A Make Slides

1. Aliquot Cells
   - Minimize exposure of Smear at room temp.
   - Aspirate drop using a Pasteur pipette adapted with a pipette tip
   - add 1 ml of 1X PBS and pipette cells up and down to mix (Do not vortex)
   - Aspirate leaving approximately 20 µl of 1X PBS
   - Mix cells by pipetting to uniformly mix cells

2. Add approx. 1 ml of cells to cells and spread evenly to distribute cells
   - let cells air dry

3. Fix in 1:1 Acetone/Methanol at -20°C for 10 min

4. Air Dry

5. Block Incubate with 20% Goat Serum 30 min at room temperature (Humidity Chamber)

6. Wash with 1X PBS

7. Incubate (Humidity Chamber) with Specific antibodies
   1: a Goat E 700-200 dilute in PBS
   2: a 1:100 700-200 dilute in PBS
   - incubate 1-2 hrs at room temp

8. Wash well with 1X PBS 4-5 5 minute washes

9. Incubate with Goat or Mouse Ig - FITC 700-2000

10. Wash well with 1X PBS

11. Keep moist

12. Add DAPI 10 µl and cover slip. Flip slide and press firmly to remove any bubbles trapped
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1. Goat Serum
2. α-Bamz or αGP11D
3. Goat x Mouse Ig-FITC

\[ \text{BSA} \rightarrow \text{BSA} / \text{Bamz}, \text{BSA} / \text{Bamz} \]
\[ 4.3 \times 2 \times 10^4 = 8.6 \times 10^5 = 860,000 \text{ cells/ml} \]

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<td>#1</td>
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<tr>
<td>Volume</td>
<td>2.0ml</td>
<td>1.5ml</td>
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<tr>
<td>Cells</td>
<td>1.7mill.</td>
<td>1.3mill</td>
<td>900,000</td>
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<tr>
<td>cells/ml</td>
<td>85,000</td>
<td>65,000</td>
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\[ 40 \mu l \text{ cells} + 40 \mu l \text{ trypan blue} = X \text{ df} \]