

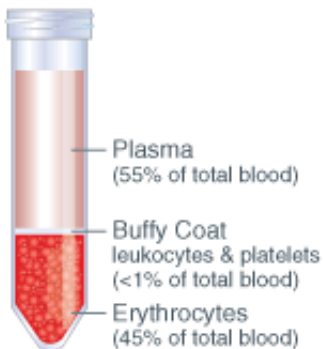


Preparing a Buffy Coat from Whole Blood

A buffy coat suspension is a concentrated leukocyte suspension. It is not mononuclear as the granulocytes are still present. The following brief protocol demonstrates how to prepare a buffy coat suspension from whole blood.

1. Collect whole blood in a blood collection tube containing anticoagulant.
2. Add 1 part PBS + 2% FBS to 1 part fresh whole blood.
3. Centrifuge the sample at room temperature and 200 x g for 10 minutes with the brake off. ([G to RPM Converter](#))
4. Remove the concentrated leukocyte band (this is the buffy coat), and a small portion of the plasma and concentrated RBCs. Platelet contamination in the buffy coat can be decreased by collecting less of the plasma.

Figure 1: Schematic of centrifuged whole blood showing different cell fractions



After centrifugation, one can distinguish a layer of clear fluid (the [plasma](#)), a layer of red fluid containing most of the [red blood cells](#), and a thin layer in between, making up less than 1% of the total volume of the blood sample. The thin layer is called the buffy coat (for its buff-colored hue), and it contains most of the white blood cells and [platelets](#).