

Baring HIV's Dependencies

New atlas reveals that HIV commandeers almost 300 human proteins to do its dirty work.

VIRUSES ARE NEEDY. EQUIPPED WITH FEW GENES, THEY LEAN HEAVILY ON their host cells to help them successfully invade. ¶ This is true of the human immunodeficiency virus (HIV), which has just nine genes that make only 15 proteins. With such a sparse molecular tool kit, it is a wonder that HIV can wreak such havoc. But its ability to take over the very immune cells intended to protect us from disease is well known. An atlas of those host cell factors the virus hijacks would provide a deeper understanding of the virus, perhaps providing potential ways to thwart it.

In January, a team led by HHMI investigator Stephen Elledge of Brigham and Women's Hospital in Boston produced just such a roadmap. Writing in the journal *Science*, Elledge's team reported that HIV requires at least 273 human proteins, called HIV-dependency factors (HDFs), to do its molecular dirty work.

"This is a tremendous resource for the entire field of HIV research," says Dan R. Littman, HHMI investigator and an HIV expert at New York University Medical Center. "It's been known for a long time that these host factors were out there, but there had never been a systematic approach to identify them. I don't think

anyone could have imagined how many would turn up."

The study greatly expands the number of known HDFs, painting a newly detailed portrait of the virus and its dependencies. Only 36 of the human proteins commandeered by HIV had been previously identified.

To produce the expanded catalog of HDFs, Elledge's group, which included postdoctoral fellow Abraham Brass and Harvard Medical School's Judy Lieberman, tapped newly available commercial libraries of what are known as small interfering RNAs. These genetic molecules can switch genes off, preventing them from making proteins.

FINDING CANCER TARGETS

IT'S NOT JUST HIV THAT STEVE ELLEDGE is probing for weaknesses. He's taken on cancer too. In a paper published in *Science* on February 1, 2008, Elledge and his collaborator, HHMI investigator Gregory J. Hannon of Cold Spring Harbor Laboratory, revealed a new screening technique to probe tumors for genes that help them thrive. **ELLEDGE AND HIS COLLEAGUES GENERATED** about 8,000 bits of short hairpin RNAs (shRNA)—single strands of RNA that fold back on themselves—that can be inserted into retroviruses. When the altered retroviruses infect either normal or cancerous cells, the shRNA binds to corresponding stretches of RNA in the cells, preventing their translation into proteins. **IF THE shRNA KNOCKS DOWN** production of a protein essential to keeping the cells alive, then the abundance of that particular shRNA quickly diminishes as cells die. If the shRNA corresponds to a gene involved in dampening cell growth, then the cells that carry that shRNA will

multiply and thrive. **BY TRACKING THE ABUNDANCE** of each shRNA from the total pool and comparing breast and colon cancers with normal tissues, Elledge and his colleagues were able to identify genes critical to tumors' growth. **"THE OVERALL IDEA BEHIND** this is that cancer cells reprogram their [molecular] networks," says Elledge. "We're interested in finding what components in these new networks are controlling proliferation." **WHILE HIS STUDIES ON HIV** rely on a slightly different method, in both HIV and cancer Elledge hopes that full-genome scanning will reveal new target proteins for drugs. **"HIV IS A LOT LIKE A CANCER CELL—**cancer cells also mutate, so it's hard to get drugs to them that kill them," he says. Both HIV and cancer are so complicated that selecting genes one at a time to test for importance would likely miss other vital ones, he says. "This is a way to take the guesswork out, because we're testing everything."
— Sarah C. P. Williams

By turning off genes in human cells one by one and then observing whether HIV could establish itself and reproduce, Elledge's team plodded through 21,000 disrupted genes to isolate those the virus required.

"We were looking for any genes that HIV needs for its life cycle," Elledge says, pointing out that the proteins those genes make have the potential to be drug targets.

Drugs now in use directly attack HIV, and they must be used in combination since the virus has evolved resistance to individual compounds, explains Elledge. Making drugs that target host proteins could bypass resistance; if a host protein the virus requires is disrupted, the virus would have to do much more to overcome the challenge than simply rearrange a few amino acids of genetic material. The new study reveals that many host proteins play coopted roles throughout the cycle of HIV infection—for example, helping the virus glom onto and enter cells, converting RNA to DNA, and creating new infectious particles.

"Many of the proteins identified in this study would probably be good candidates for screening to find new anti-HIV drugs," notes David Baltimore, a leading HIV authority at the California Institute of Technology, who agrees that the virus will have a harder time developing resis-

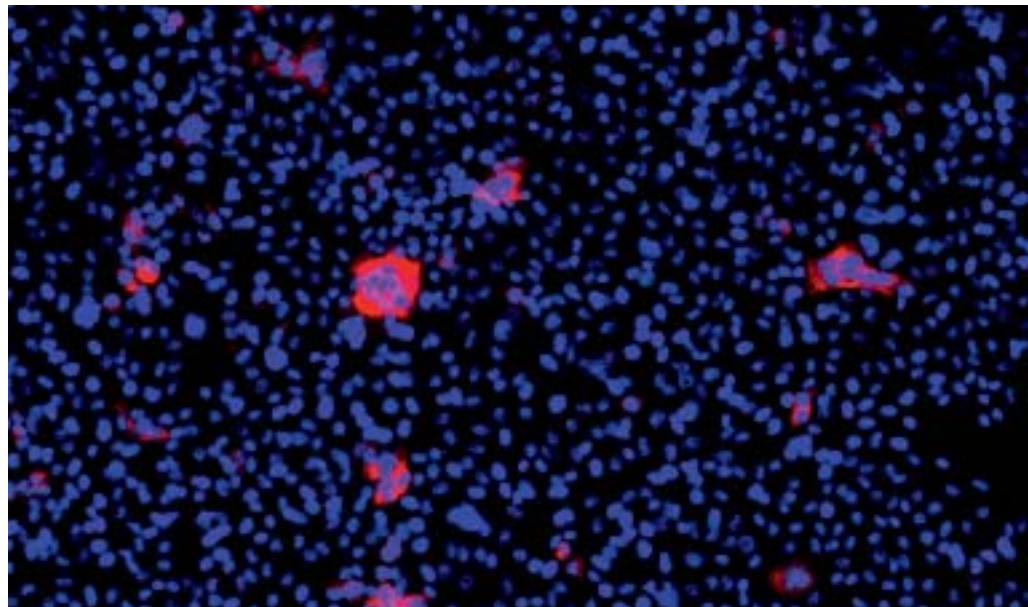
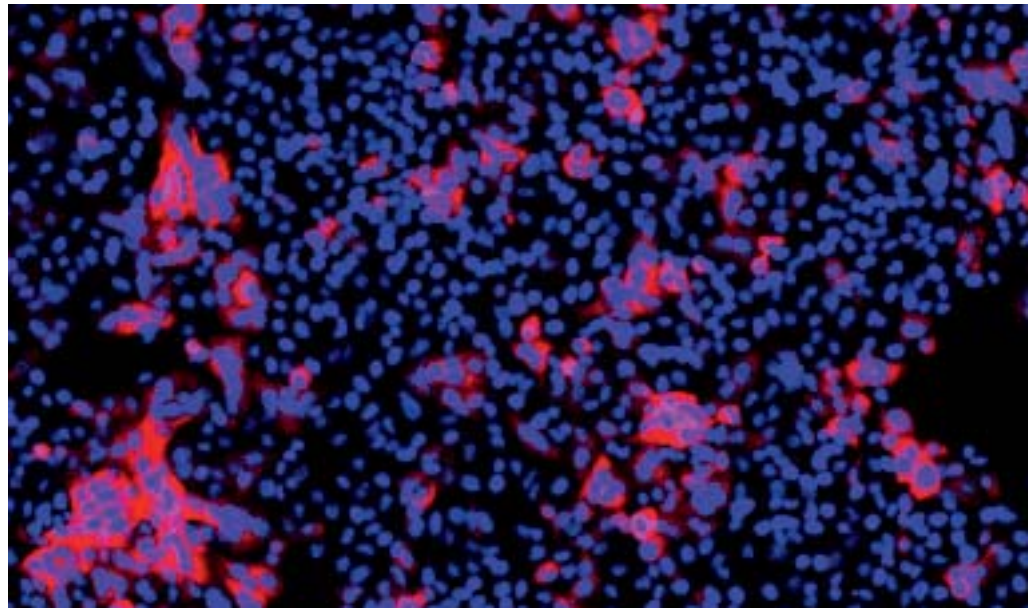
tance to drugs that target cellular proteins. "However," he adds, "for the same reason, the drugs will have to be carefully characterized for toxicity."

Elledge acknowledges that starving viruses of required host proteins could have unintended effects. "The cells do need the proteins," he says. "That needs to be worked out."

For Elledge, who is best known for his DNA cell cycle work, this new study is a

first, if dramatic, foray into HIV biology. His lab also has ongoing gene discovery projects focused on other viral pathogens, as well as cancer, stem cells, and diabetes. He plunged into HIV to spur HIV drug development by industry, which he says is lagging: "I wanted to point out using genetics that there are real targets in cells and get [drug developers] thinking about mining those pathways." ■

—TERRY DEVITT



Among the host cell proteins used by HIV when the virus infects cells is one called human transportin 3. When scientists halted production of that protein in host cells (blue), levels of HIV (red) in the cells were drastically reduced (lower panel) compared to infection under normal conditions (upper panel).