

Red Blood Cell Lysis Buffer

Catalog# BWR1003

Size: 100 ml

Lot # Check on the product label

Introduction

Red Blood Cell Lysis Buffer is specially used for lysing human erythrocytes in single-cell suspensions of peripheral blood and mouse hematopoietic tissues such as spleen. For the major effective ingredient, ammonium chloride, this buffer hardly damage the lymphocyte or other cells with nucleus during the lysis of erythrocytes. Mouse thymus and lymph gland cells do not need this lysis buffer. For the sterile filtration, the obtained blood, tissues or cells can be used for primary culture, cell fusion, nucleic acid or protein extraction and various conventional assay and detection.

Kit Components

Components	Size	Storage Instruction
Red Blood Cell Lysis Buffer	100 ml	Stored at 4°C for one year. Or store at room temperature for 3 months.

Protocol

● Tissues and cells sample

1. Digest the fresh tissues, scatter to suspension by appropriate method. Collect mouse spleen cells to prepare a single-cell suspension.
2. Pellet the cells by centrifugation at 400-500 g at 4°C and aspirate the supernatant.
3. Add Red Blood Cell Lysis Buffer into cell pellet (3-5 times than cell pellet volume), i.e. Add 3-5 ml of Red Blood Cell Lysis Buffer into 1 ml of cell pellet. Stroke slightly and mix thoroughly, and incubate on ice for 4-5 min. During lysis, shake occasionally to promote erythrocytes lysed. (This step can be operated at room temperature or 4°C).
4. Centrifuge at 400-500 g at 4°C for 5 min, remove the red supernatant.
5. Repeat the Step 3 and 4 once if the erythrocyte lysed incompletely. Usually, very small volume of erythrocyte do not affect the follow-up detections.
6. Resuspend the pellet in PBS, HBSS, normal saline or serum-free medium and wash for 1-2 times. Centrifuge at 400-500 g at 4°C for 2-3 min, discard supernatant. The volume of wash buffer should be at least 5 times than cell pellet volume.
7. Perform a cell count after resuspending the pellet.

FOR RESEARCH USE ONLY, NOT FOR DIAGNOSTIC AND CLINICAL USE.

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- **Blood sample**

1. Centrifuge the fresh anticoagulant blood at 400-500 g for 5 min at 4°C and remove the supernatant.
2. Add Red Blood Cell Lysis Buffer into cell pellet (6-10 times than cell pellet volume), i.e. Add 6-10 ml of Red Blood Cell Lysis Buffer into 1 ml of cell pellet. Stroke slightly and mix thoroughly, and incubate on ice for 4-5 min. During lysis, shake occasionally to promote erythrocytes lysed.
3. Centrifuge at 400-500 g at 4°C for 5 min, remove the red supernatant.
4. Repeat the Step 2 and 3 once if the erythrocyte lysed incompletely. Usually, very small volume of erythrocyte do not affect the follow-up detections.
5. Resuspend the pellet in PBS, HBSS, normal saline or serum-free medium and wash for 1-2 times. Centrifuge at 400-500 g at 4°C for 2-3 min, discard supernatant. The volume of wash buffer should be at least 5 times than cell pellet volume.
6. Perform a cell count after resuspending the pellet.

Note: For very small volume of blood sample, ignore Step 1 and operate Step 2 directly, add Red Blood Cell Lysis Buffer into cell pellet (10 times than cell pellet volume), and incubate at room temperature or 4°C for 4-5 min. For mouse blood sample, it is enough to lyse for 4-5 min, for human peripheral blood, increase the lysis time to 10 min, but do not exceed 15 min. During lysis, shake occasionally to promote erythrocytes lysed. And the following steps are the same.

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