

Protocol 602_Blood FACS

Reagents:

- FACS buffer
- 10x BD FACS lysis solution (BD Biosciences 349202)
- FACS tubes (VWR 352052)

Method:

1. Collect 2-3 drops of blood directly into FACS buffer containing FACS tubes.
2. Centrifuge at 4C for 5min at 1500rpm. Pour off supernatant.
3. Add 100µl of FACS antibody cocktail and incubate at 4C for 15-20min in the dark.
4. Wash 1x with FACS buffer. Centrifuge at 4C for 5min at 1500rpm. Pour off supernatant.
5. Dilute BD FACS lysis solution 1 in 10 in ddH₂O. Add 1ml of the lysis solution to the FACS tubes while vortexing. Incubate for 5-10min at room temperature until solution becomes clear (red blood cells are lysed and haemoglobin is in solution).
6. Fill up with FACS buffer and centrifuge at 4C for 5min at 1500rpm. Pour off supernatant.
7. Add 400µl FACS buffer. Cells are now ready for acquisition on FACSCalibur.

Notes

The BD FACS lysis solution lyses the red blood cells and fixes the remaining blood cells at the same time. Therefore samples can also be acquired the next day. For Blood FACS typing (eg. Smarta) one can use the quick and dirty protocol where you leave out step 4. The FACS buffer contains EDTA, therefore blood can be collected directly into the FACS tubes. If this is not possible, blood needs to be collected into EDTA coated collection tubes to avoid agglutination.