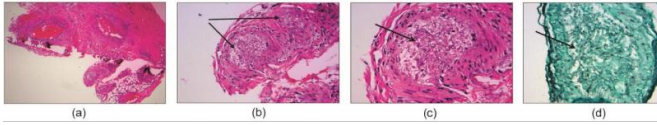


Researchers test new pathogen detection technology

18 January 2016, by Karen Kreeger



Staining of tissue section from the patient for fungus. A Hematoxylin and Eosin (a-c) as well as Grocott Methanamine Silver (d) stain were performed on the tissue sections from an AML patient. Figure (a) shows the low power view of the soft tissue, (b) shows the 20X view, (c) shows the 40X view and (d) shows the 40X view using the silver Grocott stain. Fungus in blood vessels is shown with arrows. Credit: Erle Robertson, PhD, Perelman School of Medicine, University of Pennsylvania; Cancer Biology & Therapy

Patients who are undergoing treatment for diseases such as cancer often face the added challenge of a compromised immune system, which can be a toll both of their condition and the drugs used to treat it, leaving them vulnerable to various opportunistic infections. Many of these infections are not only life-threatening, but caused by rare organisms that are extremely difficult to isolate and identify. However, the sooner an infection is pinned down, the faster and more effectively it can be treated. After developing a novel investigational technology called PathoChip that can rapidly identify elusive microorganisms, a team of Penn Medicine researchers recently succeeded for the first time in identifying a pathogen in a patient sample, demonstrating the proof of principle that this technology can be used to identify pathogens in human disease.

In a recently published study in *Cancer Biology & Therapy*, the group of Penn colleagues, led by Erle Robertson, PhD, professor and vice-chair for research in Otorhinolaryngology at the Perelman School of Medicine, and James Alwine, PhD, a professor of Cancer Biology and Michael Feldman,

MD, a professor of Pathology and Laboratory Medicine, used a version of the PathoChip microarray, which contains 60,000 probes for all known viruses, as well as a broad range of bacteria, protozoa, fungi, and helminthes, a parasitic worm, to identify the pathogenic agent in the sample of a patient. They analyzed preserved tissue samples from a middle-aged male with relapsed acute myelogenous leukemia (AML) who developed an unknown fungal infection following chemotherapy.

"We've run many tests to see if we could identify [pathogens](#) in the lab, just to see if the PathoChip has efficacy in identifying a variety of organisms, and we were able to identify all infectious agents tested," Robertson said. "But this was the first time we actually looked directly at a patient sample to identify a pathogenic agent."

The PathoChip allows for a single sample to be tested simultaneously for thousands of possibilities, dramatically reducing the time required for diagnosis. They first screened the sample and analyzed it with a type of bioinformatics probe to narrow the focus to a specific microbial family. Seventeen organisms displaying the highest signal were compared with the signals from a control sample. Molecular signals obtained from additional bioinformatics tests provided further information, which led to the identification of the specific infectious agent— in this case one of the two species of *Rhizomucor*, a rare fungus known to cause zygomycosis in humans.

A potentially fatal illness most often seen in immunocompromised patients, zygomycosis is not only difficult to treat but challenging to diagnose. Fungal species can be painfully slow or even impossible to culture in the laboratory, delaying or preventing their identification and therefore patient treatment. As Robertson explains, the PathoChip provides an efficient alternative. "With this technology, out of 60,000 possibilities and probes

that we used, in a little over 24 hours we were able to identify this particular fungi," he says.

While other techniques, such as sequencing arrays, are available to identify unknown infectious organisms, these approaches can also have significant limitations. "You could use other technologies such as next-generation sequencing, but there would have to be a high percentage of nucleic acids present in the tissue, and technicians would need enough of these materials to do the sequencing," Robertson explains.

"The analysis component of it would take more time," he adds. "We think this technology is complementary to next-generation sequencing in some ways, and even more finely tuned, because we have a much higher sensitivity in detecting agents or individual organisms present in any kind of sample, whether it's abiotic or biotic. We can identify agents in soil, for example, in plant tissue, in animal tissue, or human tissue."

Robertson and his team in the laboratory, which includes Sagarika Banerjee, PhD, and research technician Kristen Peck, M.Sc. along with bioinformatics expert Zhi Wei, Ph.D. from the New Jersey Science and Technology Institute, are looking ahead to both expanding the capabilities of the PathoChip and increasing its use in the clinic: "We are constantly updating it based on what we think are important agents to include on the chip right now." Additional work is currently being done to further develop the next steps for using PathoChip to identify infectious agents in the clinic. "This will take a great deal more research to eventually lead to approval for frontline clinical use in hospitals," Robertson said.

More information: Sagarika Banerjee et al. Identification of fungal pathogens in a patient with acute myelogenous leukemia using a pathogen detection array technology, *Cancer Biology & Therapy* (2015). DOI: [10.1080/15384047.2015.1121349](https://doi.org/10.1080/15384047.2015.1121349)

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