G: NON-TECHNICAL SUMMARY (NTS)

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

Project Title, Purpose & Duration	New Therapeutic Approaches for Inflammatory Disorders Basic Research 5 year(s) 0 months
Key Words (max. 5 words)	Inflammatory diseases. New therapies. Rheumatoid arthritis.
Describe the aims and objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)	• Identify efficacy of test drugs and any side effects not predicted by cell culture based model systems. • Study the effects of the drug on the body, and also the effects of the body on the drug. • Demonstrate that anti-inflammatory activity can be shown at specified doses. • Identify the best arthritis models to use pre-clinically that correspond with a specific therapeutic target. • Test a drug's efficacy to affect leucocyte recruitment and identify associated mechanisms that may be shared with other autoimmune diseases.
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 target of this programme of work is to provide pre-clinical supporting information for clinical trial applications. A drug requiring evaluation will be supplied to us by clients in the biotech and pharmaceutical industry, along with evidence supporting the rationale for testing the agent. We aim to investigate the efficacy and mechanism of action of a drug to help our clients make a more informed decision on whether to proceed into clinical trials. This reduces the risk of later stage failures and hopefully predict on side effects associated with a particular therapy. Information supplied by us will speed up the clinical trial process and make it less financially prohibitive. The benefit is, therefore, a reduced number of unproductive human volunteer studies (and a reduced risk of adverse effects) and most importantly the development of improved and more effective therapeutics targeting inflammatory joint diseases and potentially other inflammatory and/or autoimmune diseases with shared mechanism of action. The benefit to patients will be the identification of new anti-inflammatory drugs. This programme of work will help identify the best potential drugs early in the drug development process or aid in refining drugs that have not been efficacious in the clinic due to poor historic efficacy data and pre-clinical design.

What types and approximate numbers of animals do you expect to use and over what period of time?	We would expect to run approximately 100 studies on behalf of sponsors using approximately 4000 mice and 1000 rats over the 5 year duration 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 level of severity? What will happen to the animals at the end? | Animals injected with inflammatory stimuli to induce an arthritic disease state will, over time, develop inflammation in their hind and front paws which affect the digits, footpad area and sometimes the ankles. Joint swelling is an expected outcome of this protocol and is a primary measurement of disease progress or disease regress with a potential therapeutic. Animals may suffer from discomfort associated with inflamed joints but will be monitored daily for any additional, but unlikely, signs of discomfort associated with an arthritis diseased state such as laboured breathing, ruffled fur, weakness, diarrhoea/dehydration or a hunched appearance. These symptoms very rarely appear with such models and therefore the risk of arthritis significantly affecting an animal's wellbeing is not expected. General assessment of pain is not accurate but includes close monitoring of the animal's behaviour and feeding, the use of facial expression scoring system (mouse grimace scale) and responses upon handling. As this is a moderate protocol, it should not necessitate the use of pain relief (analgesia), further, analgesics by nature have anti-inflammatory effects which compromise on the underlying pathology of the disease, rendering the testing of potential anti-inflammatory therapeutics and the resultant need to use animals for the assessment of new therapies futile. General welfare checks and humane endpoints will be observed at all times. Whilst no weight loss is associated with arthritis models, any animal which has lost 15% of its body weight will be monitored, and if this weight loss is combined with any other signs of discomfort mentioned above, the animal will be removed from the study. If the animal loses 20% of body weight, this is an endpoint and it will be removed from the study. Advice from the resident veterinarian or senior animal technician will be sought if a mouse fails to put weight on one of its limbs for more than 1 week in order to assess the well-being of the animal and decide on a humane end point.

The white blood cell (leucocyte) migration models are short term models where the injection of an inflammatory stimulus recruits cells to the site of injection. There is no expected peripheral effects associated with these models but, depending on the route of administration, there may be low-grade systemic inflammation. However, this is not expected to affect the wellbeing of the animal due to the length of these models. Animals will be monitored regularly for any unusual signs of discomfort such those mentioned above. Such symptoms very rarely appear with leucocyte migration models and therefore the risk of significantly affecting an animal's wellbeing is not expected. Any animal which has lost 15% of its body weight will be monitored, and if this weight loss is combined with any other signs of discomfort mentioned above, the animal will be removed from the study. If the animal loses 20% of body weight, this is an endpoint and it will be removed from the study.

In all of the protocols and models described in this application, we plan to provide as much data as possible from every animal. This includes in-life assessment of disease progress (e.g. Manual measurement of joint swelling or imaging disease progress) as well as post-mortem and ex vivo analyses of whole organs and the cells/factors associated with an inflammatory response which may be specific to a particular organ (e.g. the local lymph nodes) or are systemic (blood, spleen, other organs). When possible and when confidentiality of data is not an issue, we aim to publish our results in peer reviewed journals and scientific conferences. |

Application of the 3Rs	
Replacement State why you need to use animals and why you cannot use non-protected animal alternatives	The programme requires that the models used are ones which closely mirror human disease. All compounds to be tested would have previously been screened in relevant in vitro models to determine those candidates suitable for in vivo testing. Rodents (rats and predominantly mice) are suitable for these studies as the work cannot be conducted in lower vertebrates, invertebrates or cell lines due to the poor resemblance of these options to the clinical setting. Animal models address issues which current in vitro tests cannot accurately determine.
Reduction Explain how you will ensure the use of minimum numbers of animals	Animal models will be restricted to the minimum number needed for a statistically valid result. The number of animals used will be the minimum safely necessary to allow meaningful statistical analysis of the data generated. The most important aspect of the proposed programme of work that will reduce the number of animals used is careful selection of drugs, on the basis of preclinical and in vitro data. Only those potential drugs that offer a realistic prospect of therapeutic exploitation will be investigated. Most importantly, in the past 5 years, we have validated and established several non-animal based platforms that allow a potential client to test a particular aspect of their drug, such as toxicity, the mechanism of action and / or target cell type. Such platforms are either cell culture based, relying on commercially validated and available immortal cell lines or human blood, artificial 3-D tissue equivalents, or more physiological platforms which are based on consensually or ethically derived human tissue. In fact, by installing such assays, I have managed to reduce the contract expectation under this programme from 100% use of animals (forecasted 5 years ago) to 40% (based on contracts from 2012 until today). The investment of a flow cytometer analyser 4 years ago allowed for a more thorough assessment of the inflammatory pathways and cells associated with disease, thus bolstering statistical significance by offering additional readouts of drug efficacy and reducing the number of animals required. We have also recently acquired small animal imaging technology which may allow for monitoring of disease development in each animal over time, abrogating the need to humanely kill satellite groups to examine disease progress internally, and thereby reducing total animal numbers. There are technical challenges and prohibitive cost-implications associated with the use of this technology in this programme of work, but where and when we can, I aim to validate its use in animal models of arthritis in future.

Refinement	Rodents provide a cost and time effective platform in general for most pre-clinical testing. For the purposes of drugs targeting inflammatory pathways, the use of higher species is not required because there is a wealth of knowledge on different types of models in rodents, as well as historical in-house expertise with such models. Internal expertise, and more recent technological advances, such as the use of whole body imaging and a flow cytometer, can allow for a more refined study design that will minimise the number of animals needed to achieve statistical significance. These techniques should maximise output and provide a more thorough assessment, with an aim to help in selecting the best models. Certain aspects of disease assessment, particularly in the in-life phase, are fairly subjective. Therefore, there is a demand to standardise and refine this. The use of the imager has potential to not only be beneficial in further assessment of the disease, but also in providing more measureable and standardised outcomes of disease progression, and the subsequent valuation of a therapeutic.
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.	