Institution: University College London

Unit of Assessment: 1 - Clinical Medicine

Title of case study: The development of pyrrolobenzodiazepine dimers as cancer therapeutics

1. Summary of the impact

Research at the UCL Cancer Institute into drug-DNA interactions has led to spin-out company Spirogen Ltd resulting in job creation (currently 25 employees) and significant investment from within the UK and overseas. Pyrrolobenzodiazepine dimer drug (SJG-136, SG2000) is currently in clinical trials in the USA and collaborative research and licence agreements in the area of antibody drug conjugates have been established with large pharmaceutical partners including in 2011 with Genentech, a member of the Roche group. In 2013, Spirogen was acquired by Astra-Zeneca for $200m.

2. Underpinning research

Joint research between Professor John Hartley (UCL, 1988-date) and David Thurston (UCL School of Pharmacy, 2001-11; now Kings College London) led to the rational design, synthesis and evaluation of novel pyrrolobenzodiazepine (PBD) dimers as potent anticancer agents. These drugs bind sequence-selectively in the minor groove of DNA, forming non-distorting DNA interstrand cross-links which are refractory to repair **[1, 2]**. In collaboration with the US National Cancer Institute (NCI), lead molecule SJG-136 (SG2000) was found to exhibit potent, differential cytotoxicity *in vitro,* have a novel mechanism of action through COMPARE analysis, and broad spectrum antitumour activity *in vivo*.

This drug has been evaluated in four Phase I clinical trials in the UK (UCL) through Cancer Research UK (CRUK) and in the USA through the NCI. It has completed a Phase II trial in platinum refractory ovarian cancer, and a haematological Phase I/II is currently open. The clinical trials have been facilitated through use of novel pharmacodynamics endpoints of DNA cross-linking and damage response developed at UCL **[3, 4]**.

As part of detailed structure activity relationship studies we found that the potency of PBD dimers can be enhanced by introducing unsaturation about the C2-position of the PBD C-ring and installing substituents that are directed along the floor of the DNA minor groove. The next generation of PBD dimers, which are more potent than SG2000, have been developed, including SG2057 and SG2202. They exhibit picomolar/sub-picomolar activity against a range of human tumour cell lines and demonstrate curative activity in human tumour xenograft models. SG2285, a prodrug of SG2202 is currently in pre-clinical development **[5, 6]**.

The ability to generate such cytotoxic molecules that display exquisite potency suggested a potential role in strategies aimed at targeting and releasing highly cytotoxic agents directly at a tumour site. An example is as the 'warhead' component of an antibody drug conjugate (ADC). The fully synthetic PBD dimers are ideally suited for the role of warhead in an ADC approach. They combine potency with a demonstrated therapeutic index (unlike other warheads such as calicheamycin), are not cross-resistant with widely used chemotherapy agents, and their unique mode of action sets them apart from the tubulin binders (maytansinoids and auristatins) that currently dominate the ADC arena. Several PBD dimer-containing ADCs, targeting both haematological malignancies and solid tumours, are currently undergoing preclinical and clinical evaluation.

3. References to the research

[1] Hartley JA, Spanswick VJ, Brooks N, Clingen PH, McHugh PJ, Hochhauser D, Pedley RB, Kelland LR, Alley MC, Schultz R, Hollingshead MG, Schweikart KM, Tomaszewski JE, Sausville EA, Gregson SJ, Howard PW, Thurston DE. SJG-136 (NSC 694501), a novel

rationally designed DNA minor groove interstrand cross-linking agent with potent and broad spectrum antitumor activity: part 1: cellular pharmacology, in vitro and initial in vivo antitumor activity. Cancer Res. 2004 Sep 15;64(18):6693-9. http://dx.doi.org/10.1158/0008-5472.CAN-03-2941

[2] Alley MC, Hollingshead MG, Pacula-Cox CM, Waud WR, Hartley JA, Howard PW, Gregson SJ, Thurston DE, Sausville EA. SJG-136 (NSC 694501), a novel rationally designed DNA minor groove interstrand cross-linking agent with potent and broad spectrum antitumor activity: part 2: efficacy evaluations. Cancer Res. 2004 Sep 15;64(18):6700-6. http://dx.doi.org/10.1158/0008-5472.CAN-03-2942

[3] Puzanov I, Lee W, Chen AP, Calcutt MW, Hachey DL, Vermeulen WL, Spanswick VJ, Liao CY, Hartley JA, Berlin JD, Rothenberg ML. Phase I pharmacokinetic and pharmacodynamic study of SJG-136, a novel DNA sequence selective minor groove cross-linking agent, in advanced solid tumors. Clin Cancer Res. 2011 Jun 1;17(11):3794-802. http://dx.doi.org/10.1158/1078-0432.CCR-10-2056

[4] Wu J, Clingen PH, Spanswick VJ, Mellinas-Gomez M, Meyer T, Puzanov I, Jodrell D, Hochhauser D, Hartley JA. γ-H2AX foci formation as a pharmacodynamic marker of DNA damage produced by DNA cross-linking agents: results from 2 phase I clinical trials of SJG-136 (SG2000). Clin Cancer Res. 2013 Feb 1;19(3):721-30. http://dx.doi.org/10.1158/1078-0432.CCR-12-2529

[5] Hartley JA, Hamaguchi A, Coffils M, Martin CR, Suggitt M, Chen Z, Gregson SJ, Masterson LA, Tiberghien AC, Hartley JM, Pepper C, Lin TT, Fegan C, Thurston DE, Howard PW. SG2285, a novel C2-aryl-substituted pyrrolobenzodiazepine dimer prodrug that cross-links DNA and exerts highly potent antitumor activity. Cancer Res. 2010 Sep 1;70(17):6849-58. http://dx.doi.org/10.1158/0008-5472.CAN-10-0790

[6] Hartley JA, Hamaguchi A, Suggitt M, Gregson SJ, Thurston DE, Howard PW. DNA interstrand cross-linking and in vivo antitumor activity of the extended pyrrolo[2,1-c][1,4]benzodiazepine dimer SG2057. Invest New Drugs. 2012 Jun;30(3):950-8. http://dx.doi.org/10.1007/s10637-011-9647-z

4. Details of the impact

Work on PBDs has been commercialised over the last 13 years through spin-out company Spirogen, which was set up in 2000 with Hartley as one of the founding scientists **[a]**. Initial funding came from the Bloomsbury Bioseed Fund, and laboratories were established at the UCL Cancer Institute and UCL School of Pharmacy. As at July 2013, the company has a broad intellectual property base with >40 published patents and patent filings covering the use of PBDs as stand-alone anticancer drugs and as targeted agents. The company is currently based at the Queen Mary BioEnterprise Innovation Centre, London and has 25 employees **[b]**.

Between 2001 and 2003, the company went through two rounds of funding, and SG2000 was licensed to Ipsen. This drug successfully completed Phase I Clinical Trials in the UK and US, and results were reported in 2008 **[c]**. 69 Patients were treated in multiple phase I trials, with 15 cases of stable disease and three partial responses of note **[d]**.

In October 2009, Spirogen regained development and commercialisation rights for SG2000 from Ipsen, and entered into an option agreement with Celtic Therapeutics to fund the Phase IIa trials of SG2000 in ovarian cancer, with investment of up to $15m **[e]**. Phase II trials began in 2010 **[f]**, evaluating the overall response rate of SG2000 in approximately 50 patients with recurrent, resistant or refractory epithelial ovarian, primary peritoneal, or fallopian tube carcinoma.

More recently, significant further inward investment has been obtained by Spirogen with multiple collaborations with pharmaceutical companies in the area of PBD drug conjugates. In particular,

the PBDs are beginning to have an impact in the area of antibody drug conjugates, which is fast emerging as one of the principal approaches in the field of monoclonal antibody cancer therapeutics:

January 2011: Announced a research collaboration and license agreement with Genentech, a member of the Roche Group, for the discovery and development of antibody drug conjugates involving Spirogen's proprietary PBD drugs and associated linker technology **[g]**.

March 2012: Celtic Therapeutics formed a new company, ADC Therapeutics, headquartered from Lausanne, Switzerland with a pipeline of ten proprietary ADC oncology development programs, targeting multiple major cancers, including prostate, renal, breast, lung and blood cancers and an initial budget of $50million. Celtic Therapeutics is also the majority owner of Spirogen, and ADC Therapeutics' development plan for the ADCs will use well-characterized monoclonal antibodies against these ten antigens for conjugation with best-in-class warhead and linker chemistry based on proprietary pyrrolobenzodiazepines ("PBDs") "payload" technology developed by, and licensed from Spirogen. Stephen Evans-Freke, Co-Founder and Managing General Partner of Celtic Therapeutics commented in the press release: "*We believe that ADCs will represent a significant medical breakthrough in cancer therapy over the coming decade, and that Spirogen's PBDs constitute 'best-in-class' ADC warheads. We anticipate investment of up to $50m into ADC Therapeutics to achieve clinical proof of concept in 2-3 lead oncology programs. We are committed to fully fund ADC Therapeutics and will raise additional capital if warranted*" **[h]**.

April 2012: Began a collaboration with a School of Pharmacy spin-out company, PolyTherics[1], to use their ThioBridge technology to conjugate Spirogen's potent PBD cytotoxic agents site-specifically to antibodies and antibody fragments **[i]**.

February 2013: Began a research collaboration with Ablynx to evaluate the potential of a novel anti-cancer drug conjugate combining Spirogen's proprietary cytotoxic drugs, pyrrolobenzodiazepines (PBD), and associated linker technology, with Nanobodies® generated using Ablynx's proprietary technology platform **[j]**.

[Text removed for publication].

In late 2013 Spirogen was acquired by Astra-Zeneca for a total of $440million ($200million upfront plus $240million deferred consideration on meeting defined developmental goals/milestones) **[k]**.

5. Sources to corroborate the impact

[a] http://www.spirogen.com/spirogen/history.php

[b] Claims regarding Spirogen can be corroborated by:
1. Senior Business Manager (Biopharm), UCL Business PLC. Contact details provided.
2. CEO, Spirogen. Contact details provided.

[c] The Results of the Phase I Studies of SG2000 (SJG-136) to be Presented at ASCO, Chicago, June 2008: http://www.spirogen.com/news/press-archive.php?id=210&cpg=1

[d] SG2000 Highlights http://www.spirogen.com/pdf/SG2000-Highlights.pdf

[e] Celtic Therapeutics to invest up to $15m in the development of Spirogen's cancer drug SG2000: http://www.spirogen.com/news/press-archive.php?id=196&cpg=1

[f] Commencement of a phase II clinical trial of SG2000: http://www.spirogen.com/news/press-archive.php?id=189&cpg=1

[g] Spirogen Ltd. announces a research collaboration and license agreement with Genentech for

[1] Case study on PolyTherics submitted to UoA 3.

the discovery and development of antibody drug conjugates.
http://www.spirogen.com/news/latest.php

[h] http://www.adctherapeutics.com/news/2012/03/celtic-therapeutics-launches-50m-antibody-drug-conjugates-development-company

[i] http://www.genengnews.com/gen-news-highlights/polytherics-spirogen-to-research-antibody-drug-conjugates-for/81246576/

[j] http://www.collegehill-lifesciences.com/news/2013/02/ablynx-and-spirogen-enter-into-a-research-collaboration-to-evaluate-the-potential-of-novel-toxin-nanobody-drug-conjugates-in-cancer

[k] http://www.astrazeneca.com/Media/Press-releases/Article/20131015--astrazeneca-oncology-portfolio-strengthened