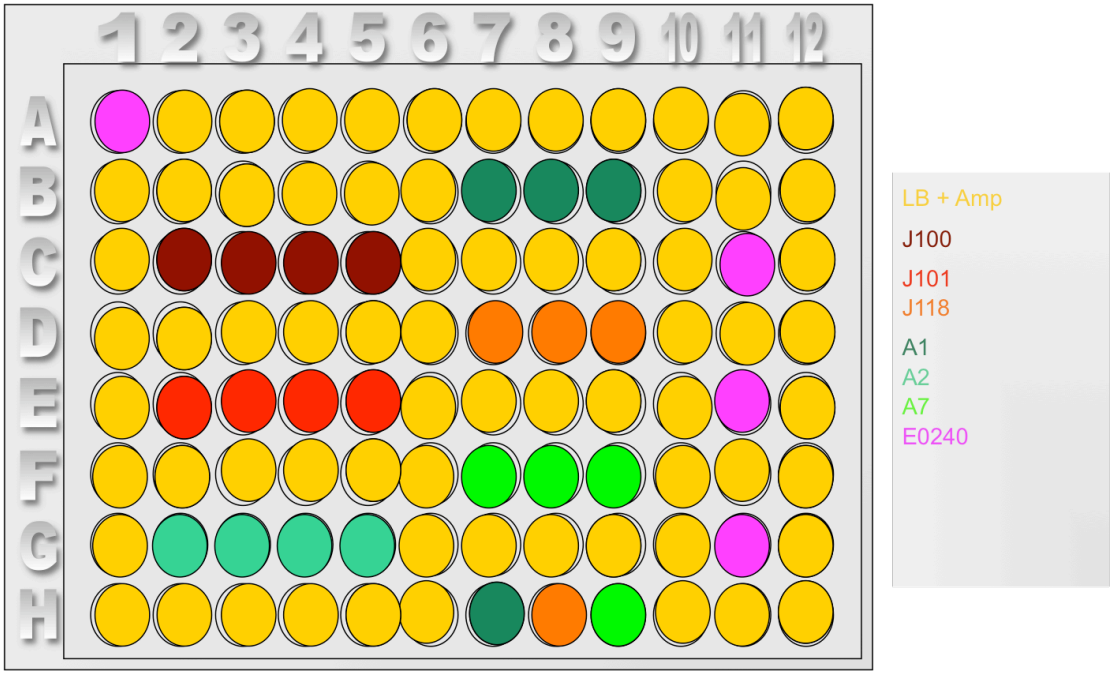
A1 A2 A7 in LB TEST 04/08/09

Description: Fluorescence in red and green for constitutive promoters
Purpose: Fluorescence test to extimate synthesis rate under the control of constitutive promoters of GFP and RFP in exponential phase of bacterial growth.
Methods:
Coltures of A1, A2, A7, J100, J101, J118 and RBS-30 have been inoculated from glycerol stock in 5 ml LB+Amp and incubated overnight. Next morning, they have been diluited 1:1000 and incubated for further 5 hours at 37°C 220 rpm.

- We measure coltures OD with a test plate and using formula:
  - ml of bacteria to keep=(OD_wanted/OD_measured)*new culture volume. Then the falcon tube is re-filled till reaching a volume equal to new culture volume.
- We have diluted all cultures till obtaining an OD of 0.5 or less.



Protocol:

- The test plate is filled to know the initial OD.
- After the dilution has been performed, the plate is filled as described in methods.
- We want to see the XFP synthesis rate in exponential phase so we sample every 3 minutes for 7 hours.
  - For 7 hours kinetic cycle of 3 minutes in wich:
    - 15 seconds shaking linear 3mm
    - 10 s wait
    - Absorbance and green and red fluorescence measure: -> GAIN SETTING: MANUAL 50

- For the rest of experiment (untill 24 hours)
    - Kinetic cycle: 20 minutes
    - Shaking 15 s linear 3mm
    - Wait 10 seconds
    - Absorbance measurement
    - GFP measurement and RFP measurement: -> GAIN SETTING: MANUAL 50