Genes Are Not Immune to Context

Examples from Bacteria

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One of the most widespread misconceptions concerning the nature of genes is that they have a defined and fixed function that allows them to operate the same in all organisms and environments. We have the picture of the robust gene determining all the characteristics an organism has. And this gene will do the same thing in a bacterium as in a corn plant or human being. It doesn't care where it is. The gene carries its set of instructions with it wherever it goes and strictly carries out its duty.

This picture informs genetic engineering. Take a gene from bacteria and put it into a plant and the plant will produce its own pesticide or become resistant to a herbicide. Since such transgenic plants exist, the proof is evidently in the pudding. Genetic manipulation works; genes are faithful workhorses. But does genetic manipulation work the way we imagine with our schematic pictures? What else may be occurring that doesn't fit into a neat mechanistic scheme?

It's somewhat ironic that precisely within the last ten to fifteen years—the period in which genetically modified crops have been developed and commercialized in the U.S. and some other countries—a wealth of research on genes in relation to environmental effects has been carried out, showing that genes are anything but automatic instruction programs immune to their context. This research has significant implications for the way we assess genetic engineering. Unfortunately, it often seems that the results of this basic research have little effect on the minds and pocket books supporting the global drive to manipulate organisms genetically. In this article I'll discuss some examples of the contextual gene in bacteria.

The Interactive Gene

With the widespread use of antibiotics in our culture, many bacteria have become resistant. They thrive even when subjected to high doses of antibiotics. As a rule, the resistance comes at a cost, since the resistant bacteria tend to grow slowly. But when they are grown in laboratory cultures, some of these resistant bacteria will experience so-called compensatory mutations—they stay resistant, but change genetically in a way that allows them to grow fast like wild, nonresistant strains. Others mutate back to the wild form and lose their resistance altogether.

The question arises whether such mutations (changes in genes or in higher-order genetic structures) are in any way dependent on the environment. The traditional view, rooted deeply in the Neodarwinian theory of evolution, holds that genes mutate spontaneously and independently of the environment. The classical experiment with bacteria by Luria and Delbrück in the 1940s gave clear evidence that such spontaneous, milieu independent mutations exist (Luria and Delbrück 1943). For decades this experiment (along with other evidence) served as the rock solid "proof" that genetic mutations, except for extreme cases involving irradiation or exposure to chemical toxins, are not influenced by their environment. But more recent research shows that mutations do in fact arise in response to changing environmental conditions.

A group of biologists in Sweden investigated whether the above-mentioned compensatory mutations and the reversion to the wild form in bacteria are influenced by the environment (Björkman et al. 2000). They grew antibiotic-resistant bacteria—in the absence of antibiotics—as laboratory cultures (in petri dishes) and also inoculated mice with the same bacteria. The researchers monitored the mutations that occurred in the bacteria in these two different habitats. They found that compensatory mutations occurred in both habitats, but, to their surprise, they discovered that the way the genetic material changed differed significantly depending upon the environment. In the case of streptomycin-resistant bacteria in mice, they found ten cases of identical compensatory mutations *within* the resistance gene. In contrast, this gene never mutated in the lab-cultured bacteria, where they found fourteen compensatory mutations in genes *outside* the resistance gene. Evidently, the environment had everything to do with what kind of mutations occurred. "Mice are not furry petri dishes," as the title of a commentary article put it (Bull and Levin 2000).

The authors conclude that the mutations are "condition-dependent" and suggest that some unknown "mutational mechanism" limited the mutations in the mice to a specific part of the resistance gene while also increasing its mutation rate. Whatever the details of cell physiology turn out to be, it is clear that the genome of the bacteria changes in relation to a specific kind of environment. The bacteria—down into their genes—interact with and evolve in relation to their environment.

Adaptive Mutations

In another recent study (Bjedov et al. 2003), a research group in France gathered wild strains of the bacterium *E. coli* from a wide variety of environments—the large intestines of humans and different animals, soil, air, and water. In the end they collected 787 different strains. These strains were given optimal conditions in lab cultures and began to grow and multiply rapidly, mimicking ideal conditions in nature where bacteria reproduce quickly. But in nature, bacteria are also exposed to times of dearth, where the substrate they live upon, for example, is suddenly used up. To mimic these conditions, the researchers withheld nutrients for a seven-day period. Most bacteria survive under these conditions, but they no longer grow and divide.

The scientists measured the rate of mutations occurring in the cultures the first day after withholding nutrients and then again at the end of the seven-day period. During this time the mutation rate increased on average sevenfold. In other words, the mutation rate increased dramatically when the bacteria no longer received adequate nutrition. The bacteria switch, in the words of the authors, "between high and low mutation rates depending on environmental conditions" (p. 1409).

Such a stress-induced increase in mutation rate has been discovered in many laboratory strains of bacteria. Does this increase in mutation rate serve the bacteria, or is it a kind of last gasp, a dissolution of the bacteria before they die of starvation? The answer is clear: the bacteria produce unique kinds of mutations during such periods of physiological stress, some of which help the bacteria survive under specifically those conditions. One speaks of "adaptive mutations." (See Wirz 1998 and Rosenberg 2001 for good overviews of the research and literature.)

For example, there are strains of *E. coli* that have lost the capacity to utilize the sugar lactose as a source of energy. If such a strain is cultured in a starvation medium with lactose as the only energy source, most of the bacteria remain in a stationary phase until they die. But under these conditions some of the bacteria begin to hypermutate, which means they rapidly create a large number of mutations and among these are ones that allow them to live from lactose. The bacteria with this ability survive, multiply and form new colonies. In at least some cases such adaptive mutations arise only in the specific medium—that is, the mutations allowing bacteria to utilize lactose don't occur when bacteria are grown in a medium with sugars other than lactose.

In other instances, *E. coli* bacteria do not hypermutate, but find another way to deal with the environmental challenge. Some of the bacteria in the medium with lactose produce multiple copies of the gene related to their inability to live from lactose. This gene amplification seems at first absurd. But scientists found that *E. coli* strains unable to grow when they only receive lactose as a nutrient do form enzymes that break down lactose, but in inadequate amounts. When the bacteria amplify the defective lactose enzyme gene, the cumulative effect is that they produce enough enzymes to break down a sufficient amount of lactose to grow slowly and survive – a remarkably active and meaningful genetic adaptation. This amplification occurs in no other genes in the bacteria. It is specific to the lactose enzyme gene and clearly induced by the environment.

Transfer of Resistance

Bacteria have a further way of adapting to new conditions. I have already mentioned antibiotic-resistant bacteria. Cholera bacteria, for example, are normally susceptible to different antibiotics. After 1993 antibiotic-resistant cholera bacteria rapidly spread around the globe. How could this occur? Scientists discovered that these bacteria are simultaneously resistant to at least five different antibiotics. They found that the genes related to this resistance were all grouped together and formed a "packet" of genes that can move from bacterium to bacterium.

A research group at Tufts University in Boston recently discovered conditions that facilitate this movement and uptake of genes (Beaber et al. 2004). When bacteria are grown in cultures with concentrations of antibiotics that are not sufficient to kill them, they go through physiological changes similar to what happens to bacteria in a starvation medium. Part of this transformation is called an SOS response. It comes about when DNA is damaged and involves DNA repair and duplication. The Tufts scientists found that during the SOS response the bacteria also increased the transfer of the resistance gene clusters to other bacteria. Evidently, the use of antibiotics promotes the spread of antibiotic resistance among bacteria. In this way, once resistance is anchored in mobile genetic elements, it can spread rapidly. *(continued on page 23)*

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The examples I have described show how strongly the environment influences the activity of genes, induces changes within genetic structures (mutations), and stimulates the movement of genes between bacteria. Bacteria are in continual interplay with their environment, actively responding to changing conditions. And this responsiveness and flexibility includes genes. If we release genetically engineered bacteria into the environment, there is little doubt that in time they will be passing their genes to other bacteria, as well as receiving genes from other bacteria and mutating according to changing circumstances. Whether the manipulated foreign genes they carry will be exchanged, or how they may affect or be affected by the dynamics of genetic responses to changing environments is completely open. But two things we can know for sure: these genes will not function immune to the changing circumstances and things will happen that no one expects or can foresee. I'm not saying this to promote fear, but to dissolve the illusion that we can keep under control what we have released into the world in this way. Genes are robust, but they are also part of the world.

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