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## Immunomodulation strategy against Respiratory Syncytial Virus infection by using lung primo-colonizing bacteria

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### Background

The respiratory syncytial virus (RSV) is the main etiological agent of bronchiolitis in infants. Few therapeutics are available to prevent or treat RSV infection. The neonatal lung environment is responsible for the high susceptibility of infants to RSV infection. Indeed, at birth the lung is a complex environment characterized by an evolving immune system continuously exposed to environmental stimuli including microbiota colonization. We assume that primo-colonizing bacteria are involved in the maturation of lung immunity and thus in the susceptibility of neonates to pulmonary infection. The neonatal period constitutes a window of opportunity to immunomodulate lung immune responses. The use of bacterial strains during this period could help directing the local immune response towards a protective immunity to RSV infection. The aim of our project is to demonstrate that primo-colonizing bacterial strains isolated from neonatal lungs could reduce the severity of RSV pathology.

### Method

In a first part, we used neonatal mouse lung explant to i) characterize the immunostimulant property of the primo-colonizing bacterial strains by measuring cytokines secretion (Luminex assays) ii) to evaluate the capacity of these strains to interfere with the replication of a recombinant RSV expressing luciferase.

Finally, we confirmed the anti-RSV activity of these bacteria by infecting Air-Liquid Interface human cell culture (Mucilair, Epithelix) with a recombinant RSV expressing mCherry.

### Result

Twenty-five primo-colonizing strains were characterized on neonatal mouse lung explants for their capacity to stimulate cytokine secretion and to interfere with RSV replication. We identified several non-cytotoxic bacterial strains that could stimulate lung explants for the secretion of type-I cytokine, such as IL-12 and IFN $\alpha$ . These strains were also able to reduce RSV viral replication in lung explants. The bacteria 17 was selected for its original cytokine signature (Type-I immunity and IL-9) in addition with its antiviral capacity on neonatal mouse lung explants. The antiviral activity of this particular bacterial strain was confirmed with Air-Liquid Interface epithelial human cell culture pre-exposed to the bacteria 17 then infected with the mCherry-RSV.

### Conclusion

The bacteria 17 exhibit anti-RSV properties on ex vivo mouse lung explants and on human airway epithelial cells. Our next goal is to establish if this particular primo-colonizing bacterial strain is able to reduce RSV pulmonary disease. To achieve it, this bacterium will be administered as a preventive treatment in a relevant neonatal mouse model of RSV infection. Such beneficial bacteria with immunomodulatory properties could provide a new potential therapeutic strategy for bronchiolitis.