

XYLENE INTERACTION WITH ECOS

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Question: Will ECOS create AmilCP in the presence of xylene? If so to what extent?

Hypothesis: If the concentration of xylene is great enough, we will get blue colonies/pellets. We predict the dark colonies possibly had issues during ligation and therefore will not properly react to the xylene. Or no color difference will be detectable if they are already producing AmilCP. The light colonies, should produce AmilCP if enough xylene is present.

Materials: xylene, plates containing ECOS ligated to AmilCP, broth containing ECOS ligated to AmilCP, alginate beads, containing ECOS ligated to AmilCP, micro pipettes, incubator shaker table, recording materials.

Variables: Manipulated- concentration of xylene and mode of delivery (plates vs Lb vs beads)
 Responding- Amount of AmilCP produced
 Controlled- time left to grow, cell cultures used, antibiotics used, amount of broth/growth allowed.

Procedure: 1) Ensure you have adequate amounts of ECOS. Spread plates with 20 microliter of LB broth an 100 micro-liters. Make eight plates from each tube. 16 plates in total. Ensure to label properly. Aliquot LB into four tubes per tube (9 mL of straight LB and 1 mL LB with ECOS). 16 tubes in total. Let grow for 3-5 hours in incubator and shaker table respectively. Create beads with LB containing ECOS

2) Add different concentrations of Xylene to plates, tubes and beads.

3) Let grow, periodically checking results.

4) Record results

Results:

Plates	Light 1	Light 2
1-control		
2-10 microliter		
3-100 microliter		
4-250 microliter		

Tubes	Light 1	Light 2	Dark 1	Dark 2
1-control				
2-1 microliter				

Tubes	Light 1	Light 2	Dark 1	Dark 2
3-25 microliter				
4-500 microliter				

Beads	Light 1	Light 2	Dark 1	Dark 2
1-control				
2-10 microliter				
3-100 microliter				
4-250 microliter				

For results, reference Notebook