Instructions for Use

ZyMōt®
MULTI (3mL)

Sperm Separation Device | ZMH3000

Important Information:
- Carefully adhere to recommended volumes for each step. Avoid over- or under-filling the device.
- Do not exceed 30 minute incubation time.
- Keep the device level during use – do not tip or rock.
- Device is single-use only and should be restricted to a single individual per device. It may not be reused.

Note on Incubation:
Good tissue practices necessitate matching media to incubation conditions. If using a bicarbonate-buffered media, incubate in a humidified, 37°C, gassed incubator. If using a HEPES-buffered media, incubate in a humidified, non-gassed incubator. If no incubator with humidity is available, add a 35mm dish of deionized or distilled water, uncovered, to the Petri dish containing the device before placing the covered dish with the device and the 35mm dish into the 37°C incubator.

Device Components:
- ZyMōt® Multi (3mL) Sperm Separation Device
- Instructions for Use

Materials/Equipment Required, But Not Supplied:
- Sperm washing solution (media): bicarbonate- or HEPES-buffered media supplemented with 2-10% protein
- 37°C incubator
- 5mL Luer-tip syringes (3)

Recommended: Norm-Ject #4050-000VZ, Henke Sass Wolf
- Sperm-safe culture tube

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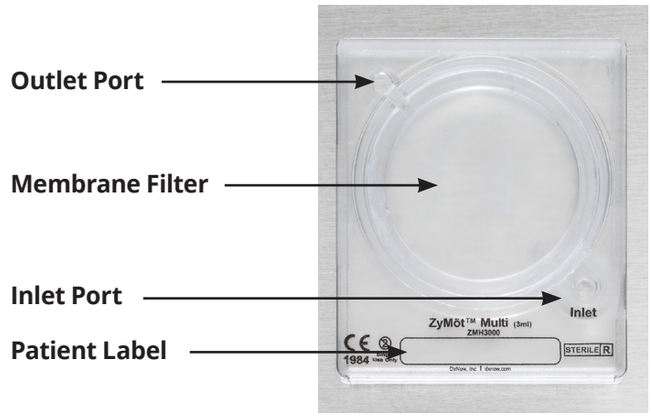

PREPARATION

1. Gather your supplies and work on a clean surface.
2. Incubate semen sample at 37°C for 20-30 minutes to allow for liquefaction.
3. Carefully open the device package without touching the device membrane.

DRAW SAMPLE

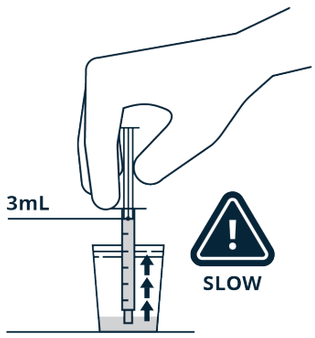

Figure 1. Draw 3mL of the sample.

4. Use a 5mL Luer-tip syringe to slowly draw a 3mL aliquot of the liquefied semen specimen. If there is insufficient volume, add sperm washing solution to give 3mL (Figure 1).

INJECT SAMPLE

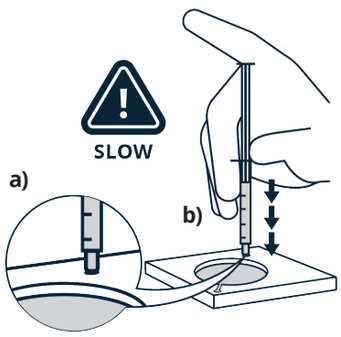

Figure 2. a) Achieve seal. **b)** Slowly inject sample.

5. Holding the syringe in a vertical position, carefully insert the tip into the Inlet Port and apply gentle pressure to achieve a seal (Figure 2a). With gentle and steady pressure, inject the sample (Figure 2b). Be careful to avoid the formation of bubbles under the membrane.

ADD MEDIA

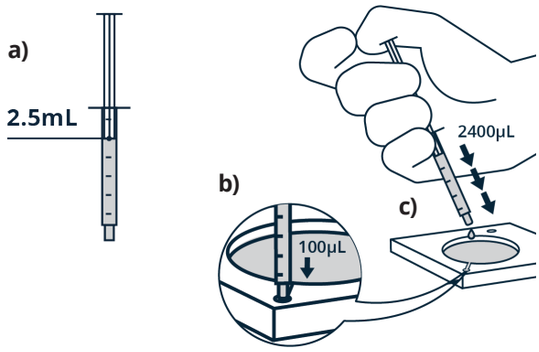

Figure 3. a) Draw 2.5mL of media. **b)** Prime outlet channel. **c)** Cover membrane surface.

6. Prepare a fresh syringe with 2.5mL of media (Figure 3a).

   a) Prime the Outlet Port/Concentration Chamber by injecting a small volume of media (approximately 100µL - Figure 3b), until the media travels through the channel to the membrane.

   b) Disconnect the syringe from the Outlet Port and apply the remaining media (2400µL) in the syringe to the surface of the upper membrane by dropping from approximately 2cm above the membrane (Figure 3c).

   c) Completely cover the upper membrane with media, making sure media touches all the edges of the upper chamber and connects with the droplet of media that was used to prime the Outlet Port.

   Note: Do not tilt the device to spread the media.

INCUBATE SAMPLE

7. Place device into a Petri dish and cover. Keep the ZyMōt device horizontal and covered at all times during the incubation. Incubate at 37°C for 30 minutes.

COLLECT SEPARATED SPERM

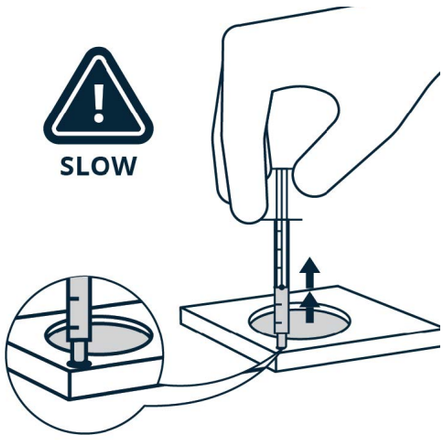

Figure 4. Slowly aspirate 1.0-1.5mL.

8. Insert a fresh 5mL syringe into the Outlet Port of the device. Slowly aspirate 1.0-1.5mL of the sperm containing fluid (Figure 4).

SAMPLE HANDLING AFTER COLLECTION FOR ICSI AND IUI

9. Transfer the collected sample to an appropriate culture tube: a 4mL round bottom culture tube with a snap top or into the bottom of a 15mL conical tube. Tubes using HEPES-buffered media may be held on the benchtop or tightly capped in an incubator. Tubes using bicarbonate-buffered media should be stored in a CO_2 incubator with the lid loosely closed.

SAMPLE HANDLING AFTER COLLECTION FOR IVF

9. Transfer the collected sample into a 15ml conical tube. Add 3ml of bicarbonate-containing media (whatever media is usually used for the final suspension of sperm for conventional insemination) to the conical tube. Mix gently. Centrifuge the conical tube for 5 min at 300 x g. Remove the supernatant, being careful to not disturb the lower pellet. Perform count and motility as usual and dilute if needed to achieve appropriate final insemination concentration. Store tube in a CO_2 incubator until insemination. Insemination should occur more than 1 hour, but less than 4 hours after preparation.

Tips, Warnings and Precautions:
- Caution: Sale of this device is restricted to sale by or on the order of a physician.
- Device should be used only by properly trained operators.
- Practice universal precautions when handling human body fluids.
- Do not use if the packaging is damaged.
- Do not clean device with any solutions. Spraying or wiping with cleaning solutions can damage the device.

Device Description:
ZyMōt ICSI and ZyMōt Multi are sperm separation devices used to prepare motile sperm for assisted reproductive technology (ART) procedures. Both devices separate sperm based on motility. The ZyMōt ICSI and the ZyMōt Multi are sterile and single use only. The mechanism of action for both is separation of sperm based on motility within a microenvironment created by the micro channels of the ZyMōt ICSI or the micropores in the filter of the ZyMōt Multi. The primary difference between the devices is the processing volume. The ZyMōt ICSI has a processing volume of 2µL per micro channel. The ZyMōt Multi is manufactured in two (2) processing volumes, 850µL and 3mL.

The ZyMōt Multi (provided with 850µL and 3mL collection chambers) has an inlet port that communicates with the lower sample chamber. The sample chamber is separated from the upper collection chamber by a microporous filter. Untreated semen is added through the inlet port. After 30 minutes, the separated sperm are collected from the upper chamber through the outlet port/concentration chamber.

Indications for Use:
The ZyMōt Multi (850µL) Sperm Separation Device is intended for preparing motile sperm from semen for use in the treatment of infertile couples by intracytoplasmic sperm injection (ICSI), in vitro fertilization (IVF), and intrauterine insemination (IUI) procedures.

Sterilization:
The sterilization method used for the ZyMōt devices is gamma radiation, at a dose level of 25kGy to 45kGy by the VD_{max}^{25} method to meet a Sterility Assurance Level of 10^{-6}.

Storage:
Store at 60°F - 77°F (15°C - 25°C).

Disposal:
Discard the used device and materials as medical waste.

Testing Performed for Devices Used in Assisted Reproduction:
Specific testing was performed for toxicity and functional screening appropriate for products used in assisted reproduction. As required by USFDA 21 CFR 884.6160, the following Special Controls were conducted (all tests were passed): human sperm survival assay (replacing the mouse embryo assay) and endotoxin testing.

Endotoxin Testing Results:
Using the Limulus Amebocyte Lysate (LAL) Analysis by the Gel-Clot Method, results were <0.0729 EU per device, which meets the acceptance level of ≤20 EU per device.

Human Sperm Survival Assay Results:
Using the Human Sperm Survival Assay, results were 96.2% for ZyMōt ICSI and 97.7% for ZyMōt Multi; both results meet the acceptance level of motility ≥80% of control at 24h after exposure for 30min. Note: The above results are from testing required prior to USFDA 510(k) clearance. These tests are conducted on each manufacturing lot of devices as part of the lot release program. A CoC can be provided upon request.

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USA Patent US10422737B2; US1100944B; Japan Patent JP6524082B; Canada Patent CA2931201C; Australia Patent AU2014353050B2; Brazil Patent BR1120160112334. Additional USA and other international patents pending.

ZyMōt FERTILITY