G. NON-TECHNICAL SUMMARY (NTS)

NOTE: The Secretary of State considers the provision of a non-technical summary (NTS) is an essential step towards greater openness and requires one to be provided as part of the licence application in every case. You should explain your proposed programme of work clearly using non-technical terms which can be understood by a lay reader. You should avoid confidential material or anything that would identify you, or others, or your place of work. Failure to address all aspects of the non-technical summary will render your application incomplete and lead to it being returned.

This summary will be published (examples of other summaries can be viewed on the Home Office website at www.gov.uk/research-and-testing-using-animals.

Word limit; 1000 words

Project Title	Gene regulation of cardiovascular disease
Key Words	Angiogenesis, VEGF (Vascular Endothelial Growth Factor), Neuropilins, Cardiovascular Development, Cardiovascular repair and regeneration, Cardiometabolism, Diabetes
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;
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.

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

This project aims to identify molecules and mechanisms that play important roles in blood vessel formation and the development of cardiovascular diseases, and to develop approaches such as gene or cell therapy that can effectively target these molecules to achieve a therapeutic effect in human cardiovascular disease. Cardiovascular disease is one of the major causes of death in the developed world and is rapidly increasing in developing countries. However, the mechanisms that cause, or protect against this disease are poorly defined, and there is a continued need for new therapeutic approaches. The work planned under this project licence will lead to the identification of molecules and mechanisms with important roles in heart disease, disease-related angiogenesis and cardiometabolism thereby potentially discover new therapies or therapeutic targets.

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)?

This work will improve understanding of mechanisms and the key molecules involved that maintain cardiovascular health. VEGF and Neuropilins-linked signalling pathways, which are the focus of this application, are already known to be important for human cardiovascular health and in human diseases such atherosclerosis. Since many of these mechanisms and molecules are conserved between vertebrate species, the work proposed here will have direct relevance for analogous process and disease states in humans. This work will therefore advance knowledge and understanding of important processes underlying human health and disease. Furthermore, by identifying key novel molecules in these processes we will be able to identify novel targets for the development of therapeutic drugs, which may lead to the development of novel therapies for heart disease and vascular disease.

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

We anticipate to use no more than 5,000 mice and 10,000 zebrafish over the course of the 5 year PPL.

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?

Our protocols are based on well-established procedures that have already gone through a considerable amount of refinement. Most animals will not undergo procedures that will inflict harm. Instead these animals will be used for phenotyping the effects of mutations using minimally invasive imaging or analysis. Some animals will undergo procedures that include minor damage to the lining of a small region of a single artery or ligation of an artery in the mouse that will restrict blood flow to the hindlimb, or in the zebrafish, injury to a small

region of the heart or in complete resection of the caudal fin. Based on our experience, adverse effects are anticipated to be very limited in all our protocols and where they do occur to be very brief in duration. Adverse effects that may occur in rodents include lethargy, hunched posture, loss of appetite, weight loss, and in fish, difficulty breathing, abnormal colouration, abnormal swimming, feeding or schooling behaviour. All our protocols, have a severity level of mild or moderate. All animals will be humanely killed at the end of the relevant protocol, and/or when signs of discomfort or pain are manifested. All animals undergoing surgical procedures are expected to recover quickly and will be given appropriate painkillers and post-operative care. At the end of a procedure, animals will be killed by a humane method and tissues taken for analysis after death. All animals will be humanely killed at the end of the relevant protocol, and/or when signs of discomfort or pain are manifested. All animals undergoing surgical procedures are expected to recover quickly and will be given appropriate painkillers and post-operative care. At the end of a procedure, animals will be killed by a humane method and tissues taken for analysis after death.

Application of the 3Rs

Replacement

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

Replacement

While cell culture models have been helpful and we continue to use them extensively, there are no computer, tissue or cell culture models that successfully mimic human cardiovascular disease or angiogenesis. Two major reasons for this are: these diseases develop in complex multi-tissue environments in living animals, which cannot be mimicked by non-animal models; they occur over long time periods which make it difficult to perform similar studies in non-animal models.

Reduction

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

Reduction

Where necessary, pilot studies involving small numbers of animals will be performed to establish the proof-of-concept, and only if these small studies are encouraging, will we proceed to larger studies. Since protocols are already well-established in the chosen species, the minimum numbers of animals needed can be determined more accurately, and unnecessary pilot work can be avoided. Studies will be performed only using animal numbers sufficient to produce statistically robust results.

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

Animal species were chosen mainly because protocols were established in those species, avoiding unnecessary pilot work. Small rodents (mice) were chosen, as these are the simplest appropriate mammalian organisms. The choice of mouse and fish is determined by the unique ability to genetically alter these species. Use of zebrafish allows us to perform studies wherever possible in simpler vertebrate organisms.