

***TITLE: If the 4 highest fitness mutations from an ancestral organism in an environment only containing the sugar Equose are placed respectively in new identical environments containing all sugars but Equose, compared to an environment without Equose, will their highest fitness mutations and average fitness rates be similar if not higher than their original mutated organism?***

**Introduction:** As previously researched in microbiology, I have performed an experiment to determine what factors contribute to the evolution of separate but identical fitness organisms in separate but identical environments. In this case my main focus was to understand whether or not an organism that mutated in an environment with only one sugar, could mutate at the same if not higher fitness rate in an environment containing all sugars but the one in its original birthed environment. The ultimate question to this experiment is to understand whether these mutated organisms could survive by chance, or adaptation. Scientist Richard Lenski has been researching similar hypothesis' by experimenting with the evolution of bacteria primarily *Escherichia coli* for multiple years.

In a long-term evolution experiment with *E. coli*, we founded 12 replicate populations from the same ancestor, and these populations have evolved for more than 30,000 generations in identical environments. We have performed competition experiments to quantify changes in organismal

fitness, analyzed whole-genome expression arrays to find beneficial mutations in genes encoding global regulators, and measured spontaneous mutation rates to discover changes in DNA-repair functions - among many other approaches and findings. Even after more than a decade of study, we continue to find fascinating evolutionary changes in these evolving populations. <sup>1</sup>

In comparison to Lenski I am performing a similar experiment on a much smaller scale. U. Munster and R.J Chrost also researched the affects of sugars on *E. coli* in different environments to determine whether the *E. coli* could evolve despite lacking certain sugars than their previous environments.

Today, more than three decades after the discovery of bacterial permeases, it is still difficult to predict the utilization pattern of sugars by *E. coli* despite the fact that much is known about the genetics and biochemistry of sugar transport and the subsequent metabolism. One can conclude from our work that the extent of catabolite repression is lessened or even absent at low glucose concentrations. As a consequence, cells growing under such conditions should be able to utilize many other carbon substrates in combination with glucose, and also immediately replace one growth substrate with another. This is particularly relevant for understanding microbial growth under environmental conditions where concentrations of individual carbon sources are usually in the  $\mu\text{g l}^{-1}$  range and the total available carbon rarely exceeds a few  $\text{mg l}^{-1}$ . <sup>2</sup>

In a review done by John Roth on parallel molecular evolution with *E.*

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<sup>1</sup> Lenski, Richard Experiment Evolution research interests, <http://myxo.css.msu.edu/Researchinterests.html>. Accessed 12-Mar-2010.

<sup>2</sup> Munster, U. & Chrost, R.J (1990). Origin, composition, and microbial utilization of dissolved organic matter. In *Aquatic Microbial Ecology Biochemical and Molecular Approaches*, pp. 8-46. Edited by J. Overbeck & R.J Chrost. New York : Springer.

coli, he documents the results of a study done by Wichman on parallel evolution.

In a landmark study, Wichman *et al.* (24) examined parallel evolution at the genetic level in  $\phi$ X174, a DNA virus with 11 genes and a 5.4-kb genome. Two populations were propagated for 10 days on a novel host strain, and the viral genomes were sequenced before and after the experiment. That study found 29 mutations, of which 14 were identical in the two populations. Qualitatively similar results were obtained by Bull *et al.* (19) with several additional populations of  $\phi$ X174 by using a more complex experimental design. However, it is not known whether much larger genomes, encoding more complex organisms and having potentially many more targets of selection, would show similarly strong parallelism at the sequence level.<sup>3</sup>

These studies are interesting and similar to my hypothesis in that mutation rates could be identical in different populations under similar circumstances.

**Methods:** Using the Avida-Ed program simulator, I began my study with a single ancestral organism placed in a Petri dish with a 3.0% per site mutation rate in a world size of 60x60 cells with the single reward of Equose in the environment. I then ran the simulator for 500 time (updates) and watched the petri dish evolve. After the simulation ended, I froze the 4 highest fitness organisms and stored them in my organism's bank (labeled a,b,c,d). Next I configured an identical environment size and mutation rate but instead, all sugars were included except

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<sup>3</sup> Roth, John R. Tests of parallel molecular evolution in a long-term experiment with *Escherichia coli*, [http://www.pnas.org/content/103/24/9107.full?maxtoshow=&HITS=10&hits=10&RESULTFORMAT=1&author1=WOODS&andorexacttitle=&andorexacttitleabs=&andorexactfulltext=&and&searchid=1&FIRSTINDEX=0&sortspec=relevance&date=1/1/2006&resource\\_type=FWCIT](http://www.pnas.org/content/103/24/9107.full?maxtoshow=&HITS=10&hits=10&RESULTFORMAT=1&author1=WOODS&andorexacttitle=&andorexacttitleabs=&andorexactfulltext=&and&searchid=1&FIRSTINDEX=0&sortspec=relevance&date=1/1/2006&resource_type=FWCIT), Accessed 12-Mar-2010.

Equose. I then placed the original a,b,c,d organism respectively in this new environment 6 different times for each organism, always starting with the original mutation. After each simulation, I froze and recorded the highest fitness organism from the dish in my organism's bank and froze the petri dish so I could analyze all it's data.

**Results:** Organisms a,b, and d all proved my hypothesis as shown in Appendix 2a and 2b. Each organism once placed in a new environment lacking the sugar Equose, mutated at a steady, similar rate and produced an almost identical average fitness rate throughout all six simulations. The average fitness rate continuously proved to be higher than the original fitness rate in the ancestral environment containing only the sugar Equose. The highest fitness organism from each simulation varies greatly as shown in Appendix 1a and 1b. However, once put in a new environment lacking the sugar Equose, each organism *still* and always produced a higher fitness organism result after 500 time (updates) than its original mutation.

The only exception that disproves my hypothesis is Organism c, which did not mutate no matter how many times it was put in an environment lacking the sugar Equose.

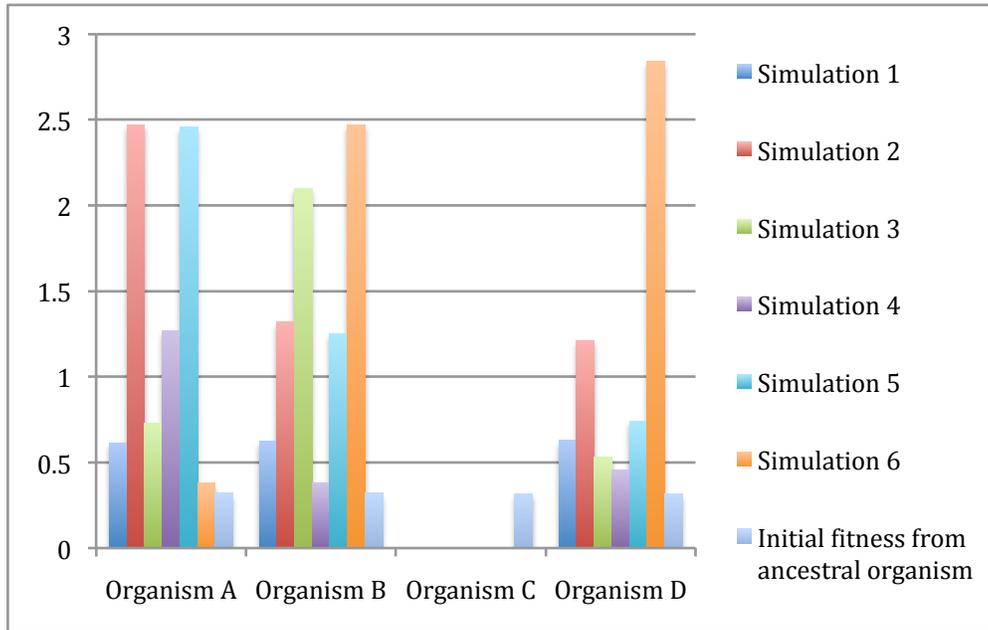
**Discussion:** The results of this experiment and tested hypothesis were very interesting. The meaning of this data proves that Organisms can almost indefinitely adapt to an environment that does not contain the resources in its original birthed

environment. It also proves that, not only will these organisms mutate at a higher fitness rate than its original environment, but will also mutate at almost identical fitness rates to each other even though they're all placed in separate but identical featured environments. Organism c was an exception to this hypothesis because it did not mutate at all in any of the six simulations. This proves how chance still plays a role in evolution.

It would be interesting to repeat this experiment on a larger scale by simulating an ancestral organism in an environment without Equose, then putting the top highest fitness mutations and simulating them multiple times in an environment only containing Equose. Then, repeating the experiment I just did and comparing the affects of Equose on an organisms ability to produce a high average fitness rate in environments that do not contain the sugars the organisms mutated in.

**Appendix:**

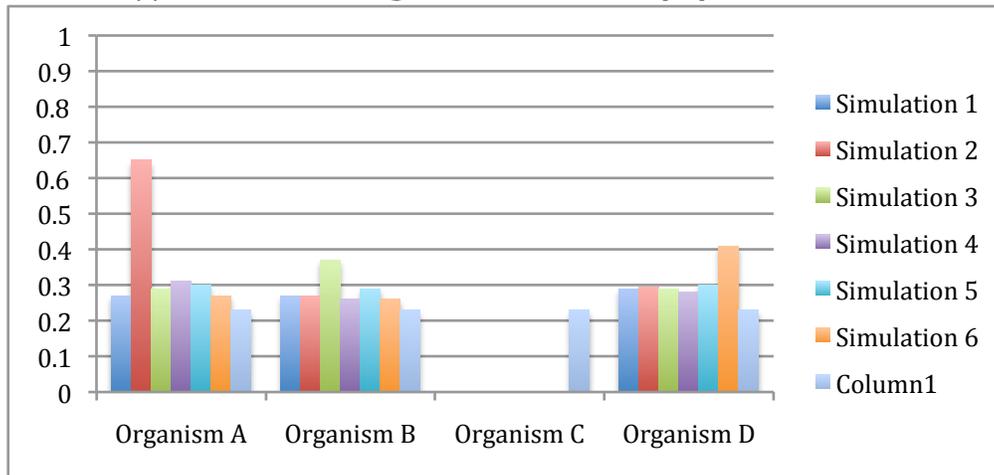
**Appendix 1a: Highest Fitness organism from each population simulation**



**Appendix 1b: Highest Fitness organism from each population simulation**

	Initial fitness from ancestral organism (environment with Equose)	Simulation 1	Simulation 2	Simulation 3	Simulation 4	Simulation 5	Simulation 6
Organism A	0.32	0.614	2.47	0.73	1.27	2.457	0.378
Organism B	0.32	0.6216	1.318	2.1	0.379	1.25	2.47
Organism C	0.315	0	0	0	0	0	0
Organism D	0.38	0.6274	1.21	0.53	0.456	0.74	2.84

**Appendix 2a: Average fitness of each population dish**



(Column 1:Initial average fitness from ancestral dish containing Equose)

**Appendix 2b: Average fitness of each population dish**

	Simulation 1	Simulation 2	Simulation 3	Simulation 4	Simulation 5	Simulation 6
Organism A	0.27	0.65	0.29	0.31	0.3	0.27
Organism B	0.27	0.27	0.37	0.26	0.29	0.26
Organism C	0	0	0	0	0	0
Organism D	0.29	0.294	0.29	0.28	0.3	0.41

## Bibliography

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