Metabolic imaging of erythrocyte and leukocytes plasma membrane fluidity for a quantitative biological evaluation of cardiovascular risk score in type 2 diabetes

<u>M. Daniele</u>^a, G. Bianchetti^a, F. Di Giacinto^a, C. Cefalo^b, T. Mezza^b, M. De Spirito^a, A. Giaccari^b, G. Maulucci^a

 ^a Istituto di Fisica, Università Cattolica del Sacro Cuore, Rome, Italy and Fondazione Policlinico Universitario A. Gemelli IRCSS, 00168, Rome, Italy
^b Fondazione Policlinico Universitario A. Gemelli IRCSS, 00168 ,Rome, Italy
e-mail: <u>maddalena.daniele@unicatt.it</u>

Keywords: metabolic imaging, phasor analysis, generalized polarization.

Beyond the average amount of plasma glucose, currently evaluated through the HbA_{1c}, also plasma lipids levels and oxidative stress are linked to the development of cardiovascular risk disease (CVD). To account for the influence of this complex network of factor, here we present a metabolic imaging approach to test whether changes in membrane fluidity and composition of Red Blood Cells (RBC) and polarity and composition of Polimorphic Mononuclear Cells (PMC) may be used as an indicator of cardiovascular risk in type 2 diabetes subjects (T2D).

We studied blood samples from a cohort of health and T2D patients. Among the diabetic subjects, we identified a subgroup of patients who have experienced a main cardiovascular event (T2D+CVD). Whole blood were stained with a fluidity sensitive probe (Laurdan), while PMC were stained with a polar sensitive probe (Nile Red) and with Laurdan. Spectrally-resolved images of erythrocytes and leukocytes were acquired with a confocal microscope [1].

Through the analysis of spectrally-resolved changes in Laurdan and Nile Red's emission spectra we separately quantified the different contributions affecting membrane composition, depending on fluidity, environmental polarity and on membrane lipid content. We evaluated differences among controls, T2DM patients and T2DM+CVD patients.

These preliminary data suggest that this spectral analysis could be potentially applied in assessing T2DMassociated CVD risk, complementing HbA1c in the definition of the quality of long-term management of diabetes.

[1] G. Maulucci et al., Molecular and Cell Biology of Lipids 1863 (2018) 783-793