Home Office

NON-TECHNICAL SUMMARY

Brain mechanisms underlying cognition and emotion

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.

Key words

No answer provided

Retrospective assessment

> The Secretary of State has determined that a retrospective assessment of this licence is required, and should be submitted within 6 months of the licence's revocation date.

Reason for retrospective assessment

This may include reasons from previous versions of this licence.

- Required at inspector’s discretion

Objectives and benefits

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

What's the aim of this project?

The overarching aim of this project is to identify the neural, genetic and neurochemical circuitry in the brain that underlies the cognitive and emotional impairments that are important symptoms of psychiatric disorders such as depression, anxiety and schizophrenia. This includes identifying how genes and physiological stressors impact upon the development and subsequent functioning of this circuitry **in adulthood**, how this affects cognitive and emotional processes, and how current therapies (ie antidepressant drugs) interact with this circuitry to treat these symptoms. These are important questions because over 40% of patients suffering from neuropsychiatric disorders are not helped by current therapies for reasons that are unknown, and when the therapies are effective, we don't understand why and thus can't predict which patients will do well on which treatments. This severely limits treatment options and treatment development. It is recognised that this is because we have very little understanding of the different brain mechanisms that can cause these symptoms, and until we understand how the neural, genetic and neurochemical circuitry within the brain contributes to the normal and symptomatic cognitive and emotional processing we will not be able to improve treatment strategies for the sufferers of psychiatric disorders.

A retrospective assessment of these aims will be due by 02 November 2023

The PPL holder will be required to disclose:

- Is there a plan for this work to continue under another licence?
- Did the project achieve it's aims and if not, why not?

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.

What are the potential benefits that will derive from this project?

This project will provide a basic understanding of how some frontal brain areas contribute to a variety of cognitive and emotional behavioural impairments (for example, compulsivity, anxiety and loss of sensitivity to rewards) that are common in patients with neuropsychiatric and neurodegenerative disorders. It will provide an understanding of how damage to different brain mechanisms contributes to the different cognitive and emotional processes that cause these impairments, such as problems in switching attention away from negative stimuli or problems in predicting when negative events may occur. By identifying the underlying psychological, neural, genetic and neurochemical causes this will not only help stratify patients but also improve their chances of getting personalised therapy. For example, if you are anxious because you find it difficult to switch attention away from negative things due to dysfunction in one region of prefrontal cortex, this will require different treatment than if you are anxious because you can't predict when negative things will happen due to dysfunction in a different part of the prefrontal cortex. It is this basic knowledge that is currently lacking. Thus, **understanding how** the different brain circuits, that mediate different aspects of such psychiatric symptoms, **interact in development and adulthood** will help us to identify particular symptoms, and combining it with

information about how particular therapies interact with such circuits, eventually target existing therapies more effectively.

Species and numbers of animals expected to be used

What types and approximate numbers of animals will you use over the course of this project?

We expect to use approximately **415** marmosets over 5 years

Predicted harms

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

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?

All animals are housed in either stable male/female pairs or live in their family groups in cages that exceed the UK and EU guidelines and contain an extensive array of environmental enrichment aids. Animals may occasionally be housed without a partner in the event of an argument with their cage mate or if their cage mate is humanely euthanased upon study completion (e.g., in cases where timing of brain assessment is critical). A new partner will be provided at the earliest possible opportunity, depending, e.g., on availability of opposite-sex partners, experimental status, etc. A typical study lasts between 18 months to 2 years. During that time marmosets are likely to receive behavioural testing 5 days a week in the home cage or specialised testing apparatus on a range of cognitive and emotional tests that either last 15 minutes or 40 minutes. The rest of the time they are in their home cage with their partner or family. Over that 18 month to 2 year period they are likely to have between 3-5 general anaesthetics, 2-3 involving a surgical procedure such as brain surgery and implantation of a measuring device, the remaining for restraint purposes only in order to e.g. perform brain scans. Normally, the animals recover well from their surgery or general anaesthesia and are back in their home cage within 2 hours of coming around from the anaesthetic. With all surgical procedures, animals will be fully recovered from one surgical procedure before undergoing another, with a minimum of 2-3 weeks between procedures. At the end of a study the animals are euthanased. Across the 5 days of a week during which they may receive daily testing, animals may also receive, prior to testing, injections of a substance, either peripherally (primarily under the skin or into the muscle) or into the brain via previously implanted metal tubes (see below) in order to study the brain mechanisms underlying behaviour and cognition. In either case they are habituated to being held by an experienced handler for the procedure which typically takes between 5-10 minutes. Over the course of an 18 month-2 year study the average animal will receive either between 20-40 infusions across several regions of the brain or between 20-40 peripheral injections. The specialised testing apparatus is a purpose built test box including a computer and touchscreen. It allows animals to be presented with positive stimuli (e.g. food rewards and visual and auditory stimuli predictive of food rewards) and mildly negative stimuli (e.g. mildly aversive loud noise (0.3-0.7sec) or darkness and visual and auditory stimuli predictive of these negative stimuli) to study learning, attention and emotion. Animals learn to voluntarily enter a transport box for transfer to the testing apparatus, to which they have been gradually acclimatised to minimise

stress. Testing away from the home cage is limited to 40 min, typically once, but very occasionally twice a day, and is halted if the animal exhibits signs of distress. No adverse effects are associated with behavioural testing, and even when mildly aversive stimuli, such as brief loud noises, are used, animals enter the transport box for testing. Animals undergoing restricted access to water during more intellectually demanding experiments utilising a liquid reward receive 2 hours of unrestricted water 5 days a week in addition to rewards received during testing and 48 hours each week of unlimited access to water. Water restriction does not affect the weight of the animals, who often ignore the water when it is returned to their cage, suggesting that they are not very thirsty. All selective surgical procedures are carried out under anaesthesia. Animals are gently caught from their home cage by an experienced handler and carried to the surgical suite. Premedication with a sedative is achieved via an injection into the muscle which causes only mild, momentary discomfort. A gas anaesthetic is used thereafter to ensure no pain is experienced during the surgical procedure (typically lasting 3-6 hours depending upon the procedure). Through small holes made in the skull, we can infuse substances that permanently or temporarily alter brain function in a discrete region or insert an implant that allows the later injection of substances to the implanted region. The latter is fixed in place using screws attached to the skull and dental adhesives. We may also temporarily implant devices to measure local brain function. Animals are monitored closely throughout the procedure and during recovery, and are usually fully recovered and back in their home cage eating, drinking and behaving normally within 2-3 hours. Long-lasting pain relief is given prior to surgery via an injection under the skin, and for several days after as an oral treatment delivered in marshmallow to minimise the need to catch them. Extra care is taken during the first week after surgery to observe any changes in normal behaviour or appearance. Long term implant sites are cleaned regularly throughout the life of the animal to prevent infection. Surgical procedures (lasting 90-120 mins) are also performed in some animals to implant a small radio transmitter into the abdomen to record physiological measures of emotion (heart rate and blood pressure) in animals that move freely during behavioural testing. Brain imaging (typically lasting 90 mins) may be carried out using anaesthesia to keep the animal still so as to ensure good quality images. Animals receiving certain brain scans may have an intravenous access device implanted under the skin to allow the injection of a radioactive substance without the stress of injecting directly into a vein. Following these surgeries animals typically return to the home cage within two hours.

A retrospective assessment of these predicted harms will be due by 02 November 2023

The PPL holder will be required to disclose:

- What harms were caused to the animals, how severe were those harms and how many animals were affected?

Replacement

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

The aim of the proposed work is to investigate how neural circuits in the brain control cognition and emotion. To do this, functional brain circuits are required. Furthermore to be able to determine the contribution of a particular brain region or circuit to the expression of a certain behaviour it is essential to be able to alter its function. As such interventional experiments cannot be done in humans for ethical

reasons, and cell cultures are unable to contribute to a functional, behaving circuit, animal models are indispensable for this work.

A retrospective assessment of replacement will be due by 02 November 2023

The PPL holder will be required to disclose:

- What, if any, non-animal alternatives were used or explored after the project started, and is there anything others can learn from your experience?

Reduction

Explain how you will assure the use of minimum numbers of animals.

We are very aware of the need to minimise the number of animals that we use in a year while optimising the validity of our scientific results. For this reason, and to keep numbers to approximately 70 per year, we screen all our animals for their suitability for studying particular behaviours and for their genetic background to optimise which animals go into which study. We also use brain scanning to ensure the precise targeting of the location within the brain which are of interest, and plan to investigate the use of imaging as a way of measuring brain structure, connectivity, chemistry, and function, allowing individual animals to act as their own control rather than requiring both control and experimental animals. All new surgical techniques are piloted in rodents first where possible, and any new techniques are tested first in one or two animals to ensure the experiment is optimised. We repair surgical implants, when possible and when there is no risk to the animal, rather than implanting additional experimental animals. We regularly consult with local statisticians to ensure that we are using the optimal group size for the results that we see, to ensure that we use the minimal number of animals while optimising the mathematical power of our analyses.

A retrospective assessment of reduction will be due by by 02 November 2023

The PPL holder will be required to disclose:

- How did you minimise the numbers of animals used on your project and is there anything others can learn from your experience?

Refinement

Explain the choice of species 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.

Primates are used specifically because their brains, in particular, those brain regions most important in regulating our cognitive abilities and emotions, such as the prefrontal cortex, are far more similar in structure and function to that of humans than lower species, e.g. rodents. To illustrate this, the cerebral cortex, that region of the brain with the most sophisticated processing abilities, makes up 80% of the

brain mass in humans and 60-70% in primates, compared to just 26% of the brain mass in rodents. Marmosets are a particularly valuable species to use for the proposed work as their relatively small primate brain makes it possible to target cortical and subcortical structures and to make regionally selective neurochemical interventions with relative ease, with little risk to the animal. Often, the same approaches cannot be used in larger primates, such as the macaque, because the surgical procedure involves too many brain entries, which increases the risk of collateral problems such as damage to major internal blood vessels. Having a breeding colony in the same establishment as the experimental program affords us considerable experimental control over the entire lifetime of the marmoset. This is an important factor, particularly when studying negative emotion and its regulation, since it is known that stress and early life experiences can have an enormous impact on the cognitive and emotional regulatory processes under study. The on site breeding colony means that animals do not have to experience the stress of transport to the laboratory and allows us to separate some of the environmental and genetic influences on behaviour. We constantly review all of our behavioural and surgical techniques to ensure that we refine procedures in order to minimize potential animal suffering.

A retrospective assessment of refinement will be due by by 02 November 2023

The PPL holder will be required to disclose:

- With the knowledge you have now, could the choice of animals or model(s) used be improved for future work of this kind? During the project, how did you minimise harm to the animals?