Institution: The University of Manchester

Unit of Assessment: 3

Title of case study: Molecular markers and drug development leading to improvements in radiotherapy. (*ICS-08*)

1. Summary of the impact

40% of all cancer patients, who are cured of their disease, receive radiotherapy as part of their treatment. The number of cancer cures could be increased if the application of radiotherapy could be improved. Research at the University of Manchester (UoM) has: led the way in identifying, validating and exemplifying the value of predictive/prognostic biomarkers of response to radiotherapy; and demonstrated, in clinical trials, the therapeutic efficacy of combining molecularly targeted agents with radiotherapy. Further, the pharmaceutical industry has incorporated these concepts into drug development programs, accelerating clinical drug development, and thus saving them time and money.

2. Underpinning research

See section 3 for references [1-6]; see section 5 for corroborating sources (S1-S7); UoM researchers are given in bold. In REF3a and REF5 this case study is referred to as ICS-08.

The impact is based on research undertaken by the following key contributors:

- **Ian Stratford** (Professor, 1996-to date)
- **Kaye Williams** (Research Assistant, 1996-2000; Research Fellow, 2001-2005; Senior Lecturer, 2006-2012; Professor, 2012-to date)
- **Catharine West** (Research Associate, 1996-2002;, Senior Lecturer, 2002-2008; Professor, 2008-to date)
- **Caroline Dive** (Research Fellow, 1995-1998; Reader, 1998-2002; Professor, 2002-to date)
- **Rachel Airley** (PhD student, 1998-2002)
- **Amanda Eustace** (PhD student, 2004-2007; Research Associate, 2008-to date)
- **Aoife Shannon** (Post-Doctoral Fellow, 2005-2008)

The overall basis for the work is that tumour cells residing in conditions of low oxygen tension (hypoxia) are resistant to radiation. However, hypoxia is a physiological abnormality of tumours that can be exploited to **improve radiotherapy**. Research aligns with four central themes:

1. **Developing methods to measure the depth of and severity of hypoxia in tumours**: During 2000-2006, the group was the first to combine the use of oxygen electrodes and molecular markers to define the presence of hypoxia in tumours and was the first to combine the use of hypoxia-activated bio-reductive drug markers, such as pimonidazole, together with molecular markers of hypoxia and demonstrate their *adverse* prognostic significance for outcome of radiotherapy [1].

2. **Understanding the biological role of hypoxia in tumours**: the hypoxia-selective expression of the molecular markers having prognostic significance in cancer radiotherapy is driven by the transcription factor hypoxia-inducible factor-1 (HIF-1). Through research (1997 to date) **Stratford/Williams** demonstrated that HIF-1 drives tumour growth and influences response to radio- and chemotherapy [2] and identified potential downstream targets (for example vascular endothelial growth factor; VEGF) as determinants of radiation-response.

3. **Exploiting the presence of hypoxia by the rational application of bio-reductive drugs or radiosensitisers when combined with radiotherapy**. Hypoxic cells in tumours can be directly targeted using hypoxia-activated bioreductive cytotoxins or by using hypoxia-selective radiosensitising agents. Work from 1996 to date has exemplified the benefit of many classes of bioreductive and radiosensitising agents in combination with radiotherapy to

improve both local tumour control and impact on the development of metastases [3]. This has underpinned clinical development and subsequent adoption of the radiosensitiser nimorazole into **standard-of-care** treatment in European centres.

4. **Optimising the use of molecular targeted therapies with radiotherapy to exploit hypoxia/HIF-1 mediated processes.** From 2002 to date, **Stratford/Williams** have placed a major emphasis on analysing the impact of new, so called, molecularly targeted drugs on tumour hypoxia, HIF-1 mediated processes and response to radiotherapy. The ultimate goal was to expedite appropriately-designed early clinical studies to maximise the likelihood of improving patient response. Among the drugs that have been examined are the pan-VEGF receptor antagonist cediranib (AZD2171) [4], the oncogenic (MEK1/2)-signalling inhibitor selumetinib (AZD6244) [5] and a variety of inhibitors of the DNA-repair protein poly-ADP-ribose polymerase (PARP) [6]. Positive interactions between agents and radiation response, optimised scheduling approaches and the underlying mechanistic basis of interaction were determined. This included the unique discovery of a vascular perfusion effect of PARP inhibitors that improves oxygenation. These observations have led to ongoing and proposed clinical trials nationally and internationally, combining cediranib, selumetinib and PARP inhibitors with radiotherapy.

3. References to the research

1. **Airley RE, Loncaster J,** Raleigh JA, Harris AL, **Davidson SE, Hunter RD, West CML, Stratford IJ.** (2003) GLUT-1 and CAIX as intrinsic markers of hypoxia in carcinoma of the cervix: relationship to pimonidazole binding. *International Journal of Cancer*. 104:85-91. DOI: 10.1002/ijc.10904
2. **Williams KJ, Telfer BA, Xenaki D, Sheridan MR,** Desbaillets I, Peters HJ, Honess D, Harris AL, Dachs GU, van der Kogel A, **Stratford IJ**. (2005) Enhanced response to radiotherapy in tumours deficient in hypoxia-inducible factor-1. *Radiotherapy &. Oncology.* 75, 89-98. DOI: 10.1016/j.radonc.2005.01.009
3. **Williams KJ,** Albertella, MR, **Fitzpatrick, B,** Loadman, PM, Shnyder, SD, **Chinje, EC, Telfer, BA,** Dunk, CR, Harris, PA, **Stratford IJ** (2009) In vivo activation of the hypoxia-targeted cytotoxin AQ4N in human tumor xenografts *Molecular Cancer Therapeutics*. 8: 3266-3275. DOI: 10.1158/1535-7163.MCT-09-0396
4. **Williams KJ, Telfer BA, Shannon AM, Babur M, Stratford IJ**, Wedge SR. (2007) Combining radiotherapy with AZD2171, a potent inhibitor of vascular endothelial growth factor signaling: pathophysiologic effects and therapeutic benefit. *Molecular Cancer Therapeutics*. 6:599-; DOI: 10.1158/1535-7163.MCT-06-0508.
5. **Shannon AM, Telfer BA**, Smith PD, **Babur M**, Logie A, Wilkinson RW, **Debray C, Stratford IJ, Williams KJ,** Wedge SR. (2009) The mitogen-activated protein/extracellular signal-regulated kinase kinase 1/2 inhibitor AZD6244 (ARRY-142886) enhances the radiation responsiveness of lung and colorectal tumor xenografts. *Clinical Cancer Reearch*.15:6619-29. DOI: 10.1158/1078-0432.CCR-08-2958.
6. Calabrese CR, Almassy R, Barton S, Batey MA, Calvert AH, Canan-Koch S, Durkacz BW, Hostomsky Z, Kumpf RA, Kyle S, Li J, Maegley K, Newell DR, Notarianni E, **Stratford IJ**, Skalitzky D, Thomas HD, Wang LZ, Webber SE, **Williams KJ**, Curtin NJ. (2004) Anticancer chemosensitization and radiosensitization by the novel poly(ADP-ribose) polymerase-1 inhibitor AG14361. *Journal of the National Cancer Institute.* 2004 96:56-67. DOI: 10.1093/jnci/djh005.

4. Details of the impact

See section 5 for numbered corroborating sources (S1-S7).

Pathways to impact.

Tumour hypoxia has been recognised as a barrier to successful radiotherapy since the 1950s. However, translating this knowledge into more beneficial outcomes for patients has been a slow process. To expedite this, we have developed and applied therapies to target or overcome

hypoxia-mediated resistance; initially by using hypoxic-cell radiosensitisers and hypoxia-activated cytotoxic drugs, and subsequently by utilising molecular targeted approaches based on understanding the biological rationale for combination. As the impact of these approaches is beneficial primarily in patients with hypoxic tumours, we have coincidentally led biomarker-based research programmes towards enabling personalised therapy based on tumour oxygenation.

Impact on prediction and diagnosis

Basic research into the biology of HIF-1 underpinned evaluations of downstream targets, such as CA-IX and Glut-1, as surrogate (protein) biomarkers of tumour hypoxia. The work has led to the patenting and commercial development (via the SME, ALMAC) of a robust *genetic* signature, based on CA-IX and Glut-1 that is highly prognostic across multiple cancer types and can predict benefit from hypoxia-modifying therapy (S1). The prognostic significance of these protein biomarkers/gene signatures has led to their use in world-leading research-based treatment centres to influence and guide the choice of treatment for many thousands of patients. A letter from the University of Toronto Princess Margaret Cancer Centre asserts, "*The international impact of this work is that these biomarkers are used in world leading Centres, to influence how patients are treated in the new era of personalized cancer medicine*". (S2).

Impact on patient treatment

(a) Head and neck (H&N) cancer*:*

Worldwide, there are 400,000 cases of H&N cancer every year of whom, 300,000 will die of their cancer. Our early work led to clinical trials showing that combining a hypoxic radiosensitiser, nimorazole, with radiotherapy increases 5-year local regional control of all H&N cancer patients from 34 to 49% (S3,4). In hypoxic tumours (identified using the molecular markers above) the control rates changed from 18% to 49% (S3,4). Nimorazole is now standard-of-care in Denmark and Sweden "*with some 800 patients a year receiving drug*"; meaning an additional 90 people per annum survive their disease. Impact within the UK has been confounded by the perceived lack of robust markers of hypoxia. Thus, the Phase III NIMRAD trial has been developed incorporating the hypoxic gene signature (above) into the evaluation of nimorazole with radiotherapy to sensitise hypoxic tumours and avoid over-treatment of aerobic tumours (S4).

(b) Development of molecular targeted therapies combined with radiotherapy*:*

We have been instrumental in driving a paradigm shift in anti-cancer drug development in the pharmaceutical industry leading to "*combining drugs with radiotherapy … at a much earlier stage in the drug development process*" , as endorsed by the Head of AstraZeneca cancer iMed, and subsequent acceleration of early phase clinical testing of drugs with radiotherapy (S5). Previously, changes in clinical practice in radiotherapy would take decades from the initial scientific observation to becoming standard-of-care (S4). However, based on our supporting preclinical data, two early clinical trials have been initiated: The MEK-RT trial combining selumetinib with radiotherapy for the treatment of Non Small Cell Lung Cancer (PI, Faivre-Finn); and the DREAM-therapy trial of cediranib or selumetinib combined with preoperative radiotherapy in rectal cancer (PI, Saunders). These trials began one year [2010] after the **Stratford/Williams** preclinical publication. "*The DREAM-therapy trial is near completion and interim analysis shows significant improvements in patient outcome with complete pathological response rates that are at least twice the national average*"(S6). This means that more patients are free of their disease. Multi-centre, international, Phase III trials are being developed in order to define these treatments as standard-of-care (S5,6).

Impact on Pharmaceutical decision making

Our work has facilitated go/no-go commercial decisions for combining drugs with radiotherapy. This is illustrated by the trials above, but a corollary of this has been that industry has also made the decision for "*drugs not being taken further in clinical development with radiotherapy*". Given the estimated >$500 million investment required to progress drug through clinical evaluation, early project termination yields significant cost saving to the pharmaceutical industry (S5).

Impact on national radiotherapy research environment

Stratford and **Williams** helped develop and are the Scientific Executive members of the NCRI

Clinical and Translational Radiotherapy Research Group. The aim of this group is to develop practice-changing clinical trials. Since the initiative began in 2009 the number of radiotherapy trials has increased by 66% and the number of patients in trials has doubled. Further, recent investment in staff has been made by the NCRI to build on the Manchester paradigm and develop a national translational radiobiology network to facilitate early clinical trials of drugs in combination with radiotherapy. The overall impact has been to increase the number of establishments within the UK that are recognised as centres of excellence for radiation research and to increase the international profile UK-based radiation-related research (S7).

5. Sources to corroborate the impact

S1 **Eustace A** *et al* (2013) A 26-Gene Hypoxia Signature Predicts Benefit from Hypoxia-Modifying Therapy in Laryngeal Cancer. *Clin Cancer Res* 19:4879-4888.

S2 Letter from a Senior Professorial Clinical Scientist at the University of Toronto and Princess Margret Cancer Centre, to confirm impact of the **Stratford/Williams** work on the subsequent use of biomarkers to guide personalized treatment with radiotherapy.

S3 Letter from Professor and Head of Department of Experimental and Clinical Oncology, Aarhus, Denmark, and Editor-in-Chief of Radiotherapy and Oncology, to confirm metrics on the impact of using hypoxic sensitizers with radiotherapy in H&N cancer and how important it is to identify (using a gene signature) those patients most likely to respond to treatment.

S4 Published data illustrating the impact of hypoxia modification in tumour identified as being "hypoxic". Toustrup K *et al* (2012) Gene expression classifier predicts for hypoxic modification of radiotherapy with nimorazole in squamous carcinoma of the head and neck. *Radiother Oncol*. 102:122-129.

S5 Letter from the Head of the AstraZeneca Cancer iMed. This endorses the importance of the **Stratford/Williams**' work for changing commercial thinking about developing molecularly targeted drugs and combining them with radiotherapy. In addition, it verifies that their work was used to decide **not** to take drugs into clinical trial with radiotherapy and hence the money this would have saved industry.

S6 Letter from the Principal Investigator of the DREAM trial indicating that the **Stratford/Williams** preclinical work has led directly to patient benefit.

S7 Letter from the Chief Scientist CR-UK to verify the impact of the **Stratford/Williams** work for developing the NCRI Clinical and Translational Radiotherapy Research Group, with consequent doubling of the numbers of patients in radiotherapy-driven clinical trials.