New study overturns orthodoxy on how macrophages kill bacteria

April 27 2009

Medical microbiology professor James Slauch led a study that challenges decades-old assumptions about how immune cells kill bacteria. Credit: Photo by L. Brian Stauffer, U. of I. News Bureau.

For decades, microbiologists assumed that macrophages, immune cells that can engulf and poison bacteria and other pathogens, killed microbes by damaging their DNA. A new study from the University of Illinois disproves that.

The study, published in the journal *PLoS ONE*, shows that macrophages focus their most potent poisons, known as reactive oxygen species

(ROS), on targets outside the cytoplasm.

Macrophages are voracious eaters that "swallow" cellular debris and invading organisms. They kill microbes with ROS. All aerobic cells inadvertently produce ROS that can, if left unchecked, damage DNA and other cellular components and cause cell death.

Bacteria and animal cells contain special enzymes, called superoxide dismutases, which neutralize an important ROS, called superoxide.

Macrophages have harnessed these lethal compounds, dumping large quantities of superoxide onto engulfed bacteria to kill them.

Although macrophages direct ROS against invading bacteria, *Salmonella typhimurium*, the microbe used in the study, is adept at evading these defenses. The most virulent strains of *S. typhimurium* can survive and even propagate inside macrophages, eventually emerging to infect more cells.

"It's been assumed that reactive oxygen species kill the bacteria by going into the cytoplasm and causing DNA damage," said medical microbiology professor James Slauch, who led the study. "You can find this idea over and over again in review articles and many immunological textbooks, but with no real data to back it up."

To test this hypothesis, Slauch and graduate student Maureen Craig looked at the superoxide dismutases that are part of the bacterial defense against ROS. There are two such enzymes in the cytoplasm of *S. typhimurium*, called SodA and SodB, and another, SodC, in the periplasm, the space between the bacteria's inner and outer membranes.

One way to understand the role of an enzyme is to see what happens when it is absent, so the researchers looked at mutant *S. typhimurium* that

had the genes for SodA, SodB, or both enzymes, deleted. Deleting the gene for SodA seemed to make no difference, but the SodB mutants were less able to survive and cause disease in a mouse. The double mutants were even more impaired. They were much, much less likely to survive in the mouse than bacteria with only the SodB gene missing. These findings "offer genetic proof" that both enzymes "are involved in the same process," Slauch said.

The fact that the bacterial mutants were less likely to survive in a mouse did not prove, however, that the missing enzymes were protecting the bacteria from ROS generated in the mouse macrophages, Slauch said.

"You get the same result if you grow these mutants in the laboratory in aerobic conditions," he said.

Furthermore, the SodA/SodB mutant bacteria were profoundly weakened - even in a mouse that was unable to produce the potent ROS superoxide in its macrophages. These results suggest that the superoxide dismutases in the bacterial cytoplasm are most likely protecting the bacterium from its own, naturally occurring ROS, Slauch said.

In contrast, deleting the gene encoding the periplasmic superoxide dismutase, SodC, conferred the same defect regardless of whether the cytoplasmic SodA/SodB were present or absent, showing that its function is independent of the cytoplasm.

Moreover, strains lacking SodC were impaired only in the presence of superoxide produced in macrophages; there was no impairment in laboratory media or in mice lacking the ability to make superoxide.

This suggests that the superoxide and other reactive oxygen species are not making it from the macrophage into the bacterial cytoplasm, Slauch said.

"We conclude from all this data that the most sensitive target of ROS in the macrophages lies outside the cytoplasm," Slauch said. "We don't know what that target is, but it's clearly not in the cytoplasm."

Source: University of Illinois at Urbana-Champaign (news : web)

Citation: New study overturns orthodoxy on how macrophages kill bacteria (2009, April 27) retrieved 24 April 2024 from https://phys.org/news/2009-04-overturns-orthodoxy-macrophages-bacteria.html

This document is subject to copyright. Apart from any fair dealing for the purpose of private study or research, no part may be reproduced without the written permission. The content is provided for information purposes only.