Project title: Epigenetic control of mammalian development and genome function
Duration of project - years: 5
Duration of project - months: 0

Purpose of the project (as in ASPA Section 5C(3)):
(a) basic research: YES
(b) translational or applied research with one of the following aims:
   (i) avoidance, prevention, diagnosis or treatment of disease, ill-health or other abnormality, or their effects, in man, animals or plants: YES
   (ii) assessment, detection, regulation or modification of physiological conditions in man, animals or plants: NO
   (iii) improvement of the welfare of animals or of the production conditions for animals reared for agricultural purposes: NO

(c) development, manufacture or testing of the quality, effectiveness and safety of drugs, foodstuffs and feedstuffs or any other substances or products, with one of the aims mentioned in paragraph (b): NO
(d) protection of the natural environment in the interests of the health or welfare of man or animals: NO
(e) research aimed at preserving the species of animal subjected to regulated procedures as part of the programme of work: NO
(f) higher education or training for the acquisition, maintenance or improvement of vocational skills: NO
(g) forensic inquiries: NO

Keywords:
Epigenetic, imprinting, genome, mechanisms, development

Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed):
Our genes are the DNA code that is the blueprint for life. Epigenetics is a layer of chemical marks that sits on top of our DNA that make our different cells (which all contain the same DNA and genes) behave in a specific way. For example, it makes liver cells behave like liver cells and brain cells behave like brain cells etc. Genes are present in two copies. One copy is inherited from the mother and one copy is inherited from the father. Some genes are epigenetically marked whereby only one copy is switched on depending on whether it is inherited from the mother or the father. These important processes contribute to mammalian development and their failure can influence human health.

The aim of the research in our laboratory is to understand the function of epigenetic marks and the DNA and genes that they regulate. We want to understand how they control growth and development of the baby, the placenta and the brain and how these genes regulate metabolic processes associated with diseases such as obesity and diabetes and how their altered regulation causes disease and aging. We also aim to understand epigenetic marks controlling the amount of gene product present in the body which when altered can cause cancer and other diseases. From our studies, we have discovered that the amounts of some genes is also involved in tissue regeneration. From these results we can investigate the regeneration process in our genetically altered animal models.
This project will generate genetically altered mice in which these processes are perturbed and compare them with normal mice. We wish to understand important epigenetic marks and how their control contributes to all stages of development. To help us to understand these mechanisms we will study the effects of these alterations on the well-being of the mice and their offspring. We will also study environmental factors which affect the epigenetic marks such as diet and aging. To complement our studies we will also use zebrafish, which have many of the same genes and pathways as mice but are simpler to study.

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 findings from our research will provide more information on how epigenetic marks contribute to normal processes in all stages of development from pre-implantation to perinatal to the adult. This includes the regeneration process which can occur in our cells following tissue damage. This information will also help us to understand how these processes are perturbed causing disease states such as obesity, diabetes and cancer. This in turn will help in the development of therapies in tissue regeneration and to target epigenetic changes that cause disease.

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

We will use two animal models to conduct our experiments; mice and zebrafish. Over a period of 5 years our projected use of mice is 18,700 and 20,500 zebra fish (larvae, juveniles and adults). These include a number of protocols and as we continue to work on our reduction and refinement we believe that we will work below these numbers.

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?

We have protocols that are classed as mild and moderate severity. However, many of our animals actually only experience a mild or less than mild severity level. New genetically altered mice strains that we generate are closely monitored for adverse effects. An adverse effect that they may encounter is restricted growth during development in the mother, which is sometimes recoverable after birth as the mouse gets older. We also study the regeneration of cells in animals. In mice we administer a substance into the muscle to cause cell damage. This is done using the lowest dose possible to achieve an effect and is performed under anaesthetic. The mice may only suffer from mild inflammation and they are expected to make a full recovery. It is this recovery process that we will investigate.

Application of the 3Rs

Replacement:

We cannot conduct all our experiments on cultured cells as we are assessing the health of the whole organism. Epigenetic states in vitro are also very different to their natural states in vivo. We are studying mechanisms and pathways in the developing organism and we therefore need to look at different time points during development. As cultured cells are exact copies of each other this means that changes occurring in developing cells cannot be seen. However we use in house and public databases and reanalyse existing data instead of rerunning experiments wherever possible. We perform initial experiments in cultured cells to test some of our hypothesis and choose important genes from these results before moving into animal experiments.

Reduction:

We have been working with mice for over 25 years and we have expertise in statistical analysis and optimal experimental design to determine the minimum number of animals needed to achieve robust, meaningful data. We collaborate with other groups, share mice tissues and data. We statistically analyse mouse numbers for use in experiments, plan experiments responsibly and communicate
between lab members to make the best use of our resources. We use databases to find candidates
genes and conduct studies first in cell lines whenever possible. We are collaborating with local
colleagues who are experts in theoretical and mathematical modelling who will provide added value
and novel insights to the animal work. Where appropriate we use control litter mates or use control
tissue from the same animal. Where appropriate we randomly assign animals to control and test
groups and analyse samples blind to avoid bias. Breeding colonies are generally kept small following
good colony management strategies.

Refinement:
The mouse is the best model for these studies because a catalogue of all the genes in the mouse
exists and there are well-established procedures that are not harmful for the animals which can be
used to mutate the genes or change their regulation. Many of the genes found in mice are found in
humans too. In addition we can breed the mice selectively and follow the effects in their offspring for
multiple generations.

We keep up-to-date with new technologies and developments that allow us to refine our experiments.
We continuously monitor our animals and work closely with the vet and the staff in the animal unit to
ensure the animals reach a humane end point and receive the best welfare possible.