Potential new drug target to fight tuberculosis identified

July 30 2008

With antibiotic resistance on the rise, tuberculosis is emerging as a bigger global health threat than ever before. But now, innovative research at Weill Cornell Medical College suggests that *Mycobacterium tuberculosis* has an as yet unsuspected weakness -- one that could be a prime target for drug development.

"Using novel techniques, we have identified a key membrane protein that's essential to the defense that *M. tuberculosis* mounts against the acidic environment of immune cells called macrophages. Without this protein, called Rv3671c, the bacterium becomes vulnerable to acidification and is killed," explains lead author Omar H. Vandal, a postdoctoral fellow in the lab of study co-senior author Dr. Sabine Ehrt, associate professor of microbiology and immunology at Weill Cornell Medical College.

"*M. tuberculosis* does not depend on Rv3671c under standard growth conditions in the test tube, so it has been overlooked as a candidate drug target," says Dr. Carl F. Nathan, also a senior author of the study and the R.A. Pritchett Professor of Microbiology. He is also chairman of the Department of Microbiology and Immunology at Weill Cornell.

Drs. Ehrt and Nathan co-supervised Dr. Vandal in this work while Dr. Vandal was a student at the Weill Cornell Graduate School of Medical Sciences. "However, when *M. tuberculosis* infects the host, then the Rv3671c protein becomes essential," added Dr. Ehrt. "This is an example of a new class of potential targets for anti-infective agents,"

continues Dr. Nathan, "those that the pathogen only needs in order to survive in the host environment."

The research was just published in *Nature Medicine*.

In numerous papers published in leading journals, Dr. Nathan has long pushed for an innovative approach to the development of anti-infective agents that goes beyond the traditional antibiotic paradigm. "That's exactly what we sought to do in this research," he says.

One of the study's innovations involved the examination of *M. tuberculosis* as it interacted with bone marrow–derived macrophages during the infective process.

"That's a huge change from standard anti-infective research, which typically deals with the pathogen simply replicating in culture," explains Dr. Vandal. "In our experiments, we wanted to see if biochemical actors would emerge in the infective process that might be inoperative in the usual in vitro setting."

The team specifically focused on changes in the pH (acidity) of the phagosome -- a structure that macrophages use to consume and destroy pathogens, including bacteria.

"As part of this process, the phagosome becomes acidic, which is thought to contribute to its ability to break down and destroy the pathogen," Dr. Ehrt explains. "However, *M. tuberculosis* appears to survive the acidification process, keeping its own internal pH stable."

How does the bacteria do this, despite being surrounded by the more highly acidic phagosome? To find out, the team used a kind of genetic tweaking that effectively disabled M.tuberculosis' ability to produce a key protein lying at its membrane -- a protease (enzyme) called

Rv3671c.

They then watched how the organism fared without it.

"What we observed was pretty amazing -- without functioning Rv3671c, the mutant bacterium was easily destroyed in a low-pH environment, both in culture and inside the more acidic environment of the macrophage," says Dr. Vandal. "This revealed a new point of vulnerability for the bacterium."

The experiment also broke new ground because the researchers were able to accurately gauge the bacterium's internal pH with the organism lying inside a host cell.

"The ability to make those kinds of measurements will expand research into this type of host-pathogen interaction," Dr. Nathan believes.

The next step is to find out why Rv3671c is so crucial to *M. tuberculosis*' defense.

"Right now, we have very little idea of the mechanism at work here. Perhaps as an enzyme Rv3671c cleaves a transcription regulator that then turns on some kind of defensive program within the bacterium. Only further study will reveal those secrets," says Dr. Ehrt.

"What is clear is that by targeting an element involved directly in the infective process, we may develop a line of drugs that work in collaboration with, rather than in difference to, the host environment, including host immune responses," Dr. Nathan says. "Hopefully, this kind of approach can help solve the ongoing problem of bacterial drug resistance."

The new study is also another example of an interdisciplinary approach

-- this time among biochemists, microbiologists, immunologists and cell biologists.

"In the ideal collaboration, each participant brings key insights from their particular discipline to the table," Dr. Nathan says. "The results are discoveries like these."

Source: New York- Presbyterian Hospital