Home Office

NON-TECHNICAL SUMMARY

Improving treatments for brain tumours

Project duration

5 years 0 months

Project purpose

- (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

Brain cancer, Glioma, Radiotherapy, Radiosensitisers, Toxicity

Animal types	Life stages
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?

This project aims to test the effects of new treatments that may improve the outcome for patients with hard to treat brain tumours, especially aggressive primary brain tumours in adults.

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?

The most common primary brain tumours in adults are aggressive, high-grade gliomas. They remain extremely challenging to treat and do not respond to conventional treatments including surgery, radiotherapy and chemotherapy

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

Publications describing the effects of new treatments or new combination treatments that may be useful to test in patients.

New information about how different treatments may be best combined to improve how well they work.

New information about how to avoid toxicity of new treatments or new combinations.

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

In the long term (5-10 years) patients with brain tumours may benefit from having access to new treatment approaches.

In the short and medium term the specialist community in brain tumour research will benefit from increasing knowledge of which new treatments may work and how they work for brain cancers.

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

Effective dissemination of new knowledge through local, national and international meetings.

Working with national and international collaborators including in established consortia to bring new treatments to the clinic as quickly as possible.

Feedback to the relevant research community about challenges and unsuccessful aspects of the project.

Species and numbers of animals expected to be used

- Mice: 1200

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.

We are using mice for these experiments as they are the smallest species that can be used in our pre-clinical imaging and treatment equipment. Mice models of glioma, especially using injection of tumour cells into brain, have been well validated as effective models of the human disease in terms of tumour and normal tissue responses. We will use adult mice for the majority of these experiments. In some cases, particularly when we are assessing the effect of treatment on normal brain we may also use younger animals to model the effect of treatment on survivorship in young patients.

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

In a typical experiment animals will be injected with tumour cells direct to the brain while under anaesthesia using a stereotactic guidance technique. Animals will then be monitored over a number of weeks (typically 3-6) until tumour growth is expected. At this point animals may be imaged, for example by MRI or bioluminescent imaging to assess tumour size. In each case animals will be under anaesthesia for the period of imaging. Animals will then be allocated to treatment groups, commonly a short course of radiotherapy (3 doses per week over 1 or 2 weeks) and/or drug treatment given by gavage or ip or tail vein injection. Following treatment animals will be closely monitored for signs of ill-health over the subsequent weeks and may also undergo further imaging (MRI, bioluminscence) at specific time points. We have set limits for interventions so that there is at least 24 hours between imaging sessions and no more than twice per week and no more than 6 imaging sessions will be performed within any one month period. A typical experiment would last 2-3 months.

Less commonly we will implant tumour cells under the skin and monitor tumour growth directly by caliper measurement across treatment groups described above.

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

The expected effects of surgery on these animals include pain from craniotomy or subcutaneous injection and/or from problems with wound healing, which may last from hours to a few days after surgery.

The expected effects of tumour growth on these mice include poor feeding and weight loss, reduced or abnormal mobility, reduced interaction with cage mates, undergrooming. The expected duration is a few days, after which animals will be sacrificed if these signs do not resolve or worsen.

The expected effects of treatment include poor feeding, pain (from injection) and after-effects of anaesthesia including reduced mobility and low body temperature. These effects are expected to be short lived (hours).

The expected effects of other investigations include pain from injections of contrast for imaging, which is expected to be short-lived (minutes or hours)

Animals with subcutaneous tumours may also experience discomfort from tumour ulceration.

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)?

The expected severities for mice are moderate in the majority of animals. Predicted severe in <1% due to exceptional circumstances

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?

Although in vitro (2D and 3D) as well as new organoid models provide useful means to test specific hypotheses and/or validate findings from screening experiments, in vivo models are a vital addition in clinical translation. They are the only means of assessing the impact of tumour micro-environment on response to treatment, which can often have a powerful modifying effect. In addition these models bridge the gap between biological end-points such as tumour cell killing and clinically relevant outcomes including symptom-free survival. They also permit investigation of clinically relevant biomarkers, for example new imaging approaches to monitor tumour responses which cannot be applied outside of a full organ model. In vivo work is also critical to assess the effects of treatment on normal tissue since this depends on the interplay between different cell populations that cannot be recapitulated in vitro.

For some glioma models, especially patient-derived tumour models, cells cannot be maintained in vitro so direct implantation to mouse brain is necessary to maintain them.

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

We have used 3D spheroid models for initial proof of concept and would also consider tumour/normal brain organoids, which may allow us to focus the in vivo work on fewer agents and reduce animals needed for pharmacodynamic or tissue based end-points of drug/combination effects.

We are working with collaborators to introduce a combined normal brain/tumour organoid which may permit assessment of effects on tumour and normal tissue simultaneously and further reduce the numbers of mice needed for definitive experiments.

We also have links to labs developing tissue on chip or tumour on chip devices which would support efficient and relatively high throughput assessment of novel agents. We will utilise all of these models

when appropriate.

Why were they not suitable?

3D spheroid models do not incorporate normal tissue components and cannot be used for clinically relevant translational end-points.

Organoid and tumour on chip technologies are still in 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?

This estimate is based on numbers used previously and the likely balance of work going forwards, which is unlikely to change significantly. I have taken in to account the reduced experimental work carried out 2020/21 during the Covid pandemic when access to in vivo work was severely restricted.

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

We always use models with high take rates and reproducible growth characteristics (eg the mouse tumour CT2A) unless there is a specific indication for a different model, for example due to expression of a specific target or an indication to use patient derived material. This very significantly reduces the number of animals required per treatment group (usually 5) compared to when we use patient-derived material to establish tumours (usually 12).

As far as possible we design experiments so that we can cross-refer between control/standard treatment groups, reducing the groups necessary across a series of similar experiments using the same model. We use standard radiotherapy/chemotherapy regimes across experiments to facilitate this approach.

We also use data from previous experiments, including serial tumour imaging/monitoring data to inform study time points, for example for histological assessment so that the numbers of animals included for these end-points are minimised.

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

We use efficient breeding in house as well as use of external suppliers to prevent animal wastage.

As above, we use models that we are familiar with and for which we have significant prior data as far as possible. For experiments using new or unfamiliar models we routinely use data from the literature to design small pilot experiments before embarking on large treatment/efficacy work to ensure efficient experimental design.

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.

Mice are the lowest sentient animal that can be used for these experiments for clinical translation.

We use established syngeneic and patient-derived xenografts for much of this work, for which we have high quality data on tumour growth, expected symptom-free survival and treatment effects. This allows us to design experiments in which mice undergo treatment at early time points prior to tumour-induced symptoms and to monitor them closely to ensure minimal duration of tumour-induced symptoms.

We also use state of the art technology to administer treatment, including highly targeted radiotherapy, to minimise the side effects of treatment since only the tumour and small volume of normal tissue receives a high dose. We routinely design experimental protocols to ensure that post-treatment effects can be monitored during the normal working day, for example by treating cohorts early in the day rather than spreading treatment through the day.

Whenever possible we will use imaging-based end-points (MRI, bioluminscent imaging) in place of survival/symptom based end-points to reduce animal distress and suffering.

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

Mice are the smallest animal species that can be use in our pre-clinical imaging and radiotherapy equipment. We need to use models that fully recapitulate the human disease as far as possible and we have selected models that reflect this. These models include a relevant tumour micro-environment and allow assessment of effects of treatment on both tumour and normal tissue.

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

We will work with local and national groups who are developing relevant technologies for these experiments. This includes local collaborators who are working on novel imaging approaches for tumour monitoring including small animal PET.

We also work with the CRUK Rad-Net pre-clinical radiotherapy group who are developing an integrated radiotherapy-MRI imaging bed set up to allow MRI-based radiotherapy planning to further refine the accuracy of this treatment and further reduce unnecessary radiation exposure.

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

We will follow current guidelines including:

The updated (2020) guidance on animal testing and research.

UKCCR guidelines for the welfare and use of animals in research (British Journal of Cancer (2010) 102: 1555-1577).

LASA guiding principles for preparing and undertaking aseptic surgery

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

We run a local user group for all researchers involved in in vivo work and for our husbandry and in vivo unit staff. This reports to the institution- wide committee who are responsible for in vivo work across the whole campus. New approaches to procedures/husbandry etc are discussed at these meetings. All researchers all also circulated regular updates from for example the NC3Rs unit (Replacement, Reduction and Refinement guidance).

All practitioners are encouraged to maintain/improve their skills through regular courses as well as meetings with researchers in other units.

Specifically for this project we work closely with other institutions doing similar work through radiotherapy research networks and are members of the pre-clinical drug- radiotherapy working group. This ensures that developments in technology, in model development and in experimental approaches are shared across the UK.