

Disputed islands

Biologists debate whether “genomic islands” are real and mark a first step in the formation of new species

By Elizabeth Pennisi

When the genome of a major malaria-transmitting mosquito was published 12 years ago, headlines touted the potential of that sequence to eventually help curb the devastating disease. But Matthew Hahn was more excited at the prospect of solving a key puzzle in evolutionary biology: how new species form. The mosquito, *Anopheles gambiae*, has two physically identical forms, dubbed M and S, that live in distinct but overlapping parts of Africa and behave differently in some ways. For example, whereas the S strain thrives in temporary pockets of water, such as puddles, the M form prefers rice fields and habitats with more permanent water. To researchers such as Hahn, such contrasts suggested they were seeing the early stages of a species splitting into two.

Using DNA probes made from the newly available genome sequence data, Hahn, now a population geneticist at Indiana University, Bloomington, and his colleagues decided to take a closer look at the two forms' DNA, expecting to find many small differences. Instead, the genomes proved almost

identical except for three spots—pockets of divergence the researchers called “genomic islands of speciation.” In all other parts of the genome, interbreeding apparently still led to the exchange of DNA, or gene flow, between chromosomes of the two forms, keeping their DNA similar. But these islands likely emerged, Hahn and his colleagues suggested, because they contained particular sequences



that improved the survival of one form over the other, making them beachheads for the divergent selection that, under the right conditions, can turn populations into species.

The discovery promised to help answer a question that has challenged evolutionary biologists for decades: how new species

form when there is no geographic barrier to keep separate populations from interbreeding. And it set off a stampede of other researchers looking in their own favorite plants and animals for similar genomic islands—and finding them, in aphids, stickleback fish, *Heliconius* butterflies, and more. “People came around to it being the way they saw speciation” when gene flow remained, says Chris Jiggins, an evolutionary biologist at the University of Cambridge in the United Kingdom.

Lately, however, a smoldering dispute over the islands has flared into open warfare. In one blow to the idea, Hahn has recently become one of its biggest critics, arguing last year at an evolution meeting that most islands are essentially statistical artifacts of the way DNA differences are assessed. “There does not seem to be much evidence for islands,” he now says. But other research groups studying organisms from sunflowers to stick insects have presented new results indicating that genomic islands do exist and are at the heart of some speciation.

Indeed, to some evolutionary biologists, the concept of genomic islands is more compelling than ever. “The controversy is almost

over,” argues Loren Rieseberg, a botanist at the University of British Columbia, Vancouver, in Canada. “It’s a metaphor that everyone is going to continue using.”

SPECIATION WAS ONCE THOUGHT to require some physical feature, such as a river or mountain range, that divides a species into two populations so they can start evolving in different directions. Over time, random mutations and natural selection will change the genomes of the separated populations to the point that should they come into contact again, their DNA will no longer be compatible. Without that kind of geographic barrier, interbreeding should keep the population genetically mixed, preventing speciation.

Yet species do form without a physical barrier. Some of this “sympatric” speciation

is ecological speciation, which starts when two populations adapt to different environments and thus develop different behaviors or preferences—one insect population laying its eggs in apples while another does so in hawthorn fruit, for example. But how do such small differences in behavior spiral into complete separation, if the populations can still interbreed?

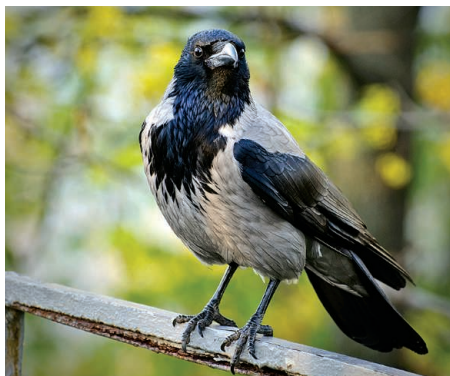
In the 1970s and 1980s, studies of hybrid zones, where two species or populations do successfully mate, indicated that some parts of genomes might diverge, even with gene flow. In 2001, Chung-I Wu of the University of Chicago proposed speciation happened gene by gene, such that parts of a genome could diverge while the rest continued to be recombined and homogenized during reproduction between interbreeding populations.

Without explaining exactly how this could come about, he suggested that slight variation in the way some individuals used their environment or picked mates could lead to a little genetic divergence that could eventually snowball into full speciation. But not until Hahn, working with Thomas Turner, now at the University of California, Santa Barbara, and Sergey Nuzhdin, now at the University of Southern California, compared the M and S mosquitoes did hints of this process come into view.

The three islands they found represented DNA that was more differentiated between the two forms than the rest of their genomes. Shortly later, Nora Besansky, a malaria researcher at the University of Notre Dame in Indiana, did her own analysis of the mosquitoes’ chromosomes. She also found evidence of regions of differentiation, and she used a statistical tool for identifying such regions. Called F_{ST} , it measures, for a particular stretch of DNA, the proportion of genetic variance contained in a subpopulation relative to the total genetic variance. “We all thought this was really cool,” recalls Jeffrey Feder, an evolutionary biologist also at Notre Dame. He and others quickly began using F_{ST} to hunt for islands in other species.

In 2008, a study of two populations of pea aphids, one that lived on red clover and another that depended on alfalfa, uncovered a connection between genomic islands and genes experiencing divergent selection. Through breeding and DNA mapping experiments, Sara Via, an evolutionary biologist at the University of Maryland, College Park, identified the genomic regions that help the aphids adapt to their different food sources and showed that they were located in areas with high F_{ST} .

Genomic islands seemed here to stay. The concept, says Luke Harmon, an evolutionary biologist at the University of Idaho in Moscow, “was viewed by many people as perfect for understanding ecological speciation.” Part of the appeal was the beautiful metaphor: Water represented gene flow, and islands emerged, like volcanoes poking up from the ocean, where gene flow fails and divergent selection occurs. The islands would expand just as volcanic islands grow, until reproductive isolation locked in and a species had split into two. At first, the islands extend beyond the beneficial mutation because DNA right next to the advantageous DNA tends to be transmitted as a unit, due to reduced recombination in this region. Over time, other beneficial mutations in the vicinity would add to this recombination coldspot, expanding the island further. And over subsequent generations, more islands would pop up, as the effects of even small survival advantages accumulated.

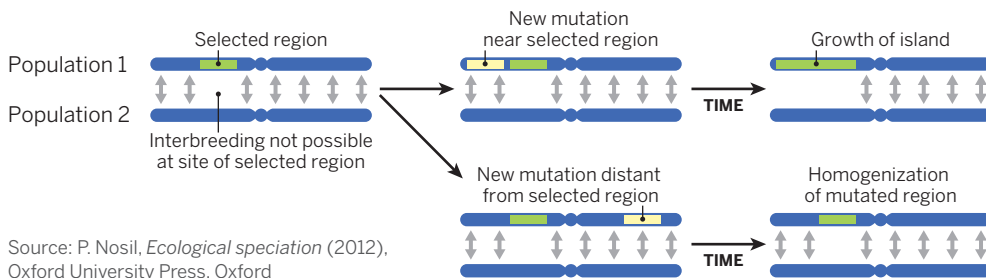


Islands everywhere? By comparing the DNA of divergent populations—such as prairie sunflowers (*top right*) adapting to dunes (*top left*) or stick insects matching different host plants (*middle*)—and closely related species, such as carrion crows (*bottom left*) with hooded crows (*bottom right*), researchers are exploring genomic islands of speciation.

PHOTOS: (TOP) JAMES HERNDON (2); (MIDDLE, LEFT TO RIGHT) MORITZ MUSCHICK; AARON CORNEAULT; (BOTTOM, LEFT TO RIGHT) JAN KIRK; PONA FOTKAS

How to make a species?

In theory, DNA differences between two populations that offer a survival advantage (green) can sometimes accumulate in genomic islands, though interbreeding tends to homogenize changes.



Source: P. Nosil, *Ecological speciation* (2012), Oxford University Press, Oxford

YET STORM CLOUDS QUICKLY gathered over the islands hypothesis. In 2009, Mohamed Noor and Sarah Bennett of Duke University in Durham, North Carolina, suggested in a review in *Heredity* that islands were “mirages in the desert.” They pointed out that divergent spots in the genome with high F_{ST} values might be left over from when a species had begun to split once before but stopped. Or they may have simply arisen because something about the DNA sequence or structure impeded gene flow at those spots, even though they don’t contain advantageous DNA.

Researchers looking for islands started getting different results depending on how they scanned their genomes or what combinations of species or races they studied. Feder started out a fan of the genomic islands but began to question the concept as he continued to study a classic example of ecological speciation, the apple maggot and hawthorn flies. In North America, most hawthorn flies lay their eggs in the fruit of the hawthorn tree, but about 150 years ago, a subset of these insects began to switch to apples, a tree newly introduced to the continent. Flies tend to lay eggs and look for mates on their natal fruit, so today there’s reduced interbreeding—and gene flow—between the two “races.” Eventually, they may speciate.

In 2003, Feder and others identified what seems to be a key genetic difference that may nudge speciation along—a very large stretch of DNA is oriented in opposite directions in the two fly races. This seemed to be a quintessential genomic island, as it includes genes important for adjusting the fly’s life cycle to the timing of fruiting in its host tree.

But when Feder and his colleagues applied more sophisticated tools to the DNA of hawthorn and apple maggot flies, they picked up dozens of other differences between the two races—so many that the group suggested that instead of islands, there were “continents,” large regions where selection was occurring, with some “mountain peaks” representing

DNA where selection was operating most strongly. Subsequently, other research groups found similarly widespread patterns of divergence among various populations of sticklebacks, whitefish, sunflowers, walking sticks, and butterflies. One group even revisited *A. gambiae*, the mosquito that spawned the island concept, doing sophisticated whole genome comparisons of the M and S forms, instead of just sampling a subset of the genomes as Hahn had done. This survey found many thousands of fixed differences between the two forms. In short, speciation seemed to arise across the whole genome from the very beginning, rather than from a few, small islands.

Last spring, Hahn turned against his own brainchild. He and his postdoc Tami Cruickshank analyzed genomic data from mosquitoes, house mice, European rabbits, the butterfly *Heliconius*, and birds called flycatchers, substituting a different measure of genetic divergence for F_{ST} . The result: The islands other teams had reported in those species vanished, Hahn and Cruickshank reported online in June in *Molecular Ecology*.

Yet other studies are shoring up the concept. Rieseberg and his postdoc Rose Andrew recently compared the DNA of sunflowers that normally grow on prairies with that of a population that has specialized to grow on dunes. The study, published in the September 2013 issue of *Evolution*, showed “gene flow was very important in creating large islands,” he says.

Jochen Wolf, an evolutionary biologist from Uppsala University in Sweden, and his colleagues have sequenced the genomes of two European crow species that can still successfully interbreed in certain parts of the continent. The carrion crow is all black, whereas the hooded crow has a gray body;

Wolf’s team found only 82 single-base differences between the two species—out of a possible 8.4 million examined, almost all the differences in one very large island. Wolf’s team reported in June that the island contains genes related to feather color and to visual perception—features that help preserve the two species by causing black crows to prefer black mates and hooded crows to prefer their kind (*Science*, 20 June, p. 1410).

Patrik Nosil of the University of Sheffield in the United Kingdom went further in stick insects, plant-eating insects that resemble twigs

or leaves, identifying genomic islands in different forms of a single species and showing that they are the handiwork of divergent selection. He and colleagues, including Feder, recently did a two-pronged study—sequencing wild populations of two stick insect forms found in southern California and then manipulating their environment experimentally to recreate the selective pressures they face living on different host plants.

The sequencing of wild populations revealed numerous, small islands, between 2000 and 5000 bases long, some shared by all the populations of one form or the other. In a field experiment, they collected the two forms of stick insects, then transplanted some of each onto the other’s host plant and the rest onto their natural home as a control

group. They collected the offspring a year later. By sequencing the transplants and their offspring, they could compare how natural selection had reshaped the genome in real time. The greatest shifts in the frequency of certain versions of a

gene occurred in the islands, they reported (*Science*, 16 May, p. 738). “We’ve shown that many genomic islands in these stick insects were created by natural selection [and] natural selection has a repeatable role in generating these islands,” Nosil says. “It removes some of the hand waving in the debate.”

Even though Hahn continues to look for islands, he and other skeptics are not convinced. But others say Nosil’s and Wolf’s results, and similar recent work, are keeping the idea of genomic islands afloat. “Things will become clearer as we get more sophisticated about the analysis and get more samples,” says Nicholas Barton, an evolutionary biologist at the University of Edinburgh in the United Kingdom. “But it will be a long time.” For now, fans and foes of genomic islands remain oceans apart. ■

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Loren Rieseberg, University of British Columbia, Vancouver