Influenza in Phagocytic Cells: A Story of Direct Infection or Exogenous Uptake of Antigen

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• Cells are shown to be directly infected with Influenza A Reporter Virus
• If macrophages and dendritic cells are reporter+, is that due to direct infection, phagocytosis of the fluorophore, or a combination of the two?
Influenza A Virus in Phagocytic Cells

• Aims
  • Demonstrate macrophages can take up exogenous fluorophore becoming reporter+
  • Determine Kinetics of tdTomato+ Macrophages and Dendritic Cells (DPI: 3, 5, 7, 10, or 21)
  • Determine if reporter+ macrophages and dendritic cells are reporter+ due to direct infection or exogenous uptake of fluorophore using BM Chimera
Aim 1: Demonstration of Exogenous Uptake of Fluorophore by Macrophages
Aim 2: Determine Kinetics of tdTomato+ Macrophages and Dendritic Cells

Time Points
Day 3, 5, 7, 10, and 21 post infection

Populations of Interest:
Alveolar Macrophages
CD11b+ DCs
CD103+ cDCs
Aim 2: Determine Kinetics of tdTomato+ Macrophages and Dendritic Cells

* Minimal detection of reporter+ cells in the spleen
Aim 2: Determine Kinetics of tdTomato+ Macrophages and Dendritic Cells
Aim 3: Bone Marrow Chimera

Generate Bone marrow chimeras to determine direct infection vs exogenous uptake

Day 5 was chosen to harvest these mice.
Aim 3: Bone Marrow Chimera

MΦs

CD11b⁺ DCs

CD103⁺ DCs

Lung

Frequency of tdTomato⁺ Cells

Frequency of tdTomato⁺ Cells

Frequency of tdTomato⁺ Cells

PR8

PA_Cre

PR8

PA_Cre

PR8

PA_Cre
Conclusions

• Bone Marrow Derived Macrophages in culture can phagocytose tdTomato \textit{in vitro} and \textit{in vivo}

• Macrophages and Dendritic Cells in IAV infection have greatest abundance at approximately D5

• Unlike epithelial cells, these cell populations do not remain in these tissues past D10

• Phagocytic immune cells can phagocytose IAV antigen and traffic to secondary lymphoid organs.

• These results have implications to IAV vaccination demonstrating that subunit vaccinations are a viable option for priming adaptive immune cells in secondary lymphoid organs.
Future Direction

• BM Chimera Experimentation
  • WT transplant with iRED Immune System – reporter+ due to direct IAV infection alone
  • Assess activation levels of CD80/86 in direct infection versus exogenous uptake of antigen

• tdTomato Detection In Tissues Over Time
  • In exogenous uptake of tdTomato, are changes in tdTomato+ populations due to trafficking to and from tissues, degradation of the tdTomato+ fluorophore, or cells dying?
Questions or Comments?

The Langlois Lab Enjoying the Eclipse 8/21/17

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