

Cell Survivability Testing

(Prepared by Wisconsin 2010 iGEM team)

Parts used in this experiment

Part Number	Function	Expression Type	Zip File
BBa_K318500	Produces Trascription Factor RcsB	Inducible - IPTG	500
BBa_K318501	Produces Trascription Factor RcsA	Inducible - IPTG	501
BBa_K318502	Produces Trascription Factor RcsA & RcsB	Inducible - IPTG	502
BBa_K200021	Empty Vector/Contol	Inducible - IPTG	NA

Materials

1M HCl

1M NaOH

LB

1M IPTG

Protocol for Acid Survivability Test

1. Grow cells overnight in LB.
2. Inoculate a new sets of samples which have same starting OD (OD=0.05) by using the overnight culture.
Add 1mM IPTG to induce.
3. Harvest cells at OD between 1 to 2
Wait until the slowest one is at OD=1.5
4. Make LB+ITPG cultures that are OD=1.5, and the volume should between 1ml to 5ml.
5. Spin down the cells at 3000g for 2-3mins.
6. Re-suspend the cells in pH LB medium (pH=2, 4, 7)
7. Plate cell samples at time zero.
8. Place all the samples into the 37C shaker and let them grow for 4hs.
9. Remove the samples from the shaker, and record the OD.
10. Make a series dilution for each sample.
10E-3, 10E-4, 10E-5, 10E-6, 10E-7, 10E-8, 10E-9
11. Plate 20ul of each dilution of each sample on a CM mini-Petridish plate.
12. Put all the plates in to 37C incubator and let the cells grow overnight.

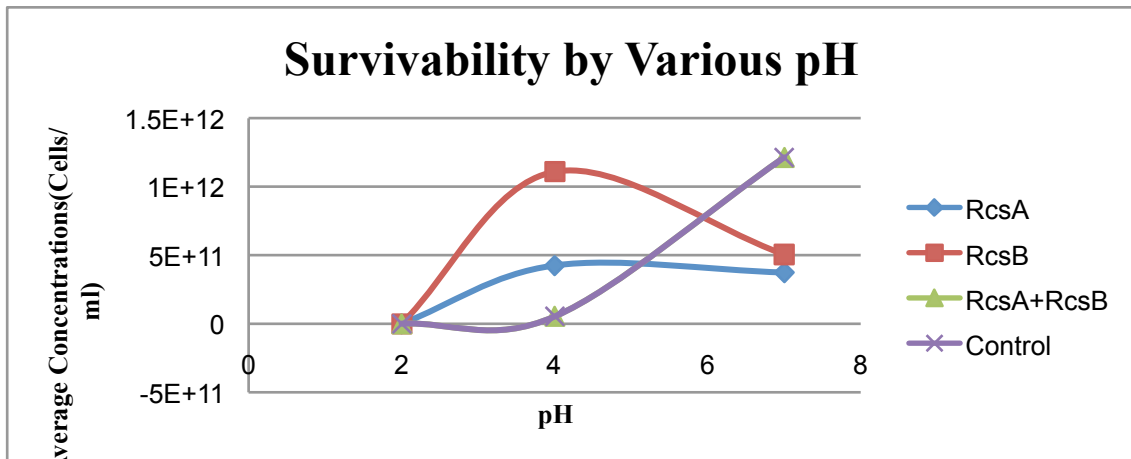


Figure 1 (Created by Wisconsin-Madison 2010 iGEM team)

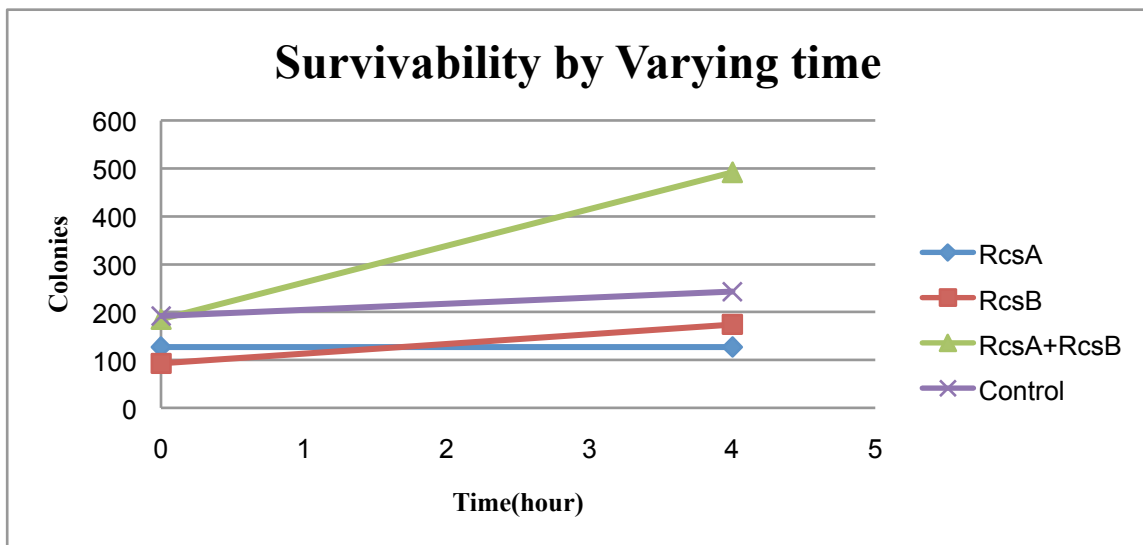


Figure 2 (Created by Wisconsin-Madison 2010 iGEM team)

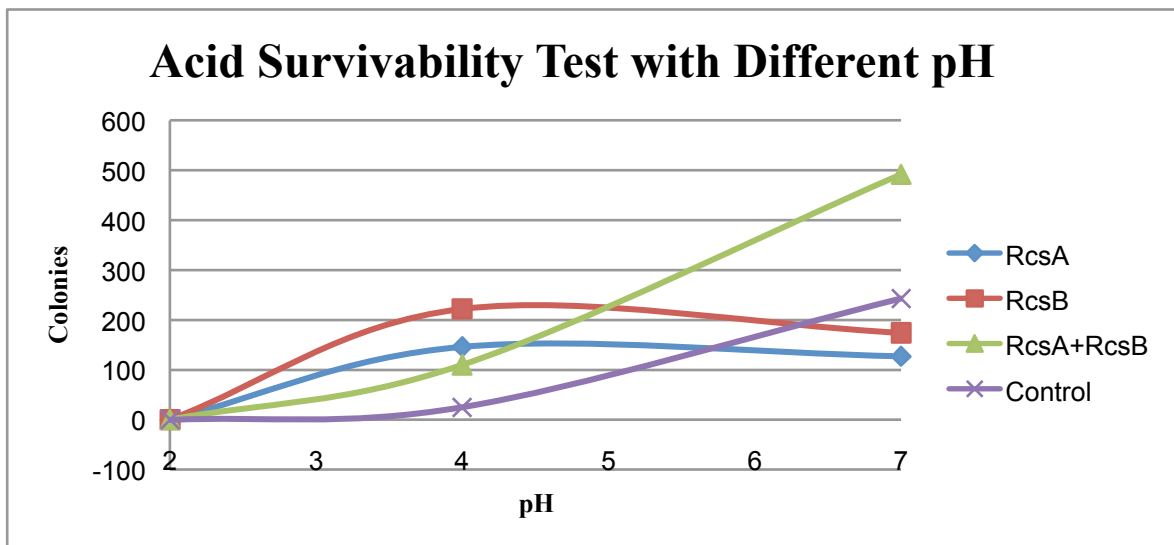


Figure 3 (Created by Wisconsin-Madison 2010 iGEM team)