Evolutionary Dynamics

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Lecture 52

Hi everyone, welcome back. Let us continue our discussion on elevated mutation rates. So, we have a population. which is evolving in a particular environmental context. Evolving in an environmental context.

And what is being selected for, as in LTEE and many other evolution experiments, is a higher growth rate. That is the feature or the phenotype that is being selected for. And in this context, we saw last time that what ends up being selected is higher mutation rates. Some of these lines exhibit elevated mutation rates when the selection was actually for higher growth rate.

And let's just conceptually try to understand how this was accomplished. So this population, the ancestor population, has a very low mutation rate. As a result, access to beneficial mutations is difficult. Because errors happen so infrequently that finding the adaptive mutation is difficult. In such a scenario, what happens is that one individual can acquire a mutation such that its error rate increases.

So this individual now has a higher error rate than everybody else in the population. than everybody else in the population. And as a result, the chances that this individual is going to divide and because of the elevated error rate, produce an offspring which has a higher fitness. Because fitness Because as this individual is dividing, its fitness is increasing.

As this individual is dividing, its progeny are acquiring many different types of mutations. This is because it has a higher error rate. And one of those progeny, because it's sampling these mutations much more frequently than the ancestor, the chances are that one of those progeny is going to hit upon a mutation which increases fitness. this sampling of mutations is being done much more poorly in the ancestral genotype as compared to this higher error rate genotype because errors are occurring much more

frequently in it and hence it's sampling much better. As a result of this, what gets selected is this individual.

So if we sort of draw upon a, if we draw a cell and its genotype, then what is happening is the first mutation that happens does not change fitness, it only changes mutation rate. The first mutation only changes mutation rate. So this has no effect on fitness Only changes mu.

After that, because in this genotype mutations are happening much more frequently, chances are that one of the progeny is going to find a mutation such as this. So this mutation was the one which facilitated higher error rate. And now because of the higher error rate, this mutation, a beneficial mutation, can happen more rapidly in this genotype as compared to the ancestral genotype where mutation rate is low. So this is a highly beneficial mutation. By beneficial, we mean that it facilitated a higher growth rate.

So again, what is important to keep in mind is that selection was for this. This was the feature that was being selected for. As we wrote down here, what happened is this elevated mutation rate, this elevated mutation rate facilitated this process by better sampling of mutations that could occur in a genotype. So this mutation rate, elevated mutation rate facilitated higher growth rates.

Such an idea that selection was not for higher mutation rates, yet higher mutation rates happened because they facilitated adaptation, this idea is called secondary level selection. that primarily the selection that was operating in LTEE was for higher growth rate. It wasn't on mutation rate. Yet mutation rate went up, not because higher mutation rate was selected for, but because higher mutation rate facilitated the increase towards higher growth rates via better and faster sampling of all sorts of mutations in the process, also sampling these beneficial mutations faster. And this has been seen repeatedly in different independent lines of the long-term evolution experiment.

In another interesting study from LTEE, what happened was, at the start of the experiment, we started with one genotype. This is the starting genotype. When this experiment was fairly early in its progress, at about 500 generations, it was noticed that in this flask, as we have discussed many times now, the population is so high that it's comprised of several genotypes and not just one genotype—the idea of clonal interference. This flask contains two different versions. Let's indicate them with different colored DNA.

And as the experiment proceeded further, it was noticed that one of these two variants was eliminated from the population. So in this context, the green variant was eliminated as the evolution experiment continued, whereas the red variant became fixed in the population. So what are the features we have discussed in this experiment? At generation 500, two genotypes were present in the population. Two genotypes present in the population.

Neither of these two genotypes was the ancestral genotype. Both carried some mutations relative to the ancestral genotype. So let's indicate the ancestral genotype with this. And as time progressed, it was noticed that one of the two genotypes was lost. One of the two genotypes was lost—in this case, it became extinct—and along with that, the second genotype.

reached fixation which is the red genotype. So as far as this information is concerned, there isn't really anything surprising in this. We expect clonal interference to be present in these microbial populations in flasks. We've seen simple calculations show that. So the fact that there were two variants present in a flask is not surprising at all.

And eventually, if there are two beneficial mutations in the same flask, they are obviously competing against each other. And what's going to happen is that one of them is going to go extinct and the other one is going to go fixation. It's also possible that there is also a third likelihood that both of them might get eliminated and that might happen in the scenario if this is happening, that there was an ancestral individual here and this ancestor, along the process of evolution, this ancestor picks up this great blue mutation And the benefit that this blue mutation confers is just so much higher than the green and the red mutation, such that by the time we give it more time, this individual is able to eliminate both the green and the red genotypes from the population and blue reaches fixation. That is also possible.

So in that manifestation of experiment, both green and red would be eliminated. However, that was not what was seen here. What was seen here was that there were two genotypes at generation 500, and if you move forward a little bit, one of the two was lost, the other one reached fixation. So there's nothing surprising here. Next, when these two mutations were characterized, what was noticed was that the green individual and the black genotype

the red genotype, both carried one mutation. And this mutation was in the same gene, which is called, so carried one mutation each in the same gene. called topA. This stands

for topoisomerase A. And we don't need to get into the details of the function of this enzyme, but what it does is it controls how tightly DNA helix is packed. And that has implications on the gene expression profiles that we see in an organism.

So if this is the gene length, if this is the length of the gene topA, then the green mutant carried one mutation only, which changed amino acid. which means that this mutation was a non-synonymous mutation, whereas in the red mutant, there was also only one mutation in topoisomerase A, and this mutation also only changed one amino acid. Changed one amino acid. So both the green mutant and the red mutant are carrying one non-synonymous mutation each in the same gene in E. coli. Now, when the relative fitness of these two individuals, of these two genotypes was characterized, what was found that the fitness of green mutant, so

Let us say this is the green mutant and this is the red mutant. What was found was that fitness of green was greater than fitness of red. Now here is where we have a little bit of a surprise take place. That now what we are saying is that we had a population, let's get rid of this ancestor one. We had a population in which we had two mutants, each one of which carried a single amino acid mutation in the same gene.

Both of them were beneficial in nature. So the fitness of both of these individuals was greater than that of the ancestors. So both these mutations were beneficial mutations. However, the green mutation conferred a higher benefit compared to the benefit conferred by the red mutation. If that is the case and we have these populations—as these mutations have established themselves in the population—they have overcome drift, and it is only selection that is dictating their fate from here onward.

Despite that, what was seen was that the red individual, the less fit of the two, won this competition between the two beneficial mutations in this experiment. Our null expectation, given all this information, would have been that the green individual should reach fixation and the red individual should be made extinct from the population. However, what was observed in this study was the opposite. The genotype that had the lower fitness eventually reached fixation. So in the paper, this was published in Science.

So in the paper, the notation—they give these two a name. So the green one, which has higher fitness but is eventually made extinct, is called the eventual loser. or EL, and the red one, which despite being less fit reaches fixation, is referred to as the eventual winner. So now the question is: how do we explain this fact that a less fit genotype reached fixation? As a first test of this observation, what was done was to repeat this

experiment and see if the same outcome happens every time or if it was a freak incident—a very rare chance event that resulted in the less fit genotype taking over the population.

So when we mix these two genotypes in a ratio of 1:1, that means the same number of cells of each kind, and we re-perform the evolution experiment many times—this can be done any number of times. So every time we do this experiment, what is seen is that the less-fit individual which is the red one, and the other is green—what we see in independent runs of this experiment is that the less-fit reaches fixation. That's the eventual winner. And the higher-fitness background goes extinct.

This is the eventual loser. So now we have a problem to explain: why is it that in this context, the less-fit individual reaches fixation and the higher-fitness genotype goes extinct? And what was found is the following. So let me just sketch these two cells. Let's make the DNA.

Let's sketch the gene topoisomerase A. So let's say this is topA. And this is the mutation that the less-fit individual picked up. And in green is the mutation that the higher-fitness individual picked up. So as far as the situation shown in this cartoon is concerned, the green individual should win. However, there is another gene called spoT.

And SPO-T is a gene which is called a global regulator gene. And it is particularly important in starvation conditions for the cell. So a global regulator is a gene that controls the expression of many, many genes across the genome. We studied the lactose utilization system in E. coli at the beginning of this course. And there we saw that lacI is a gene regulatory protein that only controls the expression of lacZ and lacY.

So that is an example of a local regulator. A global regulator, on the other hand, controls the expression of hundreds, sometimes many hundreds of genes spread across the genome. And spoT is one such global regulator in E. coli. And its role in cellular physiology becomes increasingly important once there are limited nutrients available in the cell. And remember, this experiment started with the condition that there was only a low amount of glucose available for E. coli cells to utilize.

So both these cells have this gene spoT. However, what happened in all these manifestations where the less fit reaches fixation was that After the acquisition of this red mutation, another mutation happened in Spoti. And this mutation, when coupled with this red mutation, these two together created a genotype of very high fitness. However, when

that same mutation was introduced in the higher fitness genome, these two mutations were not able to confer that high fitness.

This is another example of the concept that we discuss called epistasis, where we say that the effect of a mutation is dependent on what is present elsewhere in the genome. So what we are seeing here is that when we have this orange mutation taking place in spore T in conjunction with a red mutation in top A, then the fitness gains that a cell makes are very, very high. As compared to if that same mutation in spore T was to happen in conjunction with the green mutation in topA, then the benefit that is conferred to the cell is not very high. This is an example of epistasis, and it's just present everywhere in every manifestation of evolutionary processes. So what happened in that experiment was why the red mutation was winning.

Because once you have—so let's imagine a context like this: we have an ancestral population, and out of this ancestral population, some pick up the red mutation and some pick up the green mutation. The fitness of the green mutation is higher, and the red mutation's is lower. However, both these fitness levels are higher than the ancestral fitness, so this is the current state. However, now there exists this mutation in the spore T gene, which, when it happens in this background, takes fitness to this level, whereas when the same mutation happens in the green individual, it is only able to increase the fitness to this level. As a result, the red mutation makes this fitness level accessible to the cell via acquisition of that spore T mutation, whereas this higher fitness level is not accessible to the green cell. The same spore T mutation does not do it, and so on and so forth. As a result of this, this red population, which was originally lower in fitness as compared to the green individual, is able to fix in the population.

So this was another important lesson: that evolution is dictated not just by the mutations that have already happened, but it is also dictated by what a mutation that has happened can potentiate in a population. For instance, in this case, the red mutation potentiated access to much higher fitness levels as compared to the green one. And hence, that potential was realized by acquisition of a mutation in spore T and cells reaching much higher fitness levels. So that concludes our discussion of this particular study. Next, I will leave you with a question which I want you to think about before we start the next video.

We started this discussion of fitness changing with number of generations as the follows that as time progresses. Time being measured in number of generations. And on the y-axis, we are plotting fitness. And we saw that this trajectory, this has been seen through

many, many experiments, looks like this. Starts off fairly sharply and then slows down as it's moving forward in time.

Now this trajectory has two fates. One is that there is a plateau here above which it cannot go. Let me redraw this. The first fate of this trajectory is that maybe there is a fitness level here which I will call F max beyond which it cannot go. So it will just approach F max and never cross F max.

In which case we have a problem because what this tells me is that does evolution grind to a halt? Because you can no longer increase. So in that case. does that mean that at some point running the LTEE experiment is not going to serve any purpose because fitness cannot increase beyond a threshold. Alternatively, the other possibility is that this will continue to increase forever but get increasingly slower but it will increase forever.

This leads us to a problem: if this is the case, then does this mean that there is no fitness peak? If evolution continues forever, then what does the concept of a fitness landscape mean, where we saw that evolutionary trajectories can be visualized as climbing this mountain, which is defined by sequence space and fitness as the dimensions of this hyperdimensional space? So both these views lead us to the following two problems. So what I would like is for you to think about these issues and intuitively see which of these two viewpoints you are comfortable with.

One is that you cannot cross a fitness threshold. You can't go beyond Fmax. And the other is that you will keep on increasing, but the rate of increase will just keep getting slower and slower. Which of these two viewpoints is closer to your intuition of how evolutionary processes will proceed in these microbial populations? And we'll continue this discussion in the next video.

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