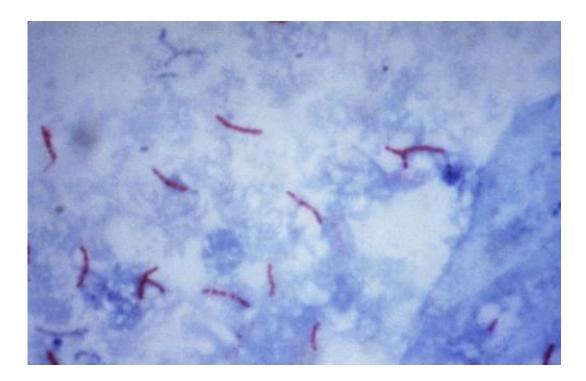


Blood test could transform tuberculosis diagnosis, treatment in developing countries

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This photomicrograph reveals Mycobacterium tuberculosis bacteria using acidfast Ziehl-Neelsen stain; Magnified 1000 X. The acid-fast stains depend on the ability of mycobacteria to retain dye when treated with mineral acid or an acidalcohol solution such as the Ziehl-Neelsen, or the Kinyoun stains that are carbolfuchsin methods specific for M. tuberculosis. Credit: public domain

A simple blood test that can accurately diagnose active tuberculosis could make it easier and cheaper to control a disease that kills 1.5 million people every year.



Researchers at the Stanford University School of Medicine have identified a gene expression "signature" that distinguishes patients with active tuberculosis from those with either latent tuberculosis or other diseases.

The technology fills a need identified by the World Health Organization, which in 2014 challenged researchers to develop better diagnostic tests for active TB.

A paper describing the work will be published online in *Lancet Respiratory Medicine* on Feb. 19.

Globally, tuberculosis infects 9.6 million new patients each year and kills 1.5 million. Yet the disease remains difficult to diagnose. "One-third of the world's population is currently infected with TB. Even if only 10 percent of them get active TB, that's still 3 percent of the world's population—240 million people," said Purvesh Khatri, PhD, assistant professor of medicine and senior author of the paper.

Traditional diagnostic methods, such as the skin prick test and interferon assays, can't separate patients with active TB from those who are no longer sick or have merely been vaccinated against TB (and most countries vaccinate everyone against TB). These older diagnostics can miss a case of TB in patients with HIV.

A sensitive test

A common way to test for TB is to look for the disease-causing bacterium in sputum samples coughed up by patients. But sometimes it's hard for people to produce sputum on demand, said research associate Tim Sweeney, MD, PhD, first author of the paper. "If someone can't produce adequate sputum, or if you have a kid who can't follow directions," it's hard to diagnose them, he said. And the sputum test is



almost useless for monitoring how someone is responding to treatment. As people start to get better, they can't produce sputum for the test.

The new test developed in the Khatri lab works on an ordinary blood sample and removes the need to collect sputum. It can signal a TB infection even if the individual also has HIV. And it won't give a positive response if someone only has latent TB or has had a TB vaccine. It also doesn't matter which strain of TB has infected a person, or even if it has evolved resistance to antibiotic drugs. The test works in both adults and children.

WHO has called for a test that would give a positive result at least 66 percent of the time when a child has active TB. The Khatri test is 86 percent sensitive in children. And if the test comes up negative, it's right 99 percent of the time. That is, of 100 patients who test negative with the Khatri test, 99 do not have active TB.

The requirements of the test are simple enough that it can potentially be done under relatively basic field conditions in rural and undeveloped areas of the world. Any hospital should be able to perform the test. Villages without electricity could likely use ordinary blood samples and a solar-powered PCR machine, which multiplies strands of DNA, to accurately test people for active TB.

Chain reaction

When pathogens infect the cells of the body, the infection sets off a chain reaction that changes the expression of hundreds of human genes. Khatri's team identified three human genes whose expression changes in a consistent pattern, revealing the presence of an active tuberculosis infection.

The team validated the new three-gene test in a separate set of 1,400



human samples from 11 different data sets, confirming the diagnostic power of the test.

The new test not only accurately distinguishes patients who have <u>active</u> <u>tuberculosis</u>, it could also be used to monitor patients to see if they are getting better and how well they are responding to different treatments. Thus, it can be used not only for diagnosis and to inform treatment, but also to study the effectiveness of different treatments. The test's hugely accurate negative response would be especially helpful in monitoring the effectiveness of treatments during clinical trials, said Khatri.

He has already begun collecting funding to develop the <u>test</u> for widespread use, both to diagnose TB in <u>patients</u> and to monitor recovery in clinical trials, allowing for more rapid development of better and cheaper treatments.

The work is an example of Stanford Medicine's focus on precision health, the goal of which is to anticipate and prevent disease in the healthy and precisely diagnose and treat disease in the ill.

Provided by Stanford University Medical Center

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