Dirofilariosis as a new zoonotic disease to control - analysis of the immune response induced by *Dirofilaria repens* worm.

Subcutaneous dirofialriosis is a disease caused by the parasite *Dirofilaria repens*. Mosquitoes transmit the parasite to the definitive host, mainly dogs, but also humans causing new infections. Until recently, dirofilariosis was considered an "exotic" disease, primarily occurring in warmer climates. However, due to climate change and human activity, it has spread to Central and Eastern Europe. Year by year, we can observe an increased number of dog and human infections, and Poland is already considered an endemic area of the occurrence of *Dirofilaria repens*. Managing this zoonosis is difficult as there are no evident symptoms of the infection and undiagnosed dogs become a new parasite reservoir, making it easier to spread to humans.

Parasites are known to have co-evolved with their hosts for millennia. The principal goal of the adult parasite is, arguably, not to kill the host but to survive and establish a chronic infection. Worms (helminths) developed multiple ways to regulate the host immune system. When the parasite enters the host body, the immune response starts to fight the parasite. But worms secrete unique molecules designed to modify the immune response so that it is beneficial to the parasite. Professional antigen-presenting cells (macrophages and dendric cells) are the main orchestrators of immune system reaction to pathogens. Therefore, they are the main target for the molecules produced by parasites.

Unfortunately, knowledge of the immune response and modulation induced during *Dirofilaria repens* infections is minimal, with no single research concerning immunomodulatory properties of parasite molecules. A better understanding of the interplay between host and parasite at the level of immune response orchestrators, mainly dendritic cells and macrophages, would be advantageous in light of potential anti-parasitic vaccine development in the future.
Thus, in our project, we will characterize the immune response induced by the *D. repens* and identify parasite molecules with immunomodulatory functions. We hypothesize that *D. repens* somatic antigen from various developmental stages would have different immunomodulatory effects on innate and adaptive immune responses, and the most successful immune modulator will be the L3 stage.
Therefore, the project aims to characterize the impact of parasite native somatic extract from different parasite stages (L3 larvae, microfilaria, adult) and ES20 (L3 stage-specific molecule) on the immune cells. In vitro analysis of the immunomodulatory function of *D. repens* molecules will be the first step for potential vaccine development.
In the project we will assess the impact of parasite molecules on immune responses using a combination of experimental approaches :

- human monocyte-derived human DCs (moDCs) and macrophages (moMACs) including DC-allogenic T cell co-culture and moDCs/moMACs phenotyping through transcriptomic, proteomic and FACS analyses
- mouse bone marrow-derived DCs and macrophages and co-culture with CD4+ OTII T cells to define the impact on the antigen presentation process
- infected/ non-infected dog PBMCs cytokine response, proliferation assays

The results obtained during the project will give new knowledge about parasite biology and molecular mechanisms involved in host-parasite interaction, which will allow for the development of an effective vaccine to combat the spread of this parasite in the future.