measured by Dr is $I_r = |E_r|^2$. Here, I_r acts as a reference signal. Finally, these two signals are sent to a lock-in amplifier and the phase difference between them can be measured.

2.1 Heterodyne light source

The heterodyne light source consists of a linearly polarized laser light source, a half-wave plate H, and an electro-optic modulator EOM as shown in Fig. 2. EOM is driven by an external saw tooth voltage signal with angular frequency ω and amplitude $V_{\lambda/2}$, the half-voltage of EOM. That signal comes from a function generator FG and a linear voltage amplifier LVA. For convenience, the +z-axis is chosen to be along the light propagation direction and the x-axis is along the direction perpendicular to the paper plane. Let the laser light be horizontally linearly polarized, the fast axis of EOM and H be 45° and 22.5° with respect to the x-axis, respectively.

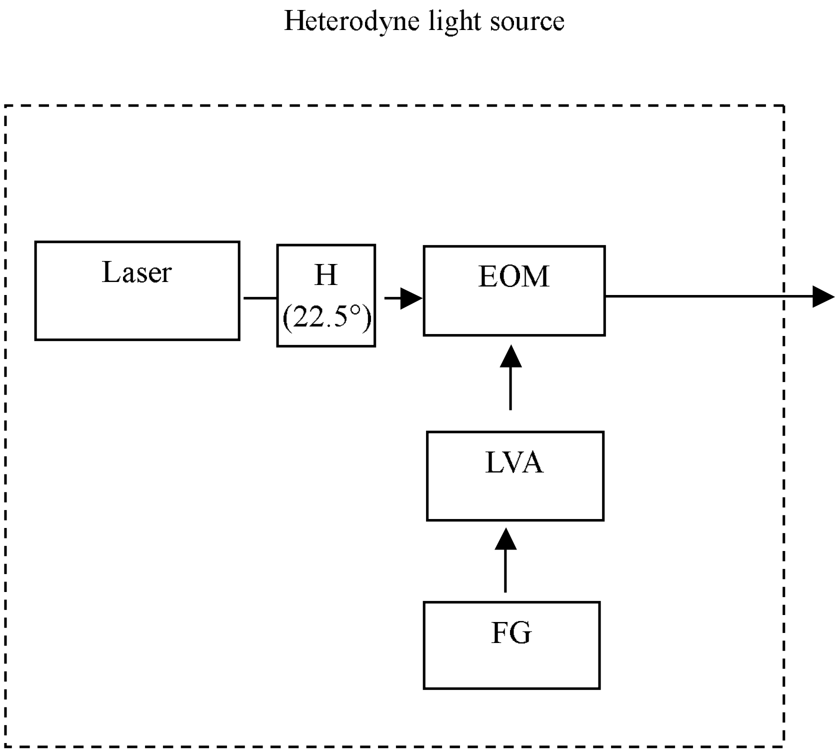


Fig. 2 Schematic diagram for the heterodyne light source. EOM: electro-optic modulator; LVA: linear voltage amplifier; FG: function generator.

2.2 Phase-difference between s- and p-polarizations of reflected light

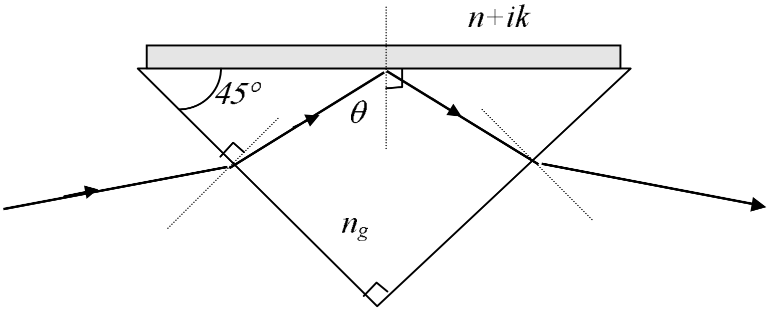


Fig. 3 The reflection at the boundary between a prism and a tested medium.

A ray of light is incident at θ on the base surface of a right-angle prism with refractive index n_g as shown in Fig. 3. At the base surface of the right-angle prism, there is a boundary between the prism and the turbid medium. Its complex refractive index is $n + ik$, where n is the refractive index and k is the extinction coefficient. According to the Fresnel equations, the amplitude reflection coefficients of s- and p- polarizations can be express as

$$r_s = \frac{\cos\theta - n_g(u+iv)}{\cos\theta + n_g(u+iv)} = |r_s|\exp(i\delta_s), \tag{1}$$

$$r_p = \frac{N^2\cos\theta - n_g(u+iv)}{N^2\cos\theta + n_g(u+iv)} = |r_p|\exp(i\delta_p), \tag{2}$$

respectively, where

$$u^2 = \frac{1}{2}\left\{\left(n^2 - k^2 - n_g{}^2\sin^2\theta\right) + \left[\left(n^2 - k^2 - n_g{}^2\sin^2\theta\right)^2 + 4n^2k^2\right]^{\frac{1}{2}}\right\}, \tag{3a}$$

$$v^2 = \frac{1}{2}\left\{-\left(n^2 - k^2 - n_g{}^2\sin^2\theta\right) + \left[\left(n^2 - k^2 - n_g{}^2\sin^2\theta\right)^2 + 4n^2k^2\right]^{\frac{1}{2}}\right\}, \tag{3b}$$

δ_s and δ_p are the phase shifts of s- and p- polarizations, and they can be expressed as

$$\delta_s = \tan^{-1}\left(\frac{2v\cos\theta}{u^2 + v^2 - \cos^2\theta}\right), \tag{4a}$$

$$\delta_{\mathrm{p}} = \tan^{-1}\left[\frac{2v\cos\theta\left[n^2 - k^2 - 2u^2\right]}{u^2 + v^2 - \left(n^2 + k^2\right)^2\cos^2\theta}\right], \tag{4b}$$

respectively. Hence, the phase difference of s-polarization relative to p-polarization can be written as

$$\phi = \delta_{\mathrm{s}} - \delta_{\mathrm{p}} = \tan^{-1}\left(\frac{ad - bc}{ac + bd}\right), \tag{5a}$$

where

$$\left.\begin{aligned} a &= 2v\cos\theta, \\ b &= u^2 + v^2 - \cos^2, \\ c &= 2v\cos\theta\left(n^2 - k^2 - 2u^2\right), \\ d &= u^2 + v^2 - \left(n^2 + k^2\right)^2\cos^2\theta. \end{aligned}\right\} \tag{5b}$$

2.3 Estimation of complex refractive index

If both the transmission axes of the analyzers AN*r* and AN*t* are located at 45° with respect to the x-axis, then we have

$$I_{\mathrm{r}} = \frac{1}{2}\left[1 + \cos\left(\omega t + \phi_{\mathrm{r}}\right)\right], \tag{6a}$$

and

$$I_{\mathrm{t}} = \frac{1}{2}\left[\frac{\left|r_{\mathrm{s}}\right|^2}{2} + \frac{\left|r_{\mathrm{p}}\right|^2}{2} + \left|r_{\mathrm{s}}\right|\left|r_{\mathrm{p}}\right|\cos\left(\omega t - \phi\right)\right], \tag{6b}$$

where ϕ_{r} is the phase difference between *s*- and *p*- polarizations that is due to the reflection at BS. The reference signal I_r and the test signal I_t are also the sinusoidal signals of a frequency difference ω. These two signals are sent to a lock-in amplifier, the phase difference

$$\phi' = \phi - \phi_{\mathrm{r}} \tag{7}$$

can be obtained. In the second measurement, let the transmitted light at BS enter photo detector Dt directly without the reflection in the right-angle prism, as shown in Fig. 1(b). The test signal still has the form of Eq. (6b) but this time with $\phi = 0$. Therefore the lock-in amplifier in Fig. 1(b) represents $-\phi_{\mathrm{r}}$. Substituting $-\phi_{\mathrm{r}}$ into Eq. (7), we obtain the phase difference ϕ.

From Eqs.(1)~(5), it is obvious that the phase difference ϕ is the function of *n, k,* and θ, and ϕ can be experimentally measured for a given θ. To evaluate the values of *n* and *k* we require two phase differences ϕ_1 and ϕ_2 that correspond to two incident angles, θ_1 and θ_2. Hence a set of simultaneous equations

$$\phi_1 = \phi_1\left(n, k, \theta_1\right), \tag{8a}$$

$$\phi_2 = \phi_2\left(n, k, \theta_2\right). \tag{8b}$$

is obtained. Because θ_1 and θ_2 are given, the two parameters, *n* and *k,* can be estimated by using the Numerical analysis[10]

$$\begin{pmatrix} n \\ k \end{pmatrix}_{m+1} = \begin{pmatrix} n \\ k \end{pmatrix}_{m} - \left.\begin{pmatrix} \frac{\partial \phi_1}{\partial n} & \frac{\partial \phi_1}{\partial k} \\ \frac{\partial \phi_2}{\partial n} & \frac{\partial \phi_2}{\partial k} \end{pmatrix}^{-1}\right|_{n,k} \cdot \begin{pmatrix} \phi_1 - \phi_1' \\ \phi_2 - \phi_2' \end{pmatrix}, \tag{8c}$$

where m is the number of calculated times. The complex refractive index of a turbid medium can be estimated by using this analysis.

3. EXPERIMENTS AND RESULTS

material	θ_1	θ_2	ϕ_1	ϕ_2	n	k	Δn	Δk
Milk	49.5°	50°	-9.92°	-15.68°	1.3464	0.0009	0.0002	0.0007
	50°	50.5°	-15.64°	-19.38°	1.3465	0.0008	0.0002	0.0007

Table. 1 Experimental conditions and measurement results

In order to show the feasibility of this method, we measured the complex refractive index of commercial whole milk at 25°C. The heterodyne light source consisting of a He-Ne laser with 632.8 nm wavelength and an electro-optic modulator EOM driven by a function generator FG and a linearly voltage amplifier LVA was used. The frequency difference between p- and s- polarizations was 1 kHz. A lock-in amplifier with resolution 0.01° (Model SR850, Stanford Research System) was used to measure the phase difference, and a personal computer was employed to record and analyze the data. A right-angle prism made of SF11 glass with refractive index n_g = 1.77862 and the tested medium were mounted on a high-precision rotation stage (SGSP-160YAW, Japan Sigma Koki Ltd.) with the angular resolution of 0.0025°. The experimental conditions and measured results are summarized in Table 1.

4. DISCUSSIONS

Because ϕ is the function of θ, we should choose two optimal incident angles to get better resolution. According to Ref. 11, the measured resolution is almost proportional to the integration result of the following integral

$$S = \int_0^\infty \int_0^\infty \left| \frac{\partial \phi_1}{\partial n} \frac{\partial \phi_1}{\partial k} - \frac{\partial \phi_2}{\partial n} \frac{\partial \phi_2}{\partial k} \right| dndk. \tag{9}$$

Substituting our experimental conditions into Eq. (9), we get that when the incident angles are in the neighborhood near the critical angle θ_c, the high resolution measurement can be achieved. Here the critical angle θ_c is defined as

$$\theta_c = \sin^{-1}\left(\frac{n}{n_g} \right). \tag{10}$$

In our experiments, we have $\theta_c = 49.2°$. Hence two conditions $\theta_1 = 49.5^\circ$ and $\theta_2 = 50.0^\circ$ and $\theta_1 = 50.0^\circ$ and $\theta_2 = 50.5^\circ$ were chosen.

From Eqs. (6a) and (6b), we can get

$$\Delta n \cong \frac{\left|\frac{\partial \phi_2}{\partial k}\right|\left|\Delta \phi_1\right| + \left|\frac{\partial \phi_1}{\partial k}\right|\left|\Delta \phi_2\right|}{\left|\frac{\partial \phi_1}{\partial n}\frac{\partial \phi_2}{\partial k} - \frac{\partial \phi_2}{\partial n}\frac{\partial \phi_1}{\partial k}\right|}, \tag{11a}$$

and

$$\Delta k \cong \frac{\left|\frac{\partial \phi_1}{\partial n}\right|\left|\Delta \phi_2\right| + \left|\frac{\partial \phi_2}{\partial n}\right|\left|\Delta \phi_1\right|}{\left|\frac{\partial \phi_1}{\partial n}\frac{\partial \phi_2}{\partial k} - \frac{\partial \phi_2}{\partial n}\frac{\partial \phi_1}{\partial k}\right|}, \tag{11b}$$

where Δn and Δk are the errors in *n* and *k,* and $\Delta\phi_1$ and $\Delta\phi_2$ are the errors in the phase differences ϕ_1 and ϕ_2, respectively. The angular resolution of a lock-in amplifier, second harmonic error, and polarization-mixing errors are the factors that may influence the accuracy in the phase difference errors in this method. So the total phase difference errors of $|\Delta\phi_1|$ and $|\Delta\phi_2|$ can be decreased to 0.03°[12] in our experiments. Substituting the conditions $|\Delta\phi_1| = |\Delta\phi_2| =$ 0.03° into Eqs. (11.a) and (11.b), the measurement errors Δn and Δk of this method are calculated and listed also at the last two columns in Table 1.

In our experiments, two random incident angles were chosen firstly to obtain the approximate value of *n*. Secondly the incident angles were changed to in the neighborhood near the critical angle θ_c derived from the approximate value of *n*, the phase differences were measured again. So the measured results with high resolution can be performed. Because the reflected light from the boundary between a prism and the tested turbid medium is measured, so this method can be applied to test the opaque turbid medium with scattering property. Even a drop of the test turbid medium can be measured, too. Owing to its common path configuration and heterodyne interferometric phase measurement, it has many advantages such as high stability against air turbulence and environmental vibrations, high resolution, and rapid measurement.

5. CONCLUSION

An alternative method for measuring the complex refractive index is proposed. First, the phase difference between s- and p- polarizations of the reflected light at boundary between a prism and the tested turbid medium are measured at two different incident angles. The measured data are substituted into specially derived equation from Fresnel equations, and then the complex refractive index of turbid can be obtained. It has both merits of the common path interferometer and the heterodyne interferometer. And its feasibility was demonstrated.

6. ACKNOWLEDGEMENT

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