

FACS analysis

Fixation:

Need 2×10^7 trypanosomes for each analysis.

Harvest cells by centrifugation at 1,500 rpm, 10 min at 4°C

Wash twice with cold PBS (PF) or TDB (BF) +2 mM EDTA.

Resuspend cells in 200µl PBS (PF) or TDB (BF) +2 mM EDTA in a 15 ml conical tube

Fix by adding 2 ml of ice-cold 70% EtOH dropwise while vortexing on high speed

Store at 4°C for at least overnight: cells are stable for weeks

Staining:

Spin cells for 10 min at 1,500 rpm at 4°C

Resuspend pellet in 0.5 ml staining solution: per ml:	950µl PBS/2 mM EDTA
	20µl RNaseA 10 mg/ml
	50µl Propidium Iodide (1 mg/ml)

Incubate at 37°C for 30 min

Proceed with FACS analysis