glycemia of infancy (FHHI), is associated in several families with mutations in the second nucleotide-binding domain of SUR, suggesting that this fold may have something to do with insulin secretion by analogy with a similar CFTR mutation (*7*, *17*). The mechanism of this defect, a putative increase in the ability of ATP to close K_{ATP} channels, suggests that the ß cells would be chronically depolarized and therefore hypersecretory. If the SUR mutations actually cause a loss of open K_{ATP} channels, this explains why the syndrome is autosomal recessive, because only a few open K_{ATP} channels are required to set the resting membrane potential (*18*). [It also explains the well-known leucine sensitivity of these patients, because leucine stimulates ß cell oxidative phosphorylation, which would produce more ATP to further depolarize the membrane (*19*).] Gene therapy may therefore afford an interesting opportunity for treatment of FHHI. Mental retardation in FHHI, ascribed to repeated hypoglycemia, should now be reconsidered to be complicated by defective SUR expression in the nervous system, and cardiac abnormalities might be uncovered in these patients as well. Insulin hypersecretion due to an SUR mutation raises the possibility that insulin hyposecretion syndromes related to defects in SUR might also exist, although diabetes genes have not been localized to the region of FHHI.

Whither K_{ATP}? Inward rectifier–related channels and SUR certainly seem destined for each other, assuming the SUR is not itself a channel, but the right match remains elusive. An exhaustive combinatorial approach with pairs of inward rectifier–like channels may be illuminating. Additional bridging subunits may yet turn up to couple sulfonylurea binding to channel inhibition. The isolation of cDNAs for these putative membrane proteins will, we sense, fuel future studies of metabolic coupling to cell excitability.

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Adaptive Mutation: Who's Really in the Garden?

James A. Shapiro

Neo-Darwinists teach that mutations arise independently of biological needs. Physical insults, chemical fluctuations, and replication errors lead to stochastic changes in DNA sequences. Random mutations mean that the evolutionary watchmaker is blind (*1*). To bolster their argument, neo-Darwinists cite an experiment by Luria and Delbrück in which bacterial mutations occur prior to selection for the mutant phenotype (*2*). However, this neo-Darwinist position has been challenged for over a decade by the discovery that certain mutations occur much more frequently when they are selected, and thus adaptively useful, than they do during normal growth (*3*–*6*). The principal difference between these so-called "adaptive mutations" and the phage resistance mutations studied by Luria and Delbrück is that selection for adaptive mutations is not lethal. Thus, nonmutant cells can survive to undergo DNA changes under selective conditions. Not surprisingly, the evolutionary significance of adaptive mutation is highly controversial, and there is great curiosity as to its mechanism, a curiosity that will be stimulated by two new reports in this issue of *Science* (*7*, *8*).

A popular system in which to study adaptive mutation uses the lacI-Z33 mutation, a fusion of lacI (encoding repressor) and lacZ (encoding ß-galactosidase), which prevents bacterial growth on lactose medium because it includes a frameshift mutation that blocks lacZ translation (*9*). Strains carrying lacI-Z33 display delayed kinetics of Lac^+ colony appearance when plated on lactose medium, indicating that many more frameshift events occur after selection than during prior growth, that is, adaptive mutation (*9*). Several laboratories have begun unraveling the molecular basis of lacI-Z33 reversion. RecA and RecBCD homologous recombination functions are necessary for adaptive lacI-Z33 reversion but not for background reversions (*9*, *10*). Moreover, adaptive frameshifts are different in sequence from those occurring nonselectively (*11*).

The two reports in this issue of *Science* add spice to the lacI-Z33 pot by demonstrating that plasmid transfer functions are also required for adaptive reversion (*7*, *8*). The lacI-Z33 mutation used in the studies described above happens to be carried on an F_{lac} plasmid. Peters and Benson have recently shown that F_{lac} transfer is frequent under conditions comparable to selection for lacI-Z33 reversion (*12*). While investigating the significance of these facts for adaptive mutation, Radicella and co-workers (*7*) find that selection-induced reversion and plasmid transfer are inseparable: A chromosomal lacI-Z33 mutation (in which transfer does not happen) displays a 25-fold reduction in selection-induced reversion, adaptive Lac^+ revertants carry plasmids that have been transferred, and nontoxic inhibition of plasmid transfer blocks selection-induced reversion. Most striking is the finding that streptomycin-sensitive lacI-Z33 bacteria can produce adaptive Lac^+ revertants in the presence of lethal concentrations of streptomycin if and only if they can transfer the F_{lac} plasmid to streptomycin-resistant recipient cells. In the accompanying report, Galitski and Roth demonstrate that a regulatory molecule that inhibits expression of F_{lac} transfer functions also blocks adaptive lacI-Z33 reversion (*8*). They also find that

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lacI-Z33 strains express low levels of ß-galactosidase activity sufficient for slow bacterial growth on selection plates. Foster and Trimarchi confirm that adaptive lacI-Z33 reversion depends on conjugation functions (*13*). Peters, Bartoszyk, and Benson provide new mechanistic insight by demonstrating an unexpected correlation between the effects of recA and recD mutations on adaptive lacI-Z33 reversion and on the frequency of plasmid transfer under selective conditions (*14*).

A major conclusion from all these results is that adaptive lacI-Z33 reversion represents more than malfunction of the basic replication machinery under stress. Additional specific molecules participate in selection-induced mutations that restore growth on lactose (namely, homologous recombination and plasmid transfer functions). The participation of DNA-processing activities is reminiscent of another well-studied case of adaptive mutation. Selection-induced formation of araB-lacZ coding sequence fusions requires phage Mu transposase and proteins needed for transposase function (*15*).

The blind watchmaker or the genetic engineer—which is the best model for evolutionary change? The adaptive mutation debate introduces a high tech element into evolutionary theory. [Photograph B. Llewellyn/Uniphoto]

Molecular genetics has taught us that cells contain many biochemical complexes capable of reorganizing DNA sequence information that act as major agents of genetic change (*16*, *17*). Both the lacI-Z33 and araB-lacZ models illustrate how these natural genetic engineering systems play an important mutational role. Because biochemical complexes that act on the genome are subject to physiological regulation like all cellular functions, it is logical that they display much more activity under certain conditions. For example, plasmid transfer and RecA function are regulated by cellular control circuits responding to oxidative metabolism, DNA damage, and carbohydrate starvation (*12*, *18*).

A second conclusion from the lacI-Z33 results is that genetic change in bacteria is often multicellular. DNA rearrangements can occur in one cell and be transferred to another before a clone of "mutant" bacteria proliferates on selective medium. Such "altruistic" mutational events had been predicted by Redfield and Higgins (*19*), and Radicella *et al.* now prove their existence (*7*). Are such lateral genetic hand-offs peculiar to bacteria, well-known as virtuosos of intercellular DNA transfer? Probably not. Viruses and other parasites capable of carrying genetic material between cells are ubiquitous in biology, and the cells of most organisms can take up and integrate foreign DNA under certain circumstances. In stress situations, likely to prevail at critical episodes in evolution, intercellular genetic exchange may well be more common than we have believed.

One meandering of the adaptive mutation debate followed the suggestion that selective substrates "direct" mutations to particular regions of the genome (*4*). Although not inherently implausible (signals can direct the transcription apparatus to particular regions of the genome, and the transcription apparatus can be coupled to DNA-processing activities) (*20*), this "directed mutation" hypothesis remains unsubstantiated. Selection-induced recombination activities and plasmid transfer functions are sufficient to explain specific adaptive reversion of lacI-Z33 without recourse to directed mutation. Directed mutation can also be excluded in the araB-lacZ system, where indirect sib-selection and other techniques have unambiguously established that selective substrates are not necessary to induce adaptive fusions (*21*).

Regulation of biochemical complexes responsible for genetic change and DNA transfer [combined, in some cases, with background growth (*8*)] can explain both quantitative and qualitative aspects of adaptive mutation. Thus, adaptive mutation has returned to the mainstream of molecular genetics. Does anything novel then remain for evolutionary theory? Most emphatically, yes. The discovery that cells use biochemical systems to change their DNA in response to physiological inputs moves mutation beyond the realm of "blind" stochastic events and provides a mechanistic basis for understanding how biological requirements can feed back onto genome structure (*17*, *21*). An ability to increase the frequency of useful mutations under selection can be evolutionarily quite significant even if other mutations occur as well. This is especially true if the adaptively useful changes can be incorporated into the genome of another cell, as happens with lacI-Z33 (*7*, *8*). To paraphrase an earlier commentator in this debate (*23*), recent results on molecular events in both the lacI-Z33 and araB-lacZ systems indicate that there is no unicorn in the genomic garden. But we have found a genetic engineer there, and she has an impressive toolbox full of sophisticated molecular devices for reorganizing DNA molecules. The years to come will teach us many fascinating lessons about her work habits and how they change in response to the myriad challenges of growth and survival.

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