Home Office

NON-TECHNICAL SUMMARY

Provision of an outsourced drug discovery platform for the development of therapeutic drugs for neurological disorders

Project duration

5 years 0 months

Project purpose

- (a) Basic research
- (b) Translational or applied research with one of the following aims:
  - (i) Avoidance, prevention, diagnosis or treatment of disease, ill-health or abnormality, or their effects, in man, animals or plants
- (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 following aims mentioned in paragraph (b)

Key words

neurodegeneration, stroke, brain, neuron

Animal types	Life stages
Rats	adult
Mice	adult

Retrospective assessment

> The Secretary of State has determined that a retrospective assessment of this licence is not required.

Objectives and benefits

Description of the projects objectives, for example the scientific unknowns or clinical or scientific needs it's addressing.

What's the aim of this project?

The aim of this project is to test the efficacy of drugs (how well they produce a desired effect) designed for use against human diseases of the brain, such as Alzheimer's and stroke.

Potential benefits likely to derive from the project, for example how science might be advanced or how humans, animals or the environment might benefit - these could be short-term benefits within the duration of the project or long-term benefits that accrue after the project has finished.

Why is it important to undertake this work?

Neurological disorders involve damage to the brain, spinal cord or nerves; they can be progressive or sudden, intermittent or unpredictable. They can cause significant burden to the patient, their families, and society as a whole; 14% of the social care budget in England is spent on people living with neurological conditions, so it is a group of conditions that need significant investment and patient care.

Conditions involving neurodegeneration (when brain cells are progressively or immediately damaged or lost) are particularly devastating for older people. The leading cause of death in the UK in 2018 was dementia and Alzheimer’s disease, accounting for 12.7% of all registered deaths (Office for National Statistics; ONS). This is expected to rise year-on-year as the population grows and lives longer. Patient quality of life is usually affected more as the degeneration progresses. Life expectancy changes with age of diagnoses, but on average 4.5 years is the median (average) for survival from onset of dementia.

Ischemic stroke is where a blockage (a blood clot) causes a sudden loss of blood supply to an area of the brain, and accounts for 85% of strokes. A stroke can cause various changes to movement, speech, or cognition (the way your brain understands, organises and stores information). The number of deaths through ischemic stroke have almost halved in the last twenty years, most likely through greater wider public knowledge of spotting early signs and therefore providing early intervention. Despite this, the incidences of stroke are set to rise due to an aging population; age-related diseases, such as heart disease or diabetes can increase the risk of having a stroke. Furthermore, quality of life for stroke patients is significantly affected and, in a large proportion, permanently life-changing with the significant risk of a second stroke also a factor in the continued management of the disease in these patients.

Our work aims to use animals, in combination with experiments that take place in the laboratory, to support the development of new treatments for neurological disorders and address the unmet clinical

needs for treatments. The use of animals is currently an essential part in the process of developing new treatments and enables us to replicate complex aspects of the disease in the entire biological system. For neurological disorders this includes the interaction of brain cells directly affected by damage, with other nearby cells, and the effect on the animal as a whole (for example how its movements are changed by the disease). They enable us to test the effect of new treatments in a relevant and intact biological system. This work will enable key decisions to be made regarding which treatments will continue to be developed, and ultimately, which will make it into clinical trials, where they are tested in humans. Our approach to the work ensures that this is done in the most efficient way possible, and that the benefit gained from every animal is maximised.

What outputs do you think you will see at the end of this project?

Work carried out under this licence will involve the testing of new drugs to treat neurological disorders (diseases of the brain) in a way that is closely related to human disease. As part of this project we expect to test the effects of at least 5 new drugs for neurodegeneration (diseases like Alzheimer's), and at least 3 new drugs for treating stroke. Our clients are experts in developing their potential (candidate) drugs, and our bespoke studies that closely replicate both disease and potential ways of treating the disease. Combining these two approaches, the candidate drugs have significant potential to enter clinical trials. The way the drug works (mechanism of action), or the kinds of biological processes we look at (targets) will be different depending on the client we have, but the aim of treating the specific diseases will remain the same.

This project will provide important information to progress new treatments through the phases of drug development. New test agents will be evaluated for their ability to treat animals with symptoms that closely match those in human neurological disorders. The information gathered will enable us to identify the most appropriate treatments to take forward to human clinical trials. The information gathered will also enable us to quickly determine which drugs should not be progressed any further.

In addition, this work will increase our knowledge of how new drugs work and will help us to identify changes in the body that occur in response to the drug. We can use this information further down the line to monitor responses in humans during clinical trials.

Data from studies under this project may be used to support patent applications and applications by clients for additional funding. Data produced may also support the design of regulatory studies for clients.

Who or what will benefit from these outputs, and how?

This project will generate important data in the development of new drugs. To assist with this, we will utilise our test agent dosing project licence to allow us to understand how drugs distribute in the body after dosing, how quickly they are eliminated, and the dosages that are well tolerated in rodents. This will aid in the design of dosing strategies for studies under this licence. Ultimately, work carried out under this licence is expected to result in the progression of new neurological disorder treatments through various stages of drug development and ultimately into the clinic to treat patients.

Our focus on a science-led approach will enable key decisions to be made at each development stage on whether a test agent is likely to become a successful drug. This allows unsuitable drugs to be

abandoned at an early stage and enables us to use the fewest number of animals possible per drug development programme. The identification of test agents as unsuitable for use in humans at an early stage of development will ensure a better success rate in the drug discovery process than has been seen previously in the pharmaceutical industry. Ultimately, this project will contribute to the successful development of new drugs, which will benefit patients with various neurological disorders who may otherwise have their quality of life greatly reduced by their condition.

How will you look to maximise the outputs of this work?

All studies are designed such that the outputs from each animal are maximised. Expert knowledge is gathered not only from within the preclinical (animal) team performing the animal studies, but from other teams within our company, or our clients' companies. This ensures that all relevant work that has been performed in the laboratory is taken into consideration when designing animal studies. The in vitro (in the test tube) and bioanalysis teams at our company are experts at analysing tissue and blood samples collected from animals, and they help with details of sample collection and storage to ensure that the samples are collected and stored in the best way possible. They are also experts at working with small quantities of samples, particularly small volumes of blood samples, meaning that they can often analyse lots of different biomarkers (a measurable indicator of a disease state or other physiological state) and test agent levels from each animal.

In addition, we will seek expertise from our established networks, to ensure that we make use of any new knowledge or incorporate better methods of performing animal studies. We will also use these networks to provide information and training to others on the models and techniques we use in our research. We will maintain good communication with managers of the animal facilities to ensure that any tissues from animals being killed that are not required for our work can be made available to other researchers if suitable.

Although there are times where we will not be able to share animal model information (for example, where it would put us at a competitive disadvantage), we aim to publish or share our findings (especially control data) wherever possible. Likewise, we hope to pass on any 3Rs progress we make in real time to colleagues, but this will ultimately be available locally to other researchers through the AWERB retrospective review for this project.

Species and numbers of animals expected to be used

- Mice: 300
- Rats: 800

Predicted harms

Typical procedures done to animals, for example injections or surgical procedures, including duration of the experiment and number of procedures.

Explain why you are using these types of animals and your choice of life stages.

Mice and rats are the most common type of animal used for developing new treatments for neurological diseases. These rodents are well characterised, meaning that a lot is already known about how their bodies work, and the techniques used to mimic the human diseases are well developed. This means that for the brain diseases we wish to study, using rats and mice will produce data that is very close to how the diseases behave in humans.

Adult rats and mice will be used for the work outlined in this project licence as we want the biology of the animals to be fully developed as this will better represent the patients we aim to treat. Both rats and mice are needed are needed for this project; each species has different characteristics that suit different types of experiments (for example rats are better suited to studying behaviour). Also there are sometimes differences in how each species reacts to the substances used to bring about the diseases (for example one drug we use is tolerated by mice but is toxic to rats).

Typically, what will be done to an animal used in your project?

The majority of animals will be part of studies that aim to test whether new drugs can be used to prevent or halt neurological damage associated with stroke or neurodegeneration. Therefore, each animal will be given some kind of new drug at some point during a study, either once or several times. Drugs will be administered more commonly intraperitoneally (into the body cavity), subcutaneously (under the skin) or orally (by mouth), and less frequently intravenously (into a vein), via a slow-release device under the skin, or very rarely into the nose. For intraperitoneal, subcutaneous and oral administration, mice will be held securely by a trained researcher for the dose to be administered. For intravenous dosing, mice will be placed briefly in a specially designed rodent restrainer (a ventilated Perspex tube where the animal can be observed), the tail will be warmed using warm water or more commonly by being placed into a warming cabinet to dilate the blood vessels and make them easier to see before administering the dose. For intranasal dosing, mice will be briefly anaesthetised and placed onto their backs to allow the dose to be administered directly into the nostrils.

The dosing of new drugs should not cause significant harm or distress overall; dosing might be once a day for a week, or once a week and cause only brief discomfort each time.

All animals will either have surgery on one occasion or be given certain chemicals to induce the disease we will be studying. Some of these animals will have the surgery but not have the vital step; these will act as control animals (to show what would have happened if they experienced all the steps without inducing the disease). Surgery is performed under anaesthesia (the animals will be asleep for the duration of the surgery and will feel no pain). Recovery from the types of surgery in this licence should be unremarkable. Most importantly, anything administered or surgery to induce disease should only cause minor impairments in the way the animal moves, only detectable by special tests.

Most of the animals will undergo some tests to assess how well they can coordinate their movements to complete tasks such as grasping for a treat, or removing a piece of tape from the top of their paw. Sometimes, tests looking at more general movements, such as grip strength, balance, and gait analysis (how the animal walks) will be used. These tests will generally not cause any stress or discomfort.

Sometimes, taking blood samples may be necessary to look at levels of indicators of disease (biomarkers) or how much drug is in the blood. Blood samples are usually small in volume and are

taken from a superficial (near the skin surface) vein.

At the end of the studies, most animals will be put under terminal anaesthesia (made unconscious permanently) while a large blood sample is taken, and a stain or fix is injected to mark the brain for further analysis ex vivo (in the lab). The animals will be humanely killed while still under anaesthesia.

What are the expected impacts and/or adverse effects for the animals during your project?

In most cases neurodegeneration will be induced under anaesthesia (while unconscious) if surgery is needed to block blood vessels or give chemicals directly into the brain. Sometimes chemicals may be administered into the body through common injection routes previously detailed (only transient discomfort) if it is known to reach the brain. Stroke will always be induced under anaesthesia.

The symptoms felt by inducing neurodegeneration or stroke, after recovery from any surgery, will be very minimal, and should only be noticeable by special tests. Some of these tests may cause temporary anxiety while the animals get used them, but overall the tests will not cause any lasting harm.

Sometimes the chemicals used to induce neurodegeneration might cause abnormal movements, but these are expected to be mild and will not cause lasting stress to the animal.

The test drugs given are not expected to cause lasting harm, although sometimes the animal may lose some body weight while adjusting to them. Blood sampling is not expected to cause any lasting discomfort.

Expected severity categories and the proportion of animals in each category, per species.

What are the expected severities and the proportion of animals in each category (per animal type)?

Mice:

Mild 35%

Moderate 65%

Rats:

Mild 20%

Moderate 80%

What will happen to animals at the end of this project?

- Killed

Replacement

State what non-animal alternatives are available in this field, which alternatives you have considered and why they cannot be used for this purpose.

Why do you need to use animals to achieve the aim of your project?

The work to be performed in this project cannot be fully replaced by using models in a laboratory. Wherever possible, work is performed either using cells (a large proportion of a typical work program will be done in tests done with cells grown in the lab) or tissues taken from a dead animal before moving into live animal studies. This is to make sure that the animal studies are well designed and that the maximum amount of data is gained from each animal study.

Which non-animal alternatives did you consider for use in this project?

Our company regularly uses a range of in vitro (taking place in a test tube in the laboratory dish) methods utilising cells to understand how a novel test agent might affect the cellular functioning of those cells. From these experiments we can prioritise test agents and only take forward those that have the desired effect, and therefore those that look the most promising for the treatment of neurological disorders.

Why were they not suitable?

Cell-based methods are useful to test the impact that novel test agents have on a cellular level, for one particular type of cell, but they do not model how cells interact with other types of cells and organs, which is particularly important with how brain cells communicate. In addition, cell-based methods do not test the effects that the body might have on a test agent, for example, how it is absorbed, distributed and removed from the body, all of which can alter how effective the test agent might be. Testing a new drug in a well-understood, whole biological system enables us to study the complex interaction in the body, as well as monitoring how the test agent performs in a whole animal. None of the alternatives mentioned can replicate this, though cell-based testing enables us to prioritise drugs and only take the best candidates forward for further development.

Reduction

Explain how the numbers of animals for this project were determined. Describe steps that have been taken to reduce animal numbers, and principles used to design studies. Describe practices that are used throughout the project to minimise numbers consistent with scientific objectives, if any. These may include e.g. pilot studies, computer modelling, sharing of tissue and reuse.

How have you estimated the numbers of animals you will use?

The number of animals to be used has been estimated by analysis of the number of animals used on previous projects, looking at the number of animals required for each type of study. This was then combined with a prediction of likely demand of future projects to give the numbers in this project.

What steps did you take during the experimental design phase to reduce the number of animals being used in this project?

We have extensive experience in the design of experiments of the types in this project, which has given us confidence in the number of animals required to ensure that no animals are used unnecessarily, but also that the data generated is reliable.

We regularly refer to the PREPARE and ARRIVE guidelines and make use of the NC3Rs Experimental Design Assistant to ensure that we are using the correct number of animals for every study.

Where information is not available in published literature or from contact with other researchers, pilot studies in a small number of animals will be used first where appropriate to assess the action of test agents, and well as the variability in replicating the disease.

What measures, apart from good experimental design, will you use to optimise the number of animals you plan to use in your project?

Data from pilot studies and previous experience are used to ensure that the numbers used are both as low as possible, but also large enough to generate reliable data. Within our company, a member of the wider team has generated a tool for performing power calculations (a way of telling whether the number of animals in a study is enough to produce data that can be trusted) and can be consulted as necessary to assist with study design.

Sometimes animals are produced on our breeding project that don't have mutations (wild-type) that would be suitable for this project, so these animals will be used whenever possible. This will reduce the need for obtaining animals externally. Where animals are obtained from external sources, only the number of animals required for the study will be purchased or imported.

Wherever possible, our in vivo (animal work) scientists will be blinded to the treatment status or even which animal has had which kind of surgery, reducing bias. This enables more reliable information to be gathered from a smaller number of animals.

Baseline data (e.g. bodyweight) are recorded and animals spread across treatment groups to ensure there are no differences between the groups at the start of the study (pseudo-randomisation).

Good planning ensures that within any series of studies we can control for variability that might be introduced. To limit this variability we look at using animals of a similar age/weight range, testing different batches of test agent in the lab first, using the same source of reagents (chemicals used during the experiments), keeping records of observations made and standardising as many components of an in vivo model as is possible.

Refinement

Give examples of the specific measures (e.g., increased monitoring, post-operative care, pain management, training of animals) to be taken, in relation to the procedures, to minimise welfare

costs (harms) to the animals. Describe the mechanisms in place to take up emerging refinement techniques during the lifetime of the project.

Which animal models and methods will you use during this project? Explain why these models and methods cause the least pain, suffering, distress, or lasting harm to the animals.

Animals will be housed in a purpose-built, state-of-the-art facility, that is specific pathogen free (SPF), which excludes certain disease-causing organisms such as bacteria, viruses and parasites that would compromise the health of the animals and the quality of the study output (how good the data is). They will have access to food, water and items that enhance their environment, such as tunnels, chew sticks and mezzanine levels to climb on. Our company staff and the animal care staff are competent in rodent welfare and will ensure that animal suffering is minimised. We aim to house mice and rats in social groups to promote normal behaviour. However, aggressive behaviour can occasionally result in animals being singly housed to prevent injury.

All animals will be wild-type; this is because the human diseases will be mimicked by making changes to the brain, either with changing blood flow to different areas, or by giving chemicals that change how the brain functions.

Why can’t you use animals that are less sentient?

Adult rodents such as rats and mice are the lowest species of mammal that allow us to adequately study the complexities of human neurological disorders. Mice are used wherever possible for studies not involving behavioural tests more suited to rats. Due to the need to conduct simple behavioural tests to assess how the animals move and behave, terminal anaesthesia is not a possibility. It is also important that we are able to monitor the behaviour of the animals both after the neurological disease is induced, and while giving test agents as this allows us to monitor for adverse reactions.

How will you refine the procedures you're using to minimise the welfare costs (harms) for the animals?

Husbandry (animal care) after procedures involving surgery is very important; we will make sure that the animals are kept as comfortable as possible after surgery, and will include pain relief, soft bedding, and a warm cage to help recovery. In-depth daily monitoring of body weights and general condition will be in place for approximately 10 days after surgery. Where possible, pain relief will be given by putting it into a palatable food like Nutella; this means that the animals will take the medication by choice rather than an injection.

When blood sampling or dosing, we will use the smallest needle size possible to minimise any pain and distress to the animal. We also give drugs via drinking water instead of by mouth where possible.

Where it is hard to predict how a drug will affect an animal, a tolerability or safety study will be performed beforehand on our related project licence to ensure that the drug is tolerable for long-term dosing in our surgical animals.

When testing how the animals move, they will be trained to get used to the tests before data is recorded. This reduces anxiety over the course of the tests. Likewise, animals will be acclimatised to

(allowed to get used to) new environments and handling before the studies start.

The choice of strain of rat or mouse will be considered carefully for each study. Based on published work and our own experience, we will consider how each strain is more or less susceptible to different chemicals or potential treatments. Likewise, we will consider the known behavioural characteristics and husbandry needs of the strain; for example, how easy it is to train a rat to perform the behavioural tests because they are naturally less stressed.

What published best practice guidance will you follow to ensure experiments are conducted in the most refined way?

We follow the PREPARE guidelines for the planning of studies (Smith et al., PREPARE: guidelines for planning animal research and testing. 2020. Laboratory Animals). LASA (Laboratory Animal Science Association) also has a range of published guidance documents with principles that can be applied to our animal studies which are found at https://www.lasa.co.uk/current_publications/.

Specific to ischemic stroke, we will refer to the IMPROVE guidelines where possible. Sert NP et al (2017) The IMPROVE Guidelines (Ischaemia Models: Procedural Refinements Of in Vivo Experiments). JCBFM 37(11): 3488-3517. doi: 10.1177/0271678X17709185.In addition we will also refer to the following when conducting our work:

Prescott MJ, Lidster K (2017) Improving quality of science through better animal welfare: the NC3Rs strategy. Lab Animal 46(4):152-156. doi:10.1038/laban.1217

LASA 2017 Guiding Principles for Preparing for and Undertaking Aseptic Surgery. (E Lilley and M. Berdoy eds.). http://www.lasa.co.uk/publications/

Smith D, Anderson D , Degryse A, Bol C, Criado A, Ferrara A, Franco NH, Gyertyan I, Orellana JM, Ostergaard G, Varga O, Voipio H (2018) Classification and reporting of severity experienced by animals used in scientific procedures: FELASA/ECLAM/ESLAV Working Group report. Lab Animal 51(1S): 5-57. doi: 10.1177/0023677217744587

How will you stay informed about advances in the 3Rs, and implement these advances effectively, during the project?

When designing animal studies we consider the appropriate guidelines, including the guidance from LASA, the NC3Rs, and the PREPARE guidelines. This guidance will influence our study design.