

## Letter to the Editor

### Point spread function in a confocal microscope with trigonometric pupil filters

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In this paper the distribution of point spread function was examined versus the spatial frequencies of the filters of  $\cos(N\rho)$  type modulating the aperture of the confocal CSM for different values of numerical aperture. In particular, the following relations were determined: i) PSF as dependent on the pupil modulating spatial frequency  $r$  for  $N = 1, 2, 3, 4, 5, 6, 7, 9, 11$  and numerical aperture  $NA = 0.8$ ; ii) PSF for filters of  $\cos(4\rho)$ ,  $\cos(10\rho)$  type and  $NA = 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4$ ; iii) cut-off spatial frequencies  $r_c$  for the aperture modulated by  $\cos(N\rho)$  for  $N = 0, \dots, 20$ ,  $NA = 0.2, 0.5, 1.0$  and  $\lambda = 0.6328 \mu\text{m}$ .

In paper [1], the point spread function (PSF) was determined as a function of spatial frequency  $r$  in a CSM microscope of apertures modulated by  $\rho^n$  for  $n = 2, 4, 6, 8, 10, 12, 14, 16$  and for  $NA = 0.5$  and  $NA = 0.8$ . Also a characteristic of the cut-off spatial frequency  $r_c(n)$  was examined as dependent on parameters  $n$  for  $NA = 0.5$ .

In the present paper, the distribution of the PSF is examined as a function of spatial frequency  $r$  in a confocal CSM microscope of aperture modulated by the filters of  $\cos(N\rho)$  type (for  $N = 1, 2, 3, 4, 5, 6, 7, 9, 11$ ) and  $NA = 0.8$ . Here,  $\rho$  is the absolute value of the radius-vector in the pupil plane. Additionally, the PSF has been examined for different  $r$  and  $\cos(4\rho)$ ,  $\cos(10\rho)$  and for  $NA = 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4$ . The characteristic of the cut-off frequency  $r_c$  has been determined for the aperture modulated by filters of  $\cos(N\rho)$  type (for  $N = 0, 1, \dots, 20$ ) and  $NA = 0.2, 0.5, 1.0$ , with  $\lambda = 0.6328 \mu\text{m}$ .

The resultant point spread function (RPSF)  $h_r$  in a confocal microscope is defined by the PSFs  $h_1, h_2$  of the first and second objectives, respectively, *i.e.*

$$h_r = h_1 h_2.$$

For the case of two identical nonmodulated circular objectives the image of the point object is defined by

$$I(w) = \left[ \frac{2J_1(w)}{w} \right]^4$$

where:  $J_1$  – Bessel function of the first kind and first order,  $w = k\rho r/f$  – reduced coordinate in the image plane,  $k = 2\pi/\lambda$  – propagation constant (wave number).

The PSF is a Fourier transform of the pupil function

$$\text{PSF} = \text{FT}\{P(\rho)\}.$$

For apertures modulated by the trigonometric filters we obtain [1]

$$h_N(r) = 2\pi \int_0^{\rho_0} \cos(N\rho)\rho J_0(k\rho r/f) d\rho$$

where:  $J_0$  – Bessel function of the first kind and zero order,  $\rho_0$  – rim value of  $\rho$ . In the numerical calculations  $f = 1 \mu\text{m}$  and  $\lambda = 0.6328 \mu\text{m}$  have been assumed.

In Figure 1, the pupil function modulated by  $\cos(N\rho)$  for  $N = 1, 4, 10$  and  $20$  is shown. In Figure 2, the PSF is presented as a function of spatial frequencies

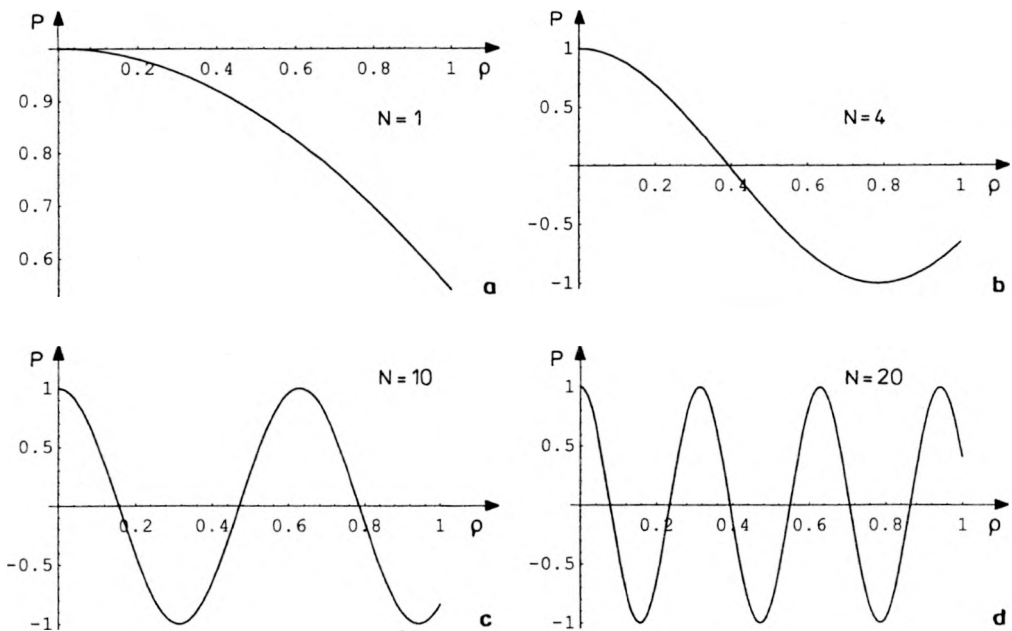


Fig. 1. Distribution of the pupil function  $\cos(N\rho)$  for:  $N = 1$  (a),  $N = 4$  (b),  $N = 10$  (c),  $N = 20$  (d).

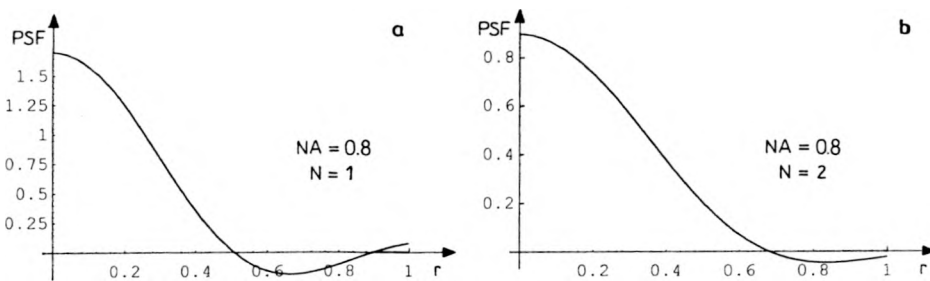


Fig. 2.a,b

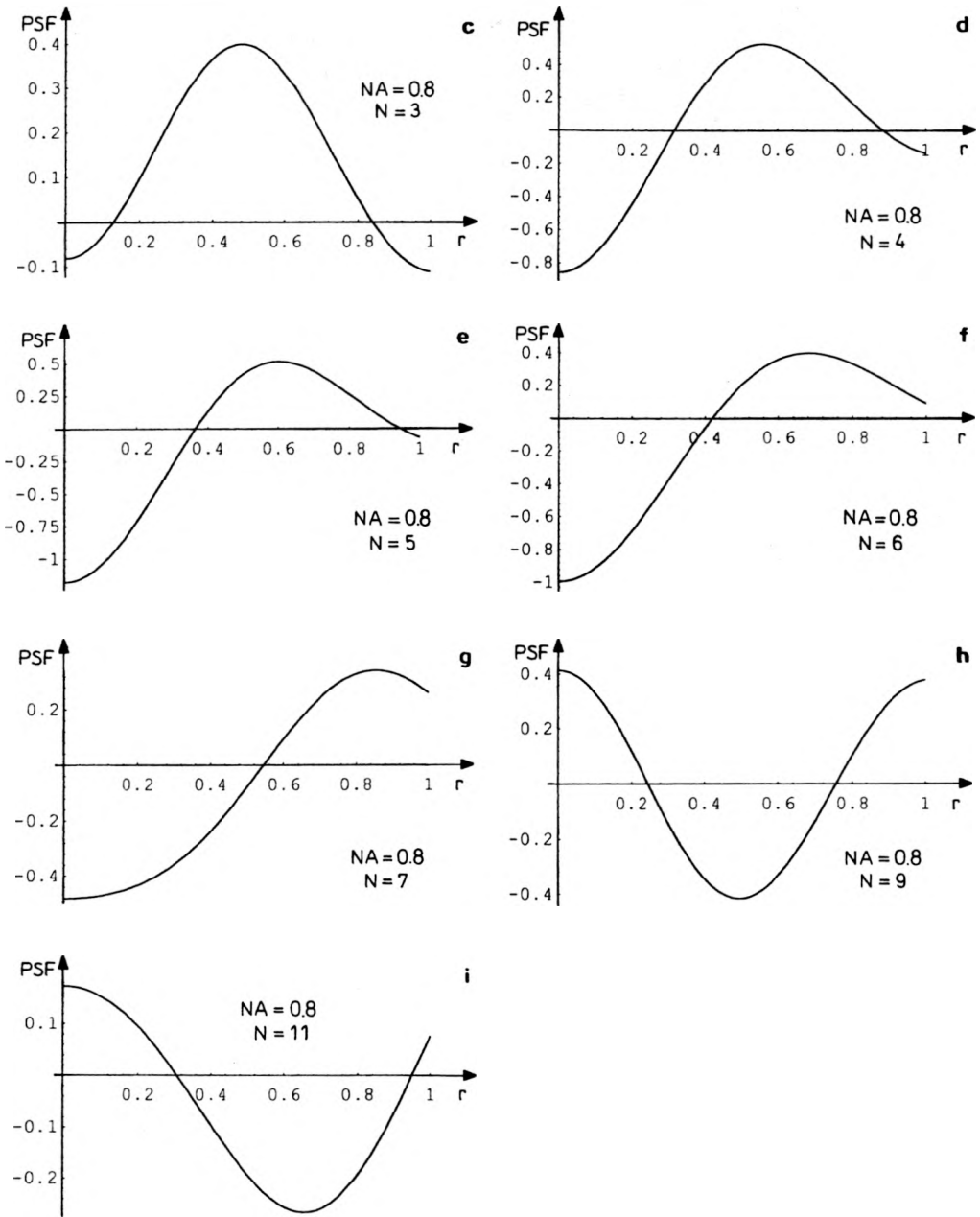


Fig. 2. Point spread function versus the spatial frequencies  $r$  [ $\mu\text{m}$ ] for the pupil filter  $\cos(N\rho)$  and the numerical apertures  $NA = 0.8$  for:  $N = 1$  (a),  $N = 2$  (b),  $N = 3$  (c),  $N = 4$  (d),  $N = 5$  (e),  $N = 6$  (f),  $N = 7$  (g),  $N = 9$  (h),  $N = 11$  (i).

$r$  for pupil filters of  $\cos(N\rho)$  type in CSM and for  $NA = 0.8$ , while  $N$  takes the values: 1, 2, 3, 4, 5, 6, 7, 9, 11.

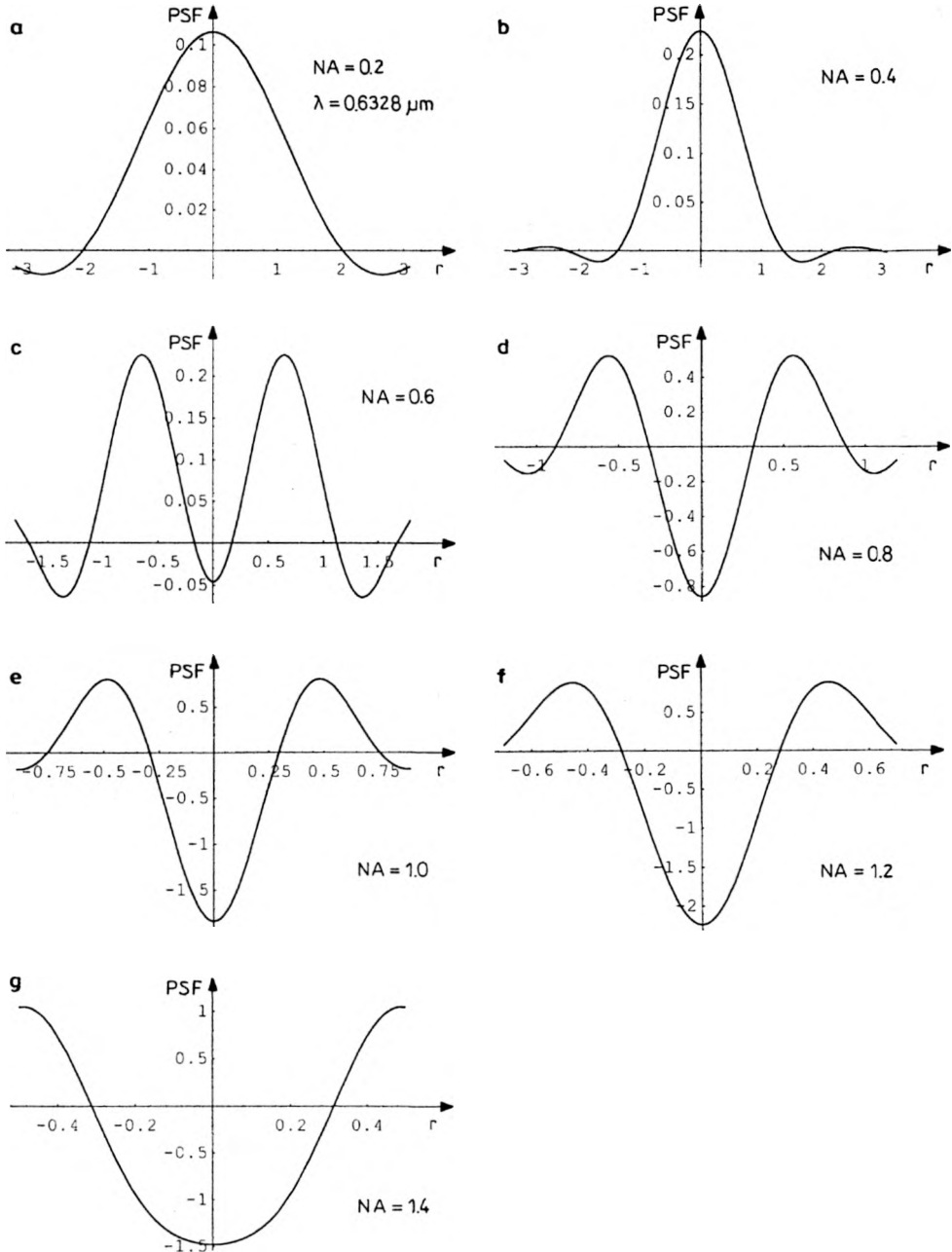


Fig. 3. Point spread function versus the spatial frequencies  $r$  [ $\mu\text{m}$ ] for the pupil filter  $\cos(4\rho)$  and the numerical apertures  $NA = 0.2$  (a),  $0.4$  (b),  $0.6$  (c),  $0.8$  (d),  $1.0$  (e),  $1.2$  (f),  $1.4$  (g).

In Figure 3, the PSF is presented as a function of spatial frequencies  $r$  for the pupil filter  $\cos(4\rho)$  and the following values of the numerical aperture:  $NA = 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4$ . In Figure 4, the PSF is presented as a function of spatial

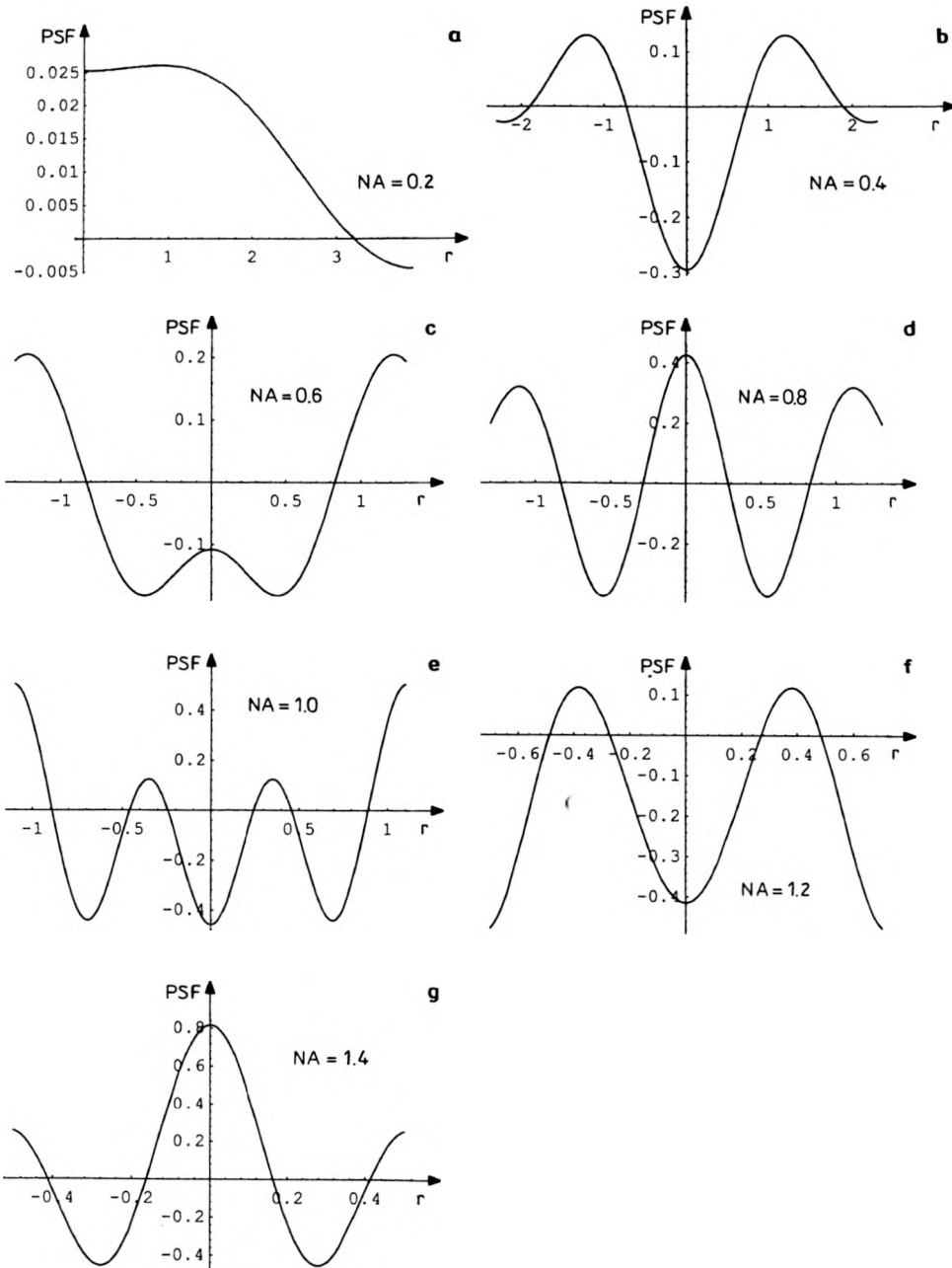


Fig. 4. Point spread function versus the spatial frequencies  $r$  [ $\mu\text{m}$ ] for the pupil filter  $\cos(10\rho)$  and the numerical apertures  $NA = 0.2$  (a),  $0.4$  (b),  $0.6$  (c),  $0.8$  (d),  $1.0$  (e),  $1.2$  (f),  $1.4$  (g).

frequencies  $r$  for the pupil filter of  $\cos(10\rho)$  for numerical apertures:  $NA = 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4$ . In Figure 5, the characteristic of the cut-off frequency  $r_c$  [ $\mu\text{m}$ ] versus the parameter  $N$  ( $N = 0, 1, \dots, 20$ ) of the pupil filter  $\cos(N\rho)$

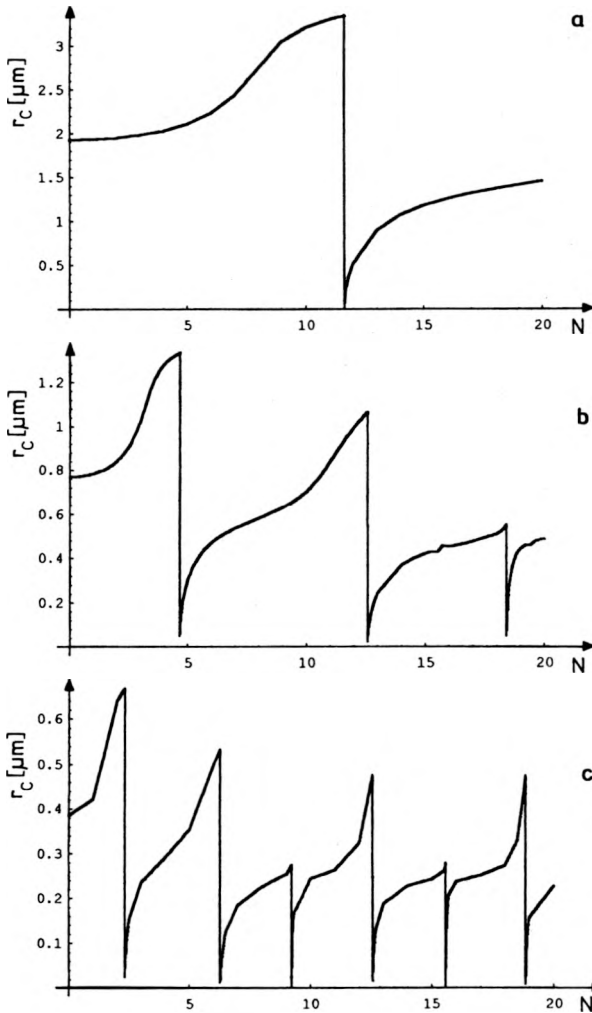


Fig. 5. Cut-off spatial frequencies  $r_c$  [ $\mu\text{m}$ ] for different values of the parameter  $N$  of the pupil filters of  $\cos(N\rho)$  type ( $\lambda = 0.6328 \mu\text{m}$ ,  $f = 1 \mu\text{m}$ ) for numerical apertures:  $NA = 0.2$  (a),  $0.5$  (b),  $1.0$  (c).

has been determined for numerical apertures:  $NA = 0.2, 0.5, 1.0$  and  $\lambda = 0.6328 \mu\text{m}$ . These frequencies have been determined by solving the equation

$$h_N = 0.$$

For a circular nonmodulated pupil ( $N = 0$ ) and the numerical aperture of  $NA = 0.5$ ,  $\lambda = 0.6328 \mu\text{m}$ ,  $f = 1 \mu\text{m}$ , the cut-off frequency is equal to  $r_c = 0.771807 \mu\text{m}$ . For the aperture  $P(\rho) = \rho^n$ , as reported in [1], the cut-off frequency  $r_c$  ranges from  $r_c = 0.772 \mu\text{m}$  for the nonmodulated circular frequency ( $n = 0$ ) to  $r_c = 0.43 \mu\text{m}$  for high values  $n = 16$ . In the case of aperture  $P(\rho) = \cos(N\rho)$  and  $NA = 0.5$  the characteristic  $r_c(N)$  is shown in Fig. 5b. The results obtained are shown in the next page.

$N$	$r_c$ [ $\mu\text{m}$ ]
0 (nonmodulated aperture)	0.7718
4.66	1.33425
4.66225	0.001351
12.56	1.06397
12.567	0.01034
18.41	0.5506
18.42	0.0488
20	0.487

For  $NA = 0.2$ , the characteristic  $r_c(N)$  is shown in Fig. 5a, while for  $NA = 1.0$  in Fig. 5c.

### Reference

- [1] HAMED A. M., *Optik* 107 (1998), 161.

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