Electrical Impedance Spectroscopy of Single Cells

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A new design of a microfluidic device combining hydrodynamic trapping and impedance spectroscopy measurements of single cells is presented. In a microfluidic trap structure, four microelectrodes are integrated to enable electrical impedance spectroscopy (EIS) measurements. Impedance measurements were carried out on single mouse oocytes with and without the surrounding glycoprotein matrix, the so-called zona pellucida. Higher impedance values were obtained for zona pellucida-free than for zona pellucida-intact oocytes which reflect the high electrical conductivity of the zona pellucida.

Microfluidic trap structures are frequently reported for the characterization of single cells. Suitable trapping mechanisms involve hydrodynamic, dielectrophoretic and negative pressure traps, respectively. Electrical impedance spectroscopy (EIS) as a non-invasive and label-free detection tool is often used for single cell analysis. Impedance measurements in microfluidic devices make use of integrated microelectrodes to sense impedance variation caused by changes of the dielectric properties in the small detection volume.

We developed a microfluidic system for impedance spectroscopy characterization of single cells [1]. As shown in figure 1, a hydrodynamic trap in combination with four integrated electrodes in the capture chamber provides a versatile setup.

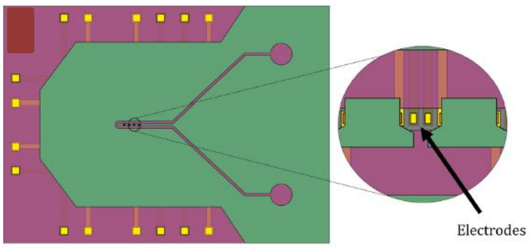

Fig. 1: Concept of microfluidic hydrodynamic traps with integrated micorelectrodes for electrical impedance spectroscopy measurements.

One pair of electrodes was placed on the bottom and two parallel facing vertical electrodes on the sidewalls of the trap, respectively. The microfluidic chip had an overall footprint of 10.6 mm x 14.3 mm and was made of several layers of lithographically structured SU-8 photo resist. The microchannel was 100 µm and the four trapping sites 86 µm wide each, respectively. Before the measurements, the interface impedance of the gold electrodes was reduced by electrochemical deposition of polypyrrole-polystyrene sulfonate *(*PPy*:*PSS*).*

As a proof-of-concept, mouse oocytes were characterized in the traps. In general, the conditions of oocytes are important in vitro fertilization (IVF) which is an established procedure in human and animal assisted reproduction. IVF is routinely employed when natural mating fails, or when livestock and genetic strains of mice must be cloned and propagated. IVF critically depends on the quality of oocytes, especially on the state of the zona pellucida (ZP), a gelatinous outer layer of extracellular matrix that spontaneously hardens and becomes impenetrable to sperm during oocyte isolation and culture.

Single oocytes were successfully loaded into the microfluidic channel. Figure 2 shows oocytes before and after trapping in the hydrodynamic trap. The ZP appeared as a visible layer of extracellular matrix, several micrometers thick.

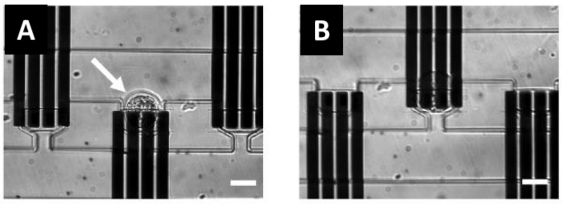

Fig. 2: Trapped oocytes. (A) With intact ZP, (B) without ZP. Scale bar: 50 µm.

EIS-measurements of ZP-intact and ZP-free oocytes were performed in one trap site using the same buffer solution. This approach minimized impedance variations caused by the buffer and thus allowed the comparison of the oocytes under identical temperature, pH, and conductivity.

As a proof-of-concept, the influence of the ZP on the impedance response of the oocytes is presented.

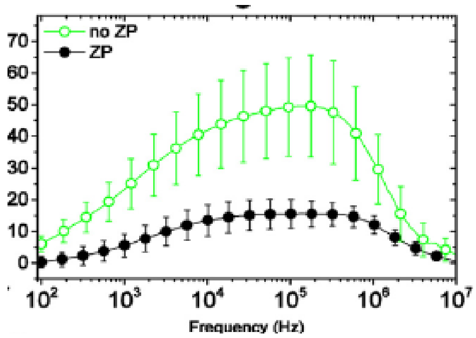

Fig. 3: Normalized impedance change of ZO-ree and ZP-intact mouse oocytes.

ZP-intact oocytes had overall smaller increase in relative impedance compared to the ZP-free oocytes.

In conclusion, electrical impedance spectra clearly distinguished zona pellucida-intact from zona pellucida-free oocytes suggesting EIS as a novel, non-destructive criterion to judge the quality of oocytes meant for in vitro fertilization.

The proposed setup may be used to determine automatized, non-destructive and label-free the quality of oocytes destined for in vitro fertilization thus obviating the need for individual microscopic inspection.

Project partners

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References

[1] A. El Hasni, C. Schmitz, K. Bui-Göbbels, P. Bräunig, W. Jahnen-Dechent, U. Schnakenberg: Electrical impedance spectroscopy of single cells in hydrodynamic traps. Sensors and Actuators B 248 419-429 (2017), doi: 10.1016/j.snb.2017.04.019