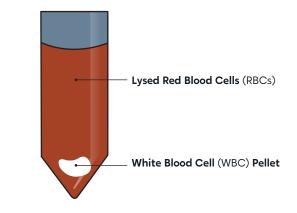
Isolation of White Blood Cells from Whole Blood

For many genomic applications it is desirable to isolate white blood cells (WBCs) from the other constituents of whole blood. This protocol provides a rapid and effective method to isolate WBCs by lysing red blood cells (RBCs) and pelleting the WBCs. The WBCs can then be processed with the lonic[®] Cells to DNA Kit (Product #: 33005).

REAGENTS REQUIRED

- 1 RBC lysis buffer: 1x Red blood cell lysis buffer (ThermoFisher Scientific Cat No: 00-4333)
- 2 PBS: Dulbecco's phosphate-buffered saline no calcium, no magnesium (ThermoFisher Scientific Cat No: 14190144)



PROTOCOL

- 1 Add 10 mL of 1x RBC Lysis Buffer per 1 mL of whole (human) blood.* *This can be scaled up to 3x, e.g.: 30 mL of 1x RBC Lysis Buffer per 3 mL of blood.
- 2 Incubate for 10 minutes at room temperature (RT) with shaking at 300 RPM. Do not allow the reaction to exceed 15 mins.
- **3** Centrifuge the cells down at 500 RCF for 5 minutes. Remove the supernatant and observe the pellet. It should be white.
 - If there is residual blood present, the pellet will appear red, if this is the case, repeat RBC Lysis using another 10 mL of 1x RBC Lysis Buffer. Repeat until cell pellet is white.
- 4 Resuspend the cell pellet in 10 mL of 1x PBS and perform a cell count at this time.
- **5** Aliquot cells at desired density (maximum of 5 x 10⁶ total cells) and pellet cells by centrifugation at 500 RCF for 5 minutes.
- **6** Aspirate off the supernatant and proceed with Lysis or freeze and store these dry cell pellets at -20°C for future use.
 - It is essential that as much as possible of the supernatant is removed as carryover supernatant can interfere with purification.

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