

« Multiphoton Microscopy for visualizing free virions and revealing the invisible enemy »

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During current pandemic times, we barely convince people to protect themselves against an invisible enemy such as SARS-CoV-2. By the use of multiphoton microscopy (MPM), our aim is to show the virus in 3D in order to simplify the sample preparation and to observe the pathogen without chemical modification or marking by endogenous fluorescence. However, the resolution limit does not allow the observation of a target of 120 nm [1]. Therefore, we are applying a computational strategy for giving a crucial improve of image quality. In this specific context we are using the pipeline FAMOUS (fast algorithm for three-dimensional multiphoton microscopy of biomedical structures) [2]. The use of fluorescent microsphere as deconvolution model allows the software to adapt the received signal as an image for each individual setup. The point-spread-function (PSF) is estimated; image quality and thus axial resolution are increased, while reducing noise and blur with a deconvolution solution [3]. 3D images are the presenting a highly improved quality. This way of computational image processing cross sub-micron resolution limits and opens up the possibilities of imaging the shape of the virion in the image by an optical solution. We are first testing our instrumental and computational pipeline with the image of cytomegalovirus (CMV), a virion with a diameter of about 300 nm. With an initial preparation in an adapted lab for virion culture (P3), and thanks to a protocol of UV inactivation, Figure 1 shows the raw acquisition (left side) and the resulting image (right side) from such strategy which allows to identify the presence of free virions. Thanks to this nice initial result, we are currently testing this strategy on SARS-Cov-2 virions but we have no significant results for the moment.

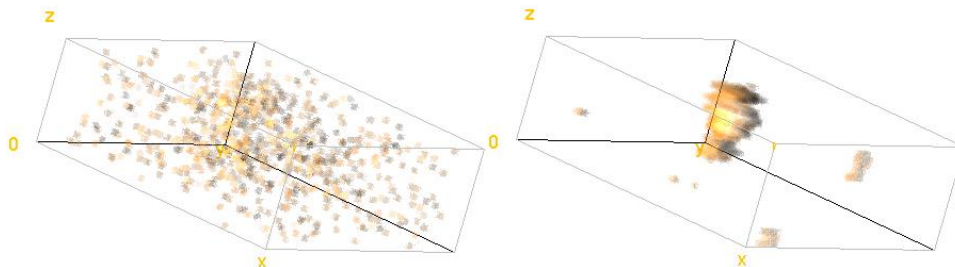


Figure 1: 3D image of viral particles of cytomegalovirus before and after FAMOUS processing. The box dimensions is $1.5 \mu\text{m} \times 1.5 \mu\text{m} \times 0.9 \mu\text{m}$

Such protocol will help to follow visually and in live the efficiency of anti-viral treatments directly within living cells by an optical microscopy technology. Thus, we could demonstrate with ease to a lay audience the virus specificities and to convince them to respect logical sanitary measures with simple, explicative and transparent explanation. Moreover, these findings might allow to rebuilt trust between scientists and collectivity and rationalize the virus threat across the laymen's socially constructed image of SARS-CoV-2 [4]. Additionally, these findings that are crucial for identifying the precise representation and viral strategy of SARS-CoV-2 might allow to alleviate laymen's distress related to the currently invisible viral enemy.

[1] - R. Witte et al, "Concepts in Light Microscopy of Viruses", *Viruses*, 10, 202 (2018)

[2] - C. Lefort et al *J. Phys. D: Appl. Phys.* **54** 274005 (2021)

[3] - B. de Monvel et al, *Mats. Biophysical journal.* **85.** 3991-4001 (2004)

[4] - P Pelletier et al. "COVID-19: From Biological Reality to the Social Construction of a Virus", International Conference on Innovative Studies of Contemporary Sciences (2021)