

INTEGRITY OF SPERMATOZOA AND THEIR MITOCHONDRIAL ENERGETIC STATUS IN RELATION TO MOTILITY OF SPERMATOZOA

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The purpose of our research was to determine the relationship between viability of spermatozoa and their mitochondrial membrane potential. Studies were done on ejaculated sperm of ART Lab. patients (n=57). The routine parameters of semen were determined according to the WHO criteria (1992). The morphological characteristics of Papanicolaou stained spermatozoa were analysed following strict criteria [Kruger *et al.*, Fertil Steril 46 (1986) 1118]. Sperm vitality and their mitochondrial membrane potential ($\Delta\Psi$) were evaluated by flow cytometry measurement using a FACSCalibur cytometer (Becton Dickinson, San Jose, CA, USA). Sperm viability was assessed with propidium iodide (PI) staining basing on the principle that dead cells with damaged plasma membrane take up PI. $\Delta\Psi$ was evaluated by means of a mitochondrial specific probe JC-1 exhibiting potential-dependent accumulation.

The flow cytometry evaluation of the PI stained spermatozoa revealed three subpopulations of cells: a group which did not absorb PI (live cells); a group which only partly absorbed the fluorochrome used (intermediate subpopulation); and a group which absorbed PI (dead cells). A fluorometric analysis of the JC-1 stained spermatozoa allows to determine the % of spermatozoa with functional mitochondria with high $\Delta\Psi$ (polarized mitochondria with orange JC-1 aggregates). There was a positive correlation between the % of live spermatozoa and the % of spermatozoa with functional mitochondria ($r_s=0.300$, $p<0.03$) or with the % of spermatozoa with progressive motility (rapid and slow) ($r_s=0.421$, $p<0.002$). A negative correlation was found between the % of live spermatozoa and the % of immotile cells ($r_s= - 0.295$, $p<0.03$). The correlation between the % of intermediate subpopulation of cells or the % of dead cells and the % of spermatozoa with high $\Delta\Psi$ was not significant. Moreover, there was a negative correlation between the % of intermediate cells ($r_s= - 0.328$, $p<0.02$) or the % of dead cells ($r_s= - 0.332$, $p<0.02$) and the % of cells with progressive motility.

Our results indicate that live spermatozoa reveal a high mitochondrial membrane potential resulting in their motility. In turn, the impairment of integrity of the sperm cellular membrane is not necessarily connected with disturbances of mitochondrial membrane potential. Generally, it may be concluded that motility of spermatozoa could be related to the normal function of mitochondria expressed as high $\Delta\Psi$ and to the normal integrity of the cellular membrane.