On the Many Origins of Species

Researchers thought it took mighty barriers like mountains to make new species. Now they are learning that the process can rest on something as slight as a taste for a new fruit

The making of a new species is one of evolution's most heralded feats, and scientific understanding of the process has itself been evolving at a rapid pace. Many advances were showcased this spring at a conference, "Endless Forms: Species and Speciation," held 19 to 23 May in Asilomar, California; some are presented in this Special News Report. New species that form without the benefit of geographic isolation are the topic of our first story, accompanied by a piece exploring a mysterious case of biogeographic species-making. Our second story details the many mechanisms, from third-party intervention to slight changes in a sperm surface protein, that add new plants and creatures to the planet.

Guy Bush's scientific debut was not an auspicious one. In 1966, Bush was a newly minted Ph.D. biologist from Harvard University, nervously presenting his first paper at a scientific meeting. He had reason to be anxious. His data pointed to a controversial finding—that a new species of fruit fly was forming in trees right beside its parent population, a process called sympatric speciation. The conventional view was that such things don't happen: New species form only after two populations are separated by a physical barrier. And after moments of dead silence from the audience, the famed geneticist Theodosius Dobzhansky spoke: "That's very interesting, but I don't believe it. Sympatric speciation is like the measles. Everybody gets it, but they all get over it."

Today, 30 years later, Bush is still infected. Now a professor at Michigan State University, he quips: "I've had this rash that's never gone away." And it has turned out to be catching. By applying new biochemical and molecular tools to their fruit fly, the apple maggot *Rhagoletis pomonella*, Bush and his students have convinced a lot of scientists that the choice of a new host plant can separate populations just as a mountain or a river can. "I'm delighted to say that I think they've developed a convincing case," says Douglas Futuyma, an evolutionary biologist at the State University of New York, Stony Brook. Evolutionary ecologist Dolph Schluter of the University of British Columbia, who has spent years arguing with Bush, now sees signs of sympatry even among the stickleback fish he studies. "I felt a bit like the president of the flat-Earth society when shown the first photograph of a round Earth from space," he says.

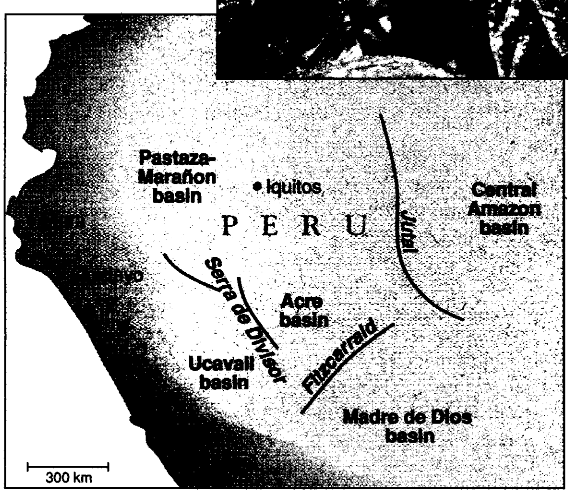
The case for sympatry is now so strong, says John Endler, an evolutionary biologist at the University of California, Santa Barbara, that the debate is less over whether sympatric speciation can take place than over how

Amazonian Diversity: A River Doesn't Run Through It

For explorers of the Amazon River basin, no feature stands out more than the winding, intricate river systems. Biologists, too, have been impressed by the Amazon and its tributaries, viewing them as barriers that created a great diversity of plants and animals. As rivers changed courses, they would have cut populations into two, creating new groups that followed separate evolutionary paths. But rivers may not run throughout Amazonian diversity. Instead, the engine behind much animal speciation may be ancient ridges now lying buried and nearly invisible.

James L. Patton, an evolutionary biologist at the University of California, Berkeley, reported at the speciation meeting that he has found a surprising correlation between the 3-million-year-old history of one hidden ridge and genetic differences among the small mammals he studies. Richard Harrison, an evolutionary biologist at Cornell University, notes that the work offers an unusually clear view of this process, known as allopatric speciation, in which the geographic barriers responsible for it are often difficult to pinpoint. "Patton's is a classical and elegant example," he says. "It's giving us a window on an evolutionary event we had no idea of."

JAMES L. PATTON

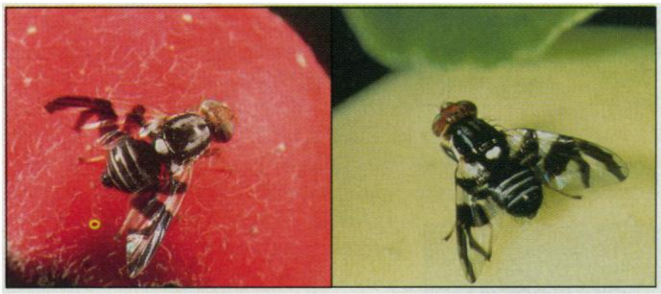


SOURCE: M. E. RASANEN *ET AL.*, *SCIENCE* 238, 1398 (1987)

Hidden barriers. Now nearly invisible, ridges *(purple)* segmented the Amazonian basin millions of years ago and may have divided the spiny tree rat into different species.

Patton and his Brazilian colleagues from the Instituto Nacional de Pesquisas da Amazonia originally set out to verify the river hypothesis, which hasn't been seriously challenged since it originated with Alfred Russell Wallace, the noted 19th century evolutionist. Wallace had observed that the geographic ranges of the basin's many primate species coincided with boundaries set by its rivers: the Amazon, Negro, and Madeira. The idea is especially appealing as an explanation for the region's diversity, notes Patton, for these rivers are not only among the largest in the world but the most dynamic. "It's easy to imagine that they would influence speciation," says Patton, because they are prone to flooding and changing course, severing a single population.

Patton and his team collected gene samples from 52 species of mammals, including tamarins, rodents, and marsupials, from both banks of the 1000-kilometer Río Juruá, a tributary of the Amazon in western Brazil. The researchers examined the anioften and under what conditions. The noted Harvard biologist Ernst Mayr, the most influential skeptic of the notion, now says sympatry could be real but is an "unimportant" process in nature, responsible for only a small number of new species when compared to the geographic process, known as allopatry (see box). But others such as Bush argue that sympatry could have helped create the diverse array of plant-feeding insects and freshwater fish that exists today.



G. BUSH AND J. JENKINS

A little difference. Related hawthorn maggots *(left)* and apple maggots *(right)* live side by side, but fruit and mating preferences keep the two populations separate.

Proposals without proof

Although most biologists have only recently changed their minds about sympatric speciation, Charles Darwin thought it was possible back in the 1850s, and said so in a manuscript for his book *Natural Selection* (published after his death). "I do not doubt that over the world far more species have been produced in continuous than in isolated areas," he wrote. But he did not spell out the details.

Others stepped in to try to solve the mystery. One was Benjamin Walsh, a minister and avid insect collector who eventually became state entomologist for Illinois. Walsh was the first to be inspired by the apple maggot *Rhagoletis*, a native American species whose natural host is the hawthorn tree. A local newspaper recorded that by 1862, some of the flies had lighted on apple trees in an orchard in the Hudson River valley of New York and acquired a taste for the fruit, which had been introduced from Europe, ignoring the small, red fruit of the hawthorn. Walsh proposed in 1864 that when the two groups of fruit flies had started eating and laying their eggs on different host plants, the sister populations effectively isolated themselves from each other enough to become separate species—a process that eventually became known as sympatry (from the Greek *sun* for together and *patra* for fatherland).

It was a provocative proposal, but one without proof. There was no firm evidence that the two groups of fruit flies were separate species: They looked and acted alike, and could well have been interbreeding. And there was no mechanism to explain how adapting to different plants would prevent the two populations from interbreeding. So the notion of sympatry rattled around until 1947. In that year, Mayr took it on and showed that none of the cases for sympatry proposed by Darwin or others was supported by evidence. Meanwhile, the argument for allopatry as the main mode of speciation was getting stronger. Case after case—ranging from Darwin's finches in the Galápagos Islands to beetles in North Africa—showed that new species formed when two populations were cut off from one another by the sea, a mountain range, or other physical barrier. By the early 1960s, influential scientists such as Mayr thought sympatry was an unimportant process. "One would think that it should no longer be necessary to devote much time to this topic ..." he wrote

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mals' mitochondrial DNA (mtDNA) to identify genetically distinct groups. Because mtDNA is inherited only from the mother, is not scrambled by sexual recombination, and appears to mutate at a regular rate, differences in the mtDNA of two related groups can also serve as a "molecular clock" to date their separation.

For the tamarins, Wallace's river theory appears to have been on target. At the Juruá's mouth and widest points, where the barriers to interbreeding should be greatest, there are two distinct subspecies of saddle-back tamarins. At the Juruá's narrow headwaters, however, the two interbreed. "It's exactly what the riverine barrier predicts," says Patton.

But the rodents and marsupials presented a surprise. Instead of diverging across the river, as do the tamarins at the river mouth, these smaller animals were separated genetically into upriver and downriver lineages. "Eleven of the 17 species we sampled show this kind of divergence," says Patton. And while current taxonomy lumps upriver and downriver groups into single species, Patton says the genetic differences between these populations are so strong—some of their mtDNA differs by 13%—that they indeed may be separate species, although morphologically the animals cannot be told apart.

What is really striking, he says, is that all 11 are separated at almost the same geographical point on the river, although there is nothing remarkable about the spot—no bend, no hill, no valley. "When I saw that repeated pattern, I thought, 'Wow! What is going on here?' " Patton recalls. Because the pattern applied to species with a wide range of lifestyles—from treetop specialists such as the spiny tree rat to ground-dwellers such as the spiny mouse—Patton was certain that some exterior force was at play. "When various species have the same pattern of geographic distribution, it's unlikely that it's due to their biology," he says.

Only recently has Patton come up with a possible explanation. While acknowledging that the geological history of the Amazon basin is poorly understood, Patton has uncovered some tantalizing correlations between geologic events and the evolutionary history of the small mammals. Today, the Amazon basin appears to be a relatively flat landscape. But according to a study of its tectonic history (*Science*, 4 December 1987, p. 1398), the basin is actually composed of several subbasins, separated by ancient ridges or arches that were formed when the Andes were uplifted 2 million to 5 million years ago. "One of these," called the Iquitos arch, "cuts perpendicularly across the middle section of the Juruá," says Patton—precisely at the point where the small mammals apparently break into distinctive genetic groups. It matches up in time as well as geography: The mtDNA analysis suggests that the species diverged between 1 million and 3 million years ago.

Patton speculates that the uplift of the arch separated the small mammal populations about 3 million years ago. Those divided populations began accumulating distinctive genetic changes. Later, as the Andes began to erode, the Amazonian subbasins filled in, forming today's vast, flat basin. No longer divided, the mammals have come into contact again, although Patton is not sure if they are interbreeding: "That question will have to wait for nuclear DNA data."

There are similar ancient—and hidden—arches throughout the Amazon basin. Have they influenced the speciation of the small mammals in these regions? If so, says Patton, "they ought to have affected every other organism as well." To answer that question, Patton's team is heading back to the field this summer, to collect the small mammals along another tributary of the Amazon, on both sides of another hidden arch. **–Virginia Morell** in his book *Animal Species and Evolution*, published in 1963.

But a graduate student in Mayr's class—Bush—had studied *Rhagoletis* and could not dismiss it. "It firmly planted the seed of uncertainty about the conventional wisdom," he recalls. What he needed to explain was how a taste for apples could provide reproductive isolation. And as he watched the tiny flies perform a courtship dance on the apples, the explanation hit him: Mate choice—which governs how genes are passed among organisms—and fruit choice overlapped.

The flies, Bush noted, used apples as a stage. Sometimes a male put himself on display, waving his wings until he attracted a female (although other times, a male would lurk under the apple until an unsuspecting female landed on it). If she was interested, they would mate on the apple. And if another male lighted upon his territorial fruit, a tussle ensued, with one male rearing up on its hind legs to push the other off the apple. The same drama was played out by other fruit flies but on another stage: the hawthorn fruit.

PHOTOS BY U. SCHLIEWEN

Species in a small place. Lake Bermin in west Cameroon, just 0.5 square kilometer in area, has no geographic barriers to force speciation, yet has nine endemic fish species, including this *Tilapia bemini (top)*.

"It was clear that both male and female *Rhagoletis* were using their larval host fruits as rendezvous sites for courtship and mating," Bush says. "The idea suddenly struck me that if mate and host choice were tightly correlated, new host races and eventually distinct species of *Rhagoletis* could evolve sympatrically after all." By mating on either apples or hawthorn fruit (but not both), the fruit flies effectively isolated themselves from each other reproductively—a process called assortative mating.

Again, however, here was a model without proof. Bush had to show that the choice of different host plants really did isolate the flies when it was time to mate. So Bush and University of Massachusetts entomologist Ron Prokopy logged many hours in orchards watching fly courtship. Apple maggots, they learned, mated almost exclusively with apple maggots, while hawthorn flies preferred other hawthorn flies. More recently, Jeff Feder, a former Bush grad student now at the University of Notre Dame, marked fruit flies with a colored dye and found that only 6% of the apple and hawthorn flies in his study area were interbreeding with one another. "This confirmed that habitat-specific mating did exist and that it was causing reproductive isolation," says Feder.

That isolation ties in with genetic differences. In 1988 papers published in *Nature*, Bush, Feder, and co-workers Stewart Berlocher, Bruce McPheron, and David Courtney Smith of the University of Illinois, Urbana-Champaign, identified differences in three different chromosomes in the two populations. Moreover, Feder has found that genetic markers within these three chromosomal regions correlate with differences in timing in fly development, which in turn helps keep the fly populations distinct. Larvae of the two groups leave their fruit before winter and burrow into the ground at different times. And the researchers have also shown how the apple maggots usually emerge from the ground as adults earlier in the summer than do their hawthorn-dwelling brethren. These differences in timing generally allow apple maggots to take advantage of the earlier fruiting time of apples, while hawthorn-feeders emerge later when hawthorn fruit are ripe—and these differences further bolster the reproductive isolation of the two groups.

The making of a species

Although *R. pomonella* isn't there yet, the ultimate effects of that kind of separation—the creation of a new species—can be seen in other, related fruit flies, according to Berlocher and Feder. The duo has studied *R. mendax*, a species that has close genetic ties to the apple-feeders and shares almost all of the same range, but has adapted to a different host—blueberries. Even where blueberry bushes grow right up to the trunks of apple trees, the two species of fruit flies stick to their own plants and do not interbreed. This occurs despite the fact that they can mate and produce viable offspring when brought together in the lab. Because there is no evidence that the two were ever geographically isolated from one another, the simplest explanation is that they diverged after mating on and adapting to different host plants, says Feder.

Lab experiments also indicate that genes and habitat preference can sort out flies without physical barriers. At the University of California, Santa Cruz, evolutionary geneticist William R. Rice put *Drosophila melanogaster* in a maze, where they started out so close together that they were crawling over each other. Although they were all members of the same species, they quickly sorted themselves out by apparent genetic predispositions for slightly different habitats in the maze. Some preferred dark instead of light areas. Others chose high instead of low nooks. Still others were drawn to regions with different fruit odors. And they mated in their preferred habitats, leading to complete reproductive isolation, with no backcrossing—a key step toward the formation of separate species. Rice concludes: "The lab work, the field work, and the theory clearly demonstrate that sympatric speciation is genetically feasible."

Corroboration is also coming from an entirely different quarter—from fish that live in newly formed lakes in such far-flung places as Africa and Canada. In several tiny crater lakes in Cameroon, researchers have studied cichlid species that have evolved in the past million years. The mitochondrial DNA from the cichlids shows that the species within each lake are more closely related to each other than to fish in nearby lakes or rivers, which indicates that the new species must have emerged within the lakes—not by lake-hopping or by repeated invasions of fish from the same ancestral stock in nearby rivers. Yet there are no geographic barriers, such as intermittent rock and sand shorelines, within these small, deep lakes to separate individuals in the founder population from each other and set allopatric speciation in motion, says Ulrich Schliewen, an evolutionary biologist at the Max Planck Institute for Behavioral Physiology in Munich, Germany.

Instead, he noticed that the different cichlids have different diets—small cichlids feed on zooplankton, while larger ones feed on insect larvae or fish. He thinks these genetically based food preferences separated the first cichlids in the lake and led to their different sizes. Then nature and assortative mating took their course: Small zooplankton-feeders mated with small zooplankton-feeders, while the large insect-feeders chose other insect-feeders, and new species were formed.

A similar story seems to have unfolded a continent away, where the University of British Columbia's Schluter has been studying stickleback fish that speciated in tiny lakes carved out by glaciers just 13,000 years ago. Like the cichlids, the sticklebacks prefer different foods within the same lake and mate with sticklebacks of the same size—although they don't mate where they eat, so the assortative mating seems to be based on fish size and shape rather than habitat preference (but could be the indirect result of different food choice). And again, genetic studies by Eric Taylor of the University of British Columbia show that pairs of species found within the same lake are more closely related to each other than to any fish outside the lake, which indicates that the species formed in sympatry, says Schluter.

A frequent occurrence?

Just how often this kind of process creates new species is still a matter of dispute. "I think that … after a major episode of extinction or other event that creates lots of empty niches, sympatric speciation is common," says Rice. Plant-feeding insects may be particularly adept at finding new opportunities to diverge from each other sympatrically when a new plant is introduced to their range, for example. And fish may go through similar bursts of speciation when a founder population finds a newly formed lake where there is little competition for resources. By contrast, when ecological niches are already full, most organisms may require geographic isolation to form new species.

Mayr, who thinks the cichlids in Cameroon are the most convincing case of sympatric speciation, is nonetheless skeptical that it occurs often: He thinks that sympatric speciation is responsible for "less than 1%" of all new species. Population genetics models show that it takes very little interbreeding to erase differences between two populations, such as the apple maggots and hawthorn flies.

But new studies of other creatures, ranging from ermine moths and *Heliconius* butterflies to aphids, sockeye salmon, and sea urchins, suggest that sympatric speciation has occurred more than a few times. These studies are also allowing biologists to refine ideas about the conditions under which sympatry might occur (see following story). While those refinements are being made, Bush now has the satisfaction of knowing that the debate is seldom over whether sympatry exists at all—but over how often and why it happens.

–Ann Gibbons

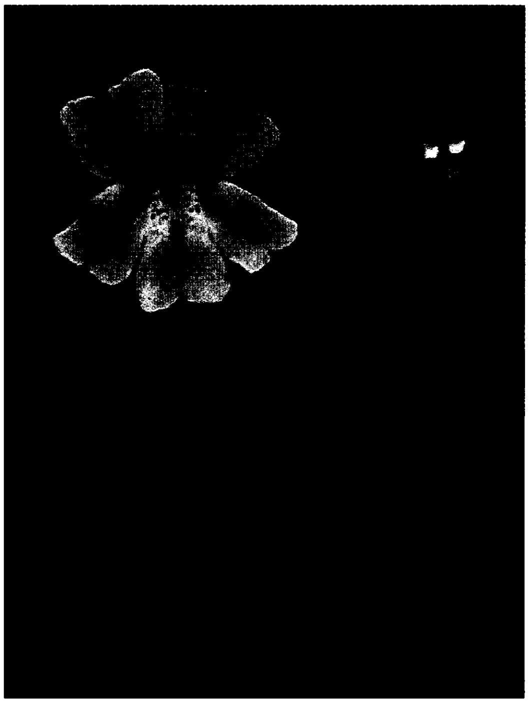
EVOLUTIONARY BIOLOGY

Starting Species With Third Parties and Sex Wars

It often takes a powerful barrier to make a new species. For some kinds of finches, isolated on islands, it was the open ocean that set them on separate evolutionary paths; for certain grasshoppers, it was mountain ranges. But three wasp species may have been pulled apart by nothing stronger than three distinct strains of bacteria living in their guts. Somehow, University of Rochester biologist John Werren has found, the bacteria in the females destroy male DNA from different species, keeping cross-species matings from producing offspring.

And perhaps the wasps aren't so unusual. Evolutionary biologists are fond of noting that in *On the Origin of Species*, Charles Darwin actually wrote very little about how species originate. To fill the theoretical vacuum, scientists have invoked geographic barriers as the usual way populations achieve reproductive isolation, the classic definition of a species. While this is still seen as a major mechanism, researchers now acknowledge that groups standing shoulder to shoulder can split apart as well, in a process known as sympatric speciation (see previous stories). And that process has more than one face, as the speciation conference in California this spring showed.

"The meeting clearly illustrated the diversity of ways that species can diverge," notes Guy Bush, an evolutionary biologist at Michigan State University, in whose honor the conference was held. Researchers described how such factors as bacterial agents, mimicry, changes in a single pigment gene or in the song of a bird, or the rapid evolution of a sperm surface protein can all trigger cascades of events leading to a new species, without any geographic barrier. Meeting participants also heard how another phenomenon that occurs within a single territory, the formation of hybrids—which have often been dismissed as the "mistakes" or "mules" of evolution—may play a much bigger role in the establishment of new taxa than has been previously believed. What has opened researchers' eyes to all these species-making processes, says ecologist Dolph Schluter of the University of British Columbia, are modern methods entailing molecular markers and genetic analysis, as well as time-honored, detailed observation. Researchers have been using these tools to take a much sharper look at populations in the wild, organisms' mating behavior, the basic biology affecting their fertility, and the flow of their genes. "That's what's been missing from speciation research—the whole natural history side of the organism," he says.



D. SHEMSKE

Flower power. *(Top) Mimulus lewisii* and *M. cardinalis* are closely related. But bees are attracted to the contrasting colors in *M. lewisii (bottom left)*, so they pollinate it rather than *M. cardinalis (right)*, which helps keep the species apart.

Third-party intervention. Often, this observational picture has to include a third party—another organism that helps create a barrier between two species. That is especially true of plants, many of which need an insect or bird pollinator to cross-fertilize. University of Washington evolutionary ecologist Doug Shemske and his colleague, plant geneticist Toby Bradshaw Jr., are investigating how pollinators can influence speciation in a study of two sister species of monkey-flowers (*Mimulus lewisii* and *M. cardinalis*). "It's clear that the pollinators contribute to their reproductive isolation," says Shemske, noting that in his lab the two plants easily produce fertile hybrids. In the monkey-flowers' native western mountain