

Lab Notebook: June 18, 2016

E. Coli Viability, Experiment

Group: Quantitative Bio Team

The purpose of the lab is to determine the viability of *Escherichia Coli* when exposed to different concentrations of sodium perchlorate and sodium chlorite. The results of this experiment will be compared with another lab in the future involving *Escherichia Coli* that have been transformed with a plasmid coding for the production of chlorite dismutase.

Procedure:

Preparation of the Inoculated LB Broth Master Solution:

1. Pipette 80 mL of LB Broth into a sterilized jar.
2. Inoculate the LB Broth with 320 μ L of the prepared *E. coli* stock culture.
3. Gently swirl the contents of the jar.

Investigation 1:

Preparation of Sodium Perchlorate Stock Solution:

1. Measure 0.489756 g of sodium perchlorate.
2. Dissolve sodium perchlorate in 10 mL of LB Broth to make a 0.4 M stock solution.

Serial Dilution of Sodium Perchlorate Solution with Stock Culture (Treatment Groups):

1. In each of 5 culture tubes, pipette 2.5 mL of the inoculated LB Broth master solution.
2. Pipette 2.5 mL of the sodium perchlorate stock solution into one culture tube.
3. Dilute solution by mixing contents.
4. Take 2.5 mL of diluted sodium perchlorate solution and pipette into the next culture tubes. Repeat for the remaining 3 culture tubes to achieve a serial dilution.
5. Label each tube according to its concentration.

Preparation of the Control Group:

1. Pipette 2.5 mL of the inoculated LB Broth master solution into the sixth culture tube.

Incubation of cultures:

1. Place prepared culture tubes in the incubator shaker for at 37°C.

Investigation 2:

Preparation of Sodium Chlorite Stock Solution:

1. Measure 0.361764 g of sodium chlorite.
2. Dissolve sodium chlorite in 10 mL of LB Broth to make a 0.4M solution.

Serial Dilution of Sodium Chlorite Solution with LB Broth (Treatment Groups):

1. In each of 5 culture tubes, pipette 2.5 mL of the inoculated LB Broth master solution.
2. Pipette 2.5 mL of the sodium chlorite stock solution into one culture tube.
3. Dilute solution by mixing contents.
4. Take 2.5 mL of diluted sodium perchlorate solution and pipette into the next culture tubes. Repeat for the remaining 3 culture tubes to achieve a serial dilution.
5. Label each tube according to its concentration.

Preparation of the Control Group:

1. Pipette 2.5 mL of the inoculated LB Broth master solution into the sixth culture tube.

Incubation of cultures:

1. Place prepared culture tubes in the incubator shaker at 37°C.

Summary:

- Culture tubes with varying concentrations (0.2M, 0.1M, 0.05M, 0.025M, 0.0125M and 0M) of sodium perchlorate and sodium chlorite are inoculated with *Escherichia Coli* cells (K12-strain).
- The experiment is designed to cover two trials for each of the sodium perchlorate and sodium chlorite investigations to reduce random error.
- Tube 1bc has relatively less volume of inoculated LB Broth, while tubes 4ap and 3bp have more volume. The volume error may have been caused by the air bubbles present in the pipette during serial dilution.
- The tubes are incubated overnight in the shaker at 37°C.

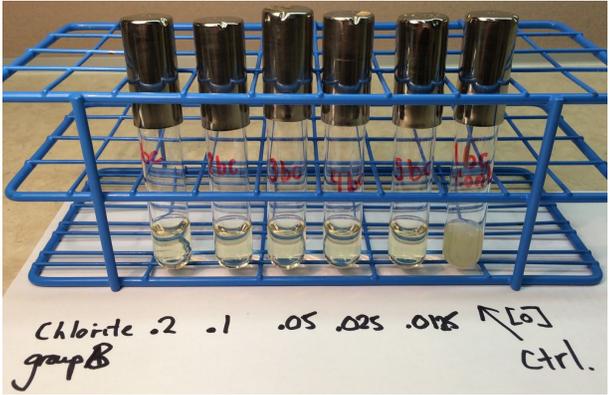
Lab Notebook: June 19, 2016

E. Coli Viability, Results

Presence of *E. coli* in Varying Concentrations of $\text{NaClO}_4(\text{aq})$

Trial	Concentration of NaClO_4	Presence of <i>E. coli</i>	Qualitative
A	0.2	No	
	0.1	Yes	
	0.05	Yes	
	0.025	Yes	
	0.0125	Yes	
	0	Yes	
B	0.2	No	
	0.1	Yes	
	0.05	Yes	
	0.025	Yes	
	0.0125	Yes	
	0	Yes	

Presence of *E. coli* in Varying Concentrations of $\text{NaClO}_2(\text{aq})$

Trial	Concentration of NaClO ₂	Presence of <i>E. coli</i>	Qualitative
A	0.2	No	
	0.1	No	
	0.05	No	
	0.025	No	
	0.0125	No	
	0	Yes	
B	0.2	No	
	0.1	No	
	0.05	No	
	0.025	No	
	0.0125	No	
	0	Yes	

Summary:

- Based on the turbidity of the LB Broth solutions, *E. coli* K12-strain is viable in a sodium perchlorate solution with concentrations of 0.1M, 0.05M, 0.025M, 0.0125M and the control tube, but not in a concentration of 0.2M. Therefore, *E. coli* can survive a range of sodium perchlorate concentrations until 0.1M. These results are consistent within two trials of the experiment.
- For the sodium chlorite investigation, however *E. coli* has only survived in the control tube with a 0M concentration. Sodium chlorite has a higher toxicity than sodium perchlorate. These results are consistent within two trials of the experiment.