

10. (October) 2019

Project: iGEM_Munich2019 Shared Project

Authors: Anja

WEDNESDAY, 9/10/2019

Johanna, Sarah

His-Tag Purification (Protocol: His-Tag-Purification (Gravity Flow, 2 mL sample)).

Constructs Tested:

- E10: CD63-L7Ae
- E11: CD63-MCP
- E16: CD63_Ser161_6xHis-L7Ae
- E17: CD63_Ser161_6xHis-MCP
- E19: CD63_Asn180_6xHis-L7Ae
- E20: CD63_Asn180_6xHis-MCP

-> All six constructs were tested in HEK293T and MIN6-K8 cells.

Alejandro

Cell culture: Transfection of HEK293T for Cytotoxicity (LDH) Assay at 24,48,72h and long-term cell-monitoring (4x 24-well plates, see entry in VLP lab book)

Joshi

HEK 293T harvesting from 24-well plate:

-600µl harvested and centrifuged (according to VLPs and exosome harvesting)

-200µl of Total Exosome Isolation reagents added to 400µl supernatant (top part after centrifugation)

-incubation in the fridge ON

Anja

EXO HiBiT Assay for MIN6-K8 cell line (same as for HEK293T, according to VLPs / Exosomes harvesting & HiBiT assay from 96-well plates)