

## Cryopreservation of Limited Sperm Using A Combination of Sucrose and Taurine, Loaded on Two Different Devices, and Thawed at Two Different Temperatures

Moloud Tahmasebi, M.Sc.<sup>1</sup>, Leila Rashki Ghaleno, M.Sc.<sup>2</sup>, Azam Dalman, Ph.D.<sup>2</sup>, Mojtaba Rezazadeh Valojerdi, Ph.D.<sup>1,2\*</sup>,

1. Department of Anatomy, Faculty of Medical Sciences, Tarbiat Modares University, Tehran, Iran

2. Department of Embryology, Reproductive Biomedicine Research Center, Royan Institute for Reproductive Biomedicine, ACECR, Tehran, Iran

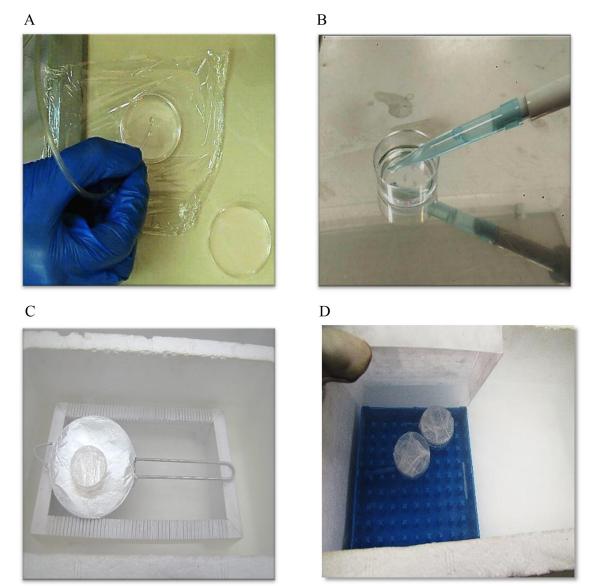


Fig.S1: The cryopreservation of limited sperm by rapid freezing method on a petri dish. A. The limited number of sperm was transferred on a medium drop in a petri dish. B. The droplets were covered by mineral oil. C. The petri dish was cooled on an aluminum sheet by nitrogen vapor. D. Transferring the petri dish into the LN tank.

Received: 11/September/2022, Revised: 21/May/2023, Accepted: 29/May/2023 \*Corresponding Addresses: P.O.Box: 14115-111, Department of Anatomy, Faculty of Medical Sciences, Tarbiat Modares University, Tehran, Iran Department of Embryology, Reproductive Biomedicine Research Center, Royan Institute for Reproductive Biomedicine, ACECR, Tehran, Iran Emails: mr\_valojerdi@modares.ac.ir, mr\_valojerdi@royaninstitute.org

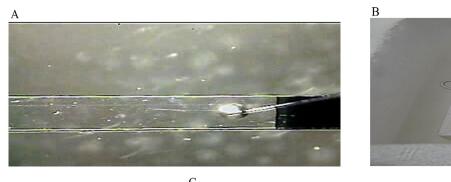
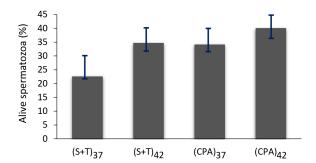


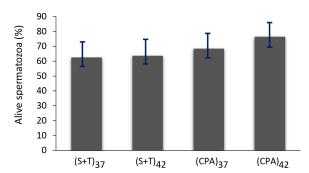




Fig.S2: The cryopreservation of limited sperm by rapid freezing method on a cryotop. A. A limited number of sperm was placed on the cryotop. B. The device was cooled on nitrogen vapor. C. Following the covering of the cryotop by the strip, the device was dropped in LN.



**Fig.S3:** Comparison of sperm viability in terms of average percentage in frozen samples with two different cryoprotectants and by microdroplet method on the petri dish and thawing at two different temperatures of 37°C and 42°C. Data represented as mean values ± SEM. The significance level was considered as P<0.05. S+T; Sucrose+Taurine and CPA; Cryoprotectant.



**Fig.S4:** Comparison of the viability of the frozen sperm with two different cryoprotectants and cryotop methods and thawing at two different temperatures of  $37^{\circ}$ C and  $42^{\circ}$ C as a percentage of the semen sample. Data represented as mean values  $\pm$  SEM. The significance level was considered as P<0.05. S+T; Sucrose+Taurine and CPA; Cryoprotectant.