

# 07. (July) 2019

**Project:** iGEM\_Munich2019 Shared Project

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## HiBit Assay:

- harvesting according to harvesting protocol at ~11:20 (72h post transfection, 24h post medium change)
- The HiBit assay was carried out according to the standard HiBit protocol for 96 well plates.
- the INVERSE measuring scheme is :

measurement scheme												
	1	2	3	4	5	6	7	8	9	10	11	12
A	1 fmol HiBit protein	1 fmol HiBit protein	1 fmol HiBit protein	1 fmol HiBit protein	1 fmol HiBit protein	1 fmol HiBit protein	1a SI	1b SI	1c...	2a...	2b	2c
B	3a	3b	3c	4a	4b	4c	5a	5b	5c	6a	6b	6c
C	7a	7b	7c	8a	8b	8c	9a	9b	9c	10a	10b	10c
D	1a Su	...	...									
E												
F				...	...	10c Su	1a CC	...	...			
G												
H										...	...	10c CC

- some samples were stored to carry out a Western Blot : conditons 4,5 and 6 ; CC, SI, Su from each one. 2µL from each replicate were pooled and diluted 1:10 as follows :  
 -> 2µL a + 2µL b + 2µL c  
 + 54µL PBS with PI 1:100  
 + 60µL Laemmli Buffer 2X
- at 95°C for 10min and stored at -20°C
- calibration curve for the HiBit Assay with the promega HiBit protein. The 20µM solution was diluted 1:1000 -> 20nM and 1:800 -> 25pM