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Biomedical Engineering

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FACS - DBCO-PEG10kDa

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1 Stock solutions

- DBCO-PEG 10 kDa in DMSO (300 μ M)
- Buffer: PBS

2 Preparation of FACS samples

- Prepare following tubes:

Tube	[DBCO]	Cells (10^9)	DBCO-PEG10kDa (μ L) (300 μ M)	DBCO/tag ratio
1	0	200 μ L		
2	30 μ M	200 μ L	22.2	300.5

- Make sure you vortex the cell well before and after adding the DBCO-PEG10kDa
- React DBCO tubes for 1h to 6h in shaking block at 4°C and 300rpm
- Prepare FACS samples after 1h and 6h:
 - Spin down the cells for 5 min at 13,400 rpm and discard the supernatant
 - Resuspend with 1 mL ice cold PBS
 - Spin down the cells for 5 min at 13,400 rpm and discard the supernatant
 - Put the pellets on ice until FACS
 - Right before FACS: resuspend with 200 μ L ice cold PBS