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Microfluidic separation of motile sperm with millilitre-scale sample capacity REZA NOSRATI, MARION VOLLMER, LISE EAMER, University of Toronto, KRISTA ZEIDAN, MARIA C. SAN GABRIEL, ARMAND ZINI, McGill University, DAVID SINTON, University of Toronto — Isolating motile from non-motile spermatozoa has been a challenge since the establishment of *in vitro* fertilization. Microfluidic approaches have been employed for this purpose, but current devices are limited by low sample volume. Here, we present a high-throughput microfluidic device that separates spermatozoa from one millilitre of raw semen sample based on the hydrodynamic characteristics of swimming sperm in a confined geometry. The device consists of two layers: an outer injection ring on top aligned with a network of radial microchannels at the bottom guiding motile sperm into an inner collection chamber. This approach (1) maximizes exposure of the sperm to the fluid channels, (2) maximizes surface area density (3) prevents fluid flow bias, and (4) employs a non-Newtonian viscoelastic medium consistent with the *in vivo* environment. Tests with human and bull spermatozoa indicate an increase in motile sperm concentration from 62.2% in raw semen to 99.2% in separated sample combined with a higher incidence of normal morphology. DNA integrity testing is currently underway. In conclusion, we present an effective one-step procedure to perform semen purification and separation on a millilitre-scale with clinically relevant numbers.

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