

## G. Non-Technical Summary (NTS)

*Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed):*

During adult life, blood cells are continuously produced by a population of unique blood cells that is generated during embryonic life. We know very little about the formation of these unique blood cells, termed stem cells. In the first part of the project, we will define the role of novel regulators of embryonic and adult blood stem cell formation.

Many blood cell diseases can be cured by injection of these unique blood stem cells in sick people. Sadly, the availability of these cells is often limited because these cells can only come from donation from healthy people. In the second part of the project, we will test for novel sources of these stem cells that do not depend on donation.

*What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?*

The work is interesting in its own right, and it will help efforts to understand haematopoietic development and the generation of blood stem cells. Blood stem cell transfer has been used in the clinic for many years now to treat multiple forms of blood cell diseases. Unfortunately, a severe limitation in treatment with blood stem cell remains the restricted availability of compatible donors. The efficient generation of blood stem cells would represent a remarkable progress, providing access to limitless sources for use in the clinic.

*What types and approximate numbers of animals do you expect to use and over what period of time?*

Mice will be used in this project. We anticipate using around 4500 animals during the 5-year period of this project licence.

*In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected levels of severity? What will happen to the animals at the end?*

The majority of the animals will undergo procedures to study blood cells; this includes blood withdrawal, immunization, irradiation and bone marrow transfer, injection of drugs affecting blood cell biology, genetic modification of regulators of blood cells. Aseptic techniques will be used to minimise potential infection and anaesthesia will be used when necessary to minimise the pain and discomfort from the procedure. The level of severity will be moderate for all these experiments. Adverse effects might include stress and mild pain from treatments such as injection and irradiation. All animals in these experiments will be closely observed for sign of poor health and behaviour. Supportive therapy such antibiotics and analgesics will be used to mitigate pain and distress or possible complications from the procedures. At the end of all experiments, animals will be humanely killed by an approved method.

## Application of the 3Rs

### Replacement

This programme of research is based on many experiments using cells *in vitro*. A large amount of pilot experiments and optimization are performed using these assays. As a result, this decreases greatly the number of experiments performed with animals. Unfortunately, no assays in a dish or a test tube are good enough to test blood stem cells. The regulation of blood cell production can also only be studied in animal and the specific structure of the body where blood cells develop cannot be replicated in a dish or a test tube.

### Reduction

We will ask for help from our statistician on a regular basis and particularly before starting new experiments. This will allow predicting the length and strength of treatments and how many animals are needed to answer the questions we are studying. We will mostly use experimental procedures that are well established. We will regularly discuss with people, at the university and in the larger community, who are experts in experimental procedures and animal well-being. We will use hygienic techniques and clean, disease-free, animals to help limit the numbers of animals required. The use of new technologies such as imaging blood cells in live animals under anaesthesia will also contribute to reducing the number of animal used.

### Refinement

Mice have been chosen for the study because they represent the least sentient species from which useful experimental data can be generated. Mice have considerable genetic and biological similarities to human with regard to blood cell formation. They are the best characterized specie to study the function of blood stem cells. Experimental protocols are particularly well established for this animal model. Data acquired over time using mouse models have translated well into clinical trials.