

A novel solution for freezing small numbers of spermatozoa by a sperm vitrification device

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Introduction:

The ability to cryopreserve a small number of human spermatozoa is essential in cases of severe male infertility, especially for patients requiring surgical sperm retrieval. Over the years, different methods for cryopreservation of small numbers of spermatozoa were proposed, however, each of them had drawbacks which prevented widespread use, mostly due to cumbersome preparation and sperm retrieval procedures. Thus, a need has arisen for a simple and convenient means of storing small numbers of spermatozoa.

The SpermVD®:

- › A novel means of cryopreservation of very low numbers of spermatozoa.
- › Aims to maximize fertility preservation potential for males.
- › Injection molded from a medical grade polycarbonate and sterilized by gamma irradiation.
- › Size: 26 x 8 x 7 millimeters
- › Weight: 0.2 grams.



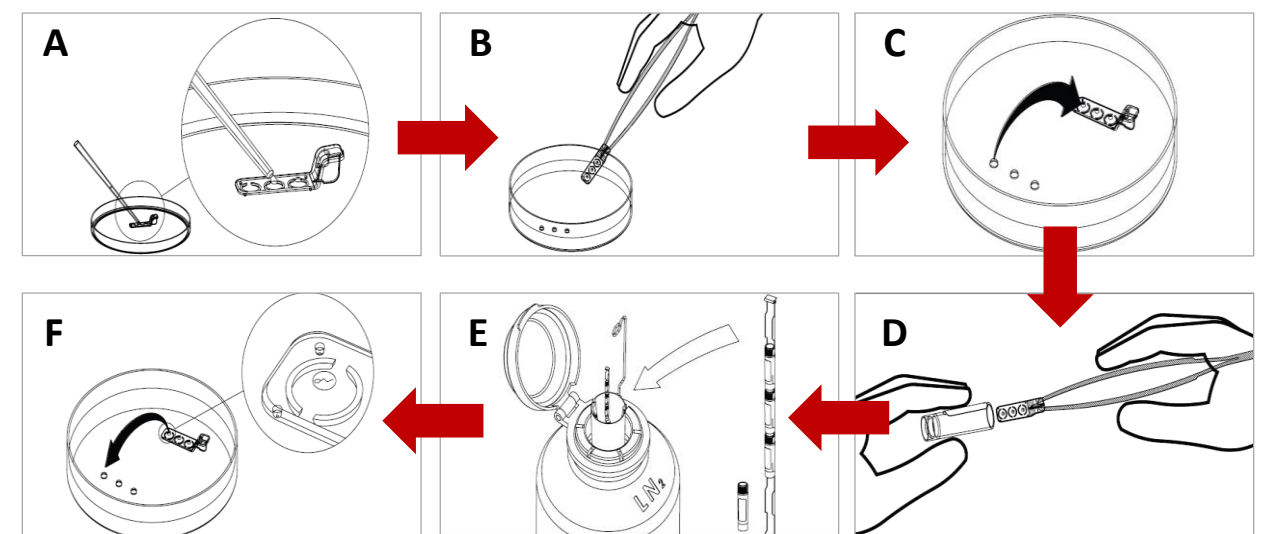
Study setting:

- › 15 patients diagnosed with virtual azoospermia.
- › 5 patients with IMSI-selected spermatozoa.
- › Prospective study 2015-2016

Methods:

After collection, sperm was centrifuged and washed. For patients diagnosed with virtual azoospermia, the re-suspended pellet was distributed into 10µL droplets on a Petri dish. The droplets were thoroughly searched and any spermatozoa found was transferred to 1µL droplet of washing medium/cryoprotectant on the SpermVD®. For IMSI patients, a morphological sorting under x6000 magnification was performed on an ICSI dish and the sorted cells were similarly transferred to the SpermVD®. The SpermVD® was then plunged into LN2. At OPU day, the SpermVD® was thawed and placed on an ICSI dish and spermatozoa were retrieved from the droplets, washed and injected.

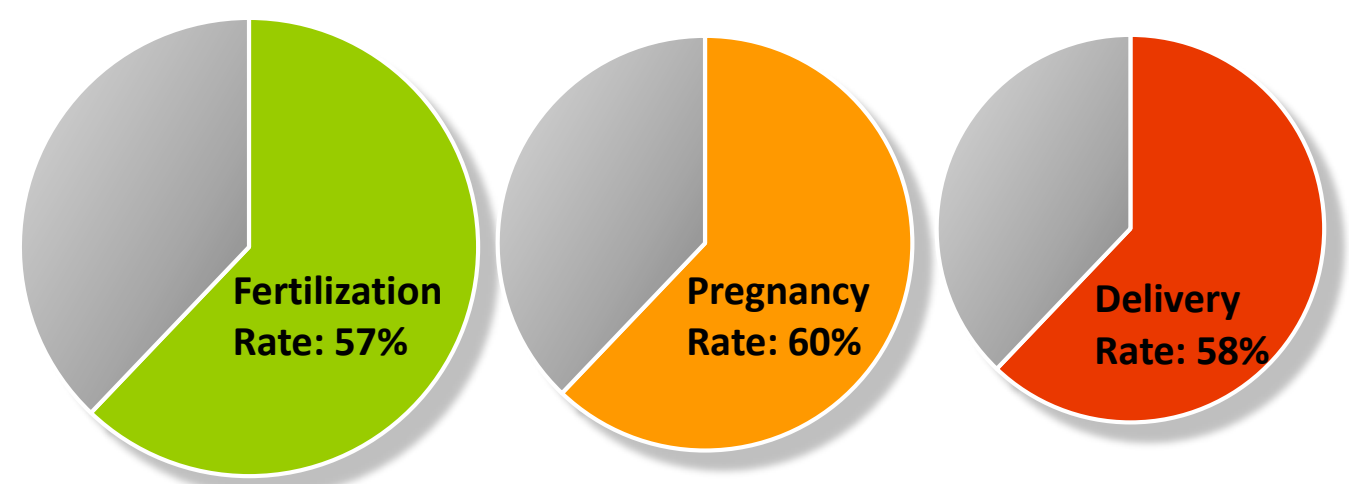
SpermVD® flowchart:



Characteristic/Outcome

Characteristic/Outcome	Average, SD, %
Male Age	37.5 ± 8.6
Female Age	33.6 ± 8.6
No. of spermatozoa frozen	17.5 ± 10.5
No. of motile frozen sperm	17.1 ± 11.1
No. of spermatozoa thawed	16.9 ± 10.2
Spermatozoa retrieval rate	96.3%
Thawed sperm motility	29%

Clinical results:



Conclusions:

- › The SpermVD® is a suitable means of cryopreservation and storage of small numbers of spermatozoa.
- › SpermVD® usage made surgical sperm retrieval at OPU day unnecessary for patients with virtual azoospermia.
- › The SpermVD® allowed optimal spermatozoa recovery and since only motile sperm were frozen, thawed immotile sperm was considered alive and suitable for ICSI.
- › The SpermVD® design allowed minimal manipulation of thawed spermatozoa, as recovered cells were immediately available for ICSI.
- › The SpermVD® allowed preliminary morphological selection of spermatozoa prior to fertilization of donor eggs abroad.

Prognosis:

- › Several SpermVD® devices could be used for storage of multiple doses of a single TESE yield and therefore reduce the need for subsequent TESE and cryopreservation of oocytes, as well as minimizing loss of excess spermatozoa.
- › The SpermVD® could make the extended sperm search a viable substitute for TESE in cases of virtual azoospermia.
- › With proper staff training, the SpermVD® could become a suitable means of transportation of frozen spermatozoa between laboratories and even countries.