

Reliable single sperm cryopreservation in Cryopiece for azoospermia management

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Objective: Our aim was to answer this question: Is a Cryopiece an efficient carrier for freezing a single or small number of human spermatozoa for ICSI?

Methods: we conducted a prospective cohort study of ICSI cycles in men suffering from severe oligospermia or azoospermia, at the Shanghai General Hospital, from 2015 through 2019. In patients with severe oligospermia or azoospermia who underwent ejaculation or surgical sperm retrieval, motile spermatozoa retrieved were subjected to cryopreservation using the Cryopiece.

Results: The prospective cohort included 27 cases. We used the Cryopiece to vitrify 573 spermatozoa, of which 573 (100%) were motile. The average number of frozen spermatozoa per patient was 21.2 ± 10.3 . After thawing, we retrieved 465 spermatozoa, of which 177 were motile, producing a recovery rate of 81.2%. The average number of thawed spermatozoa was 17.2 ± 9.2 . The recovery of 177 thawed motile sperm accounted for 38.1% of all frozen motile spermatozoa. The fertilization rate was 56.2%. Of 206 oocyte retrieval procedures, 11 (40.7%) clinical pregnancies were achieved.

Conclusion: The Cryopiece is a simple efficient carrier, optimizing the protocol for freezing a single or small number of spermatozoa. It may allow for the routine use of frozen spermatozoa after ejaculation or surgical for men suffering from azoospermia and thus avoid repeated surgeries.

| patient | Dignosis | Spermatozoa origin | No.motile spermatoza/ no.cryopiece | No. retrieved/no. vitrified spermatozoa(%) | No. motile spermatozoa/no.retrieved spermatozoa(%) | Clinic procedure | Injected MII oocyte | 2PN fertilization[n(%)] | Embryo cleaved [n(%)] | Available Embryo | outcome(pregnancy date) |
|---------|----------|--------------------|---------------------------------------|--|---|----------------------------|------------------------|----------------------------|-----------------------------|---------------------|--------------------------------|
| 1 | NOA | m-TESE | 20/2 | 18/20 (90) | 10/18 (56) | ICSI | 6 | 5 (83) | 5 (100) | 4 | Single live birth (2015-08-31) |
| 2 | Oligo | Ejaculate | 40/2 | 30/40 (75) | 12/30 (40) | ICSI | 11 | 8 (73) | 7 (88) | 3 | Twin live birth (2015-0-24) |
| 3 | NOA | m-TESE | 26/1 | 22/26 (85) | 10/22 (45) | ICSI | 8 | 5 (63) | 4 (80) | 1 | No pregnancy |
| 4 | NOA | m-TESE | 20/1 | 18/20 (90) | 6/18 (33) | ICSI | 5 | 4 (80) | 3 (75) | 2 | Single live birth (2016-09-12) |
| 5 | NOA(KS) | m-TESE | 22/1 | 18/22 (82) | 3/18 (17) | ICSI | 12 | 7 (58) | 7 (100) | 5 | Single live birth (2016-5-18) |
| 6 | NOA | m-TESE | 25/1 | 18/25 (72) | 3/18 (17) | ICSI | 2 | 1 (50) | 1 (100) | 1 | No pregnancy |
| 7 | Oligo | Ejaculate | 45/2 | 35/45 (78) | 10/35 (29) | ICSI | 18 | 3 (17) | 2 (67) | 2 | No pregnancy |
| | | Ejaculate | 39/2 | 30/39 (77) | 9/30 (30) | ICSI | 9 | 3 (33) | 2 (67) | 2 | No pregnancy |
| 8 | Oligo | Ejaculate | 21/1 | 16/21 (76) | 10/16 (63) | ICSI | 10 | 9 (90) | 9 (100) | 6 | Single live birth (2016-08-13) |
| 9 | Oligo | Ejaculate | 33/2 | 25/33 (76) | 5/25 (20) | ICSI | 5 | 3 (60) | 3 (100) | 2 | No pregnancy |
| 10 | NOA(KS) | m-TESE | 3/1 | 3/3 (100) | 0/3 | Donor semen | | | | | |
| 11 | Oligo | Ejaculate | 8/1 | 8/8 (100) | 6/8 (75) | ICSI | 6 | 3 (50) | 2 (67) | 0 | No pregnancy |
| 12 | NOA(KS) | m-TESE | 20/1 | 12/20 (60) | 0/20 | Oocyte cryopreservation | | | | | |
| 13 | NOA | m-TESE | 11/1 | 7/11 (64) | 4/7 (57) | ICSI | 4 | 3 (75) | 3 (100) | 3 | No pregnancy |
| 14 | Oligo | Ejaculate | 27/2 | 21/27 (78) | 8/21 (38) | ICSI | 8 | 6 (75) | 3 (50) | 3 | Single live birth (2017-11-10) |
| 15 | NOA | m-TESE | 6/1 | 5/6 (83) | 0/6 (0) | ICSI | 5 | 2 (40) | 0 (0) | 0 | No pregnancy |
| 16 | Oligo | Ejaculate | 21/1 | 21/21 (100) | 14/21 (67) | ICSI | 14 | 5 (36) | 5 (100) | 2 | No pregnancy |
| 17 | Oligo | Ejaculate | 22/3 | 17/22 (77) | 7/17 (41) | ICSI | 7 | 3 (43) | 2 (67) | 1 | No pregnancy |
| 18 | Oligo | Ejaculate | 14/1 | 13/14 (93) | 10/13 (77) | ICSI | 10 | 3 (30) | 3 (100) | 2 | No pregnancy |
| 19 | NOA | m-TESE | 11/1 | 9/11 (82) | 3/9 (33) | ICSI | 5 | 4 (80) | 4 (100) | 3 | Single live birth (2018-03-26) |
| 20 | Oligo | Ejaculate | 20/2 | 20/20 (100) | 14/20 (70) | ICSI | 8 | 5 (63) | 5 (100) | 2 | Single live birth (2018-07-05) |
| 21 | Oligo | m-TESE | 7/1 | 5/7 (71) | 1/5 (20) | ICSI | 1 | 0 (0) | 0 (0) | 0 | No pregnancy |

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|----|-------|-----------|------|-------------|------------|------|----|---------|---------|---|---|
| 22 | Oligo | Ejaculate | 15/1 | 11/15 (73) | 7/11 (64) | ICSI | 14 | 9 (64) | 9 (100) | 7 | Single live birth (2018-11-01) |
| 23 | Oligo | Ejaculate | 11/1 | 7/11 (64) | 3/7 (43) | ICSI | 5 | 5 (100) | 5 (100) | 2 | No pregnancy |
| 24 | Oligo | Ejaculate | 12/1 | 9/12 (75) | 1/9 (11) | ICSI | 9 | 2 (22) | 2 (100) | 2 | Singleton ongoing pregnancy (2018-11-08) |
| 25 | Oligo | Ejaculate | 22/2 | 20/22 (100) | 10/20 (50) | ICSI | 10 | 7 (70) | 7 (100) | 3 | Twins ongoing pregnancy (2019-1-8) |
| 26 | Oligo | Ejaculate | 47/2 | 38/47 (81) | 7/38 (18) | ICSI | 8 | 6 (75) | 6 (100) | 4 | No pregnancy |
| 27 | Oligo | Ejaculate | 12/1 | 9/12 (75) | 4/9 (44) | ICSI | 6 | 5 (83) | 4 (80) | 2 | No pregnancy |
