Home Office

NON-TECHNICAL SUMMARY

# Radiation Combinations for Cancer Treatment

**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**

Radiation, Oncology, Rodent

| Animal types | Life stages |
|---|---|
| Mice | adult |
| Rats | 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?**

To test anti-cancer agents in combination with ionising radiation with the aim of improving current clinical therapies.

**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?**

Radiotherapy provides significant benefit and is used in over 50% of all cancers. To combine radiotherapy with novel anti-cancer agents may significantly enhance the therapeutic outcome and has the potential to translate into highly significant clinical benefit.

In order to determine potential clinical benefit we need to have pre-clinical animal models that will demonstrate and characterise improvements in anti-tumour activity and relevant potential clinical benefits thus directing clinical development.

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

Data to support clinical trials. Improved radiation protocols combined with novel anti-cancer agents to be used in the clinic leading to improved efficacy and tolerability. We will share pre-clinical data with principal investigators to influence clinical trial designs. Publication of both successful and unsuccessful data in high impact journals.

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

Radiotherapy provides significant benefit being used in over 50% of all cancers. As the patient population treated with radiotherapy is so large, enhancing therapeutic outcome for even a relatively small proportion has the potential to translate into highly significant clinical benefit. To have pre-clinical animal models that will demonstrate and characterise improvements in anti-tumour activity will lead to more relevant potential clinical benefits thus directing clinical development.

In this license we will be testing the combination of potential anticancer drugs and ionising radiation with the aim of improving current anti-cancer therapies. With this license we will be able to investigate if new compounds sensitise human tumours to irradiation or if new dosing schedules are better tolerated and/or more active than the current ones. With all this information, new clinical trials can be designed that eventually may change clinical practice.

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

Dissemination of knowledge at conferences via poster sessions and seminars. Publication of both successful and unsuccessful data in relevant journals. We will share pre-clinical data with principal investigators to influence clinical trial designs. We work in collaboration with many scientists and groups in which we are able to share data and learning.

**Species and numbers of animals expected to be used**

- Mice: 11000
- Rats: 1100

# 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 use animal studies in mice and rats alongside many other experimental approaches and they are crucial in building up a complete picture of cancer biology. Our research using animals has helped drive advances in cancer treatment that are benefiting people with cancer all over the world today.

Our work mainly uses mice (90%), which can grow tumours which mimic those of human cancer patients. Studies of cancer in mice mimic the complex way tumours grow and spread in people with cancer.

Mice can be easily genetically altered to allow us to study the genetic causes of cancer and reproduce tumour types which naturally occur in humans in the correct tissues and body systems.

We also conduct some studies in rats (10%). Some compounds that we want to test may not have sufficient levels in the blood to have an effect on the tumour in the mouse and therefore we need to use the rat as an alternative species. The rat is also usually the species of choice for toxicity studies. These studies would be conducted under a different project licence but it may be necessary to directly compare the dose level of a drug that causes an effect on the tumour to the dose level that produces unwanted side effects. This is to ensure that there is a big enough margin between activity and safety.

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

In a typical study animals are implanted with a tumour. As our work is not focused on one area of cancer but across all cancer types with focus on lung, prostate, breast, ovarian, pancreatic, bladder and haematological cancers there are a wide variety of tumour types that may be used within this licence. The majority of tumour models are derived from human tumours. As the human tumour tissue is foreign to the animal, the animals immune system would reject the tumour tissue and therefore we need to grow human tumours in either mice or rats that have an impaired immune system. This allows the tumour tissue to grow and not be rejected.

Tumours are usually implanted as cells using a needle, this is done on the lower left or right side of the back (flank) of the animal. In some cases the tumour is not available as cells and therefore a very small piece of tumour tissue needs to be surgically implanted into this area. This is done under anaesthesia and requires a very small cut in the skin and a pocket made under the skin where the tumour tissue can be placed. The cut is then sealed using either stitches, special tissue glue or clips. This area also allows the tumour to be easily monitored and measured and does not affect the animals ability to move around.

Once the tumour has started to grow the animal's tumour will be irradiated using an x-ray machine. Only the area where the tumour is will be exposed to the x-ray beam, the rest of the animal will be protected with a lead shield. The animals will be anaesthetised to ensure that the animal does not move whilst being exposed to the x-ray beam. The animals may also receive doses of an anti-cancer drug. In a typical study an animal may receive one dose of radiation each day for 5 days. Anti-cancer drugs are usually dosed orally and the animal may receive up to two doses per day for 28 days. The animals are closely monitored every day and body weights and condition of the animals are recorded to ensure that the animals are healthy. The effect on the tumour growth is compared to an animal which does not receive any active drug and/or radiation.The hope is that the anti-cancer drug in combination with radiation significantly reduces the growth of the tumour compared to either giving the drug alone or radiation alone.

During the study blood samples may be taken. This is to check the level of the drug in the blood.

At the end of the study the animal is killed and the tumour tissue and other tissues are taken which can then be used for further investigation.

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

The tumour is continually monitored and measured and although the tumour may continue to grow this does not appear to cause the animal any pain or discomfort and they continue to behave normally. The size the tumour can grow is limited by the use of a measurement/condition/size scoring system to ensure that it does not cause any pain or discomfort to the animal.

The doses of radiation are targeted to the tumour area only and the rest of the animal is protected therefore no adverse side effects are expected.

The dosing procedure for dosing of the anti-cancer drugs does not usually cause any issues but the drug itself may have some side effects. Side effects may include weight loss or abnormal behaviours such as being less active or not socialising or interacting with their cage mates. Very strict criteria are put in place to minimise any unwanted side effects to avoid any pain, suffering or distress to the animals. The side effects usually only last for a short period of time.

**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)?**

Approximately 90% of animals used within the licence will be mice and 10% rats. It is expected that 90% of both the mice and rats will be returned within the moderate category and approximately 10% within the mild category. This will be for all strains of mice and rats used within this project licence.

**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?**

Animals are needed in our research to help us understand the mechanisms that underpin cancer, such as the growth and spread of tumours, and to develop new ways of diagnosing, treating and preventing the disease. Cancer is a very complex disease and animal studies are essential to understand these complexities within living organisms. They are also required by regulatory authorities before any trials of new drugs can be tested in humans. Animal studies are only performed after every feasible test has been conducted on cancer cells in the laboratory and where no alternative exists.

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

Multi-cellular 'organ on a chip' models are available, but as yet have not reached the reliability and multi-system complexity of the rodent model, especially when shaping the treatment of patients in the clinic. Non-animal alternatives are used in the identification and selection of compounds. These generally include measurements of the drug's activity on particular target cells. Activity in particular cell types however cannot predict the activity in humans due to a complexity of issues such as availability of the drug in the body and whether it is able to reach the target cancer cell.

**Why were they not suitable?**

They are not suitable because they cannot mimic the living organism and the processes that under-pin cancer in a living organism.

# 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 licence reflects a well established cancer research program and the numbers of animals used within this project licence are based on the diverse areas of cancer that are being investigated.

We typically run approximately two studies per month, each study usually has ~100 animals per study.

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

All studies are designed to ensure that the minimal numbers of animals are used to achieve the question being asked. This is done with help and guidance from a statistician who is a maths expert who uses huge amounts of data to figure out how likely it is that something will happen or not. They ensure that all studies are designed to ensure that we are able to use the minimal numbers of animals to see an effect of a potential anti-cancer drug if there is an effect.

Good experimental design principles such as randomisation are incorporated into all experiments. All study designs are approved by a statistician.

All experiments are performed in accordance with Good Laboratory Standards (GLS). This standard sets the minimum laboratory requirements for all our research and development. This ensures that procedures and results are accurate, reliable, traceable and reproducible and where appropriate, comply with the appropriate regulatory authorities' legislation.

All experiments are performed in accordance with the PREPARE guidelines - Planning Research and Experimental Procedures on Animals: Recommendations for Excellence.

All research that will be published will be published in accordance with the ARRIVE guidelines - Animal Research: Reporting of In Vivo Experiments.

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

When multiple project groups want to investigate the effect of their compounds in the same tumour model wherever possible we run these within one study that share the same control group therefore reducing the requirement for multiple control groups if the studies were run independently, leading to an overall reduction in numbers. Such opportunities are identified at monthly demand meetings I chair where projects are looking at exploring their compounds in combination with radiotherapy treatment.

To minimise any side effects associated with treatment of potential drugs, a small pilot study in 2- 3 animals is performed to ensure the treatment does not have any unwanted side effects before progressing into larger numbers of animals.

Wherever possible multiple tumour and/or tissue samples will be taken from the same animals and may be frozen down and used in other non-animal experiments.

# 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.**

We will use a wide variety of different tumour models in both mice (90%) and rats (10%). The majority of the tumours are implanted as cells in an area on the animal (the lower left or right side of the back (flank)) which allows the tumour to be easily monitored and measured and does not affect the animals ability to move around. The size the tumour can grow is limited to ensure that they do cause any pain or discomfort to the animal.

The doses of radiation are targeted to the tumour area only and the rest of the animal is protected therefore no adverse side effects are expected.

The dosing procedure for dosing of the anti-cancer drugs does not usually cause any issues but the drug itself may have some side effects. Side effects may include weight loss or abnormal behaviours such as being less active or not socialising or interacting with their cage mates. The drugs are tested in a very small number of animals initially (2 to 3 per group) and only drugs that do not have unwanted side effects can be used in larger numbers of animals.

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

Using less sentient animals for example a non-mammalian species such as the fruit fly, is not possible since they lack a closed circulatory system and so you cannot replicate a number of the complex processes that underpin cancer such as the growth and spread of cancer.

Earlier life stages of vertebrates such as zebrafish is an option, and early studies of human xenograft models do show promise of tumour inhibition studies . However the small size and difficulty of collecting meaningful blood samples makes pharmacokinetics essential in our work, currently impossible in that model.

https://www.sciencedirect.com/science/article/abs/pii/S2405803320301217

Xaio et al. 2020 Trends in Cancer (Online 17th April)

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6234738/

https://www.sciencedirect.com/science/article/pii/S2352396419307881 Costa et al 2020 in the EBioMedicine (The Lancet) Developments in zebrafish avatars as radiotherapy sensitivity reporters — towards personalized medicine.

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

All animals will be acclimatised for 7 days from arrival before they undergo any experimental procedure.

Animals will not be handled by the tail and will be handled by an alternative method, for example tunnel handling, modified cupping and/or ‘pinch’ scruffing. The tail will only be used for the initial catching of an animal in exceptional circumstances or when absolutely necessary (for example if an animal has escaped and priority is to regain control).

All surgery is performed in concordance with 2017 LASA Guiding Principles for Preparing for and Undertaking Aseptic Surgery. Any animals that undergo a surgical procedure will be provided with analgesia (pain killers) prior to the surgery and maintained in a warm environment until full recovery to minimise weight loss. Analgesia may be administered post-operatively within an edible jelly. The mice and rats will have access to a non-medicated form of the jelly prior to surgery to become accustomed to eating it.

Explore if there are options to apply any palliative treatments to minimise any adverse effects of dosing.

All parental dosing routes will be done using a single needle. For cell implants this will be done with a single needle unless documented exceptions are in place.

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

Guidelines for the welfare and use of animals in cancer research (Workman, P., Aboagye, E., Balkwill, F. et al.  Br J Cancer 102, 1555–1577 (2010))

Animal research: Reporting in vivo experiments: The ARRIVE guidelines. Br J Pharmacol. 2010 Aug; 160(7): 1577–1579.

PREPARE: guidelines for planning animal research and testing (Adrian J Smith, R Eddie Clutton, Elliot Lilley et al. Laboratory Animals Volume: 52 issue: 2, page(s): 135-141

LASA Guiding Principles

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

I actively participate in continuous professional development as a fellow member of the Royal Society of Biology. I receive The Biologist newsletter every month which often provides a good source of relevant 3R's initiatives. I also follow the NC3R's website to keep myself updated on relevant 3R's initiatives (https://www.nc3rs.org.uk/news/using-award-scheme-promote-3rs-innovation).  We also actively discuss and implement new 3R's initiatives and run a yearly 3R's poster session competition, sharing information across different establishments.

I am an active member of AWERB and annual refinement goals are set annually by AWERB, for example alternative mouse handling.