EFFECT OF CRYOPRESERVATION ON DNA FRAGMENTATION INDEX OF SPERM IN PATIENTS WITH OLIGOASTHENOTERATOZOOSPERMIA

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INTRODUCTION

Infertility of the male factor has gradually become a key issue in the care of infertile couples, and up to 50 percent of all cases of couple infertility have been attributed to male factor.(1)

Current literature represents growing concern and attempts to retain capacity of fertility. semen cryopreservation is a simple and realistic solution available before any cytotoxic chemotherapy or radiotherapy for patients with cancer who wish to maintain fertility capacity. It is an important way to preserve reproductive ability in patients facing a possible risk of permanent and longstanding azoospermia.(2)

It is well known that cryopreservation also decreases the motility of sperm, mostly due to osmotic and thermal injury to these cells. During the freeze-thaw process, chromatin stability also decrease, with implication for DNA integrity. This was confirmed by Donnelly et al.,(3) who demonstrated an increase in the fragmentation of single-stranded DNA during cryopreservation of human sperm..

AIM OF THE WORK

The aim of this work was to evaluate the effect of cryopreservation on DNA fragmentation index of sperm in patients with oilgoasthenoteratozoospermia.

PATIENTS AND METHODS

A prospective study started from 1/6/2021 to 30/8/2022 including 30 male patients with oligoasthenoteratozoospermia in urology department in Alexandria Main University Hospital.

Patients including must fulfilling the following criteria:

-Adult male with sperm count less than 5 million per ejaculate.

- Adult male with normal sperm morphology less than 4%.

-Adult male with progressive sperm motility less than 32% A.

- Seeking fertility with no obvious female cause of infertility.

Semen analysis was performed before and after cryopreservation on each specimen.

Semen Cryopreservation: Semen samples were cryopreserved by a standard protocol using a SAGE media as the cryoprotectant.

DNA fragmentation was done on fresh semen and then on thawed “post cryo” semen.

RESULTS

Table 1: Multiple linear regression analysis to assess significant independent predictors to change of DNA fragmentation index from baseline in percent after cryopreservation which show significant association between BMI, sperm morphology and DNA fragmentation index

	Unstandardized Coefficients		Standardized Coefficients	T	Sig.
	B	Std. Error	Beta		
Step 1:					
BMI	.121	.025	1.379	4.857	<.001*
Age	.003	.017	.051	.199	.844
Sperm count	.099	.130	.141	.764	.452
Sperm morphology	-.780	.225	-.669	-3.457	.002*
Step 2:					
BMI	.125	.011	1.429	11.353	<.001*
Sperm count	.103	.126	.146	.812	.424
Sperm morphology	-.785	.220	-.674	-3.574	.001*
Step 3:					
BMI	.128	.011	1.455	12.019	<.001*
Sperm morphology	-.649	.141	-.557	-4.600	<.001*

Table 2: Comparison between fresh and post cryopreservation DNA fragmentation (n = 30) which show significant increase in DNA fragmentation index post cryopreservation

	DNA fragmentation index		Difference
	Fresh	Post cryopreservation	
Min. – Max.	12.0 – 40.0	16.0 – 100.0	2.0 – 86.0
Mean ± SD.	22.83 ± 8.40	39.64 ± 19.37	16.81 ± 16.59
Median (IQR)	22.0 (15.0 – 29.0)	36.0 (23.0 – 54.0)	9.50 (6.0 – 25.0)
Z(p)	4.783*(<0.001*)		

CONCLUSION

Cryopreservation of semen is indicated for fertility preservation in oligoasthenoteratozoospermic patient and in young cancer patients undergoing gonad toxic radiotherapy and chemotherapeutic agents. It is also beneficial prior to performing a vasectomy and in certain medical conditions which cause loss of testicular function. In some situations, cryopreservation of semen samples is done as a backup for the intended use of the sample on the day of the IVF or the ICSI procedures. Although cryopreservation increases the DNA fragmentation level of washed sperm significantly, this does not prevent us from utilization of cryopreservation facility because benefits far outweigh the adverse effects of cryopreservation.

REFERENCES

1. World Health Organization. Laboratory manual for the examination of human semen and sperm-cervical mucus interaction. 4thed. New York: Cambridge University Press; 1999.
2. Lass A, Akagbosu F, Brinsden P. Sperm banking and assisted reproduction treatment for couples following cancer treatment of the male partner. Hum Reprod Update.2001; 7: 370–7.
3. Donnelly ET, McClure N, Lewis SE. Cryopreservation of human semen and prepared sperm: effects on motility parameters and DNA integrity. Fertil Steril. 2001; 76(5):892–900.