Private & confidential: *Please be aware that the contents of this form may be made public resulting from the "Freedom of information Act". Personal details will not be released.*

G: NON-TECHNICAL SUMMARY (NTS)

Please attach the Non-technical Summary as generated by your application in ASPeL.

Project Title Zebrafish models of haemorrhagic stroke

Key Words Zebrafish, intracerebral haemorrhage, disease mechanisms, drug studies

Expected duration of the project 5 year(s) 0 months

Purpose of the project (as in ASPA section 5C(3))

Purpose

Yes (a) basic research;

(b) translational or applied research with one of the following aims:

Yes (i) avoidance, prevention, diagnosis or treatment of disease, ill-health or other abnormality, or their effects, in man, animals or plants;

No (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.

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

The overall aim of this project is to improve our understanding of the biology that is associated with brain haemorrhage - a type of stroke caused by bleeding in the brain.

Specifically, we aim to learn more about:

- The biological mechanisms that underlie certain risk factors of the disease.
- The biological processes that worsen injury in the brain after bleeding.
- The biological processes that allow the brain to recover following a bleed.

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)?

Our research hopes to find new ways of preventing and treating intracerebral haemorrhage - a type of stroke caused by bleeding in the brain. Although the less common form of stroke, it is associated with worse death and disability rates. It is responsible for almost 6% of deaths worldwide, therefore this represents a significant public health burden. To date, other than surgical intervention and blood pressure lowering, there are no drugs available for patients. We urgently need to improve our understanding of the disease biology if we are to identify new medicines for patients in the future. My research offers a new exciting approach for studying the disease and will provide clues into how we will one day treat haemorrhagic stroke patients.

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

55,000 zebrafish over 5 years.

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?

In this project, we will induce brain haemorrhage in zebrafish through genetic modification and/or exposure to specific drugs. We will treat some older animals (beyond the embryonic/early larval stages) with specific drugs to determine whether this can induce brain haemorrhage. Such drugs will target known risk factors for brain haemorrhage in humans, such as low cholesterol. This process could incur some moderate suffering/pain associated with brain haemorrhage that may be observed as erratic swimming. Any animals that exhibit unexpected effects, such as respiratory distress or an inability to feed, will be killed immediately using humane methods. Some animals will experience brain haemorrhage during the embryonic stages (at ~2 days post-fertilisation (dpf)). Our current data is showing that these particular fish begin to recover from brain injury at 5dpf. We will continue to monitor these animals over time (beyond 5dpf) to improve our understanding of the progression and recovery processes associated with brain haemorrhage. We will monitor live, anaesthetised animals under a microscope to study the responses of brain cells in real-time. We will also monitor their swimming behaviour over time. As these animals are undergoing a recovery process, we do not expect to observe many, if any, adverse effects when the animals are in their tanks. All animals will be killed at the end of the experiments using humane methods.

Application of the 3Rs

Replacement

State why you need to use animals and why you cannot use non-protected animal alternatives

Replacement

Although experiments using non-animal systems can be informative, these types of model cannot currently replicate the complex interactions that occur between blood vessels, brain cells and the immune system in intracerebral haemorrhage. The only way we can perform the necessary experiments to understand this intricate biological relationship is to use animal models– where the natural environment within the brain remains intact.

Historically rodents have been used to study ICH in-vivo. Zebrafish are a vertebrate of lower neurological complexity than mammals. As such, the use of zebrafish embryos and early-stage larvae (aged <5dpf) can be considered as a form of replacement of the existing rodent models of ICH. Furthermore, many of the experiments performed in this proposal will be performed on fish embryos and larvae (aged <5dpf), which we know experience minimal distress and recover quickly after ICH.

Reduction

Explain how you will ensure the use of minimum numbers of animals

Reduction

We know that zebrafish embryos/larvae experience minimal distress and recover quickly after ICH. As such, where possible, pilot experiments will be performed in zebrafish embryos/larvae prior to experimentation in older animals. Efficient experimental design and statistical techniques such as power analysis will keep the number of protected animals used to a minimum.

Refinement

Explain the choice of animals and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.

Refinement

Zebrafish is the vertebrate of lowest neurological complexity that can be genetically modified to study the processes of interest. Any genetic/pharmacological manipulation that induces ICH phenotypes in zebrafish that are shown to interfere with feeding or respiration, or inducing significant behavioural or other physiological abnormality will result in immediate termination of the animal concerned and other animals sharing the genotype