New test shows promise in identifying new drugs to treat Lyme disease

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Researchers from the Johns Hopkins Bloomberg School of Public Health have developed a test they say will allow them to test thousands of FDA-approved drugs to see if they will work against the bacteria that causes tick-borne Lyme disease.

The researchers, reporting Nov. 3 in the journal *PLOS ONE*, say doctors and patients are desperate for new treatments for Lyme disease, which in many people is cleared up with a few weeks of antibiotics but, in some, lingers long after completion of the standard drug regimen. Until now, it has been very difficult to determine on a large scale which drugs work against these lingering *Borrelia burgdorferi* – the bacteria that cause Lyme disease – hampering new drug discovery for the organism.

Study leader Ying Zhang, MD, PhD, a professor in the Bloomberg School's Department of Molecular Microbiology and Immunology, and his colleagues tweaked a test typically used for simply counting DNA in samples in the lab. Using the test, they were able to quantify how many *Borrelia burgdorferi* are alive and how many are dead after each drug was added to the bacteria. The method stains the living bacteria green and the dead or dying bacteria red in a way that filters out the noise that can corrupt existing tests.

"It's superior to the current gold standard for testing *Borrelia* viability," Zhang says. "This could become be the new gold standard."

The most exciting part of the development of the test, Zhang says, is that his team has already used it to identify a series of antibiotics approved to treat other infections that show promise in the lab against the lingering *Borrelia burgdorferi* bacteria, known as persisters. In a study published in July in the journal *Emerging Microbes and Infections*, Zhang and colleagues used the new test – called the SYBR Green I/PI assay – to identify several antibiotics that showed promise against the

persistent bacteria that appear immune to the current Lyme antibiotics. That paper has been the most popular on the journal's website, and patients, doctors and researchers have been contacting Zhang interested in testing out the most promising of the newly identified drugs.

The new test enables researchers to analyze thousands of drugs at a time because its use can directly measure the viability of the bacteria after drug treatment and doesn't require each bacteria sample to be washed, as the currently used LIVE/DEAD BacLight assay does. In skipping that step, the new test is not only more accurate but also more efficient, is less expensive and takes less time, Zhang says.

Lyme disease is named after the town in Connecticut where it was first recognized in 1975. It is found from Maine to Virginia along the East Coast of the United States, peaking in June and July. The tick causes a skin reaction that resembles a bulls-eye or spider bite. If left untreated, Lyme disease may lead to systemic problems such as neurologic and arthritis symptoms weeks or months later. The antibiotic doxycycline or amoxicillin is considered the most effective treatment in resolving early signs of infection and preventing the majority of later symptoms but is ineffective for late-stage disease.

While most people with the disease are fine after a short course of antibiotics, a large subset of patients – some estimates say as high as 20 percent or more – suffer long-term symptoms including fatigue, musculoskeletal pain and memory problems. Their precise diagnosis has long been a controversial topic in medical circles. Some doctors believe that very few people experience persistent Lyme-related symptoms, known as post-treatment Lyme disease syndrome (PTLDS) or chronic Lyme disease. They say that most of the complaints of fatigue, headaches and memory problems in the weeks and months after a Lyme diagnosis, are just

the aches and pains of daily life.

Zhang says it is very likely that these symptoms occur because the standard course of antibiotics does not get all of the *Borrelia burgdorferi* and these lingering bacteria remain in the body despite treatment with current Lyme antibiotics. The problem is that while a blood test may help to confirm Lyme disease, there is no definitive test for PTLDS as no bacteria can be cultured after antibiotic treatment and there are no approved or proven treatments.

The new drugs identified by Zhang's assay appear to specifically target these persisters in the lab. The question remains as to whether they will do the same thing in humans. Some doctors are considering prescribing the drugs off-label for persistent Lyme disease symptoms, even without evidence showing it will work. Zhang cautions that animal studies should be done to see if they work before they are tested in Lyme disease patients, even though these are clinically used drugs for treating other infections, as inappropriate use of antibiotics can have side effects.

"There are a significant number of people who are sick and desperate for a cure for their Lyme disease symptoms months and even years after they have been told they are cured of the [disease](#)," Zhang says. "The current drugs we use aren't good enough for these persistent cases. This is why I have been getting so many calls and emails about our [test](#) and the drugs we have identified."

More information: "An Optimized SYBR Green I/PI Assay for Rapid Viability Assessment and Antibiotic Susceptibility Testing for Borrelia burgdorferi" by Jie Feng, Ting Wang, Shuo Zhang, Wanliang Shi and Ying Zhang, *PLOS ONE*, 2014.