Tuberculosis bacteria hide in the low oxygen niches of bone marrow stem cells

June 19 2015

A new study from the Forsyth Institute is helping to shed light on latent tuberculosis and the bacteria's ability to hide in stem cells. Some bone marrow stem cells reside in low oxygen (hypoxia) zones. These specialized zones are secured as immune cells and toxic chemicals cannot reach this zone. Hypoxia- activated cell signaling pathways may also protect the stem cells from dying or ageing. A new study led by Forsyth Scientist Dr. Bikul Das has found that *Mycobacterium tuberculosis* (Mtb) hijack this protective hypoxic zone to hide intracellular to a special stem cell type. The study was published online

on June 8th in the *American Journal of Pathology*.

Mtb, the causative organism of tuberculosis, infects nearly 2.2 billion people worldwide and causes 1.7 million annual deaths. This is largely attributed to the bacteria's ability to stay dormant in the human body and later resurface as active disease. Earlier research at Forsyth revealed that Mtb hides inside a specific stem cell population in bone marrow, the CD271+ mesenchymal stem cells. However, the exact location of the Mtb harboring stem cells was not known.

"From our previous research, we learned that cancer stem cells reside in the hypoxic zones to maintain self-renewal property, and escape from the immune system" said Bikul Das, MBBS, PhD, Associate Research Investigator at the Forsyth Institute, and the honorary director of the KaviKrishna laboratory, Guwahati, India. "So, we hypothesized that Mtb, like cancer, may also have figured out the advantage of hiding in the hypoxic area."

To test this hypothesis, Dr. Das and his collaborators at Jawarharlal Nehru Univeristy (JNU), New Delhi, and KaviKrishna Laboratory, Indian Institute of Technology, Guwahati, utilized a well-known mouse model of Mtb infection, where months after drug treatment, Mtb remain dormant for future reactivation. Using this mouse model of dormancy, scientists isolated the special bone marrow stem cell type, the CD271+ mesenchymal stem cells, from the drug treated mice. Prior to isolation of the stem cells, mice were injected with pimonidazole, a chemical that binds specifically to hypoxic cells. Pimonidazole binding of these cells was visualized under confocal microscope and via flow cytometry. The scientists found that despite months of drug treatment, Mtb could be recovered from the CD271+ stem cells. Most importantly, these stem cells exhibit strong binding to pimonidazole, indicating the hypoxic localization of the stem cells. Experiments also confirmed that these stem cells express a hypoxia activated gene, the hypoxia inducible factor

1 alpha (HIF-1 alpha).

To confirm the findings in clinical subjects, the research team, in collaboration with KaviKrishna Laboratory, the team isolated the CD271+ stem cell type from the bone marrow of TB infected human subjects who had undergone extensive treatment for the disease. They found that not only did the stem cell type contain viable Mtb, but also exhibit strong expression of HIF-1alpha. To their surprise, the CD271+ stem cell population expressed several fold higher expression of HIF-1alpha than the stem cell type obtained from the healthy individuals.

"These findings now explain why it is difficult to develop vaccines against tuberculosis," said Dr. Das. "The immune cells activated by the vaccine agent may not be able to reach the hypoxic site of bone marrow to target these "wolfs-in-stem-cell-clothing".

The success of this international collaborative study is now encouraging the team to develop a Forsyth Institute/KaviKrishna Laboratory global health research initiative to advance stem cell research and its application to global health issues including TB, HIV and oral cancer, all critical problems in the area where KaviKrishna Laboratory is located.

Provided by Forsyth Institute