

## Letter to the Editor

### Point of inflection in the coherent two-photon fluorescent microscope of annular pupil

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In this paper, the intensity distribution in the clear slit image is presented in coherent two photon fluorescent microscope (C2P) with an annular pupil outside the point of inflection as well as in the point of inflection (one dimensional case).

The point of inflection corresponds to a particular half-width of the slit for which the central peak of the intensity distribution in the slit image changes its curvature from convexity to the concavity. In paper [1], the authors determined the values of the point of inflection in a coherent annular CSM with a Lanczos apodizing filter. For the constant control parameter of the transmittance  $\beta$  of the Lanczos apodizer the values taken by the point of inflection  $u_{0\text{infl}}$  diminish within the range of obstruction of the annular pupil  $0 < \varepsilon < 0.5$ , while they increase within the range  $0.5 < \varepsilon < 1$ . For constant  $\beta$  and  $\varepsilon$ , only one value of  $u_{0\text{infl}}$  appears. In the paper [2], the point of inflection has been calculated for a coherent scanning microscope (CSM) with  $\cos^2(Nx)$  apodizer. It has been shown that for  $N = \text{const.}$  there exist several points of inflection.

In this work, it has been shown that the intensity distribution in the clear slit image obtained in a two-photon fluorescent microscope of nonapodized type with the annular pupil, the point of inflection has been calculated, and the intensity distribution in the clear slit image at the point of inflection has been shown for several chosen values of obstruction in the annular pupil.

The intensity distribution in the image in a coherent scanning microscope (CSM) is described by the relation [3]

$$I_{\text{CSM}} = |A \otimes (h_{\text{obj}} h_{\text{coll}})|^2 \quad (1)$$

where  $A$  is the amplitude transmittance of the object,  $h_{\text{obj}}$  and  $h_{\text{coll}}$  are the amplitude spread functions of the objective and the collector, respectively,  $\otimes$  denotes the convolution.

In the two-photon fluorescent microscope (C2P), the intensity distribution is described by the relation [3]

$$I_{\text{C2P}} = I^2(u/2, v/2)I(u, v) \quad (2)$$

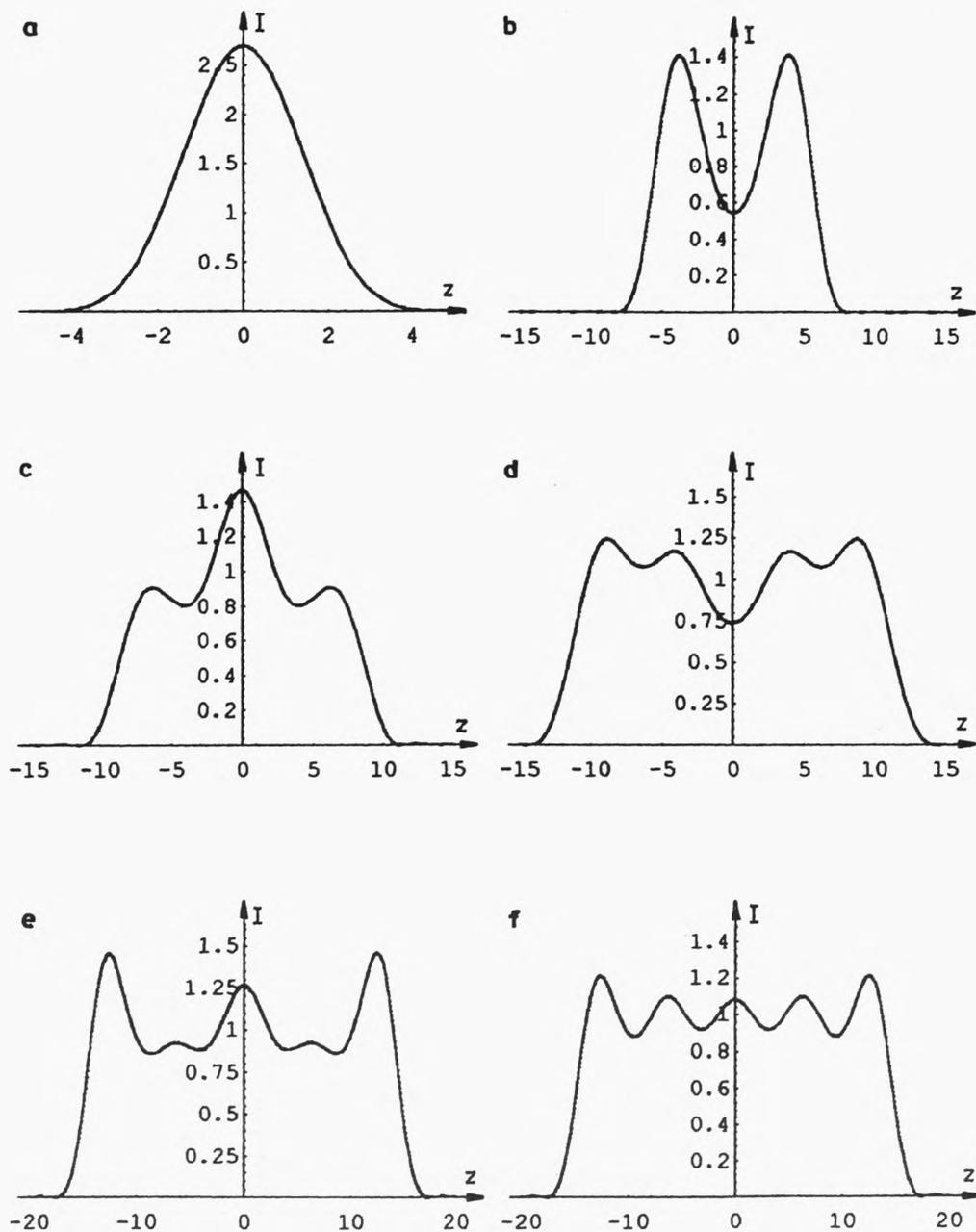


Fig. 1. Intensity distribution in the clear slit image with C2P of the circular pupil ( $\varepsilon = 0$ ), for:  $u_0 = 0.5$  (a),  $u_0 = 1.0$  (b),  $u_0 = 1.5$  (c),  $u_0 = 2.0$  (d),  $u_0 = 2.5$  (e). Intensity distribution in the clear slit image for  $u_0 = 2.5$  in the coherent CSM with circular pupil  $\varepsilon = 0$  (f)

where:  $I(u, v)$  – intensity of the point object,  
 $u, v$  – axial and radial normed optical coordinates:

$$u = 4kz \sin^2(\alpha/2), \quad v = kr \sin \alpha, \quad k = 2\pi/\lambda.$$

For an object in the form of a clear slit of halfwidth  $u_0$  and the amplitude transmittance

$$A(u, v) = \begin{cases} 1 & \text{for } |u| \leq u_0, \\ 0 & \end{cases}$$

the intensity distribution in the image of the clear slit in the two-photon fluorescent nonapodized microscope (C2P) with an annular pupil  $\varepsilon$  is described by the relation

$$I(z) = \left[ (4u_0 \int_z^1 \frac{\sin(2\pi u_0 x)}{(2\pi u_0 x)} \cos\left(z \frac{x}{2}\right) dx )^2 \right]^2 \left[ (4u_0 \int_z^1 \frac{\sin(2\pi u_0 x)}{(2\pi u_0 x)} \cos(zx) dx )^2 \right]^2. \quad (3)$$

In Figure 1, there has been shown the intensity distribution in the clear slit of the width  $u_0 = 0.5$  (Fig. 1a),  $u_0 = 1.0$  (Fig. 1b),  $u_0 = 1.5$  (Fig. 1c),  $u_0 = 2.0$  (Fig. 1d),  $u_0 = 2.5$  (Fig. 1e) in C2P with the circular pupil ( $\varepsilon = 0$ ) and for comparison that of  $u_0 = 2.5$  (Fig. 1f) in the coherent CSM. In C2P, no fringe structure appears in contrast to the case of the CSM. In Figure 2, the intensity distribution in the clear slit image in C2P with the annular ( $\varepsilon = 0.5$ ) pupil has been shown for the slit width  $u_0 = 0.5$  (Fig. 2a),  $u_0 = 1.0$  (Fig. 2b),  $u_0 = 1.5$  (Fig. 2c),  $u_0 = 2.0$  (Fig. 2d),  $u_0 = 2.5$  (Fig. 2e), while in Fig. 3, the intensity distribution of the clear slit for C2P with the annular  $\varepsilon = 0.8$  pupil has been presented for the slit width  $u_0 = 0.5$  (Fig. 3a),  $u_0 = 1.0$  (Fig. 3b),  $u_0 = 1.5$  (Fig. 3c),  $u_0 = 2.0$  (Fig. 3d),  $u_0 = 2.5$  (Fig. 3e). For  $\varepsilon = 0.5$ , a change of the ratio of intensities in the maxima is observed for  $u_0 > 1.5$  (Figs. 2c–e) and the values of  $I$  in the main maximum diminish. For  $\varepsilon = 0.8$  this ratio is practically constant and the values of  $I$  increase with increasing  $u_0$ .

The values of the point of inflection were determined from the condition

$$\Delta = \frac{\partial^2 I(u)}{\partial u^2} \Big|_{u=0} = 0. \quad (4)$$

In Figure 4, the values of the point of inflection in C2P are determined numerically from Eq. (4) correspondingly for: circular pupil –  $\varepsilon = 0$  (Fig. 4a), annular pupil and  $\varepsilon = 0.1$  (Fig. 4b),  $\varepsilon = 0.2$  (Fig. 4c),  $\varepsilon = 0.3$  (Fig. 4d),  $\varepsilon = 0.4$  (Fig. 4e),  $\varepsilon = 0.5$  (Fig. 4f),  $\varepsilon = 0.6$  (Fig. 4g),  $\varepsilon = 0.7$  (Fig. 4h),  $\varepsilon = 0.8$  (Fig. 4i).

For  $u = u_{0\text{inf}l}$  from formula (3) the clear slit image in the point of inflection can be obtained. In Figure 5,  $I(u_{0\text{inf}l})$  is shown for  $\varepsilon = 0$  (circular pupil) at points of inflection  $u_{0\text{inf}l1} = 0.715149$  (Fig. 5a),  $u_{0\text{inf}l2} = 1.22951$  (Fig. 5b), for  $\varepsilon = 0.1$ , in  $u_{0\text{inf}l1} = 0.714077$  (Fig. 5c), for  $\varepsilon = 0.2$  in  $u_{0\text{inf}l1} = 0.707219$  (Fig. 5d) and for  $\varepsilon = 0.3$  in  $u_{0\text{inf}l3} = 1.69042$  (Fig. 5e).

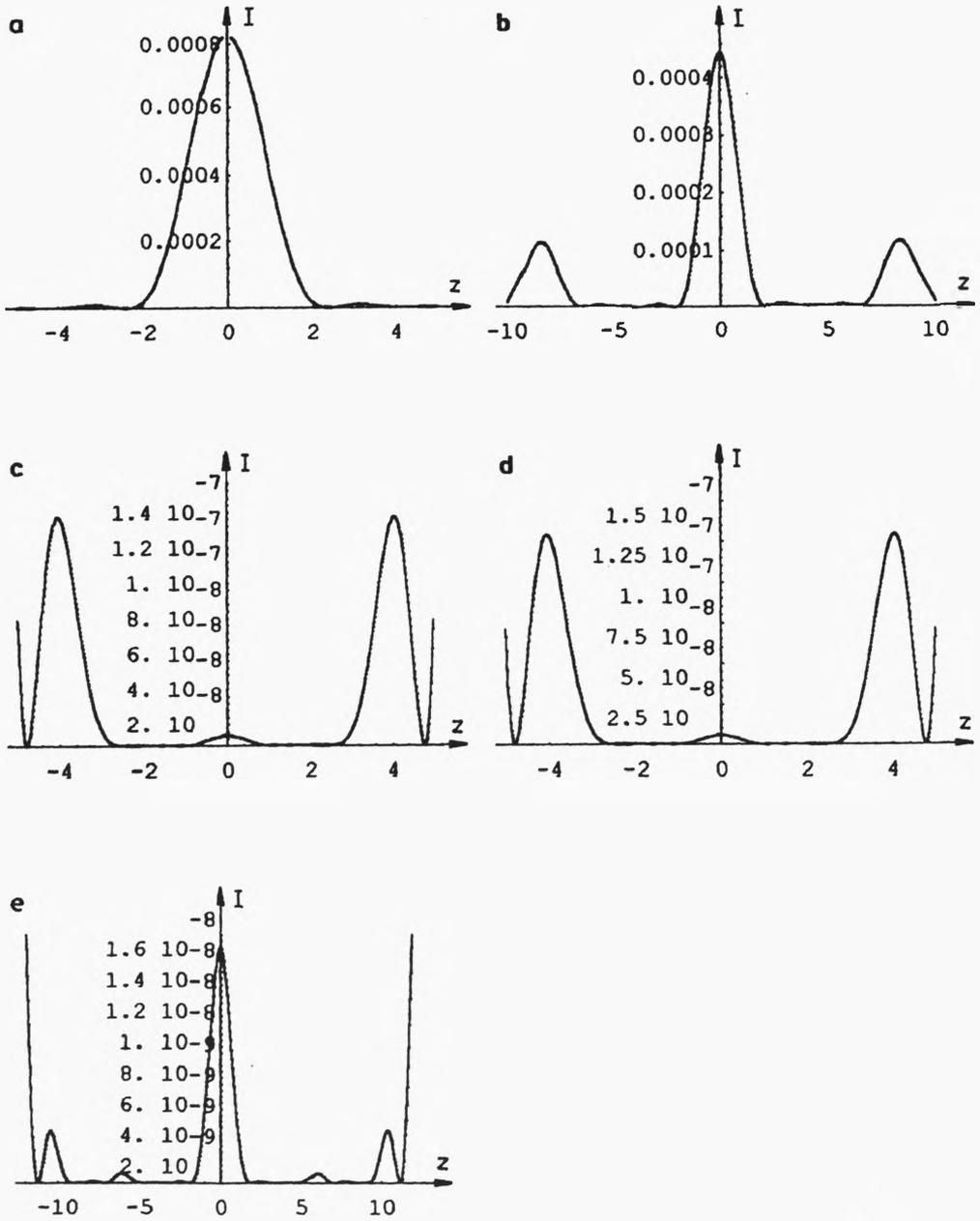


Fig. 2. Intensity distribution in the clear slit image in C2P with the annular pupil ( $\epsilon = 0.5$ ) for the slit for which:  $u_0 = 0.5$  (a),  $u_0 = 1.0$  (b),  $u_0 = 1.5$  (c),  $u_0 = 2.0$  (d),  $u_0 = 2.5$  (e)

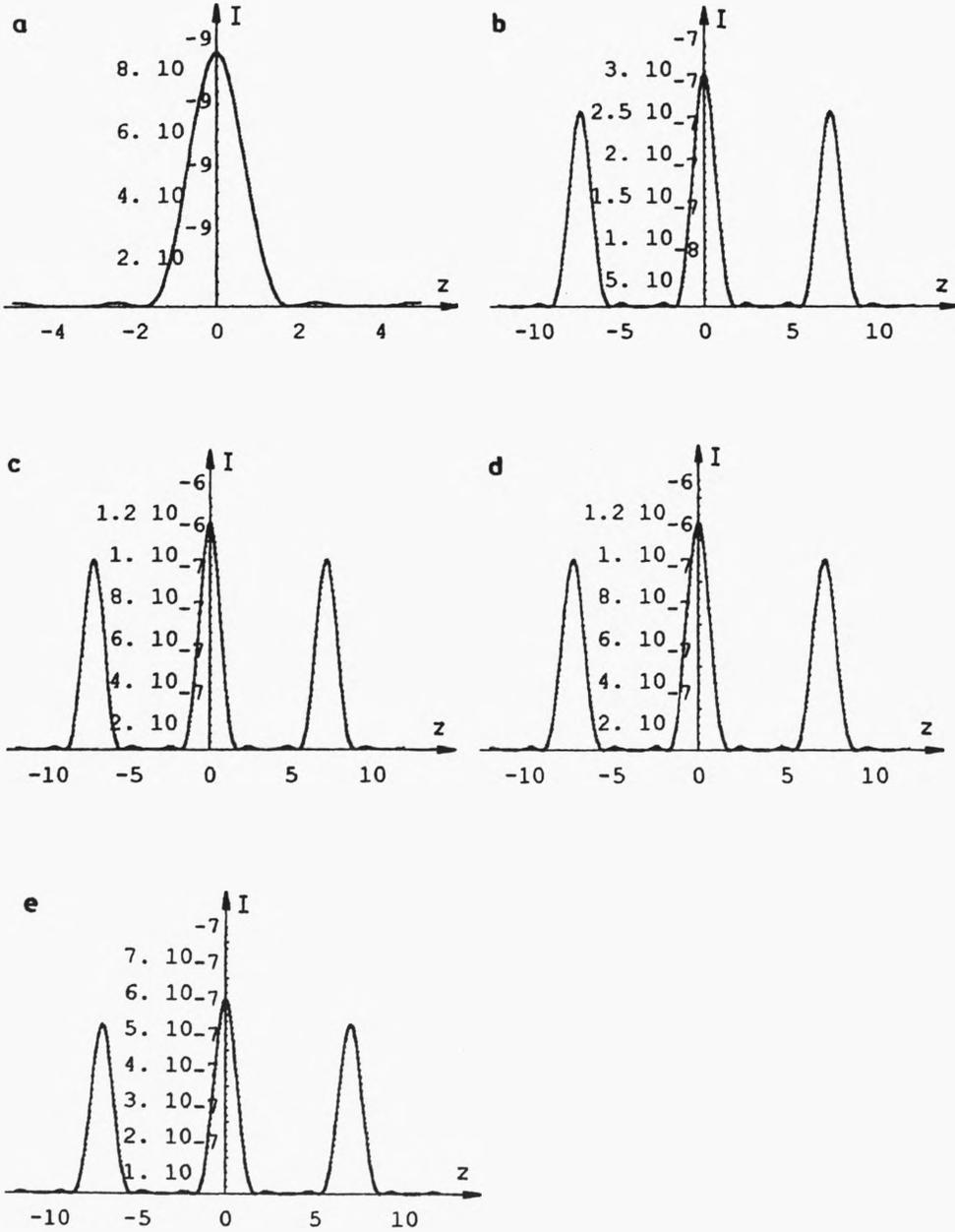


Fig. 3. Intensity distribution in the clear slit image in C2P of the annular pupil ( $\epsilon = 0.8$ ) for:  $u_0 = 0.5$  (a),  $u_0 = 1.0$  (b),  $u_0 = 1.5$  (c),  $u_0 = 2.0$  (d),  $u_0 = 2.5$  (e)

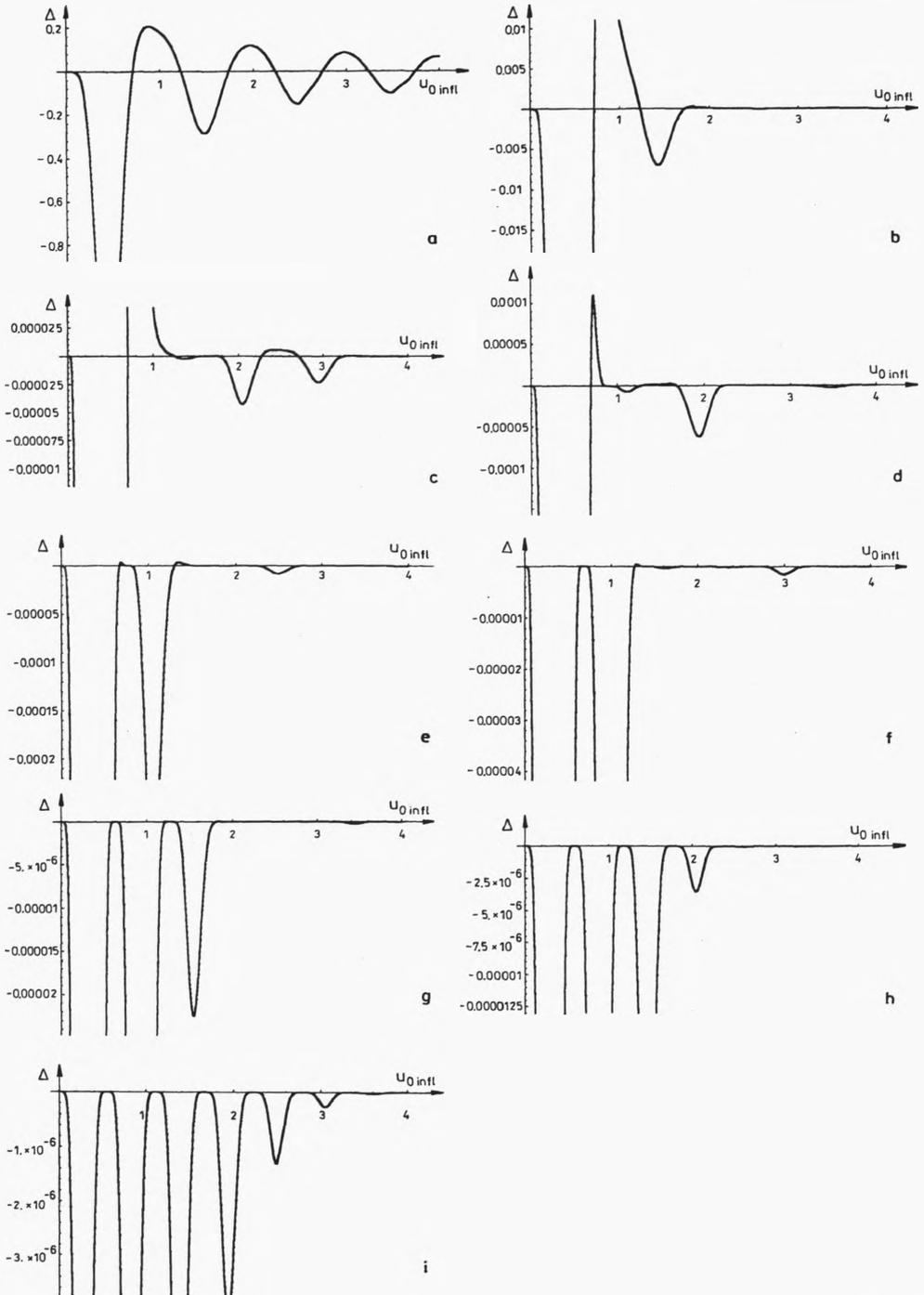


Fig. 4. Dependence  $\Delta = \frac{\partial^2 I(u)}{\partial u^2} \Big|_{u=0} = 0(u_{0\text{ infl}})$  for **a** -  $\epsilon = 0$ , **b** -  $\epsilon = 0.1$ , **c** -  $\epsilon = 0.2$ , **d** -  $\epsilon = 0.3$ , **e** -  $\epsilon = 0.4$ , **f** -  $\epsilon = 0.5$ , **g** -  $\epsilon = 0.6$ , **h** -  $\epsilon = 0.7$ , **i** -  $\epsilon = 0.8$

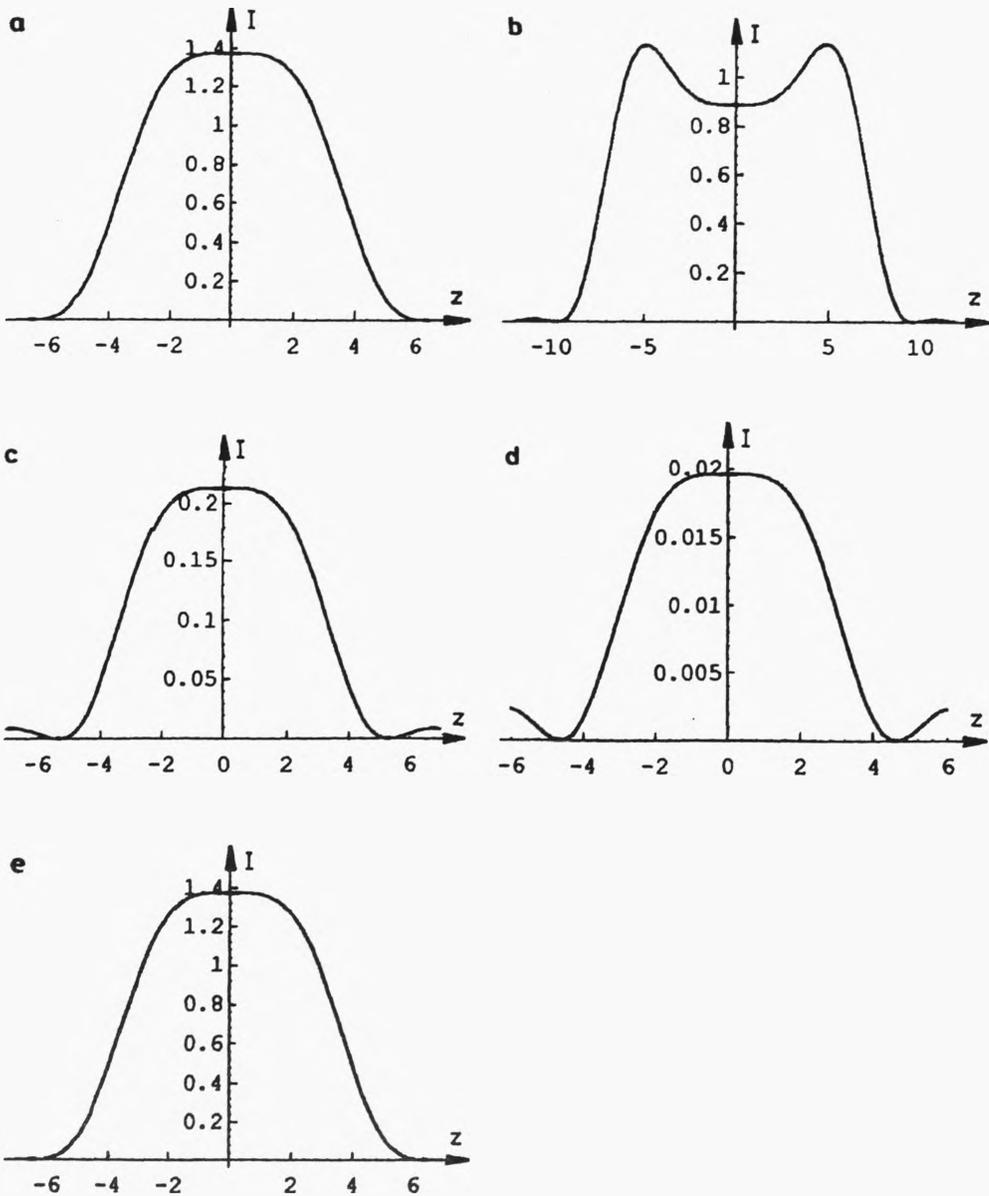


Fig. 5. Intensity distribution in the clear slit image in the point of inflection:  $u_{0inf11} = 0.715149$  (a),  $u_{0inf12} = 1.22951$  (b) for  $\epsilon = 0$ ,  $u_{0inf11} = 0.714077$  (c) for  $\epsilon = 0.1$ ,  $u_{0inf11} = 0.707219$  (d) for  $\epsilon = 0.2$ ,  $u_{0inf13} = 1.69042$  (e) for  $\epsilon = 0.3$

At the point of inflection the clear slit image of highest fidelity is created. For  $\epsilon = \text{const.}$ , many points of inflection appear which is shown in Fig. 4a–f. For  $\epsilon > 0.5$  the point of inflection does not appear which is the case in the corresponding situation in CSM.

**References**

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