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JOURNAL ARTICLE

# Selection of acrosome-reacted sperm with MH61-immunobeads

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Separation of acrosome-reacted viable sperm is an important problem in the analysis of sperm function and the micromanipulation of gametes such as the microinjection of sperm into ooplasm and subzonal sperm insertion. In the present study, we collected acrosome-reacted sperm selectively using paramagnetic immunobeads, referred to as MH61-beads, which attach themselves to the head of the acrosome-reacted sperm and

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are recovered by a magnet. The maximum number of sperm was recovered by the MH61-beads when 3 x 10 (5) sperm/100 microliters and 6 x 10(5) MH61-beads were coincubated for 1 hour. The recovery of sperm was significantly correlated with the percentage of the acrosome reaction in the capacitation medium. After incubation for 12 hours, 2.4-9.0 x 10(4) sperm could be collected, and the percentage of viable and acrosome-reacted sperm was 28-60%. To improve the recovery of the acrosome-reacted sperm, we utilized follicular fluid, which induces the acrosome reaction. Addition of follicular fluid to the capacitation medium at a final concentration of 20% induced the acrosome reaction and increased the recovery of acrosome-reacted sperm significantly. In conclusion, separation of acrosome-reacted sperm by the MH61-bead-binding method could become a useful technique in research of sperm function and gamete micromanipulation.

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