

### Erythrocyte lysis on plate

- Transfer 50-100µl of blood per well (96-well plates).
- Top up to 200µl with PBS and wash (500-600g, 2min., 4°C).
- Drain the supernatant.
- Transfer 200µl ACK solution per well and mix thoroughly (repeatedly pass through pipette tips after ACK transfer) and wash until the solution clears (the otherwise turbid solution becomes transparent - approx. 10min., depends on the volume of ACK added).
- Wash with 200µl PBS (1-3x, until supernatant is no longer red).
- If sediment is still red, repeat lysis. In this case, wash after 5 minutes after addition of 200µl ACK.
- Wash 2-3x with 200µl PBS – never perform third lysis session.

For other lysis purposes the ratio of blood:ACK added should be 1:3-4 and can be performed in two steps as described above.

#### ACK:

16.58 g	NH <sub>4</sub> CL
2g	KHCO <sub>3</sub>
0.744g	EDTA
H <sub>2</sub> O	fill up to 200ml
pH	set to 7.4