An Introduction to Evolutionary Biology

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Coping with changing environments

Hi. So, in our last discussion, we looked at how new genes originate. We remarked on the fact that most of the time the rate of origin of new genes is not very high. However, organisms have to, you know, keep up with an environment that can and often does change extremely fast. And therefore, a huge question is how exactly organisms can cope with changing environments if the underlying mechanism The rate of change, whether in terms of the birth of new genes or in terms of mutations, is actually much, much slower. So, part of this answer we have already dealt with, and part of this answer is, for example, phenotypic plasticity.

Now, although we have already dealt with it, I will quickly use two of the slides that we have already looked at. Just to refresh your memory, and then we will get into the part that we want to discuss today. So, what exactly is plasticity? Phenotypic plasticity is the ability of a genotype to produce more than one phenotype in response to different environments. And we typically show plasticity using this kind of figure called a reaction norm where you have different environments on the x-axis and each line represents the trait value for a particular genotype. So, when the lines are parallel to each other, Then that means that when you change the environment, the phenotypic expression does not change. So, there is no plasticity. Whereas, when the lines have some kind of slope, That means that the phenotypic expression is changing across environments, which indicates that there is plasticity. Now, we also saw an example; well, we saw multiple examples, actually.

Here is just one of them to refresh your memory. So, this is the larva of the geometrid moth Nemoria arizonaria. And this, you know, can grow either during the spring season or during the summer season. This grows on the oak tree. When it grows during the spring season, it resembles the inflorescence of the oak flower known as catkins.

So, you can see this is the larva and this is the oak inflorescence, but when the same larva, The same genotype is growing during the summer; then the flowers are no longer there. At that time, the larva actually represents the twig of an oak tree. So, this is what is a special case of plasticity, where instead of having continuous variation in the trait. You have discrete variations or discrete forms. This is what is known as polyphenism.

Now, one of the things that we remarked at that point is that sometimes plasticity can vary across genotypes. So, along with these two cases of no plasticity and plasticity, you can have a situation wherein, you know, for example, here, The red one, when it is going from environment A to B, is reducing in value, whereas for the other two, the value is increasing. So, in other words, how a genotype will respond to an environment will vary across genotypes. Now, if you have a situation like this, where the ability to respond to the environment depends on the genotype, Then that means that you should be able to select for plasticity. In other words, plasticity itself becomes a selectable trait on which evolution can operate, or rather, selection can operate.

And here is a very nice example of it. So, what I am showing you here is an organism called Daphnia melanica. So, these are very tiny crustaceans. Typically, you find them in, you know, clear water, and daphnia are actually a good indicator of the health of a pond or a lake. Because if the lake becomes polluted, then the daphnia typically disappear.

So, it is a water-quality indicator. Now, it turns out that mostly these are transparent things, and because of that, their internal organs They are visible from outside, as you can see over here. This is the normal form. Now, because of this, Daphnia is a very popular model organism. You can figure out what is happening inside the organism just by

placing it under a microscope.

But the same transparency also creates a problem. Because the organism is now vulnerable to the effects of ultraviolet rays, particularly UVB. Now, it turns out that in some populations of Daphnia melanica, When you expose them to UVB, they produce this dark pigment called melanin. And the moment you have melanin, this is what I am showing you here on top. The daphnia becomes opaque, dark in color, and now the UVB cannot penetrate the melanin, I mean, this is the same thing that is happening on our skin, right? Now, this phenomenon happens because of the action of two genes: one is called dopa decarboxylase and the other is called ebony. Now, the melanic forms obviously are better in terms of not letting UVB through, but they have two problems. The first problem is that if there are predators of crustaceans, who are the predators? Typically fish. So, these fish are mostly visual predators. Now, when the daphnia are transparent, it is typically very difficult for the predator to see them, particularly in clear water.

But the moment they become melanic like this, the predators ability to spot them increases quite a bit At that point, they basically get eaten a lot by these predators. The second thing that happens is that the production of melanin is energetically expensive. because of which the maturation rate of the melanic forms is actually slower than that of the non-melanic form. So, there is a cost to producing melanin. Now, there was a very nice experiment done by two people, Scoville and Pfrender, in 2010 where they looked at the plasticity of melanin production in the Daphnia melanica species. So, what they did was go to four lakes, you know, High alpine lakes in which the ultraviolet ray exposure was rather high, but it varied seasonally. So, from these four lakes, they picked up populations of Daphnia melanica, and two of these lakes were such that They, you know, the daphnia over there had never been exposed to any predators; there were no fish ever introduced over there. The two other lakes were such that daphnia had been exposed to fish for the last 53 years in one case and 91 years in another case. So, basically, long-term exposure.

Now, this is significant because, as I told you, these fish are very strong predators of

daphnia. So, in many other lakes, the fish had actually, you know, pushed the daphnia to extinction. But they were able to get two lakes where the daphnia and the fish coexisted. Now, what is the, you know, beauty of this whole setup? The beauty of this whole setup is that when you know all the lakes are high UV lakes, which means that in all the lakes it is better for the daphnia to form the melanin form. But in the lakes where the predators are present, if they form melanin, they are going to be exposed to high levels of predation whereas in the two other lakes, they ideally should not have required you know to change their melanin form. But because there is seasonal variation, You expect that there will be some parts of the year when it is good to have the UV. And there will be some other parts of the year when the UV is not, you know, a burden because it reduces the growth rate. So, accordingly, what they did was these two people brought together, I think, 13 to 17 different genotypes that were collected. They brought all of these to the lab and then created two treatments.

One in which they subjected them to UV, and another in which there was no UV. So, that should be UV, actually, UVB, not C. Now these are the results that they have got. So, here low means no UV, and high means the presence of UV. And what they are plotting on the y-axis is the amount of melanin that is being produced.

Now, first let us look at what is happening to the case when there are predators. So, you can see that there is a slight change, but this is statistically non-significant. So, for all practical purposes, when they have evolved in the presence of the predators, Then they have lost their ability to modulate their levels of melanin in the absence of UV. But look at what is happening to those Daphnia that have evolved without the predators. In their case, when they are exposed to UV, they are able to produce melanin.

But when the UV is turned off, they are able to reduce their melanin exposure. And as I told you, this plasticity is very important here because the environment is variable. And because the environment is variable, the ability to modulate the melanin level is what this Daphnia needs. The environment is variable for these guys, also in the presence of the predators. But over there, the presence of the predators is overwhelming everything else.

So, these are the two results. Number one, in the presence of predators, the Daphnia greatly reduced their plasticity. And in this particular case, these two people were also able to pinpoint. What were the molecules that caused that to happen? So, they showed that this is happening because in those that have evolved with predators, There is constant upregulation of dopa decarboxylase and ebony. And both of these proteins, when they are upregulated, actually reduce the amount of melanin in Daphnia. So, the guys who have evolved with the predators in them, you know that this correlates with the fact.

The upregulated level of these two molecules correlates with the fact that they have a lower amount of melanin. So, this particular example actually illustrates quite a few very interesting features of phenotypic plasticity. First, as I showed you, in variable environments where it is desirable to turn on and off, Or, you know, upregulating or downregulating your trait, plasticity can actually be pretty advantageous. However, the plastic expression of a phenotype many times is costly, and when I say costly, I mean it can have a negative effect on fitness. And whether a particular plasticity is advantageous or not may depend on other factors.

So, just think about the fact, you know, in the context of this Daphnia, if you evolve melanin plastically. Then that will end up affecting your fitness in terms of a reduced growth rate. But whether your ability to turn it down is advantageous or not depends on whether predators are there or not. If the predators are there, then you know this turning you know making the thing more melanic that you know you do not really have any option in turning down the melanin, right. I am sorry, but it's the other way around; you do not have an option in terms of turning on the melanin.

Because if you do, the predators are going to eat you up, right? So, that is what I mean by saying that whether plasticity is advantageous or not might depend on other contexts. And when the expression of plasticity is costly, it is possible for organisms to lose their plasticity, which might affect their future evolution. I mean what you saw over here because it was costly for them to, you know, change their melanic levels. They had to

maintain a constant melanic level; that is why they ended up losing the plasticity itself. And finally, this is not something that is coming from this experiment, but plastic responses are typically fast.

Sometimes they can happen within one generation or maybe one or two generations. and that is why this allows organisms to quickly respond to a phenotypic change. Now, if you have followed until this point, then you know you are obviously going to think that you know. Is this something that looks like Lamarckian inheritance? Because you know, here you have ended up inducing a variation through the environment, and now you are trying to select for it. Then, does it not mean that this is inheritance of acquired characteristics or selection of acquired characteristics? Not exactly, and I will tell you why.

So, this is, you know, a very famous experiment. So, this is Drosophila melanogaster, and you know that this is a dipteran, which means that you know. It has two wings, and the second pair of wings is modified into these balancing organs known as halteres. So, they are, you know, small, tiny, knob-like things. However, if you take eggs of Drosophila when they are about two and a half or three and a half hours old, then And you expose them to the vapor of ether; you know the chemical.

Then, when those eggs hatch and when the adults form, the adults tend to have, instead of, you know, three thoracic segments, I mean they have this extra segment over here and, more importantly, instead of the halter, this halter actually turns into wings. So, essentially, this thorax contains a thoracic segment which contains the wings that get duplicated as a result of which. They end up having the so-called bithorax phenotype, two thoraxes, and, more importantly, they have four wings. Now there was a very famous experiment conducted by the British embryologist Conrad Waddington. Where what he did was select flies for producing bithorax-like phenotypes.

So basically what he did was he you know exposed them to ether And those flies that ended up producing the bithorax phenotypes, not all of them did; you know, some of them did. He ended up taking those eggs and allowing them to reproduce for the next

generation, and he kept on doing this repeatedly. It turned out that within 29 generations, he was able to obtain flies in the selected lines. That showed the bithorax phenotype even when it was not exposed to ether. In other words, now they did not need either to produce the four wings.

Now this is something that Waddington said: the bithorax phenotype has now been genetically assimilated. So what did he mean by that? So the phenomenon of genetic assimilation, as Waddington thought about it, was the process by which a phenotypically plastic trait becomes genetically determined and no longer requires the environmental stimulus to be expressed. As I said, you know this on the face of it looks like the inheritance of acquired traits. But Waddington actually went out of his way, and you know he kind of looked at his entire data. And did all kinds of experiments to actually show that that was not the case.

So, I will just quote from his paper; he says that the genetic basis of the bithorax character is partly a recessive gene. which causes females to produce eggs that develop into bithorax phenotypes, a maternal effect Partly a number of minor genes on both the second and third chromosomes. Remember, this work is happening in 1942. So, people still do not have the ability to sequence the genes and tell you that, okay, you know, this position, that thing, and so on. But in spite of that, using the kind of, you know, techniques that Morgan and his lab had figured out, This person is able to pinpoint the fact that there are, you know, up to which chromosome these genes might exist.

So, Waddington was very sure that he was not talking about a phenomenon that is outside the Darwinian, you know, framework. However, there were other people who kind of interpreted it as, you know, the inheritance of acquired traits. And somehow, they became very aggravated by it. But then later, historians and biologists explicitly pointed out that, look, that is not what Waddington was trying to say. So now this leads us to a certain you know concept of cryptic genetic variation and this relates to the work of Susan Lindquist.

So, what was the work? So there is a certain mutant in Drosophila that is known as the

Hsp83 mutant. Now, when it is homozygous for that mutation, Hsp83, then that organism is not viable. So typically, you have to maintain it as heterozygous. But the heterozygous mutants exhibit a large diversity of phenotypes, including deformed legs. Deformed eyes, small wings, notched wings; you know, all kinds of variations in the wings, and so on and so forth.

So Rutherford and Lindquist explicitly showed that these phenotypes could be inherited. which indicated that they had some kind of a genetic basis. Then they ended up showing that this was related to the action of the gene. The mutation was in the gene Hsp90, heat shock protein 90, which they hypothesized was a chaperone protein. In other words, these are the proteins that are responsible for the proper folding of many proteins.

And they hypothesize that these proteins are actually taking many genetic variants and forcing them to fold in a proper way. Such that whatever variation is present is actually being buffered. Now, why should you know that protein folding leads to buffering of genetic variation? So you know that the functioning of any protein depends on its structure, right? And if it is not able to form the proper structure, you know, the tertiary structure, it will not be able to function. Now, why will it not be able to form a proper structure? Let us say there are some mutations due to which the structure does not get formed. So what they hypothesized was that the chaperone proteins are essentially binding with these proteins during formation.

And essentially, you know, kind of contorting them or kind of forcing them to form the proper structure, because of which. They are able to function properly, and whatever variation is present at the ATGC level. That is not showing up at the protein structure and hence not showing up at the protein function level. Now they said that when you mutate the Hsp90, which is, you know, what is happening in these Hsp83 mutants, When you mutate Hsp90, it can no longer function as chaperones. And when they cannot function as chaperones anymore, now all these little little genetic variants that are there in the population They are now going to form different things: different proteins, slightly different proteins.

And all this stuff, all this variation is now going to get expressed. which, in the presence of Hsp90, would not have otherwise been expressed. So, this kind of genetic variation that is present in the genome, But normally it is not seen at the phenotype level; this is what is known as cryptic genetic variation or CGV. Now you can imagine that this was mega mega news that said organisms have variation but which are normally not seen. Now people were obviously skeptical for very good reasons, but you know the Lindquist group spent a lot of time. And then, you know they were able to show the same phenomenon for Hsp90 in Arabidopsis. So, they did exactly the same thing, you know; they took an inhibitor of Hsp90. There they did not do, as far as I remember. They did not do, you know, mutation; they used geldanamycin as an inhibitor.

And they showed that the moment you inhibit the effect of Hsp90, all kinds of mutations start showing up. Similar observations were made in this particular fish. This is known as the Mexican Tetra. So, you can find this fish in two forms. There are some forms that are found at the surface of the water, you know, close to the surface of the water.

And these are the ones that I am showing here on the top, and as you can see, they have nice eyes. You know they are regular, normal fish that see things. But some of these populations of the species tend to migrate into underwater dark caves. And when they do that in multiple cases, they lose their eyesight very, very quickly and become blind. Now, can you see over here that there are no eyes? So not only are there no eyes, but also the orbit of the eye and the, you know, the.

This thing actually shrinks and becomes either like a point or is lost completely. So the question is, why exactly is this happening? Now, what Rohner et al. were able to show is that this is also related to Hsp90. So if you hinder the action of Hsp90, this time they used an inhibitor called radicicol. So if you end up hindering the production of HSP90, then the variation in eye size and orbit size increases.

So earlier, suppose it is tight like this; then the variation goes up like this. Now, if you

take those individuals who had lower, you know, eye size and orbit size and breed them with each other, Then, in the next generation, there is a very sharp drop in the mean size of the orbit and the eye, telling you that. This trait is genetically inheritable. So now the question is, okay, this is happening in the lab when you are putting them under Radisicol. Why and how does that explain why these guys are losing their eyesight, you know, under natural conditions? So what Rohner et al. showed was that if you looked at the water in the underground caves Then those waters typically have much lower conductivity. And this lower conductivity, you know, kind of acts like a stress or something because of which. It actually ends up inhibiting or mimicking the effect of radicicol. In other words, the lower conductivity again increases the variation in the, you know, variation. In the eye size and the orbit size, there are many individuals produced who have very low or very small eyes and very small orbits.

Now, of course, the question is, okay, this is leading to the production, but then how are the eyes, you know, getting lost? What exactly is happening that is causing the eyes to get lost? Now that you know the work Rohner et al. did not cover that, but there was a subsequent paper which Actually ended up showing that in the dark, obviously, these fish can no longer use their eyes for catching their prey. So, they are actually depending on certain cells and other sensory organs in their heads for catching their prey. And the expression of these things, along with the sensitivity of these things, is negatively correlated with eye size. In other words, those fish that have smaller eye sizes have much better sensitivity for these sensory organs and therefore, they are able to catch fish much better and catch their prey much better. As a result, there is a positive selection for low eye size, and as Rohner et al. commented, this loss of eye You know, for the surface fish going blind and entering the underground caves, that has actually happened multiple times. It is not a one-time situation in one population; it has happened multiple times in the Mexican Tetra. So, this leads us to two different but closely connected concepts in evolutionary biology in terms of how variation is generated.

One is, you know, there is one called canalization; the other is called evolutionary capacitance. So, as we saw, there can be a lot of genetic variation or cryptic genetic

variation for traits, and This variation is normally not expressed because the developmental system is very robust. We just talked about Hsp90, but there can be other things as well that are leading to all these variations not being expressed. And this phenomenon wherein there is a reduction in phenotypic variation despite it being present Either in terms of the environment or in terms of the genotype, this phenomenon is what is known as canalization.

Now, as you can imagine, this is a very, very important phenomenon. Why? Because all organisms are continuously exposed to all kinds of environmental variation. As we saw, sometimes there can be variations, you know, small mutations, etcetera, at the genetic level as well. Now, if the organism has no way to buffer this variation, then all you know is that everybody is going to experience continuous developmental shocks. So, at some level, you need some way to buffer as much of what you know about environmental variations to some extent.

So that is canalization. However, there has to be a limit to the buffering. If the environment changes too much such that it becomes a pressure, it becomes stress; then the organism needs a way to Generate variation, and let us say either there are changes at the genetic level or there are changes at the environmental level. And the way in which the organisms do that, you know, like what I showed you in the context of the Mexican tetra, That is what is known as evolutionary capacitance. So, the phenomenon by which phenotypic variation can be exposed to selection in response to a cue. So, this allows for a just-in-time release of variation that can help an organism face environmental fluctuations. So, you can see that phenotypic plasticity, you know, it kind of cuts both ways, or any kind of plasticity.

Sometimes you need the whole thing to be expressed, and sometimes you need the whole thing to be buffered. So, you have these two phenomena of canalization and capacitance that are kind of working in conjunction with each other. And you know the balance between the two is what maintains the stability of the organism. However, there is a slight twist in the tale over here. What is it? So, it turns out that there is a kind of RNA in the

body known as piRNA, and these are the piwi-interacting RNAs.

These are small non-coding RNAs. They are particularly involved in the epigenetic and post-transcriptional silencing of transposons, the jumping genes. Remember the ones that we talked about. We were talking about retrotransposons in our previous discussion, where mRNA is converted to cDNA for incorporation into the genome. But along with retrotransposons, you also have lots of transposons, right? which will just take segments of the gene and randomly insert them somewhere else.

So, piRNAs they tend to control the action of these transposons. Now it turns out that Hsp90 plays a very important role in the formation of piRNA. So when you end up mutating Hsp90, the piRNA level goes down. and it has been shown that this leads to a major increase in the transposon-induced mutagenesis. Now, if that happens, remember what we saw in the context of Rutherford and Lindquist's work when they mutated Hsp90, they started seeing all kinds of phenotypes. They assumed that the phenotype was the variation that was already there; the genetic variation was already there, and now. Because of the mutation of Hsp90, that variation is getting expressed, which is why they call it cryptic genetic variation. But if this is correct and people have shown that this works, then this indicates that the variation was not there to begin with. The variation was created because there was a particular form of mutation through a transposon. Activity had been kept suppressed by Hsp90, and now When you take out that break, lots of mutations happen, and that is what is showing up.

In other words, it is not pre-existing variation; it is de novo created variation. Now this obviously changes you know whether the our way of thinking about cryptic genetic variation. Now, another thing that we already know, particularly in the context of Bacteria, it has been shown again and again, are what is known as stress-induced mutagenesis. What is that? We are not talking about, you know, mutation that is induced by, let us say, UV. For some mutagen, and you know other things that are a different ball game altogether.

Those are chemical, or you know, whatever radiation-induced mutagenesis. But I am talking about the fact that in many bacteria, it has been shown that If you stress the microorganism, then the mutation rate goes up. This is what is known as stress-induced mutagenesis. Now, obviously, if you have that, then that is another source of variation and potential variation. And lots of people are now asking the question of how important stress-induced mutagenesis is. In terms of, you know, as a source of variation for organisms, and more importantly, whatever we are seeing as CGV, cryptic genetic variation, can it be explained away as a stress-induced mutagenesis? We do not know if people are still working on this.

So now let us take a step back and reflect on what we have learned. So previously we learned about how various kinds of mutations arise. In the previous discussion, we saw how new genes arise, right? But as we remarked, Both are very slow processes, and organisms very often, because of environmental changes, need faster solutions. What could those solutions be? Previously, we learned about epigenetic mechanisms; we had learned about plasticity. And we had learned that you know some of those epigenetic mechanisms; well, epigenetic mechanisms are very often. The underlying mechanisms behind phenotypic plasticity; you know that is something we have to understand.

But we had previously said that many epigenetic mechanisms are not transgenerational, although We can nowadays see more and more epigenetic changes being discovered that are transgenerationally inheritable. which means that selection can act on them. In continuation of that, we today saw that plasticity itself, the ability to change, can be subjected to evolution. And all these things are basically telling us that our, you know, genetic systems, everything that we have, All that is continuous, oh sorry, and we also saw that the environment can also induce genetic changes.

So, that basically means that our genome is actually, you know, being continuously bombarded. by all kinds of forces that are operating at various time scales. Some are operating on the short time scale, while others are operating on the long time scale, and so on. And all these changes are continuously leaving their marks on the genome. So, you

can think of our genome as, you know, a very old visitor's book on which people— you know, the kind that they keep in. Various shrines or monuments where people go to sign and express something about their feelings, and so on.

So, you know this has been accumulating all these signatures over the last several years. And in the context of the genome, you know, for millions of years. And if you are able to read and understand those patterns, Then it tells you a lot of things about how that organism's genome has evolved. And when you try to put that in the context of other species, other nearby close relatives of that organism, you can see. How the whole history of evolution has proceeded in the context of that lineage.

You can see all those patterns written in the genome's code. But how do you see that? What are those patterns? That is what we are going to do in our next discussion. See you then. Bye.