Unit 2: Paper Summary – Reed

Contribution of olfactory neural stem cells to tissue maintenance and regeneration.

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Abstract:

The olfactory neuroepithelium undergoes continual neurogenesis and, after extensive lesions, fully regenerates to maintain sensory function. The stem cell population underlying this regenerative capacity remains elusive. Here we show that mouse horizontal basal cells (HBCs) function as adult olfactory neuroepithelium neural stem cells and examine their distinct dynamics in olfactory neuroepithelium maintenance and regeneration. Fate-mapping analysis after olfactory neuroepithelium lesioning shows that HBCs are competent to regenerate both neuronal and non-neuronal olfactory neuroepithelium lineages. HBCs serve as a reservoir of long-lived progenitors that remain largely quiescent during normal neuronal turnover or even after acute, selective loss of mature neurons. Under these conditions, previously identified progenitors are largely responsible for tissue maintenance. Yet after extensive injuries that deplete resident neuronal precursors, HBCs transiently proliferate and their progeny fully reconstitute the neuroepithelium. Our data support a new model of adult neurogenesis in which distinct cell populations mediate normal neuronal turnover and neuronal replacement upon traumatic injury.

In this paper the researchers determined the identity of the adult stem cell in the olfactory epithelium. The olfactory epithelium is inside the nasal cavity of many animals, and it is the sensory tissue for smell. Inside the olfactory epithelium (OE) there are neurons that are exposed to air. These neurons have receptor that bind and detect odor molecules in the air, and then signal to the brain. The brain uses all the signals coming from different neurons in the olfactory epithelium to determine what the smell is.

Since the neurons are in contact with the air and anything floating in it, the neurons are vulnerable and can easily be damaged or destroyed. For this reason, the OE has evolved the ability to completely replace all the neurons if they are destroyed. There are also two other cell types in the OE. These are called Bowman's gland, which makes nasal mucus, and sustentacular cells, which are glial support cells (glial cells are oligodendrocytes, which wrap neurons with myelin sheaths improving transmission of nerve impulses, and astrocytes, which provide metabolic and other support for

neurons.) If these cells are also destroyed, they are then regenerated. To replace all three cell types there needs to be a multipotent adult stem cell in the epithelium.

Before this paper was published the identity of the stem cell was unknown. There were two candidate cell types, the Horizontal Basal Cell (HBC) and the Globose Basal Cell (GBC). This paper uses a powerful technique called the Cre-Lox recombination to perform lineage tracing on the HBC cells. They determine that the HBCs are the multipotent stem cell.

Cre-lox is a powerful and complicated technique that is frequently used in many scientific experiments (see attached figure). In it, two mice strains are genetically engineered to have either a Cre or a lox construction. In the Cre mouse, the genetic code for the protein Cre is put into the DNA under control what is called a tissue specific or cell specific promoter. This means the Cre will only be expressed in the specific cell type that the researcher wants. In the lox mouse two identical pieces of DNA, called loxP sites, are placed around a DNA that you want to be removed. The Cre and lox mice don't do anything on their own, but if they are breed together so that both the Cre and the lox are in the same mouse then the system becomes active. The Cre will bind to the two lox sites, make a loop of DNA, and cut out any DNA that sits between them. The change to the DNA is permanent, and any descendent of cells that this happens in will also have the DNA change.

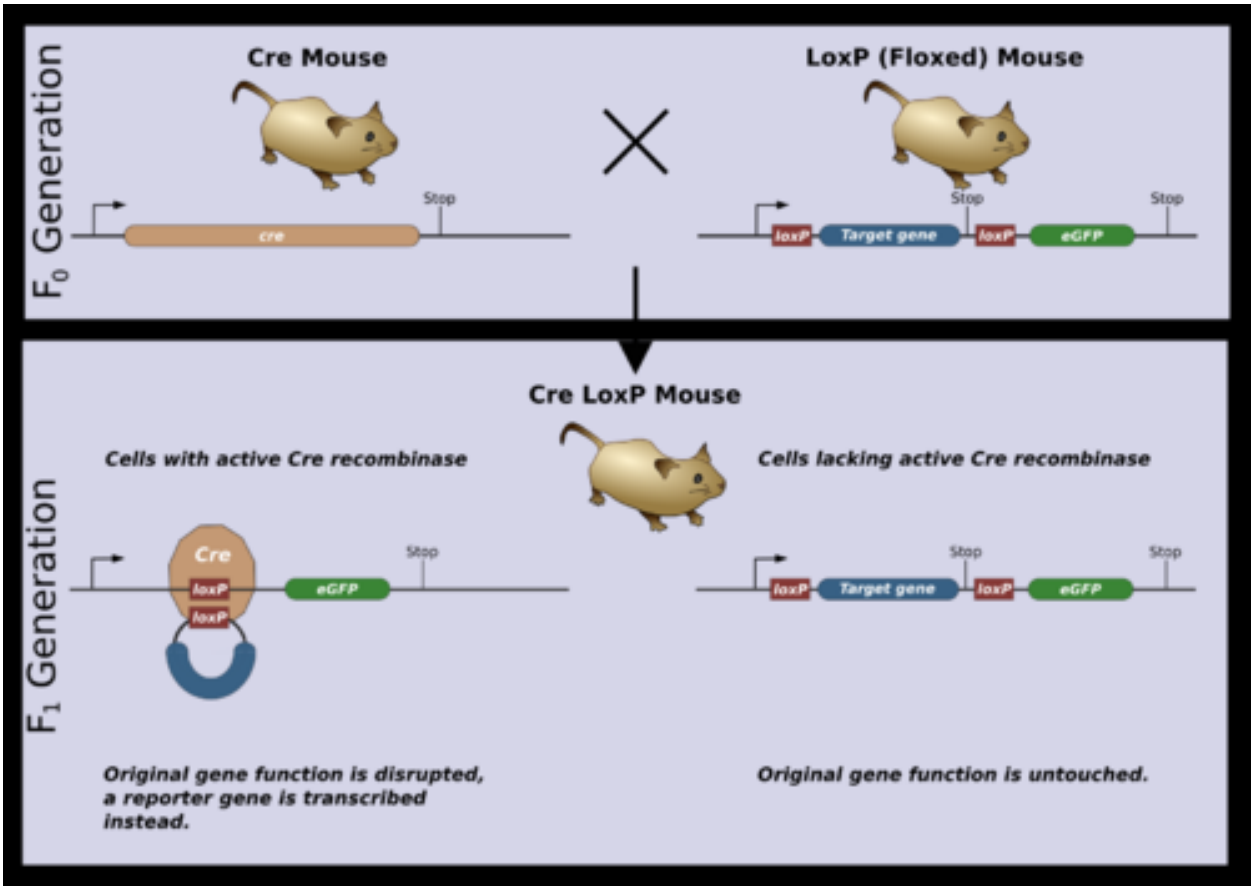
In this paper the Cre was designed to be expressed in the HBC cells. In the lox animal, the loxP sites surround a stop codon that is in front of the reporter LacZ. Without Cre the stop codon prevents LacZ from being expressed. So in the HBCs with Cre and LoxP, Cre is turned on, binds to the LoxP sites, and removes the stop codon. As a result, LacZ is expressed in the HBCs and any cell that HBCs give birth to.

The researchers found that in these Cre-Lox mice only the HBCs expressed LacZ. They then gave the mice an inhaled chemical that destroys all the cells in the OE except the HBCs. After 21 days (enough time for the epithelium to regenerate) they examined the epithelium and found many more cell types expressing LacZ. Antibody staining showed that neurons and the sustentacular cells were both expressing LacZ. Since only HBCs and cells that are born from HBCs could express LacZ, the experiment proved that HBCs must have given rise to all the regenerated cells. Also, the GBCs, the other cells thought to be multipotent stem cells, were also labeled with LacZ. This shows that the GBCs are also descendants of HBCs, and are not stem cells.

The researchers did another experiment in which they surgically removed the olfactory bulb from the brains of Cre-lox animals. The olfactory bulb is what neurons in the OE are connected to in the brain. When the bulb is removed and the connection is lost, the neurons in the OE die, but the other cells in the OE are still alive. The OE will

regenerate new neurons, but only neurons without a connection to the brain. The researchers found in this situation, that the new neurons were not labeled with LacZ. This means that to just make new neurons, the epithelium does not need to use the multipotent stem cell. The GBCs are probably early progenitors of just the neuron population and they are sufficient to regenerate the neurons. In these conditions, the HBCs are not mitotic and are inactive.

There is still much to learn about the HBC stem cell population. For example, it is unclear how these cells develop and what signals them to start dividing and differentiating. By studying how these cells work we may be able to learn more about how they and other adult stem cells function.



Wikipedia, accessed Oct 26, 2009.