

## Artemia vs. Iodine

### Purpose:

Prior studies showed that *Dunaliella Tertiolecta* produced Thyroxine from Iodine and that its predators made use of this growth hormone. We wanted to test whether similar effects were seen with *Dunaliella Salina*. *Artemia* feed on *D. Salina*, so we chose it as our predator to test for growth effects. However, first we needed to test whether *Artemia* were affected by Iodine to act as a control.

### Materials:

- 6 6 well plates capable of holding 5 mL
- 1000 *Artemia* eggs
- MM3 (link) with KI of 0 M, 0.1 M, 1 M, 10 M, 100 M, 1000 M
- p1000 micropipette
- Microscope (microscope camera optional)
- *Dunaliella Salina*
- 50°C Waterbath

### Procedure:

1. Hydrate cysts for four hours
2. Fill three wells with 5 mL MM3 of 0 M KI
3. Repeat step two for MM3 of all KI molarities
4. Transfer about 100 eggs into each well, counting as take in the eggs
5. If you prefer, take a picture of each well to count the number of eggs
6. Repeat steps two and three with new plates
7. Transfer hatched *Artemia* via pipette to the new wells of the corresponding molarity every day
  - a. Count the hatchlings as you transfer them
8. Feed the hatchlings 50 mL of heat-killed *Dunaliella Salina* every day until there are many Brine Shrimp
  - a. Feed them 50 mL of 4x heat-killed *Dunaliella Salina*. Increase the density of the food by centrifuging 4 mL of dead *D. Salina* and resuspending the pellet in 1 mL
9. Transfer dead *Artemia* away every day, counting them as transferred
10. Clean the water Monday, Wednesday, and Friday
  - a. Pipette away 2 mL of medium. Aim for the center where the waste is concentrated.
  - b. Add 2 mL of medium of the same molarity
11. After two weeks, take the eggs and count the hatches and non hatches of each well
  - a. Add a few to the microscope and crush them between the cover and the slide
  - b. Count ones with embryos as non-hatch and others as hatch. Have a partner tally.

**Data:**

Eggs in each well

KI Molarity	Trial A	Trial B	Trial C
0	101	144	101
.1	112	110	108
1	106	104	102
10	135	131	114
100	105	101	109
1000	110	114	120

Hatchlings

	7/7/2011	7/8/2011	7/9/2011	7/12/2011
0 $\mu$ M A	2	-	2	1
0 $\mu$ M B	1	-	1	0
0 $\mu$ M C	1	-	0	1
.1 $\mu$ M A	4	-	1	0
.1 $\mu$ M B	4	-	0	2
.1 $\mu$ M C	1	61	3	0
1 $\mu$ M A	5	62	1	0
1 $\mu$ M B	1	67	2	1
1 $\mu$ M C	0	62	1	0
10 $\mu$ M A	2	72	0	0
10 $\mu$ M B	4	66	3	0
10 $\mu$ M C	1	72	0	1
100 $\mu$ M A	4	56	2	3
100 $\mu$ M B	0	59	2	2
100 $\mu$ M C	2	65	4	1
1000 $\mu$ M A	1	78	1	0
1000 $\mu$ M B	0	73	4	0
1000 $\mu$ M C	0	65	9	0

7/9/2011: The hatchlings in the 0  $\mu$ M wells swim in a lively manner compared to the ones in the 1000  $\mu$ M wells.

7/12/2011: All hatchlings in the 1000  $\mu$ M wells are dead.

8/8/2011: All hatchlings are dead.

**Conclusions:**

This experiment fell apart because of a lot of errors. We know that the brine shrimp in the 1000x died before the others and that we fed them the same amount as the others. However, we did not know enough about brine shrimp to care for them properly and we did not stop to consider a more practical way to measure data. We have not repeated this experiment because we are no longer pursuing the hypothesis we would need this data for. The other experiment indicated that the strand of *D. Salina* we studied does not produce Thyroxine, at least under the constraints we put it in. Thus we would not test for whether *Artemia* are affected by Thyroxine, or Iodine, in following up our study.

To draw conclusive results, we would have to redesign the experiment.