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MCM2019

PROCEEDINGS

from the

14th MULTINATIONAL CONGRESS ON MICROSCOPY

September 15–20, 2019, Belgrade, Serbia

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We are honored to host for the first time the Multinational Congress of Microscopy (MCM2019) in Serbia. The aim of MCM conferences is to become a worldwide forum for discussion on different application of various microscopical techniques for both experts and young researchers. MCM conferences have always been a good instrument for establishment of new liaisons between laboratories interested in similar projects. Trade exhibitions also helped to gain insight into the newest development of microscopy

MCM2019 is jointly organized by 8 societies: Austrian Society for Electron Microscopy (ASEM), Croatian Microscopy Society (CMS), Czechoslovak Microscopy Society (CSMS), Hungarian Society for Microscopy (HSM), Italian Society of Microscopical Sciences (SISM), Serbian Society for Microscopy (SSM), Slovenian Society for Microscopy (SDM) and Turkish Society for Electron Microscopy (TEMD)

The bit of history

Extracted from the "Opening lecture" given at the 10th Multinational Congress on Microscopy (Urbino, 4-7 September 2011) by Giuseppe Arancia,, Department of Technology and Health, Italian National Institute of Health Past President and Honorary Member of the Italian Society of Microscopical Sciences.

"In 1990, some representatives of the Italian, Hungarian, Austrian, Yugoslavian and Czechoslovak Societies for Electron Microscopy began to have contacts in order to evaluate the possibility of organizing jointly a multinational congress on electron microscopy. The inspirer reasons of this idea were, mainly, the substitution of a number of small congresses in neighboring countries with a single multinational meeting with the aim of increasing the scientific level and reducing the organizing costs, and to favor interactions and exchange of information and experiences among researchers operating in different countries."

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Combined two photon excitation fluorescence and third harmonic generation imaging of redox ratio for monitoring metabolic state of live cells of fungus *Phycomyces blakesleeanus*

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Label-free two photon nonlinear microscopy is well established as a powerful tool for monitoring metabolic state of the various cell types due to its non-perturbative nature and fairly low phototoxicity, while application of third harmonic generation (THG) for three-dimensional (3D) cell and tissue microscopy was enabled more recently. THG occurs at structural interfaces, such as local transitions of the refractive index, most generally speaking at interfaces that are formed between aqueous interstitial fluids and lipid-rich structures. Here we present preliminary data obtained by capturing both THG and optical redox ratio signal from the same regions of the hyphae of *Phycomyces blakesleeanus*.

Label- free metabolic intravital microscopy through application of both THG and $NAD(P)H^+/FAD^+$ autofluorescence ratio was used in alternating sequence on the same field of view on a fungal cells of a model filamentous fungus *Phycomyces blakesleeanus*. The glass coverslip with collagen coating bearing unstained hyphae was mounted on custom built microscope. Laser beams for both imaging modalities were focused with the same objective lens, Zeiss Plan Neofluar 40x1.3. The autofluorescence of NAD(P)H was excited by Ti:Sa laser pulses at 730 nm, 160 fs duration and signal was collected through 479/40 filter, while for autofluorescence of FAD we used excitation by the same laser pulses at 860 nm, 160 fs duration and 530/43 filter.

For THG, we used 1040 nm, 200 fs pulses from Yb KGW laser, and detection was performed by PMT through Hoya glass UV filter with peak transmission at 340nm. As a control for perturbation of optical redox ratio, rothenone (complex I inhibitor) was applied in some experiments. Nile red staining was used to confirm that the brightest structures of round shape in THG images consist of lipids and probably represent lipid droplets that serve as energy deposits in hyphea.