Reproduction and Heredity

Why Do Some Genes Maintain More Than One Common Allele in a Population?

When Mendel did his crosses of pea plants, he knew what a pea plant was supposed to look like: a small plant with green leaves, purple flowers, and smooth seeds. But if all pea plants were like that, he would never have been able to sort out the rules of heredity—in a cross of green peas with green peas, there would have been no visible differences to reveal the 3:1 pattern of gene segregation. The variant alleles that Mendel employed in his studies—yellow leaves, white flowers, wrinkled seeds—were rare "accidents" maintained in seed collections for their novelty. In nature, such unusual kinds of peas had never been encountered by Mendel.

By the time Mendel's work was rediscovered in 1900, Darwin had provided a ready explanation of why alternative alleles seemed to be rare in natural populations. Natural selection was simply scouring the population, cleansing it in each generation of less fit alternatives. While recombination can complicate the process in interesting ways among sexual organisms like peas, asexual organisms like bacteria were predicted to be very sensitive to the effects of selection. Left to do its work, natural selection should crown as winner in bacterial population the best allele of each gene, producing a uniform population.

Why do populations contain variants at all? In 1932 the famous geneticist Herman Muller formulated what has come to be called the "classical model," explaining gene variation in natural populations of asexual organisms as a temporary, transient condition, new variations arising by random mutation only to be established or eliminated by selection. Except for the brief periods when populations are undergoing this periodic cleansing, they should remain genetically uniform.

The removal of variants was proposed to be a very straightforward process. During the periodic cleansing periods envisioned by Muller, his classical model operates under a "competitive exclusion" principle first proposed by Gause: whenever a new variant appears, it is weighed in the balance by natural selection, and either wins or loses. There are no ties. One version of the gene becomes universal in the population, and the other is eliminated.

.04 µm

These bacterial cells are dividing. As the population grows, gene variants arise by mutation. Do the new variants persist, or are they eliminated by natural selection?

Muller's classical model thus makes a very straightforward prediction: in nature, most populations of asexual organisms should be genetically uniform most of the time. However, this is not at all what is observed. Natural populations of most species, including asexual ones like bacteria, appear to have lots of common variants—they are said to be "polymorphic."

So where are all of these variants coming from? Variation in the environment, either spatial or temporal, can be used to explain how some polymorphisms arise. Selection favors one form at a particular place and time, a different form at a different place or time. In a nutshell, varying selection can encourage polymorphism.

Is that all there is to it? Is it really impossible for more than one variant to become common in a population, if the population lives in a constant uniform environment, an environment that does not vary from one place to another or from one time to another? Theory says so.

Biologists that study microbial communities have begun to report that bacteria are not aware of Muller's theory. Bacterial cultures started from a single cell living in simple unstructured environments rapidly become polymorphic.

There is a way to reconcile theory and experiment. Perhaps the variant individuals in the population are interacting with one another. Muller's theory assumes that every individual undergoes an independent trial by selection. But what if that's not so? What if different kinds of individuals help each other out? Stable coexistence of variants in a population might be possible if interactions between them contribute to the welfare of both (what a biologist calls mutualism) or favors one (what a biologist calls commensalism). In essence, cooperation would be counterbalancing the effects of competition.

Real People Doing Real Science
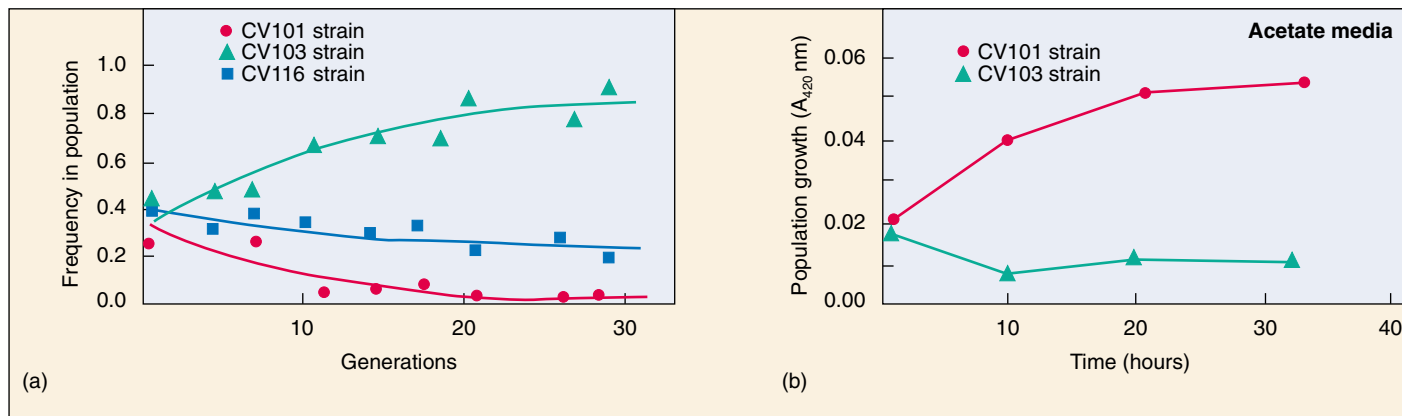
Maintaining stable polymorphism. (*a*) Three new strains emerge in culture and are maintained. (*b*) Two strains are grown on media containing acetate. The strain CV103 was found to excrete acetate, while the strain CV101 was found to thrive in media with acetate as the sole source of carbon. Population growth is measured by an increase in the turbidity of the liquid medium; turbidity is measured as an increase in light absorbance at a wavelength of 420 nm (A_{420} nm).

The Experiment

To investigate this intriguing possibility, Julian Adams and co-workers at the University of Michigan set out to see if polymorphism for metabolic abilities would develop spontaneously in bacteria growing in a uniform environment.

For a bacterial subject they chose *Escherichia coli* (*E. coli*), a widely studied bacterium whose growth under laboratory conditions is well understood. Cultures of *Escherichia coli* can be maintained in chemostat culture for many hundreds of generations. A chemostat is a large container holding liquid culture medium. A little bit of the liquid is continuously removed, and an equal amount of fresh culture medium added to replace what leaves. The growth of the *E. coli* culture is limited by the amount of glucose remaining in the culture medium to feed the growing cells.

Researchers inoculated a glucose-limited chemostat culture media with the *E. coli* strain JA122, and maintained the continuous culture for 773 generations. A sample was taken from the chemostat after 773 generations and analyzed for the presence of new strains of *E. coli*. Any variation among the cells in the sample would indicate that polymorphism had arisen.

To detect metabolic variation within the sample of growing cells, Adams's team analyzed the rate of glucose uptake and the concentration of acetate, among other variables. By examining such biochemical parameters, the researchers could determine if the different strains were filling different metabolic "niches"—that is, using the metabolic environment in different ways. Metabolic niches were characterized by looking at the normal products of aerobic fermentation, acetate and glycerol, which appear in the growth medium as a by-product of *E. coli* metabolism.

To further classify the strains, batch cultures containing two strains were established to analyze interactions between the two groups.

The Results

Three distinct variants were detected in the 773-generation *E. coli*, each being maintained at stable levels in the continuously growing culture. Clearly polymorphism *can* appear within an initially uniform bacterial population growing in a simple homogeneous environment.

When mixed together and allowed to compete, one strain does not drive the other two to extinction, as theory had predicted. Instead, the three new strains, CV101, CV103, and CV116, all persist (see graph *a* above).

The three strains were then analyzed to see how they differed. CV103 exhibited the highest rate of glucose uptake and produced the most acetate (an end product of glucose aerobic fermentation). Is this difference important? To see, the CV103 strain was co-cultured with CV101. They maintained stable growth levels, which indicated that the contribution of the third strain, CV116, was not required to maintain their growth.

What is the difference between CV101 and CV103? CV101 could grow in culture filtrate of CV103 but in the reverse situation, CV103 could not grow. This indicates that CV103 secretes a substance upon which CV101 can grow. Is CV101 utilizing the acetate produced by CV103 as its carbon source?

To test this possibility, CV101 and CV103 were grown together in media with acetate as the only carbon source. The results from this experiment are shown in graph *b* above and indicate that CV101 thrives on an acetate carbon source, while CV103 does not and requires an additional carbon source such as glucose.

These results indicate that two of the strains are maintained in polymorphism at stable levels because they have evolved different adaptations that allow them to coexist by filling different niches. One strain (CV101) is maintained in the population because it is able to use a metabolic by-product released by another strain (CV103).

To explore this experiment further, go to the Virtual Lab at www.mhhe.com/raven6/vlab4.mhtml