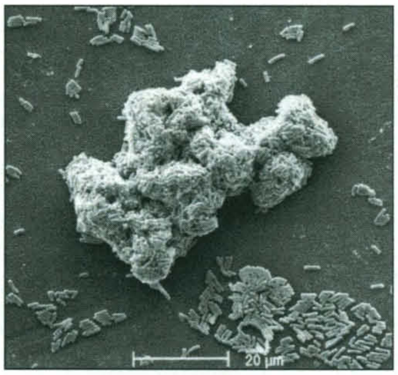
MICROBIOLOGY

A Tangled Tale of *E. coli* Virulence

Most people would do anything to avoid a bout of diarrhea. But along with 59 other brave souls at Stanford University, I lined up last year to drink a microbial cocktail the very thought of which seems guaranteed to turn the bowels to water. Sequestered in the clinical research center for 3 days, we anxiously awaited the first signs of the bacterial onslaught and clutched at our last remaining vestiges of dignity as nurses and scientists monitored the outcome. Our sacrifice for science—for which we were paid \$300—was not in vain: It turns out that our diarrheal responses revealed a subtle feature of the mechanism that helps explain the virulence of enteropathogenic *Escherichia coli* (EPEC), a common cause of diarrhea in children in developing countries.



BIEBER *ET AL.*

Togetherness. Mutant bacteria stick tightly together—perhaps too tightly.

On page 2114, Stanford microbiologist Gary Schoolnik and his colleagues confirm expectations that hairlike appendages on the surface of EPEC known as bundle-forming pili (BFP) are critical to the full virulence of these bacteria. The pili bundle together into ropelike filaments that interweave among bacteria, binding them into large aggregates. But the tests also suggest that another key to EPEC's virulence is the ability of the pili to disentangle themselves so the bacteria can go on to infect new intestinal cells. "The results are very surprising," says microbiologist Michael Donnenberg of the University of Maryland, Baltimore.

Schoolnik and members of his team have been studying EPEC's pili since they discovered them in 1991 (*Science*, 1 November 1991, p. 710). A strong hint that pili are important for virulence came when they and others mutated some of the 14 genes known to control pili formation and produced strains of bacteria lacking these appendages: The organisms couldn't attach to epithelial cells in the test tube or form bacterial aggregates. As the group now reports, the bugs proved relatively benign when fed to human volunteers. However, another mutant strain—dubbed the *bfpF* mutant—has produced more unexpected results.

At first glance the *bfpF* mutant seemed to be an overachiever. Schoolnik and Donnenberg independently showed that these bacteria aggregate into clusters and stick to epithelial cells in greater numbers than the wild-type EPEC. They also seemed to produce more pili and adhere more closely to human cells than their wild-type cousins. But when the Stanford group used time-lapse photomicrography to take a look at their behavior, they found a subtle difference: Wild-type bacteria form aggregates that disperse over time, but the *bfpF* mutants remained clumped together in a mass.

"We had a bet in the lab at that point," says Schoolnik. "Some bet that it [the F mutant] would have increased virulence, some bet that it would have no virulence." So they recruited more volunteers and returned to the clinical research center. The results were significant. Only four of 13 volunteers developed diarrhea, succumbing to doses of 2×10^{10} or 1×10^{11} mutant bacteria, compared to 11 of 13 in another set of volunteers who received doses of wild-type EPEC ranging from 5×10^{8} to 2×10^{10}. The EPEC mutant "does almost everything better in vitro, yet causes so much less disease when given to volunteers," says Donnenberg.

Schoolnik's team concludes that the *bfpF* mutant can infect and colonize the human gastrointestinal tract but fails to disperse, which severely reduces its power to cause diarrhea. "It all links in together very nicely and very logically," says Alan Phillips, a pediatric gastroenterologist from London's Royal Free Hospital. "If you can't aggregate and disaggregate, then you're not going to colonize very effectively."

Microbiologist James Kaper of the University of Maryland, Baltimore, agrees that the study provides "conclusive evidence that BFP are required for full virulence of this organism." But he is not yet convinced that the human tests prove that aggregate dispersal is critical for virulence. "The dispersal phase is a reasonable hypothesis," he says, but the mutant bacteria may colonize different sites in the intestine, or other unknown factors may contribute to reduced virulence.

Donnenberg agrees that human tests can't answer all the questions. "We consider the human model the gold standard, but there are big limitations," he says. "The volunteer is a big black box. We put bugs in one end and measure diarrhea out the other end, and what happens inside we really have no clue."

–Kristin Weidenbach

Kristin Weidenbach is a science writer in Boston. She received the highest dose of the bfpF *strain.*

ECOLOGY

El Niño Drives Spectacular Flower Show

TUCSON, ARIZONA—The serrated Tucson Mountains that rise above their namesake city are usually hospitable only to cacti and other hardy desert life-forms. This spring, however, the Tucsons' volcanic soil—along with much of the rest of the Southwestern desert—erupted in wildflowers, from golden poppies to velvet-red ocotillo and the sunflower blooms of brittlebush. Experts have proclaimed the display one of the desert's most dazzling blooms of the century.

The rainbow-hued outburst came courtesy of El Niño, the Pacific Ocean warm-up blamed for the rainstorms last winter that drenched much of the Southwest and triggered fluky weather elsewhere in the Americas. Although the capricious climate was a bane to many a mud-encrusted homeowner, it has been a boon to scientists hoping to learn more about how wildflowers survive in harsh climates—and why not every flower blooms in every rainy year.

Wonderland. Ocotillo *(inset)* and brittlebush helped fuel El Niño's floral fireworks.


PHOTOS: R. A. LOVETT

Good wildflower blooms hit the Southwest about once a decade, says botanist Mark Dimmitt of the Arizona-Sonora Desert Museum near Tucson. But the last banner year, he says—when "all you have to do is go out in the desert and see flowers everywhere"—was 1979. Nineteen relatively flower-poor years followed, until last winter's El Niño, combined with a late-September hurricane that swept inland from the Baja Peninsula.

Throughout the winter and early spring, El Niño–driven storms dumped two to four times the normal rainfall in the Arizona-California desert, where precipitation ranges from 7 to 30 centimeters a year. The rule of thumb, Dimmitt says, is that fall rains and cool, wet winters—the classic El Niño pattern—favor lupines, daisies, and other winter-germinating annuals. But total moisture is "less than half the story," he adds. During some wet years, Dimmitt says, grasses—not flowers—come out in force, along with other plants that paint the desert green but neglect the rest of the palette. Even in this year's prime wildflower conditions, some desert pockets responded only modestly.

One reason for this pattern, says Dimmitt, is that the flowers engage in "temporal niche separation," in which species occupy the same tract by each thriving in years with different climates. In 1979, for instance, owl clover reigned. But this year the fuzzy purple flower "was almost nonexistent," he says, perhaps because early rains last fall favored other species. Dimmitt is compiling photos of past blooms to compare with weather data to better understand which wildflowers thrive under which conditions.

Another reason why a wet winter might not draw a full response from some flowers is "bet hedging," says University of Arizona, Tucson, ecologist D. Lawrence Venable. He compares the tactic to investors who avoid buying a single stock: They might make a heap of money, or they might go broke. The same can happen to a flower population that bets everything on a wet year—in this case, going broke means extinction. That's because sprouts that germinate after the first rainfall stand a risk of getting parched in a later drought.

Bet-hedging models predict that the drier the environment, the more important it is to a flower population's survival that only a small fraction of its seeds germinate in any given year. A just-completed study on Indian wheat seeds suggests that prediction may not hold in Arizona, says Venable grad student Maria Clauss. After receiving comparable levels of precipitation, seeds from scorching southwestern Arizona actually appeared to have higher germination fractions than seeds from regions near Tucson that usually get up to three times as much rain. Clauss thinks she knows why: In the harsher desert areas, many winters bring no measurable rain, and then El Niño rolls in with repeated drenchings. This means that if it rains at all, it's likely to rain a lot, and the best survival strategy may be for seeds to bet heavily on El Niño: Germinate early and in large numbers.

Still, just how temperature, rainfall timing and amount, and plants' own strategies interact to cue an El Niño bloom remains a mystery. "When it starts to rain, nobody can predict if it's going to be a good wildflower year," says Dimmitt. "I don't make predictions."

–Richard A. Lovett

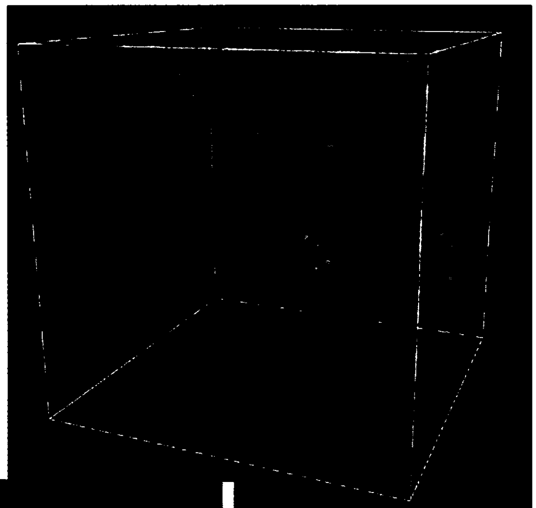
Richard A. Lovett is a writer in Portland, Oregon.

COSMOLOGY

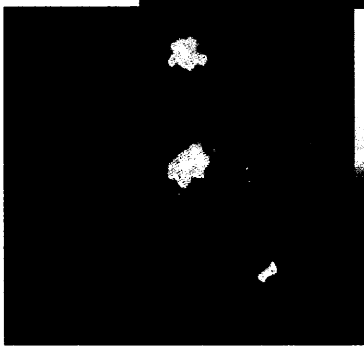
Cosmic Web Captures Lost Matter

SAN DIEGO—Like the parents of acquisitive toddlers, astronomers have gotten used to losing sight of items they know must be around somewhere. At least 90% of the matter in the universe, for instance, must be in some exotic "dark" form that has not yet been seen directly. More irritating to some astronomers is that they cannot even see most of the universe's ordinary, or baryonic, matter. Billions of years ago—in the era visible in the most distant reaches of space—the missing baryons (protons and neutrons) formed sprawling complexes of hydrogen clouds. Then they vanished.

Or did they? In a talk at an American Astronomical Society meeting here earlier this month, Jeremiah P. Ostriker, a cosmologist who is also provost of Princeton University, predicted that observers will soon find most of the ordinary matter in the universe right under their noses—resting on the kitchen table, so to speak. Computer simulations by Ostriker and Princeton's Renyue Cen show that the primordial clouds condensed over time into a vast, filamentary network of fully ionized gas, or plasma—a cosmic cobweb that now links galaxies and galaxy clusters.



R. CEN AND J. P. OSTRIKER



O. D. WANG

Gravity's gossamer. A simulation shows how primordial clouds might have collapsed into a plasma web over hundreds of millions of light-years; an x-ray observation *(left)* may show a hot spot in the network.

In the process, the simulations say, the plasma heated up to about a million degrees kelvin, so it now gives off a faint x-ray glow that is extremely difficult to detect with present satellites. If Cen and Ostriker are right, "most of the baryons could live in this warm intergalactic medium and be nearly invisible," says August Evrard, a cosmologist at the University of Michigan, Ann Arbor. Soon-to-be-launched satellites, like NASA's Advanced X-ray Astrophysics Facility (AXAF), ought to be able to see the plasma, said Ostriker: These missions "have a fair chance of finding most of the baryons in the universe." And at the end of Ostriker's talk, one x-ray observer announced that he may already have seen a hint of these hot filaments.

Cen and Ostriker's simulations take their lead from what has become the standard picture of structure formation in the universe. In this view, the dark matter's gravity slowly amplifies slight ripples in the primordial universe. As the waves steepen, the baryons collect like froth at the top of the waves, where the density is greatest.

That froth gave rise to filaments and clusters of galaxies and, when the universe was less than half its present age of roughly 13 billion years, to vast gas clouds. Neutral—non-ionized—hydrogen in the clouds absorbed the light of even more distant beacons called quasars, making the gas visible to observers today. The densely packed spikes of absorption in the quasars' spectra give these distant clouds their name—the Lyman-α forest—and let observers gauge how much baryonic matter they contained.

Astronomers can then compare the mass of the clouds with the total amount of baryonic matter that must have emerged from the big bang, which can be gauged from chemical tracers such as the amount of deuterium that survived the explosion (*Science*, 7 June 1996, p. 1429; 10 January 1997, p. 158). Although the comparison is fraught with uncertainties, the two numbers suggest that 80% to 90% of the baryons made in the big bang can be found in the ancient forest. "We have it tied pretty solid," says Michael Rauch of the California Institute of Technology in Pasadena.

In the billions of years since then, some of the gas has been consumed by star formation and sucked into the cores of great clusters of galaxies, where it is heated to hundreds of millions of degrees and gives off bright, easily detected x-rays. But all of those reservoirs account for less than half of the original bary-