Supplementary information

Correlating microscopy techniques and ToF-SIMS analysis of fully grown mammalian oocytes

Alexander Gulin^{a,b}, Victor Nadtochenko^{*a,b,c}, Artyom Astafiev^a, Valentina Pogorelova^d, Sami Rtimi^e and Alexander Pogorelov^d

^b Moscow State University, Department of Chemistry, Leninskiye Gory 1-3, 119991, Moscow, Russian Federation.

^{c.} Institute of Problems of Chemical Physics, Russian Academy of Sciences, pr. Akademika Semenova 1, 142432 Chernogolovka, Moscow Region, Russia.

^{d.} Institute of Theoretical and Experimental Biophysics, RAS, ul. Institustskaya 3, 142290 Pushchino, Moscow Region, Russia

e. Ecole Polytechnique Fédeérale de Lausanne, Institute of chemical sciences and engineering (ISIC). Lausanne, VD, CH 1015

* Phone: +7 495 9397347

E-mail: nadtochenko@gmail.com.

The 2D-molecular thin film analysis protocol for fully grown mice oocytes is described using an innovative approach. Time-of-flight secondary ion mass spectrometry (ToF-SIMS), scanning electron microscopy (SEM), atomic force microscopy (AFM) and optical microscopy imaging were applied to the same mice oocyte section on the same sample holder. A freeze-dried mice oocyte was infiltrated into embedding media, e.g. Epon, and then was cut with a microtome and 2 µm thick sections were transferred onto an ITO coated conductive glass. Mammalian oocytes can contain "nucleolus-like body" (NLB) units and ToF-SIMS analysis was used to investigate the NLB composition. The ion-spatial distribution in the cell components was identified and compared with the images acquired by SEM, AFM and optical microscopy. This study presents a significant advancement in cell embryology, cell physiology and cancercell biochemistry.

^a. N. N. Semenov Institute of Chemical Physics, Russian Academy of Sciences, ul. Kosygina 4, 119991 Moscow, Russia.



Fig. S1 Images of different oocyte sections embedded in Epon in stereomicroscope attached to ToF-SIMS. Arrow indicates the oocyte shown in Fig. 1C, Fig. 2, Fig. 4 and Fig. 5. Sections are stained by methylene blue.



Fig. S2 Mass spectra of unstained (black line) and stained (blue line) oocyte sections. (A) Positive ions. (B) Negative ions.



Fig. S3 Mass spectra of oocyte section (black line) and Epon (red line). (A) Positive ions. (B) Negative ions.



Fig. S4 m/z 30 ion image (amino acids fragment, CH_4N^+). (A) Untreated sample. (B) Sample covered by Pt layer.



Fig. S5 Detailed mass spectra of biological ions. (A) Amino acid fragment CH_4N^+ (m/z 30.0). (B) Amino acid and uracil fragments (m/z 70.1 and 70.0). (C) PO_3^- (m/z 79.0). (D) Phosphatidylcholine fragment $C_5H_{12}N^+$ (m/z 86.1).



Fig. S6 m/z 33 ions (HS⁻) image. Right side: plot profiles for line 1 and line 2.



Fig. S7 m/z 79 ions (phosphate) image. Right side: plot profiles for line 1 and line 2.



Fig. S8 m/z 86 ions (choline) image. Right side: plot profiles for line 1 and line 2.



Fig. S9 m/z 30 ions (amino acids fragment, CH_4N^+). Right side: plot profile.



Fig. S10 m/z 70 ($C_4H_8N^+$ or $C_3H_4NO^+$ or superposition of $C_4H_8N^+$ and $C_3H_4NO^+$). Right side: plot profile.



Fig. S11 Useful lateral resolution evaluation for PO_3^- ions.