Final Report to Cystic Fibrosis Australia

Report by: Stefanie Bader – PhD Candidate Pellegrini Laboratory
Project Name: Progressing a novel treatment paradigm to improve management of infections in people with CF
Date of report: 16/12/2022

Lay project description:

Bacterial infection is the most common cause of death in CF patients. The immune cells responsible for combating bacteria are often defective in people suffering from cystic fibrosis and can damage the lungs while trying to fight infections. This reaction worsens symptoms and fails to kill the pathogen, leading to chronic disease that is refractory to antibiotic therapy. We propose to use a new class of drugs that can tackle this problem in two ways: by killing infected host cells, reducing the number of bacteria, and by removing defective immune cells. Importantly, while bacteria rapidly develop resistance to antibiotics, leading to a decline in median survival, our proposed strategy promotes the death of defective host cells, as well as intracellular pathogens, and bacteria will not be able to adapt to this, completely circumventing the problem of bacterial resistance.

Research Progress:

We hypothesised that opportunistic pathogens, such as *P. aeruginosa*, promote or take advantage of a mechanism of cell death (called NETosis), creating an infectious niche and causing inflammation in the lungs of CF patients. We aimed to test if NETosis of activated neutrophils (a type of white blood cell that is an important part of the immune system), could be prevented by induction of apoptosis using SMAC or BH3 mimetics. To test this hypothesis, we proposed a collaboration combining the capabilities of the Pellegrini Lab in the use of cell death inducing compounds against infections, with the extensive experience of the Bragonzi Lab with different CF and *P.aeruginosa* pre-clinical models.

In June 2022, I travelled to Milan to undertake research in the Lab of Prof. Bragonzi where I was trained in the preparation of *P. aeruginosa* for the establishment of both acute and chronic infection in mice. To model chronic infection in pre-clinical models, bacteria are embedded in agar beads, a multistep process requiring extensive knowledge about the ideal size and concentration of the bacterial beads, which are then inserted via intratracheal operation. The combination of these methods and extensive conversations with Lab members was invaluable for me to acquire the necessary knowledge to establish a model of *P.aeruginosa* infection at WEHI.

Furthermore, during my time in the Bragonzi Lab, we tested the SMAC mimetic LCL-161, and BCL-XL inhibitor A-1331852 in acute and chronic stages of murine *P.aeruginosa* infection. The experiments demonstrated that LCL-161 could significantly reduce viral burdens in chronic stages, which is incredibly promising, as no drugs currently reduce bacterial burdens during late chronic infection in mice.

We are currently establishing the pre-clinical *P.aeruginosa* model at WEHI and have all the tools to progress the project in Australia. Additionally, Prof. Bragonzi was also very interested in our expertise on COVID-19. There is little known about the host interaction of SARS-CoV-2 in CF patients, and we plan to extend our collaboration to better understand COVID-19 in CF using our unique mouse-adapted SARS-CoV-2 strain. My time in Milan established a solid foundation for a long-lasting collaboration between the Pellegrini and Bragonzi laboratories.

Additionally to my time in Milan, the Ann Maree Bosch Career Fellowship allowed me to visit a cell death conference (ECDO) held in Bonn, Germany, where I had the opportunity to discuss my research with other scientists and leaders in the cell death community. The 2022 ECDO was a vast source of the latest cell death research, providing me with the unique chance to learn from other researchers and allowing me to become involved in the European and worldwide cell death community, a great opportunity to foster further international collaborations.

Thank you to Cystic Fibrosis Australia for supporting this important research collaboration through the Ann Maree Bosch Career Fellowship.

Revised Budget

Item	Cost
Return flights	$ 3839.00
Accommodation	$ 3226.47
Transport – to/from airports and in-country	$ 1380.56
Meals and living expenses	$ 1439.62
Total	**$9919.26**

Signed: Date: 16/12/2022

Stefanie Bader