Unique portion of enzyme fights lung infection

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An enzyme known to play a key role in the development of emphysema serves as the first line of defense against bacterial infection of the lung, according to researchers at the University of Pittsburgh School of Medicine. They also found that the antimicrobial activity comes from a small portion of the enzyme that is structurally and sequentially unique in nature.

Lead author A. McGarry Houghton, M.D., assistant professor, Division of Pulmonary, Allergy and Critical Care Medicine, Pitt School of Medicine, said that prior to this discovery scientists thought that the enzyme, called macrophage elastase, matrix metalloproteinase-12 or MMP-12, which is produced in excess in smokers, didn't do anything but degrade the lung's elastic fibers, thereby contributing to the tissue destruction of emphysema.

"But we found that mice that didn't have the gene to make this enzyme could not clear bacteria well and were more likely to die of infection," he explained. "They couldn't make this small protein, which kills bacteria by poking holes in cell membranes." The findings were described in *Nature*.

"While not the initial purpose of this study, finding novel antimicrobial mechanisms is extremely important," said senior author Steven D. Shapiro, M.D., Jack D. Myers Professor and chair of the Department of Medicine, Pitt School of Medicine, whose research teams cloned the MMP-12 gene almost 20 years ago and conducted the work that showed

its role in emphysema. "Many microorganisms have adapted to circumvent our current and stagnant arsenal of antibiotics. We must find new weapons so that we don't fall back to the public health problems we had prior to penicillin."

MMP-12 is stored in macrophages, the cells that swallow up invading bacteria. When Staphylococcus aureus was injected into the tail vein of healthy and MMP-12-deficient mice, the two-week mortality rate was about the same. However, the amount of bacteria was much greater in the lungs of MMP-12-deficient mice. In models of pneumonia and peritonitis, MMP-12-deficient mice were much less likely to survive the infection. Macrophages are present in very high numbers in the lungs and the peritoneum, which is the lining of the abdomen.

"Our experiments also showed that while the MMP-12-deficient macrophages were able to ingest bacteria, they couldn't kill them," Dr. Houghton said. "The intracellular bacteria level escalated rather than diminished."

The researchers then looked for what gave MMP-12 its antibacterial properties. While the portion of the enzyme that catalyzes, or speeds up, chemical reactions degrades lung tissue in emphysema, its tail is the portion that kills microbes. Protein fragments were tested to identify a chain of 20 amino acids that could kill Staph aureus in culture dishes. A computer-generated 3-dimensional model of the enzyme's tail, including the 20-amino acid chain, revealed there were only a few exposed places permitting interaction with bacterial surfaces, and that one of those loops had a protrusion containing a sequence of four amino acids, called KDEK, that is not present in any other enzymes of the MMP class.

"Humans, mice, rats and rabbits all have that special sequence and structure in MMP-12, but not in other MMPs," Dr. Houghton noted. He and his colleagues synthesized chains nearly identical to the 20-amino

acid sequence but substituted other segments for KDEK, and found that both the sequence and the loop structure was necessary to kill bacteria.

The team plans to study whether the same part of the enzyme is able to kill viruses and fungi, and whether there are any connections between MMP-12's roles in emphysema and infection defense.

Source: University of Pittsburgh Schools of the Health Sciences (news : web)