

Synchrotron FTIR Spectroscopy Reveals New Insights into Mouse Oocyte Maturation

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Synchrotron Fourier transform infrared (FTIR) spectroscopy was applied to investigate mouse oocyte maturation *in vivo* and *in vitro*. FTIR maps of entire intact immature Germinal Vesicle (GV) and mature (MII) mouse oocytes matured *in vivo* are compared with *in vitro* matured oocytes (IVM) and *in vivo* matured oocytes aged in culture for 13 hours (designated Aged). False-colour univariate and multivariate maps show distinct lipid regions within the oocytes that vary in size and composition between the different maturation states. GV oocytes show a small intranuclear lipid deposit and another deposit located at the periphery of the egg. MII oocytes have large centrally located lipid deposits that are predominantly composed of long chain saturated fatty acids. To assess inter-oocyte variability line scans were recorded across the diameter of the oocytes from the four groups and added together from 3 independent trials (GV oocytes n = 91, MII n = 172, IVM n = 95 and Aged n = 58). The average spectra show distinct and reproducible changes in the CH stretching region and ester carbonyl region for the different oocyte types. Aged oocytes have a pronounced band at 1120 cm⁻¹ assigned to the RNA ribose skeletal vibration, which is indicative of an increase in RNA synthesis in response to repair. MII and IVM cells have very similar averaged spectra that differ significantly to the Aged and GV oocytes. The results are corroborated by performing a Principal Components Analysis on the CH stretching region, which show distinct groupings of the GV and Aged oocyte spectra but a mixed group of IVM and MII oocyte spectra. An artificial neural network (ANN) could correctly classify all spectra using absorbance values from the CH stretching region as inputs. The technique paves the way for developing an independent assay to assess oocyte maturation status and provides new insight into lipid distribution at the single oocyte level.

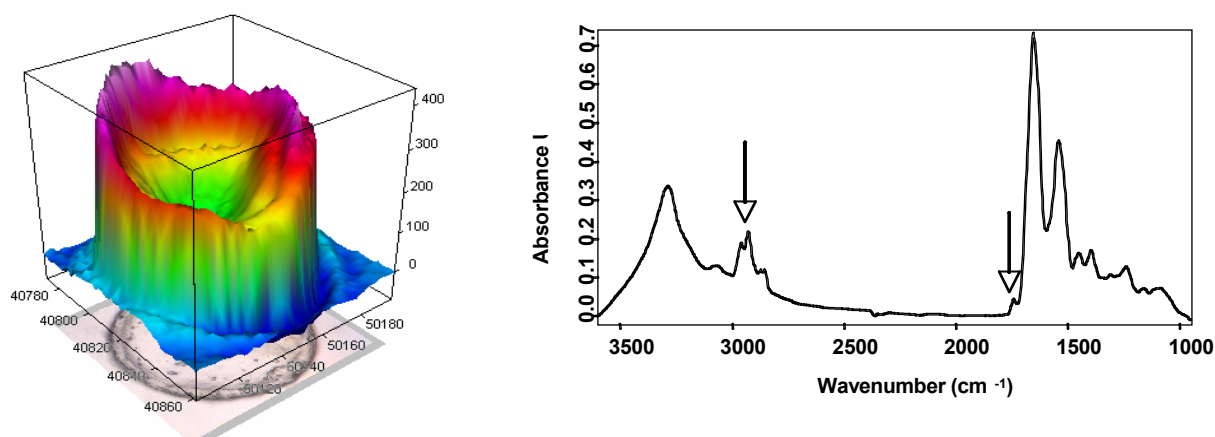


Figure 1: A. Total absorbance map of a GV oocyte showing the variable thickness of the zona pellucida and low absorbance in the nucleus. B. Mean spectra of an MII oocyte (light) and a GV oocyte (dark). Note the lipid contribution to the MII oocyte highlighted by the arrows.