

Theoretical remarks on optical coherent microscopes

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Theoretical studies are made using confocal coherent microscope. The total impulse response for two symmetric pupils having conic amplitude distribution is calculated, and analytic formulae given. Also, calculations are made using combinations of different shapes for the collector and objective lenses, and the image for a point object is calculated in all cases which show that further improvement in resolution can be attained by using pupils of conic and annular amplitude distribution for the objective and collector lenses of the coherent microscope.

Also, studies are made in a spatially coherent microscope for the purpose of the processing of the microscopic information.

1. Introduction

Coherent microscopy due to MINSKY [1] was to provide a microscope with simple objectives and to obtain at the same time a resolving power unattainable in conventional microscopic apparatus. The properties of confocal coherent microscope were first discussed by Minsky and investigated experimentally by EGGER et al. [2] and DAVIDOVITS et al. [3]. These workers were interested in the fact that the depth of field in the confocal microscope is reduced, as was also discussed by NOMARSKI [4].

Later on, many authors [5, 6] developed a scanning optical microscope for different applications and have shown that the performance of these microscopes depends mainly on the geometry of the optical system. SHEPPARD [7, 8] has stated that resolution can be improved by using the confocal coherent microscope provided with circular and annular pupils. Recently [9], he studied multiple passes through the point object which give resolution about 2.4 times sharper than that obtained in conventional microscopes for the imaging of a point object.

A further improvement in resolution of conventional confocal microscope [10] was described for the case when the detector pinhole was offset resulting in a nearly confocal operation.

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In this paper, we present some shapes of the objective and collector lenses for the improvement of resolution of the imaging system. The image of a point object or the total impulse response is calculated in the case of two symmetric conical amplitude distributions for the objective and collector lenses. Some combinations with circular and annular pupils are also made and compared with the calculations of the total impulse response made in the previous work, account being always taken of confocal microscopic arrangements.

The coherent microscope is also analysed as a correlator for the processing of microscopic information.

2. Theoretical analysis

Figure 1 is a schematic representation of a coherent microscope of type 2, like that described by SHEPPARD [11]. The difference is that the amplitude transmission of the objective and collector lenses is triangular in the case for one-dimensional pupils, and conic for two-dimensional pupils.

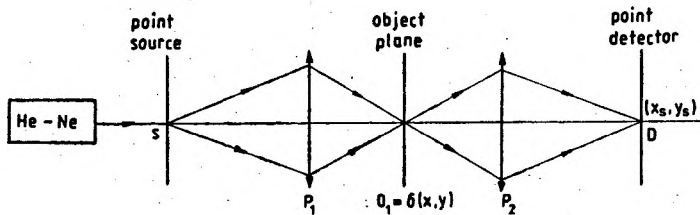


Fig. 1. Confocal coherent microscope for the imaging of a point object, by using pupils P_1 and P_2 having conic amplitude distribution

For a coherent microscope we know that the intensity distribution can be written as follows:

$$I(x', y') = |O(x', y') * h_1(x', y') h_2(x', y')| \quad (1)$$

where $*$ denotes convolution operation.

In the case of a point object, which can be represented by Dirac-delta distribution, i.e. $O(x', y') = \delta(x', y')$, the formula (1) gives

$$I(x', y') = |h_1(x', y') h_2(x', y')| \quad (2)$$

where h_1 and h_2 are the point spread function of the objective lens and collector lens, respectively, and each of these point spread functions can be calculated if their corresponding pupil distributions are known, namely

$$h_1(x', y') = \text{F.T.} [P_1(u, v)], \text{ and } h_2(x', y') = \text{F.T.} [P_2(u, v)]$$

where (u, v) are the reduced coordinates in the spatial domain (x, y) given by $(u = x/\lambda f, v = y/\lambda f)$, and F.T. denotes the Fourier transform operation.

2.1. Results from Sheppard's calculations

If the objective lens is circular, the impulse response is calculated easily to give

$$h_1(r) = \text{const } J_1(2\pi ar/\lambda f)/(2\pi ar/\lambda f) \quad (3)$$

where $r = \sqrt{x'^2 + y'^2}$.

In compact notation formula (3) can be rewritten as follows

$$h_1 = \text{const } J_1(z)/z \quad (4)$$

with $z = 2\pi ar/\lambda f$, λ is the wavelength emitted from He-Ne laser, and a/f is the numerical aperture of the objective lens. Hence, the intensity distribution for a point object in the case of two symmetric circular pupils can be easily calculated from formula (2) to give

$$I(z) = \text{const} [J_1(z)/z]^4. \quad (5)$$

For one annular pupil and the second circular one we have

$$I(z) = \text{const} J_0^2(z) [J_1(z)/z]^2. \quad (6)$$

For two annular pupils we have

$$I(z) = \text{const} J_0^4(z). \quad (7)$$

2.2.1. Calculation of the image of a point object considering one-dimensional pupil having triangular distribution

In the case of a triangular pupil with amplitude transmission given as follows:

$$P_1(x) = A(x) = \begin{cases} 1 - |x/a|, & \text{for } |x/a| \leq 1 \\ \text{zero} & \text{otherwise,} \end{cases} \quad (8)$$

the impulse response can be calculated easily with the help of Fourier transformation techniques

$$h_1(Z) = \sin^2(\pi x' \text{N.A.}/\lambda)/(\pi x' \text{N.A.}/\lambda)^2 = [\sin(Z)/Z]^2, \quad (9)$$

(N.A. is the numerical aperture of the pupil lens) h_2 gives a formula similar to (9) if the pupil has the same triangular distribution.

Hence, in the case of two triangular pupils the intensity distribution for a point object (see formula (2)) is

$$I(Z) = [\sin(Z)/Z]^8. \quad (10)$$

This factor 8 leads us to investigate a more realistic set-up with two-dimensional amplitude distribution as a function of conic distribution.

2.2.2. Calculation of the image of a point object using conical amplitude distribution

The question arises how to determine the form of the pupil function which, in some agreed sense, would give the best possible image, considering that the contrast may be enhanced at the expense of the limit of resolution.

Now, let us represent the amplitude transmission function for a conical pupil, as follows:

$$P_1(\rho) = \begin{cases} 1 - |\rho/\rho_0|, & \text{with } |\rho/\rho_0| \leq 1 \\ \text{zero} & \text{otherwise} \end{cases} \quad (11)$$

with $\rho = \sqrt{x^2 + y^2}$.

This conical amplitude distribution can be made either by depositing a partially transmitting thin film of suitable substance or by making computer-generated holograms as filters using the techniques cited by CLAIR et al. [13].

It is convenient to perform the two-dimensional Fourier-Bessel transformation in polar coordinates

$$\begin{aligned} h_1(r) &= 2 \int_0^{\rho_0} \int_0^{2\pi} (1 - \rho/\rho_0) \exp\left(-j2\pi \frac{\rho r}{f} \cos\theta\right) \rho d\rho d\theta \\ &= 2 \int_0^{\rho_0} (1 - \rho/\rho_0) J_0(2\pi\rho r/\lambda f) \rho d\rho \end{aligned} \quad (12)$$

where J_0 is the Bessel function of zero order and $r = \sqrt{r^{12} + y^{12}}$. Formula (12) can be rewritten as follows:

$$h_1(r) = 2\pi \int_0^{\rho_0} \rho J_0(2\pi\rho r/\lambda f) d\rho - (2/\rho_0) \int_0^{\rho_0} \rho^2 (2\pi\rho r/\lambda f) d\rho \quad (13)$$

by changing the variables in formula (13) we get

$$h(Z) = \frac{\pi}{2} (\lambda f/\pi r)^2 \int_0^Z z J_0(z) dz - \frac{\pi}{4\rho} (\lambda f/\pi r)^3 \int_0^Z z^2 J_0(z) dz. \quad (14)$$

These formulae (cited in [12]) yield

$$\int_0^Z z J_0(z) dz = Z J_1(Z), \quad (15)$$

and

$$\int_0^Z z^2 J_0(z) dz = Z^2 J_1(Z) - \int_0^Z z J_1(z) dz \quad (16)$$

where $z = 2\pi qr/\lambda f$, and $Z = 2\pi \varrho_0 r/\lambda f$. Formula (14) gives

$$\begin{aligned}
 h_1(Z) &= \frac{1}{2} (\lambda f/\pi r)^2 Z J_1 - \frac{\pi}{4\varrho_0} (\lambda f/\pi r)^3 \left[Z^2 J_1(Z) - \int_0^Z z J_1(z) dz \right] \\
 &= \frac{\pi}{4\varrho_0} (\lambda f/\pi r)^3 \int_0^Z z J_1(z) dz,
 \end{aligned}
 \tag{17}$$

since $J_0'(z) = -J_1(z)$, then by substituting in (17) we get

$$h_1(Z) = -\frac{\pi}{4\varrho_0} (\lambda f/\pi r)^3 \int_0^Z z d[J_0(z)].
 \tag{18}$$

Formula (18) being integrated by partition gives

$$h_1(Z) = \frac{2\pi}{\varrho_0} (\varrho_0/Z)^3 \left[\int_0^Z J_0(z) dz - Z J_0(Z) \right].
 \tag{19}$$

We shall also use the formula given in [12]

$$\int_0^Z J_0(z) dz = 2 \sum_i J_i(Z), \text{ with } i = 1, 3, 5, \dots
 \tag{20}$$

By substituting (20) into (19), we have got finally the following analytic solution for the point spread function of the lens:

$$h_1(Z) = 2\pi \varrho_0^2 \left\{ \left[2 \sum_i J_i(Z) - Z J_0(Z) \right] / Z^3 \right\}.
 \tag{21}$$

In the case of two symmetric conic amplitude distributions of the objective and collector lenses, the intensity distribution calculated for a point object is

$$I(Z) = \beta \left[2 \sum_i J_i(Z) - Z J_0(Z) \right]^4 / Z^{12},
 \tag{22}$$

with $\beta = 16\pi^4 \varrho^8$.

For the objective and collector lenses with conic and circular pupils the total squared impulse response of the optical system is the following:

$$I(Z) = \text{const } J_1^2(Z) \left[2 \sum_i J_i(Z) - Z J_0(Z) \right]^2 / Z^8.
 \tag{23}$$

In the case of conic and annular pupils we obtain

$$I(Z) = \text{const } J_0^2(Z) \left[2 \sum_i J_i(Z) - Z J_0(Z) \right]^2 / Z^6.
 \tag{24}$$

It is to be noted that computer programs are constructed to calculate the last formulae (22), (23) and (24), and the results of computations are represented graphically which we shall see in Sec. 4.

3. Image processing using confocal coherent microscope

Referring to Figure 2, consider the transparency O_1 moving in the plane (x, y) , the second transparency O_2 found at a distance ϵ from the first one being stationary. Assuming that O_2 is much closer to O_1 we may write $O_2(x_0, y_0) = O_2(x, y)$. The intensity distribution in the imaging plane is [14]

$$I(x_s, y_s) = |O_1 * h_1 h_2 O_2 \exp(jkd^2/2\epsilon)|_{(x_s, y_s)}^2 \quad (25)$$

where $d = \sqrt{x_0^2 + y_0^2}$, and $k = 2\pi/\lambda$.

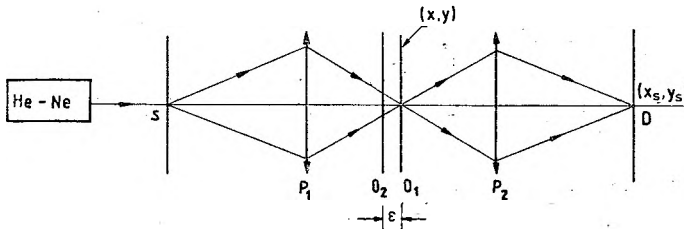


Fig. 2. Image processing using confocal coherent microscope

Formula (25) can be expanded to give

$$I(x_s, y_s) = |O_1 * h_1 h_2 O_2 \cos(\pi^2 d^2/\lambda\epsilon)|^2 + |O_1 * h_1 h_2 O_2 \sin(\pi d^2/\lambda\epsilon)|^2. \quad (26)$$

For perfect imaging, which is the case of geometrical optics approximation, we have got $h_1 h_2 = 1$, and in the case where $\pi d^2/\lambda\epsilon \ll 1$, formula (26) gives

$$I(x, y) = |O_1 * O_2|^2 + |O_1 * (\pi d^2/\lambda\epsilon) O_2|^2. \quad (27)$$

The first term of the formula (27) gives the correlation intensity between the two signals O_1 and O_2 , while the second term is considered as a noise. Hence, to improve the results of correlation it is preferable to put two transparencies as close as possible in the plane (x, y) , consequently the second term vanishes.

4. Results and discussion

Computer programs are constructed to calculate the total impulse response of the optical system for different combinations of the pupil functions cited in the theoretical analysis. The calculations are made, using the wavelength of light emitted from the He-Ne laser with $\lambda = 0.6328 \mu\text{m}$, and providing the microscope with two lenses having high numerical apertures (N.A. = 0.85).

The image of a point object in the case of a coherent microscope is presented

in Fig. 3, where curve 1 corresponds to the case of two symmetric conic pupils, curve 2 corresponds to the combination of conic and circular pupils, curve 3 is given for conic and annular pupils, and the last curve 4 is given for two circular pupils.

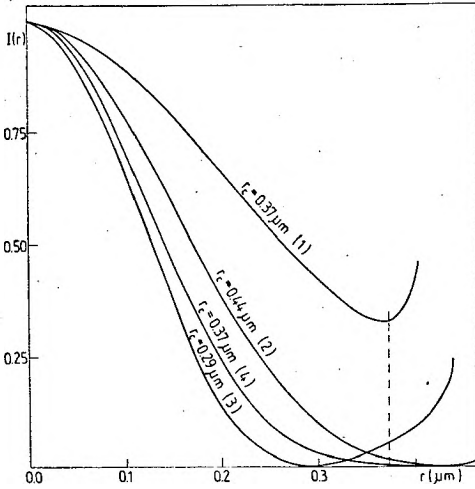


Fig. 3. Image intensity distribution of a point object, using coherent microscope with different combinations of pupil functions. $\lambda = 0.6382 \mu\text{m}$, N. A. = 0.85, and r_c is the cut off frequency corresponding to each curve. Two conic pupils - 1, conic and circular pupils - 2, conic and annular pupils - 3, two circular pupils - 4

Figure 4 represents image intensity distribution for: two circular pupils (curve 1), circular-annular combination (curve 2), and for two annular pupils (curve 3). It is to be noted that the results given in Fig. 4 are due to SHEPPARD et al. [11] and are cited only for comparison with our results given in Fig. 3.

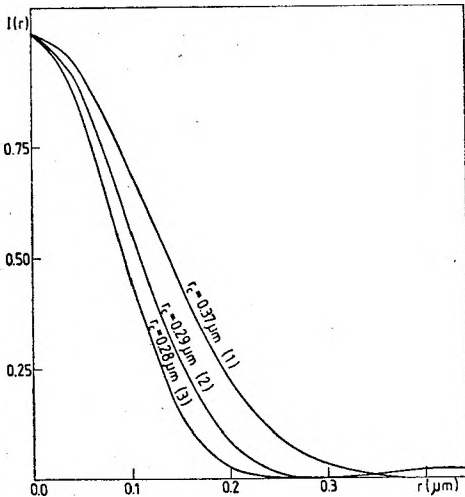


Fig. 4. Image intensity distribution of a point object, using coherent microscope with combinations of circular and annular pupils. $\lambda = 0.6328 \mu\text{m}$, N. A. = 0.85, and r_c is the cut off frequency corresponding to each curve. Two circular pupils - 1, circular and annular pupils - 2, two annular pupils - 3

From the above results it may be concluded that the best resolution can be attained either by using conic-annular pupils for the optical system or by using two annular pupils as stated by SHEPPARD [11], and this improvement of resolution is due to the obstruction of the light through the pupils.

5. Conclusion

Theoretical studies on coherent optical microscope have been performed in order to improve resolution. From the results obtained it may be concluded that further improvement in resolution may be attained if instead of two circular pupils combination of both conic and annular pupils of the objective and collector lenses is used.

We also state that the coherent microscope may be treated as a two-dimensional correlator to be used for microscopic information processing, and believe that this microscope is also suitable for the processing of the coloured information by using polychromatic light of illumination.

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Теоретические исследования когерентных оптических микроскопов

Проведены теоретические исследования конфокального когерентного микроскопа. Расчитан полный импульсный ответ для двух симметричных зрачков конусообразного распределения амплитуды, а также даны аналитические формулы. Проведен, кроме того, расчет для комбинации разных видов линз коллектора и объектива, а также расчитано изображение пунктирного объекта для всех случаев. Доказано, что можно достичь дальнейшего повышения распределения путем применения конусообразного и кольцеобразного распределений амплитуды в объективных и коллекторных линзах когерентного микроскопа.

Перевела Малгожата Хейдрих