# RBC Depletion Using Magnetic Beads

## A. Materials Provided

1. Rabbit anti-Human RBC Magnetic Beads, catalog #'s 01-1801-20 or 01-1801-50

## B. Additional Materials (not provided)

1. Binding/Wash Buffer (1x PBS; catalog #MB-008)
2. Micro-pipettes with disposable plastic tips (12-200 and 200-1000 µl)
3. 1.5ml or 2ml microcentrifuge tubes
4. Timer
5. Rotator
6. Microfuge
7. Magnetic Separator capable of holding 1.5 to 2.0 ml tubes (TMS-06 or TMS-32)

## C. Procedure

***Note:*** *Process may vary depending on nature & origin of blood sample, suggest 250-500 µl beads/ml whole blood for 80-90% removal of RBC's, but end user should test to determine optimum amount needed.*

*All procedures can be performed at room temperature or on ice if required for blood cell experiments*.

1. Resuspend beads by gently inverting bottle. Remove required amount of magnetic beads from bottle and place in a microcentrifuge tube.
2. Wash beads twice with PBS by adding 1x volume of PBS equal to the amount of bead volume. Gently invert tube to mix and then place on magnetic separator. Leave for 2 minutes or until all beads have been drawn to the magnet. While leaving tube in the separator remove the PBS, do not disturb the beads. Discard PBS. Remove the tube from the separator and repeat the wash step.
3. After washing, resuspend the beads in original bead volume of PBS. Gently invert to mix. *Do not vortex*.
4. Aliquot blood samples to new microfuge tubes. Add between 250 – 500 µl beads to blood. Incubate 10-30 minutes on a rotator. Optimal incubation time should be optimized by end user.
5. After incubation, place tubes on the magnetic separator for 5 minutes. Remove supernatant. The RBC depleted sample will be in the supernatant. If there are still RBC's remaining repeat Step 4.
6. Process RBC depleted sample as required for further experiments.