Researchers develop new method for screening drug-resistant forms of HIV

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A growing number of drug-resistant strains of HIV are a threat to the effectiveness of current treatments despite anti-HIV drug cocktails decreasing the number of HIV-related deaths and improving the quality of life for HIV patients. Existing methods of detecting drug-resistant forms of HIV are expensive, time consuming, and often fail to identify small populations of drug-resistant HIV.

Now, researchers at the University of Pennsylvania School of Medicine have developed a drug resistance screening method that analyzes multiple HIV variants at the same time, while also saving time and money.

By combining two genetic tests, Frederic D. Bushman, PhD, Professor of Microbiology, and colleagues, rapidly obtained gene sequences from multiple drug-resistant HIV samples at once. The study appeared online this month in Nucleic Acids Research.

“There is considerable interest in identifying minor drug resistant variants prior to initiating new therapy, in order to allow treatment with the most effective drugs,” explains Bushman. “Treatment of HIV infection often fails because viruses mutate to resist drugs. Under the pressure of drug treatment, small populations of resistant viruses can quickly grow to become the majority, resulting in treatment failure due to drug resistance.”

According to the Joint United Nations Programme on HIV/AIDS

(UNAIDS), approximately 40 million people in the world are currently living with HIV/AIDS. Commonly prescribed antiretroviral cocktails work to slow the debilitating effects of HIV by disrupting the virus at various stages in its replication. While combinations of antiretroviral drugs have proven effective, quickly mutating forms of HIV can complicate treatment outcomes.

Researchers estimate that up to 50 percent of individuals being treated for HIV in the US carry drug-resistant forms of HIV, caused by mutations in the virus in response to drug treatment or by being infected with a resistant form of HIV. The increased availability of antiretroviral drugs to meet the HIV/AIDS needs of developing countries in recent years will likely contribute to a global rise in drug-resistant strains of HIV.

“To overcome drug resistance, patients must be treated with drugs to which the HIV virus is still susceptible,” says Bushman.

To tailor the most effective anti-HIV treatment for a patient, antiretroviral resistance screenings are conducted before a patient begins or changes drug therapy. In developing countries where HIV/AIDS is most prevalent, resistance testing is rare due to the high costs of screening. In cases where resistance testing is available, most screening techniques are not sensitive enough to analyze small populations of drug-resistant strains of HIV. While small populations of drug-resistant HIV may go undetected by current screening methods, Bushman says that minor mutant forms of HIV often impair a patient’s response to future drug therapy.

To increase the sensitivity of HIV screening techniques and decrease the time and cost of each test, Bushman and others examined seven samples of mutated strains of HIV, including three HIV samples from patients who had experienced antiretroviral multi-drug resistance. DNA bar

coding, in which different DNA molecules are indexed using DNA sequence tags, allowed Bushman and others to map multiple sequences of HIV mutants simultaneously. Drug-resistant mutations were identified using a new DNA sequencing technique called pyrosequencing, which allows researchers to determine millions of bases of a DNA sequence in a single one-day experiment. By combining the two methods, researchers were able to quantify and characterize hundreds of thousands of HIV variants for drug resistance in a single test.

Not only did the parallel analysis help researchers to cut time, the new screening technique also uncovered four rare, minor drug-resistant mutations in the patient samples of HIV that had gone undetected by standard screening measures. These small drug-resistant populations of HIV may explain why some patients do not respond to antiretroviral treatment, because the minor alleles can rapidly grow out, generating new populations of drug-resistant viruses.

Bushman's new screening technique may open up opportunities for improved drug-resistance screening in the United States and around the world. By rapidly gathering sequencing information about drug-resistant HIV, decoding resistance in HIV and other viruses can potentially be done at a fraction of current costs and time.

In the future, Bushman plans to apply his new method to optimize drug-resistance testing in the US and the developing world.

"Thanks to DNA bar coding and pyrosequencing, clinicians should be able to optimize treatment for their patients with much more relevant information in front of them," says Bushman.

Source: University of Pennsylvania School of Medicine