The assessment and comparison of different media and methods of sperm cryopreservation (Sperm banking)

Talebian A. (M.Sc.)¹, Sadeghi M.R. (Ph.D.)², Sadri Ardekani H. (M.D.)³, Bolourzadeh M. (B.Sc.)⁴, Akhoondi M.M. (Ph.D.)².

1- Instructor, Department of Reproductive Endocrinology & Embryology, Avesina Research Institute, Tehran, Iran.

2- Assistant Professor, Department of Reproductive Endocrinology & Embryology, Avesina Research Institute, Tehran, Iran.

3- Instructor, Department of Reproductive Endocrinology & Embryology, Avesina Research Institute, Tehran, Iran.

4- Laboratory Technician, Avesina Infertility Clinic, Avesina Research Institute, Tehran, Iran.



Introduction: Cryopreservation is a branch of cryo biology concern with the maintenance of cells during prolonged storage in the frozen state at ultra low temperature. Sperm cryopreservation is routinely performed in andrology laboratory and fertility centers and industry.Cryopreservation decrease sperm quality and its Fertilizing capacity. Evaluation of different sperm cryo-media and methods have been performed for several years. Yet, it is not known the best media and technique for sperm cryopreservation. Thus it seems very necessary to set a sperm cryo-media and method base on experimental finding. Therefore, the aim of this study is the assessment of sperm survival and motility during cryopreservation by three different media including HSPM (Human Sperm Preservation Medium), TYBG (Test-Yolk Buffer) and GEYC (Glycerol Egg Yolk Citrate) and two different cryo-techniques including programmable (with apparatus) and using vapor phase (without apparatus).

Materials and Methods: 22 samples were collected of normal male into sterile container. Sperm analysis was performed on the base of WHO criteria and freezing program including programmable and vapor phase was performed. Prefreezing sperm concentration and motility was evaluated before and after freezing and subsequently survival rate (CSF) was calculated. Each sample was divided in three parts and mixed with three different medium, subsequently at least two straws filled of each sample. One straw freezed with equipment and another one in vapor phase. The SPSS (Edition 11.0) statistical program was utilized for statistical analysis and t-test was used to compare sperm motility prefreezing and post thawing with two different cryopreservation methods and ANOVA/LSD was used to compare sperm motility with three different cryopreservation media. Significant level was considered p<0.05.

Results: Base on our result, sperm motility was $\%46.13\pm8.29$ prefreezing, but considerably decreased post thawing by programmable technique in HSPM ($\%16.9\pm5$), GEYC ($\%16.31\pm4.57$) and TYBG ($\%16.04\pm4.75$) and also by using vapor phase technique in HSPM ($\%16.95\pm4.55$), GEYC ($\%14.13\pm5.14$) and TYBG ($\%14.18\pm4.47$). There was significantly difference between programmable and vapor phase technique in GEYC (p=0.001) and TYBG (p=0.007). There was also significant difference between HSPM and both other media (p=0.05).

Conclusion: According to our study, sperm thawing after its freezing by liquid Nitrogen vapor phase in three different media (HSPM, GEYC, TYBG) leads to significant reduction in sperm motility. The best result in sperm motility and survival achieved through programmable thawing with apparatus and in comparing freezing media, HSPM media significantly has higher survival rate in comparison with TYBG and GEYC media (p-value= 0.05). Since the freezing apparatus is not available in most centers therefore, utilizing HSPM freezing media and vapor phase freezing method would be recommended for normal semen samples.

Key Words: Sperm bank, Cryopreservation, Cryo-media, HSPM, GEYC, TYBG, Slow Freezing, Vapor phase.

Corresponding Author: Dr. Akhoondi M.M., Reproductive Endocrinology & Embryology Dep., Avesina Research Institute, P.O. Box: 19835-177, Evin, Tehran, Iran. **E mail:** akhondi@avesina.ac.ir