

Whole Blood Stabilization Protocol

Using Bulk-Supplied TokuKit Buffers

APPLICATION	SAMPLE AMOUNT	KIT
Human whole blood	2 mL whole blood / sample	TokuKit Buffer Pack Cat. No. TokuKit-BP-20

1 INTRODUCTION

This protocol describes how to collect and stabilize human whole blood using the TokuKit buffers, stabilizing each sample for storage and downstream analysis. The buffers are provided in bulk—one bottle of Buffer 1 and one bottle of Buffer 2—and the user measures out the per-sample volumes.

All volumes in this protocol are based on processing **2 mL of whole blood per sample**. Buffer volumes are derived from a fixed ratio of whole blood to Buffer 1 to Buffer 2 (below), so the same ratio can be applied if the user scales the blood volume.

BUFFER VOLUME RATIO

1 : 1.4 : 4

Blood : Buffer 1 : Buffer 2

PER SAMPLE (2 mL WHOLE BLOOD)

Whole blood	2 mL
Buffer 1	2.8 mL
Buffer 2	8 mL

2 MATERIALS

Bulk buffers provided as part of the TokuKit Buffer Pack, sized for **20 samples (20 × 2 mL = 40 mL whole blood)**:

Item	Bulk volume supplied	Per-sample volume
Buffer 1	56 mL	2.8 mL
Buffer 2	160 mL	8 mL

Also required (not supplied):

- EDTA or Sodium Heparin blood collection tube(s) for whole blood collection
- 15 mL or 50 mL conical tubes (one per sample depending on volume of blood)
- Calibrated serological pipettes or equivalent for measuring buffer volumes
- Dry ice and/or a –80 °C freezer

3 PREPARATION

Confirm the sample count. The bulk Buffer Pack is sized for 20 samples at 2 mL whole blood each. Confirm the user has enough Buffer 1 (2.8 mL/sample) and Buffer 2 (8 mL/sample) for each sample to be collected.

Label one conical tube per sample. Use a 15 mL or 50 mL conical tube per sample (depending on blood volume) so there is space for the added buffers (12.8 mL total, assuming 2 mL of whole blood per test).

Invert the blood collection tube. Invert the whole blood collection tube 10 times to mix immediately before use.

4 PROCEDURE

- 1 **Collect whole blood.** Collect whole blood into an EDTA or Sodium Heparin blood collection tube.
- 2 **Invert the collection tube.** Invert the whole blood collection tube 10× to mix before use.
- 3 **Add blood to a conical tube.** Add 2 mL of whole blood to a labeled conical tube.
- 4 **Add Buffer 1.** Measure 2.8 mL of Buffer 1 from the bulk bottle, add it to the blood, then close the tube.
- 5 **Invert and incubate.** Invert 10× and incubate for 15 minutes at room temperature.
- 6 **Add Buffer 2.** Open the tube, measure 8 mL of Buffer 2 from the bulk bottle, add it, then close the tube.
- 7 **Mix and freeze.** Invert 10× and place immediately on dry ice for transport to –80 °C, or place immediately in a –80 °C freezer within 15 minutes of mixing.

5 NOTES & TIPS

Anticoagulant	Use either EDTA or Sodium Heparin blood collection tubes. Avoid other anticoagulants unless confirmed with Teiko.
Mixing	Always invert—do not vortex. Inverting 10× after each buffer addition ensures even, gentle mixing.
Timing	The 15-minute room-temperature incubation after Buffer 1 is required. Add Buffer 2 promptly afterward, mix, and freeze immediately or within 15 minutes of mixing.
Freezing	Get samples onto dry ice or into a –80 °C freezer immediately or within 15 minutes of mixing. Avoid any hold time at room temperature if possible once Buffer 2 is added.
Scaling	To process a different blood volume, keep the 1 : 1.4 : 4 ratio. For example, for 1 mL blood use 1.4 mL Buffer 1 and 4 mL Buffer 2; the bulk Buffer Pack covers up to 40 mL of whole blood total.
Measuring from bulk	Measure each buffer with a clean pipette to avoid cross-contamination between the Buffer 1 and Buffer 2 bottles.
Antibody compatibility	For antibodies and clones we've demonstrated work on TokuKit-fixed samples, see the TokuKit antibody clone compatibility list on teiko-labs.com/tokukit .

Questions about your TokuKit run? Reach out to your Teiko contact or visit kits.teiko.bio.

<https://teiko-labs.com/tokukit-in-house-testing/spectra1k1-cytometry>