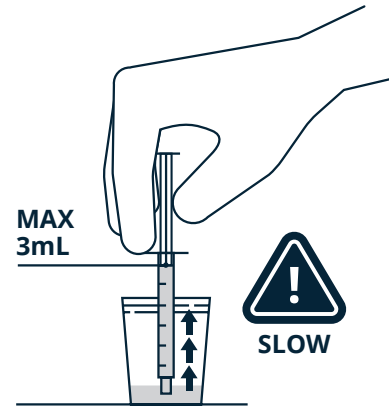
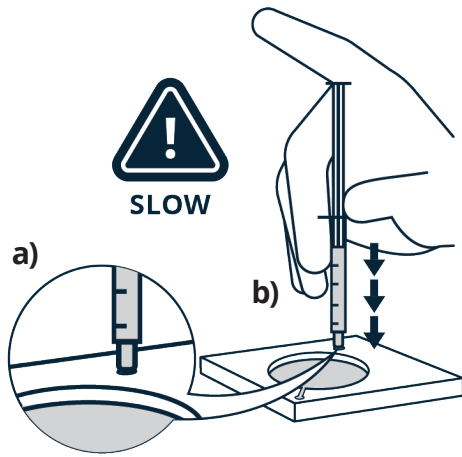


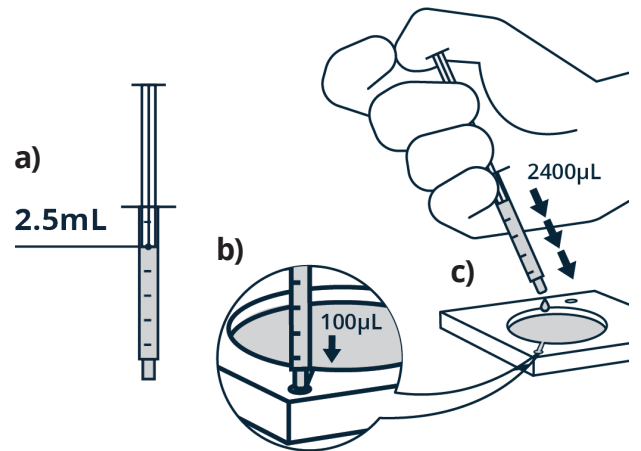
**1** Allow sample to liquify



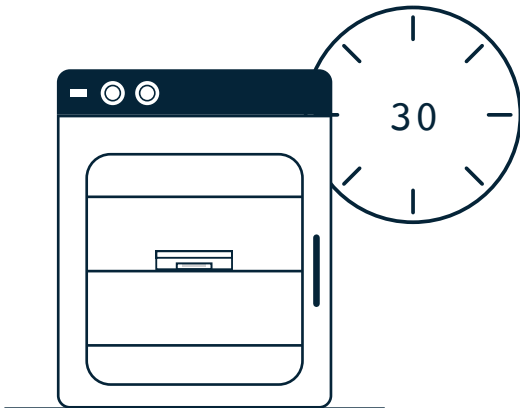
**2** Draw 3mL of the sample.



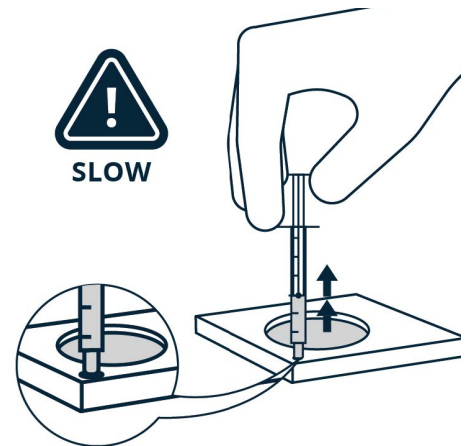
**3** a) Achieve seal. b) Slowly inject sample.



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



**5** Incubate at 37°C for 30 minutes.



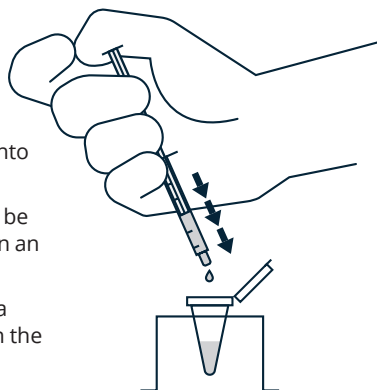
**6** Slowly aspirate 1.0-1.5mL.

#### ICSI and IUI

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<sub>2</sub> incubator with the lid loosely closed.



#### IVF

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 5min 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<sub>2</sub> incubator until insemination.

Insemination should occur more than 1hr, but less than 4hrs after preparation.

**7** Sample Handling After Collection – ICSI and IUI

**7** Sample Handling After Collection – IVF