

JULY 01, 2005

Identifying Blood Stem Cells Is a SLAM Dunk

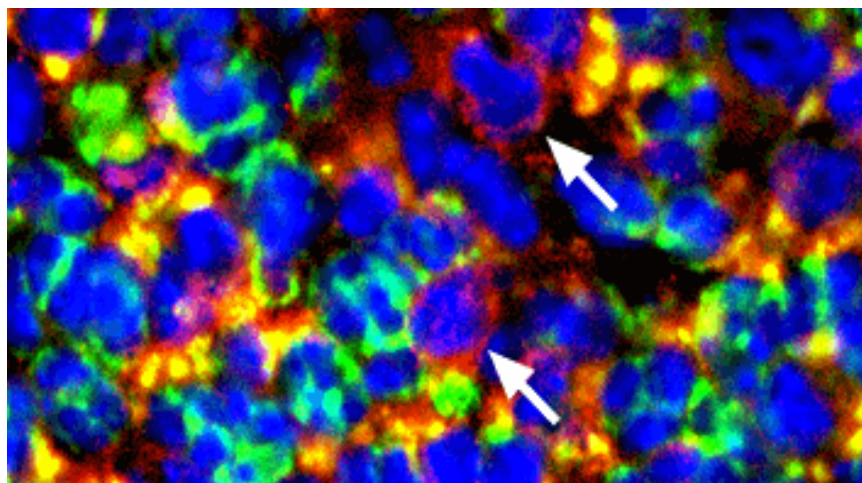


Image Title: The image shows two hematopoietic stem cells in a section through the spleen (arrows). The stem cell marker CD150 is in red, mature hematopoietic cells are stained in green, and blue reveals the nuclei of all cells. - Courtesy of Sean Morrison, HHMI at the University of Michigan.

Researchers have developed a simple technique to identify hematopoietic, or blood-forming, stem cells based on a set of characteristic markers that the cells display on their surface. The elucidation of this distinctive stem-cell code is the first time that researchers have been able to identify specific stem cells by looking at surface markers drawn from a single family of genes.

Stem cells are immature cells that can develop into a variety of adult cells. In this case, hematopoietic stem cells can develop into all blood and immune system cell types and are already used therapeutically to restore the hematopoietic system of patients after chemotherapy or radiation therapy.

The new technique to identify hematopoietic stem cells (HSCs) will enable scientists to determine where stem cells are located in blood-forming tissues and to trace the developmental routes these stem cells take as they mature into blood cells. If the researchers' studies in mice apply to blood-forming stem cells in humans, the technique may enable safer transplants of stem

cells, by improving purification of the stem cells prior to transplantation, said the scientists.

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- Sean J. Morrison

Sean J. Morrison, a Howard Hughes Medical Institute investigator at the University of Michigan, led the research team, which published its findings in the July 1, 2005, issue of the journal *Cell*. Other members of the team included graduate students Mark Kiel and Omer Yilmaz as well as postdoctoral fellow Toshihide Iwashita from the Morrison laboratory. Another co-author of the paper is from Harvard Medical School.

The method developed by Morrison and his colleagues can distinguish different types of blood-forming progenitors based on differences in their expression of members of a family of highly similar protein receptors, called SLAM family receptors. The receptors nestle in the surface of the cell membrane and detect external chemical signals and translate those signals into cellular responses.

According to Morrison, specific SLAM family members were known to be expressed in white blood cells and to play a role in their function. However, it was not appreciated that specific members of the SLAM family could be used to distinguish HSCs from other types of progenitor cells. Until now, the problem in using cell surface proteins to identify HSCs has been that the combinations of such markers have been complex and difficult to manage, said Morrison.

In their analyses of SLAM proteins on HSCs, the researchers found that they could distinguish HSCs by the presence or absence of a few specific members of the SLAM family. This provided a simpler and more robust way to identify HSCs.

The researchers demonstrated that the HSC SLAM markers appeared to be universal in mice by showing that they distinguished HSCs from different genetic strains of mice, which show variation in other HSC markers that have been used previously. The scientists also found that the SLAM markers distinguished HSCs in mice whose immune systems had been activated, or mobilized, by immune triggers called cytokines.

"Thus, these SLAM family members are so precisely, differentially expressed that we can now purify HSCs much more simply and, in some contexts, much more rigorously," said Morrison. "The SLAM markers that we have identified work in every context that we have looked so far -- different mouse strains, old mice, young mice, cytokine-mobilized mice and non-mobilized mice. Using this simple combination of markers, we have achieved very high levels of purity in these cells," said Morrison.

The simplicity of the technique enabled the researchers to tag the SLAM markers to trace the localization of HSCs in their "niches" in tissues. Niches are "microenvironments" of supporting cells in which stem cells are induced by chemical signals to produce mature blood cells, in a process called hematopoiesis.

"A major impediment to understanding hematopoiesis has been the inability to identify these niches," said Morrison. "So, in the past we needed to use very complicated combinations of markers to rigorously purify the stem cells - that is too complicated to use in staining tissue sections."

Using antibody stains that revealed SLAM markers, the researchers could visualize where in the bone marrow and spleen HSCs localized. They found that most HSCs migrated to tiny blood vessels in the bone marrow and spleen called sinusoids, where the HSCs remained in contact with the endothelial cells surrounding these sinusoids.

"That finding suggests that most stem cells are sustained most of the time by their interaction with these sinusoidal endothelial cells; implying that the sinusoidal endothelial cells may be secreting factors that help maintain stem cells or regulate their function," concluded Morrison. Such findings offer important clues to the role of such endothelial cells in maintaining HSCs, he said.

More broadly, similar vascular endothelial cells have been found to provide such niches in the nervous system and other tissues, said Morrison. "These studies suggest that vascular endothelium may be generally important in creating stem cell niches in a variety of different tissues; and that sinusoidal endothelium, a specialized form of endothelium found only in hematopoietic tissue, may be specialized to support the maintenance of HSCs," said Morrison.

The presence of HSCs in such endothelium helps explain a mystery of how stem cells - which are manufactured in the bone marrow -- can enter the blood stream within minutes of being treated with certain drugs. "Our data suggest that the reason stem cells can get into the circulation so fast might be that at least a subset of them is sitting right on top of the sinusoids that they would use to enter circulation," said Morrison.

These insights represent only the beginning of new research pathways in stem cell biology enabled by the SLAM markers, said Morrison. Researchers can now use imaging techniques to explore precisely how HSCs interact with supporting niche cells and isolate the chemical signals from these cells that regulate their maintenance and trigger their maturation.

Morrison said that future studies will aim to determine the function of the SLAM family of receptors. The researchers will also explore whether other members of the SLAM family can also be used to identify stem cells. And, they will use their technique to identify cells that create stem cell niches and the chemical signals they secrete.

Regarding clinical applications, Morrison said that "the big question is whether these SLAM family receptors are differentially expressed in human HSCs. If they are, their use might dramatically improve the purity of isolated HSCs, as they have in our mouse studies. Such an improvement in purity might improve the safety and effectiveness of HSC transplants. We are optimistic that these markers are also expressed on human HSCs, and we are now testing that possibility," he said.