MOLECULAR BIOLOGY

New Link Found Between *p53* and DNA Repair

In order for an organism to keep functioning, protecting the integrity of its genetic material is of utmost importance. The reason is simple: Accumulation of damage to the DNA can lead to harm, including cancer. To help prevent such damage, cells have evolved elaborate machinery for repairing the DNA after it is damaged (*Science*, 4 November, p. 728). The cell even has its own genomic "guardian angel," the *p53* tumor suppressor gene. Its job: temporarily halting the cycle of cell division in response to damaging insults to allow time for the DNA to be repaired before being copied. In some cases, *p53* may actually cause damaged cells to self-destruct before their damaged genetic goods can be handed down to progeny cells.

The importance of *p53* as a genetic guardian has been known for some time, but new evidence, including a report on page 1376, suggests that this gene may have even more ways of protecting the genome than was thought. In addition to blocking cell cycle progression and helping trigger programmed cell death, *p53* may directly and indirectly stimulate the DNA repair machinery. Indeed, molecular biologist Albert Fornace Jr. of the National Cancer Institute (NCI), who led the team reporting its results in this issue, says of *p53*'s many functions: "It's just amazing that evolution would put so many eggs in one basket." One of the downsides of having that many eggs in one container, however, is that if *p53* itself breaks down, the cellular machinery can go badly awry: Mutations in the gene contribute to as many as 50% of all human cancers.

The route by which Fornace and his colleagues, including Michael Kastan of the Johns Hopkins Oncology Center in Baltimore and Tona Gilmer at Glaxo Research Institute in Research Triangle Park, North Carolina, linked *p53* to the DNA repair machinery began with another gene, *GADD45*. The Fornace group has found that *GADD45* is turned on in cells exposed to stresses that stop cell growth and cause DNA damage. Thus the name *GADD*, for growth-arrest-and-DNA-damage-inducible.

Among the stresses that switch on *GADD45* is exposure to ionizing radiation, which is also a potent stimulator of the activities of the protein produced by *p53*. Knowing that, Fornace and his colleagues checked to see whether the p53 protein might turn on *GADD45*. There was reason to believe that it might, because some of p53's effects are due to the fact that it is a transcription factor—a protein that regulates the activity of other genes. It blocks the cell cycle, for example, presumably by stimulating activity of a gene variously called *p21*, *Waf1*, *Cip1*, or *Sdi1*, which makes a protein that inhibits the cyclin-dependent protein kinases (CDKs), key enzymes needed for cell cycle progression. The Fornace team found that their hunch was correct: The cell's ability to turn *GADD45* on in response to ionizing radiation depends on the presence of the normal *p53* gene, whose protein product stimulates production of the Gadd45 protein in much the same way that it stimulates p21 synthesis.

To find out what the Gadd45 protein itself does, the researchers went looking for other proteins with which it might associate in the cell. And there they hit pay dirt. They found Gadd45 in complexes with proliferating cell nuclear antigen (PCNA). Finding it there provided some clues to what Gadd45 is doing, because PCNA has two important functions in the cell. In the late 1980s, Bruce Stillman's group at Cold Spring Harbor Laboratory on Long Island showed that PCNA is a necessary component of the machinery that copies DNA so that cell division can take place. And 2 years ago, Richard Wood's group at the Imperial Cancer Research Fund Clare Hall Laboratories in South Mimms, U.K., showed that PCNA is also needed for the resynthesis of DNA after damaged portions are removed by the cell's nucleotide excision repair system, which remedies damage caused by environmental insults. (The fact that the protein is needed for DNA repair as well as replication reflects the fact that the same enzymes carry out both activities.)

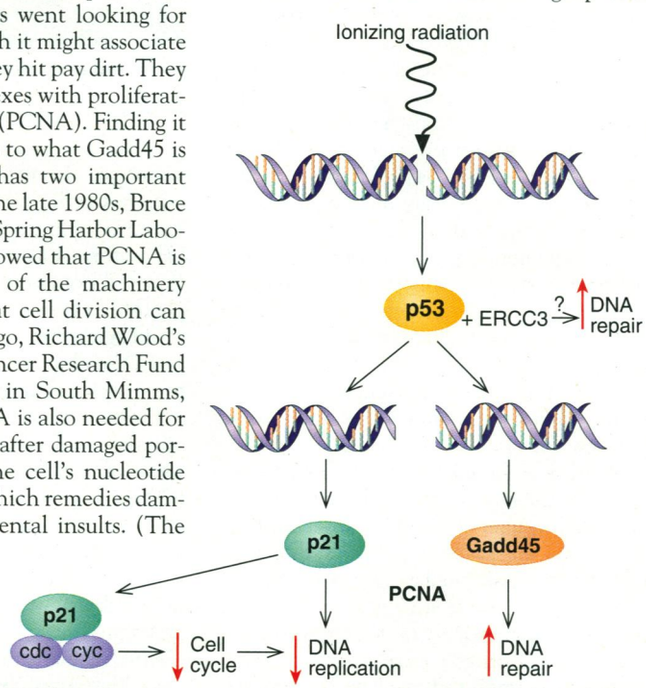
With this clue to a possible Gadd45 function, Fornace and his colleagues went on to ask whether this protein stimulates excision repair. And the answer was yes. "If we removed Gadd45, repair decreased," Fornace says, "and if we added it, repair increased." Because Gadd45 production in the cell is stimulated by *p53*, he adds, "the take-home message is that there's another role for *p53* in protecting the DNA from the onslaught of damage. It further strengthens the 'guardian of the genome' hypothesis."

Bert Vogelstein, also of Johns Hopkins, a *p53* expert, describes the finding as "intriguing. It will stimulate the community that studies DNA repair." He adds, however, that "how it all fits together is still unclear." Fornace agrees, noting that it's not yet known just how Gadd45's interaction with PCNA influences DNA repair, although it presumably affects the resynthesis step.

In addition to the finding that *p53* may indirectly stimulate DNA repair through Gadd45, there are also hints that it may play a more direct role. Earlier this year, Curt Harris's group at NCI found that the p53 protein also binds to a protein called ERCC3, one of several excision repair molecules that together recognize and remove damaged segments from DNA. The work is at a very early stage, with little known about how p53 binding to ERCC3 might affect repair, but Harris suggests that the tumor suppressor protein may be involved in the early stages of damage recognition as well as in the later stages of resynthesis. Because ERCC3 is part of a transcription factor, it might also be involved in p53's gene control activities.

While recent work is turning up some intriguing new ways that *p53* protects the genome, other research is providing additional insight into how p21, the other protein whose production *p53* is known to stimulate, carries out its activity of shutting down DNA replication so that repair can occur. One way p21 apparently does this is by inhibiting the CDKs, thereby halting the cell cycle before DNA synthesis takes place.

But Stillman's group, working with that of David Beach, also at Cold Spring Harbor, and another team, including Jerard Hurwitz of the Memorial Sloan-Kettering Cancer Center in New York City and Stephen



Center of interest. By stimulating synthesis of the proteins p21 and Gadd45, p53 may both halt DNA replication and stimulate DNA repair.

Elledge of Baylor College of Medicine in Houston, have recently shown that p21 also directly inhibits the DNA-replicating machinery by binding to its PCNA component. "p21 is doing something more than inhibiting the cell cycle," Stillman says.

DNA repair can still proceed after p21 binds to PCNA, the Cold Spring Harbor team finds, even though repair and replication are carried out by the same set of proteins. What apparently happens, Stillman explains, is that p21 prevents synthesis of long stretches of DNA by preventing the enzyme complex from sliding along the DNA, but it does not stop the synthesis of the shorter segments that must be remade for repair. The upshot is that p21 may allow DNA repair to be coordinated with replication even after replication gets under way, instead of just stopping it beforehand.

The picture growing out of all this is that *p53*, working through p21, may halt DNA synthesis both directly and indirectly, thereby allowing time for repair to take place. And then p53, working through Gadd45 and perhaps on its own as well, can stimulate the repair machinery. Still, a great many gaps in the picture remain to be filled. But given that both *p53* and DNA repair are among the hottest topics going these days, there will be plenty of interest in tackling the remaining questions.

–Jean Marx

PLANETARY SCIENCE

Baring the Secrets of Asteroid Ida

BETHESDA, MARYLAND—When planetary scientists poring over Galileo spacecraft images of Ida discovered last spring that this small asteroid has an even smaller moon, they got a big break. Dactyl, as the moon is called, is turning out to be a tattletale, spilling the intimate secrets of its mysterious companion. The latest of these secrets—a clue to Ida's mass and an inkling that its reddish tint might be only skin-deep—hint that Ida might be made of primordial solar system material, the same primitive stuff that falls to Earth in the stony meteorites called chondrites. If so, astronomers struggling to understand the class of asteroids to which Ida belongs, the S type, may once again have some rethinking to do.

Many planetary scientists once assumed that S types, the commonest kind of asteroid, had to be the source of the chondrites, implying that these asteroids are simply chondrites writ large: lumps of primitive solar system material. But recent evidence, including Galileo images showing color differences between Ida and Dactyl, suggests that S types are too varied for all of them to be chunks of the same primordial stuff. Instead, many astronomers concluded that most or all S types are made of material that was heated and transformed early in solar system history, separating it into rock and metal. Chondrite meteorites, in that case, must come from another source—perhaps from asteroids too small to have been recognized yet as chondritic.

But the new clues about Ida may cause the pendulum to swing at least partway back toward the idea that S-type asteroids can indeed be made of primitive, undifferentiated stuff. "I'm leaning pretty strongly away from differentiation," says asteroid specialist Clark Chapman of the Planetary Science Institute in Tucson, Arizona, who argued at the annual meeting of the American Astronomical Society's Division for Planetary Sciences (DPS) here in suburban Washington, D.C., that Ida and at least some other S types are primitive. And many researchers who learned of the new evidence think it reinvigorates arguments that at least some S types are

JPL/NASA

A red, red asteroid. An extreme false-color rendition of Galileo data emphasizes the redness of Ida relative to its moon Dactyl.

primitive, although they're not sure Chapman has really proved the case for Ida. As Torrence Johnson of the Jet Propulsion Laboratory in Pasadena, California, puts it, "I'm still not convinced that if we had samples of Ida and Dactyl in front of us that we could agree they were chondrites."

In the absence of samples, Chapman has to rely on such indirect evidence as Dactyl's orbit around Ida, at a distance of about 100 kilometers. Because Dactyl is so small, just 1.4 kilometers in diameter compared to the 60-kilometer length of Ida, its orbit depends almost entirely on Ida's mass, and therefore on Ida's density. The latest measurements of Dactyl's orbital motion imply, Chapman noted, that Ida's density must be between 2 and 3 grams per cubic centimeter. Any less, Galileo team member Michael Belton of Kitt Peak National Observatory reported at the meeting, and Dactyl could not actually be in orbit, only caught by chance as it flew by, a highly unlikely possibility. Any more, Jean-Marc Petit of Nice Observatory and colleagues at the University of Arizona found, and Dactyl should long ago have spiraled into a catastrophic collision with Ida.

Chapman uses this density range of 2 to 3 grams per cubic centimeter to rule out one composition for Ida: that of the highly differentiated meteorites of mixed rock and metal called stony irons. The less dense ordinary chondrites, though, fit the density range nicely, Chapman notes.

At the same time, he argues that the color difference between Ida and Dactyl no longer requires a differentiated composition. When Galileo's sensitive instruments first revealed that Ida's spectrum is a bit "redder" than Dactyl's (although both would look gray to the human eye), even Chapman took that subtle difference as a possible sign that Ida is differentiated (*Science*, 17 June, p. 1667). But when he looked at the latest calibrated version of the Galileo spectral data, which came out the week before the DPS meeting, he concluded that the similarities are more telling than the differences: "The color differences are significant, but they don't require substantial differences in mineralogy."

Instead, says Chapman, the color differences look like the product of space weathering, a poorly understood process in which micrometeorite impacts or some other agent reddens exposed surfaces. Because larger bodies, with their stronger gravity, would have been pummeled by higher speed impactors, Ida should have suffered more extensive weathering than Dactyl, which would explain the color difference. Ida, says Chapman, looks like an ordinary chondrite hiding in a red cloak; he thinks weathering may have also disguised other S types.

Most other researchers, however, believe that the case is far from closed. For one thing, as Jeffrey Bell of the University of Hawaii points out, Ida's relatively low density may rule out some differentiated compositions, but it could still be made of the lower density rock from a larger body that had differentiated into rocky and metallic parts. And Richard Binzel of the Massachusetts Institute of Technology adds that even if space weathering has disguised Ida, its true identity should be apparent in material freshly exposed on its surface by recent impacts. This fresh impact debris, notes Binzel, doesn't match the color of an ordinary chondrite—although Chapman responds that it may not be fresh enough to have escaped space weathering entirely.

Still, the hints that, among S types, appearances can be deceiving strengthens the possibility that at least a few of them could be primitive, Johnson says. Ida is still in the running, but researchers want to learn a few more of its intimate secrets before deciding.

–Richard A. Kerr