# Observed Rotifera and Acid Phenomena on Algae (PL3LC) Harvest Potential

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## Abstract

Acid Test: Cultures currently being used by AlgaeTown have shown clumping and falling out of suspension when their solutions become too acidic. This is counter to other observations of acidic conditions increasing yields in the long term. (Isom, 1986) There was demonstrated potential to use this clumping to enhance algae harvesting ability. Two sets of samples were setup and the rates of falling out of suspension were measured through turbidity. Turbidity is an abstract measurement of the ability of light to pass through a substance. Preliminary results indicate an increased rate of falling out of suspension at higher pH. However, the differences in rate are not a high enough to be useful for harvesting applications.

Rotifera Test: After a month or two of growing large 700 liter cultures demonstrate a "bathtub ring" effect of algae buildup in the photobioreactors (PBR). This may be for a number of different reasons such as low nutrients or other contaminants. (Kroen, 1984) One of the main contaminants present in these cultures are Rotifera. It was determined to test for possible harvesting applications and effects of Rotifera on culture. Results indicate no significant difference between cultures with and without Rotifera aside from different levels of Nitrate. This may have been due to bacterial contamination. There ultimately are no determined harvesting applications.

## Introduction

AlgaeTown: This project group is a collaboration between employees of Formco Inc., students, and faculty of Goshen College. The purpose of AlgaeTown is to find solutions to problems currently limiting the proliferation of algae based technology. Some current limitations are continual harvesting, continual growth, high yield strains, and growing with limited surface areas.

Recent improvements in harvesting techniques have led to collecting 30x concentrated yields without input of additional energy or chemicals to the growth system.

Acid and Rotifer tests: The primary purpose of these tests has been to look for variables in the algal cultures with the potential to increase the efficiency and effectiveness of current harvesting methods. This is to be done using various observed phenomena.

PL3LC: This is the main algal strain used in the AlgaeTown photobioreactors. The name is derived from Perrin Lake 3 Large Cell. The cells are large, round, and thick walled.



(Image 2.)

Rotifera: Small invertebrates that can grow up to 2mm in length. Often they are found contaminating AlgaeTowns 700 Liter PBRs after 2 or 3 months. They only live for about a week and feed primarily on bacteria, but, they will eat whatever they can fit in their mouths. One end of the Rotifera has 2 to 4 "toes" which are glands used to adhere to surfaces. Cilia are on the heads of Rotifera are used to fan food into their mouths.





(Images 3.)

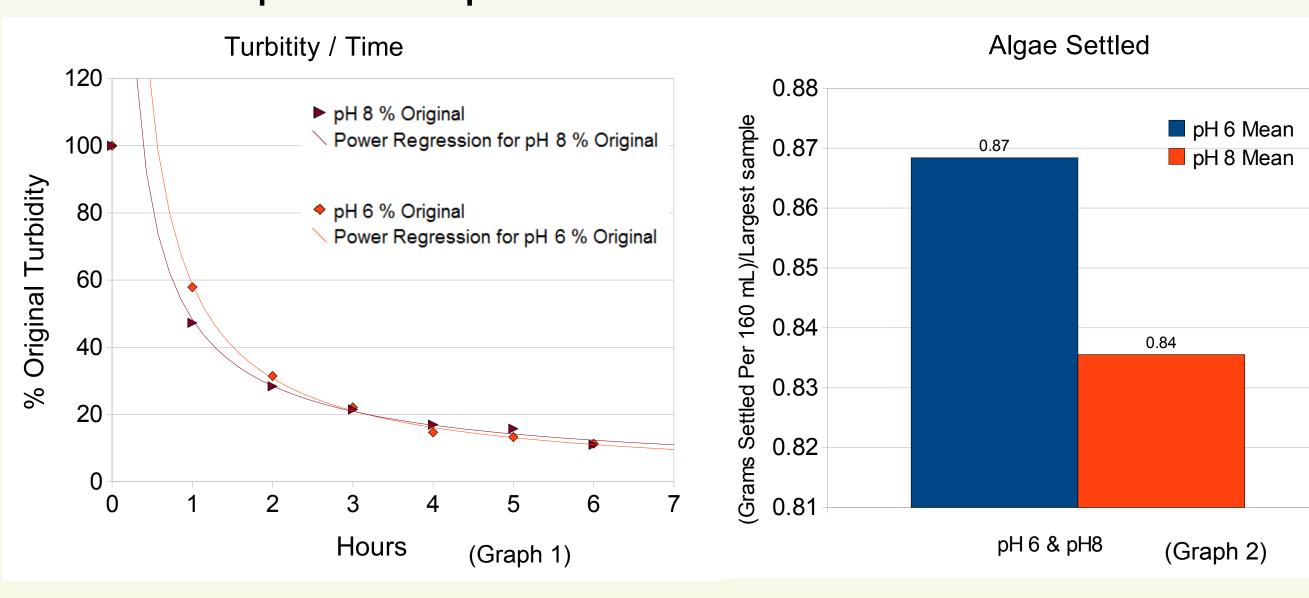
## Materials and Methods

Acid Test: The acid test consisted of two sets of five 250mL flasks. Samples tested were taken from a homogenous 2 liter stock culture of PL3LC for both trials. Five of the flasks used were adjusted to pH 8 and five flasks were adjusted to pH 6. Hydrochloric acid (HCl 2M) was used for adjusting. One set of the five flasks were measured every hour on the hour for six hours. IOmL of sample was taken out from the center of the flasks with a sterile IOmL pipette and placed in a turbidity meter. The numbers were recorded, averaged to account for random error, and later equally manipulated into ratios of the original turbidity. This was for the accurate comparison of data after different initial starting turbidites.

Rotifera Test: Rotifera were taken from a contaminated PBR and concentrated in a 500mL flask placed in the dark for 3-4 weeks. The Rotifera were feed dried algae so that all of the algae from the sample would die off. Concentrated Rotifera were rinsed in .24% bleach solution to kill adult Rotifera and any bacteria present. The mixture was filtered and resuspended in deionized water. Flasks with Rotifera were inoculated with this Rotifera egg solution. The Rotifera tests consisted of five flacks of both Rotifera inoculated and un-inoculated 250mL samples. These were grown on a shaker table for I week. (Suga, 2011)

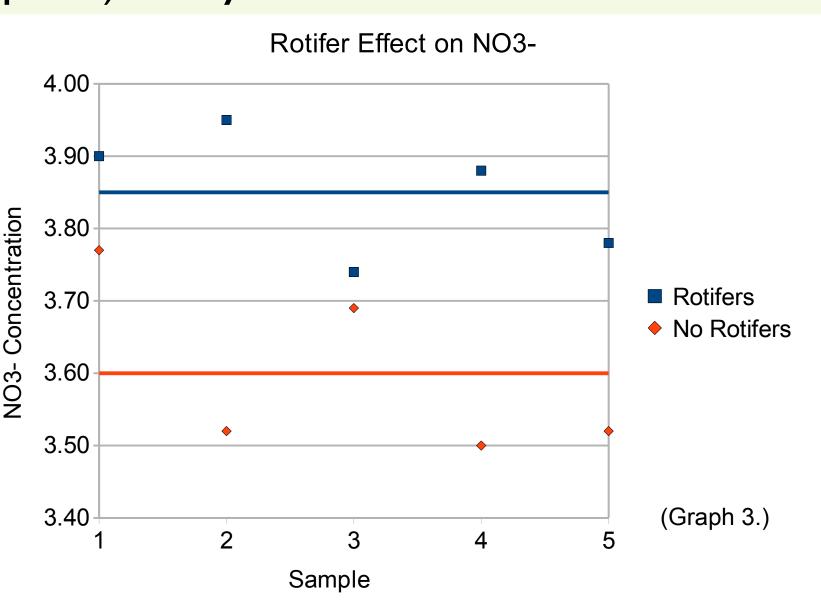
#### Results

Acid Test: Results of the settling data indicate there is no significant difference between rates of settling between different levels of tested acidity. (Graph I) A difference in volume of material that Settled between the two sample pH levels was observable. (Graph 2) The difference in volume that settled was not significant according to the Chi Square test performed.



Rotifer Test: The test for the effects of Rotifera were mostly negative. The "bathtub ring" effect was present in both cultures with and without Rotifera. Variables measured were Dissolved Oxygen (DO), DO mg/L, pH, nitrate, ammonia, dry weight and turbidity. Two tests were used to verify the significance of the results (T-test, Chi Square). Only levels of nitrate were

significant. (Graph 3)
But even with the
nitrate levels being
significant the
culture was
contaminated with
bacteria nullifying
results.



#### Discussion

Acid Test: The results of the acid test hint that it may not be the acid or the acid alone that is inducing the clumping of algae in the failing tanks.

Sources of Error: The only apparent two sources of error are related to time and scale. The time frame of the experiment might have been to short for an accurate representation of the observed phenomena in the PBRs. In addition to the scale of time the small scale of the experiment might have had an effect on the results of the experiment

Further Research: Alterations to be performed in a follow-up study would include agitation of the media, readings over the time frame of days, and larger sample flasks.

Rotifera Test: The results from this experiment in relation to use in harvest are inconclusive. The "bathtub ring" appeared in both the control and experimental.

Sources of error: There are three possible sources of error that relate to agitation, bleach, and bacteria. A shaker table was used for agitation which was unlike the PBRs mode of agitation.

Traces of bleach may have effected the results even though it is unlikely. The bleach solution from materials and methods was filtered off and then the

though it is unlikely. The bleach solution from materials and methods was filtered off and then the material was diluted. An additional source of error is that bacteria were not completely eliminated from the cultures.

Further Research: The best thing that could be done for further research here would be to start over and refine sterilization techniques until all bacteria are killed.

### References

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