International PhD Program IC-PhD

PhD thesis project

2024 Call for application

Investigating innovative dose delivery methods to optimize the effectiveness of novel radiotherapy techniques

General information

Call	2024
Reference	2024-10-PREZADO_PARSONS
Keyword(s)	Innovative radiation therapy; proton FLASH therapy; proton minibeam radiation therapy (MBRT); therapeutic index; DNA damage repair

Director(s) and team

Thesis director(s)	Yolanda Prezado & Jason Parsons
Research team	New Approaches in RAdiotherapy (NARA)
Research department	UMR3347 / U1021 - Signaling, radiobiology and cancer

Description of the PhD thesis project

Abstract

Recent studies have revealed that the way in which dose is delivered in radiotherapy (RT) has a major impact on the tumour and normal tissue responses [1, 2], which is important given that ~50 % of all human cancers are treated with RT. Non-conventional beam microstructures in time and space have been shown to confer significant clinical advantage. The first technique, "FLASH" RT exploits very high dose rate (≥ 40 Gy/s) irradiation, short beam-on times (≤100 ms) and large single doses (≥10 Gy). The second technique, minibeam radiotherapy (MBRT), relies on the use of arrays of minibeams, each with a diameter of 0.5–1 mm, delivered in a grid with a spacing of 1–3.5 mm. Both techniques have been shown to significantly protect healthy tissue whilst maintaining tumour control, and which have the potential to revolutionise the treatment of cancer patients. However, an in-depth understanding of the distinct radiobiological mechanisms that underpin the efficacy of these RT techniques, and how the underlying mechanisms change as a function of the beam parameters is still missing. The interdisciplinary PhD programme we propose will investigate the key currently under-explored cellular and molecular mechanisms, such as the DNA damage response, that are responsible for their therapeutic effects. The PhD will lay the foundations for a greater understanding and future clinical exploitation of novel FLASH and MBRT techniques.

Background

Two of the most recent and exciting developments in RT are FLASH and MBRT [3, 4]. Both approaches have been shown to significantly protect healthy tissue while maintaining local tumour control [3, 4], and are considered exciting and revolutionary RT techniques that could optimise the treatment of cancer patients. Considering ~50 % of all human cancers are treated with RT, and where acute and long term adverse side effects are common with conventional RT using X-rays, FLASH and MBRT have significant potential and scope

to improve on the effectiveness and safety of current RT treatments. Despite this, the biological mechanisms that give rise to the therapeutic effect are not well understood. FLASH appears to depend on the presence of oxygen and could proceed from the chemistry of peroxyradicals and a reduced level of DNA damage. MBRT action appears to be based on abscopal effects, cell signalling and/or migration of cells between the "valleys and hills" present in the non-uniform irradiation field, or through faster repair of vascular damage. Therefore, an in-depth evaluation of FLASH and MBRT at the cellular and molecular level (particularly DNA damage and repair [5, 6], plus influence of time and beam structure) is essential for the effective translation of these RT technologies through to the clinic for cancer patient benefit.

Objectives

This PhD thesis will contribute to the global effort to bring innovative FLASH and MBRT techniques closer towards clinical practice. The concrete objectives are the following:

1. *In vitro* evaluation of the potentially distinct molecular mechanisms (such as DNA damage and repair pathways under different oxygen conditions) responsive to these innovative RT techniques, under the guidance of Prof Parsons from the University of Birmingham, UK. The focus will be on glioblastoma and head and neck cancer models, in which the participant teams have long standing experience, but also cancers that would significantly benefit from the normal tissue sparing and potentially enhanced doses to the tumour that are enabled by these new techniques.
2. *In vivo* validation of the main results obtained in the first objective. The student will profit from the experience of Dr Prezado and her team on translational radiobiology studies.

Experimental approaches

A. *In vitro* evaluation of the distinct molecular mechanisms of FLASH and MBRT

The analysis will be done under the supervision of Prof Parsons, who is highly experienced in fundamental radiobiology research. Different head and neck cancer and glioblastoma cell lines (in addition to appropriate non-tumour cells) will be irradiated with conventional photon (X-ray) RT, or with proton FLASH and proton MBRT using the MC-40 cyclotron in Birmingham, which is dedicated for research. Irradiations will be performed in different configurations, including normoxic and hypoxic (0.1-1 % oxygen) conditions to examine for oxygen dependence. Direct assessment of DNA damage (single and double strand breaks, plus complex DNA damage by comet assays) as well as surrogate markers (e.g. γH2AX/53BP1/RAD51 by fluorescence microscopy), at different times post-IR and the efficiency of their repair through cellular pathways (e.g. single strand break repair, non-homologous end-joining and homologous recombination) will be comparatively assessed.

In addition to the above, a more expansive transcriptomic analysis will be performed to comparatively assess the main genes and cellular pathways upregulated/downregulated in response to X-rays, proton FLASH and proton MBRT under the different oxygen conditions and through temporal evaluation. Preliminary data has already been acquired and which will be a starting point for the PhD student to explore using bioinformatic analysis. Further validations at the protein level (e.g. by immunoblotting and/or immunofluorescence) will be conducted.

B. *In vivo* validations of FLASH and MBRT molecular mechanisms

This part of the Ph.D. will be carried out at Prezado's lab, who has extensive experience in translational radiobiology and *in vivo* experiments. Using the data acquired in Part A (DNA damage repair, and transcriptomics), *in vivo* experiments will be carried out using proton FLASH and proton MBRT to validate the main findings.

For example, analysis of γH2AX/53BP1/RAD51 protein levels in glioblastoma tumours will be evaluated in response to the different RT configurations and tissue oxygenation, and which will be correlated with the results obtained *in vitro*. Different hypoxia markers will be used to assess the impact of the comparative irradiation modes. Survival curves at different oxygen concentrations will also be assessed to evaluate the impact of oxygenation on treatment response.

International, interdisciplinary & intersectoral aspects of the project

The PhD student will split their time between France (I. Curie) and the UK (University of Birmingham). They will have the unique opportunity of working in both academic labs and groups that have significant and highly complementary in vitro and in vivo proficiency. This combination of multidisciplinary expertise in radiation biology and physics will enable the establishment of a new vibrant collaboration that is uniquely placed in driving forward FLASH and MBRT as an exciting new field of innovative RT techniques, which is currently being investigated worldwide.

The teams have highly complementary experience and expertise ranging from accelerator physics, engineering, medical physics to radiobiology, which will be an asset to successfully guide the PhD student in this exciting and interdisciplinary PhD programme. The student will be trained in basic radiobiology techniques, particularly cell culture and DNA damage repair analysis, and to utilise the MC-40 cyclotron and associated radiobiology facilities by Prof Parsons, who leads a multidisciplinary team (~15 staff/PhD/MD students). The PhD programme also includes training in novel RT techniques and delivery (proton FLASH and proton MBRT), and knowledge both from the medical physics and radiobiology point of view will be provided by Y. Prezado (I. Curie). The team of Prezado is itself interdisciplinary.

Recent publications

1. L. Iturri, A. Bertho, C. Lamirault, M. Juchaux, C. Gilbert, J. Espenon, C. Sebrie, L. Jourdain, F. Pouzoulet, P. Verrelle, L. de Marzi and **Y. Prezado**. Oxygen supplementation in anesthesia can block FLASH effect and anti-tumor immunity in conventional proton therapy. Nature communications Medicine 2023

2. L. Iturri, A. Bertho, C. Lamirault, M. Juchaux, C. Gilbert, J. Espenon, C. Sebrie, L. Jourdain, F. Pouzoulet, P. Verrelle, L. de Marzi and **Y. Prezado**. Proton FLASH Radiation Therapy and Immune Infiltration: Evaluation in an Orthotopic Glioma Rat Model. Int. J. Rad. Oncol. Biol. Phys. 2022.

3. A. Bertho, L. Iturri, E. Brisebard, M. Juchaux, C. Gilbert, R. Ortiz, C. Sebrie, L. Jourdain, C. Lamirault, G. Ramasamy, F. Pouzoulet and **Y. Prezado**. Evaluation of the role of the immune system response following minibeam radiation therapy, Int. J. Rad. Oncol. Biol. Phys. 2022.

4. Thongchai a M Masilela, **Yolanda Prezado**. Monte Carlo study of the free radical yields in minibeam radiation therapy. Medical Physics, 2023, 50, pp.5115 – 5134

5. Sifaddin M R Konis , Jonathan R Hughes , Jason L Parsons TRIM26 Maintains Cell Survival in Response to Oxidative Stress through Regulating DNA Glycosylase Stability, Int J Mol Sci 2022

Expected profile of the candidate

We are looking for a motivated student, with a background in biology, preferably. Some competences in cell culture, or in vivo experiments will be an asset.