Institution: Newcastle University

Unit of Assessment: UoA5

Title of case study: Discovery of a major drug-drug interaction that led to important changes in the regulation of drug development by the pharmaceutical industry

1. Summary of the impact

Researchers at Newcastle University discovered interactions *in vitro* between the widely prescribed cholesterol-lowering drug rosuvastatin and cyclosporine, and between rosuvastatin and gemfibrozil, at the liver transporter protein OATP1B1. Subsequent clinical trials showed that the interactions occurred in patients and slowed clearance of rosuvastatin from the body. The research findings not only had direct implications for the safe prescribing of rosuvastatin when it came to be marketed but also more far-reaching impact. US Food and Drug Administration and European Medicines Agency guidelines published in 2012 stipulate that pharmaceutical companies must investigate potential drug-drug interactions in the pre-clinical development phase of all candidates that bind that transporter.

2. Underpinning research

Key Newcastle University researcher

- Dr Colin Brown, a Lecturer in the Department of Physiology from 1990 – 2000, and since then a Senior Lecturer.

Underpinning research

In the late 1990s, Brown's research was concerned with identifying the transporter proteins that mediated the uptake and secretion of particular compounds across epithelia in kidney and intestine (R1 and R2). As part of that programme of research, Brown's group had generated a series of clones of different transport proteins expressed in *Xenopus* oocytes (frog egg cells), which provided a useful model for studying the interaction of compounds with individual transporters.

Because of his innovative research in the area, Zeneca Pharmaceuticals (which merged that year with Astra AB to form AstraZeneca plc) approached Brown in 1999 and requested that he investigate the mechanism of clearance by the liver of one of their lead compounds, ZD4552 (later named rosuvastatin), a statin drug that reduces the risk of cardiovascular events by reducing blood cholesterol. In his first set of experiments, Brown identified OATP-C (OATP1B1 in the new nomenclature) as the main uptake transporter of rosuvastatin in hepatocytes (liver cells). He then tested the effect of a number of clinically relevant compounds on rosuvastatin uptake into oocytes expressing that transporter and found that two compounds had a particularly intense effect: gemfibrozil (used, like statins, for the treatment of high cholesterol), and cyclosporine (used to control inflammatory disorders and suppress rejection of organ transplants). Experiments showed that the maximum inhibition of uptake of 5 µM rosuvastatin by gemfibrozil was 50%, with an IC_{50} of 4 µM (R3). Cyclosporine was an even more potent inhibitor of rosuvastatin uptake: the maximum inhibition of rosuvastatin uptake by cyclosporine was higher than 90%, with an IC_{50} of 2.2 µM (R4).

Brown's *in vitro* data were commercially confidential and hence not published immediately, but they were passed via AstraZeneca to the US Food and Drug Administration. Having examined the data, the US Food and Drug Administration mandated that clinical trials be run to test for the drug-drug interactions in patients. AstraZeneca led the trials with Brown as an important collaborator. In a two-period cross-over trial of 20 healthy volunteers, co-administration of gemfibrozil was found to increase the total exposure of patients to rosuvastatin on average 1.88 fold and increase the maximum plasma concentration 2.21 fold (R3). The effect of cyclosporine was determined by comparing rosuvastatin pharmacokinetics in a group of 10 transplant recipients on cyclosporine with pharmacokinetics in a historical group of 21 healthy controls. Co-administration of cyclosporine was associated with a 7.1 fold higher total exposure and a 10.6 fold higher maximum plasma concentration of rosuvastatin. The *in vitro* and *in vivo* data were paired and published in 2004 (R4).

3. References to the research

(Newcastle researchers in bold. Citation count from Scopus, July 2013)

R1. **Dudley AJ, Bleasby K** and **Brown CDA** (2000) The organic cation transporter OCT2 mediates the uptake of β-adrenoceptor antagonists across the apical membrane of renal LLC-PK1 cell monolayers. *British Journal of Pharmacology* 131(1):71-9. DOI: 10.1038/sj.bjp.0703518. **35 citations.**

R2. **Bleasby K, Chauhan S** and **Brown CDA** (2000) Characterization of MPP^+ secretion across human intestinal Caco-2 cell monolayers: role of P-glycoprotein and a novel Na^+-dependent organic cation transport mechanism. *British Journal of Pharmacology* 129(3):619-25. DOI: 10.1038/sj.bjp.0703078. **21 citations.**

R3. Schneck DW, Birmingham BK, Zalikowski JA, Mitchell PD, Wang Y, Martin PD, Lasseter KC, **Brown CD**, **Windass AS** and Raza A (2004) The effect of gemfibrozil on the pharmacokinetics of rosuvastatin. *Clinical Pharmacology & Therapeutics* 75(5):455-63. DOI: 10.1016/j.clpt.2003.12.014. **161 citations.**

R4. Simonson SG, Raza A, Martin PD, Mitchell PD, Jarcho JA, **Brown CD, Windass AS** and Schneck DW (2004) Rosuvastatin pharmacokinetics in heart transplant recipients administered an antirejection regimen including cyclosporine. *Clinical Pharmacology & Therapeutics* 76(2):167-77. DOI: 10.1016/j.clpt.2004.03.010. **140 citations.**

Note on R3 and R4. *Brown (and Windass, his PhD student) had a significant role in producing both outputs. They carried out all the in vitro rosuvastatin transport experiments reported in the papers, which make up half the figures in each. Brown also had a significant role in drafting and intellectually critiquing the manuscripts. (Corroboration by AstraZeneca, Ev a.)*

Funding

AstraZeneca. 1999-2004. £112,000. *Studies on the in vitro interactions of rosuvastatin and other compounds with OATP transporters.*

4. Details of the impact

Pathway to impact: regulatory approval of Crestor (rosuvastatin)

Newcastle demonstrated strong interactions *in vitro* between gemfibrozil and rosuvastatin, and between cyclosporine and rosuvastatin, at the transporter OATP1B1. Those findings were passed to AstraZeneca (Ev a) and considered during the drug development process. This eventually led to two clinical trials that confirmed drug-drug interactions in patients. In August 2003, rosuvastatin was approved for marketing in the United States, accompanied by an FDA-approved drug medication guide that referred specifically to the safety implications of the drug-drug interactions.

The current rosuvastatin medication guide states:

"Rosuvastatin is a substrate for certain transporter proteins including the hepatic uptake transporter organic anion-transporting polyprotein 1B1 (OATP1B1)"

"Concomitant administration of CRESTOR [rosuvastatin] *with medications that are inhibitors of these transporter proteins (e.g. cyclosporine, certain HIV protease inhibitors) may result in increased rosuvastatin plasma concentrations and an increased risk of myopathy"* (Ev b).

Rosuvastatin is now commonly prescribed in Europe and the United States for those at risk of cardiovascular disease. In 2012, 1.9 million prescriptions of rosuvastatin were written by the NHS in England, and it was the third most prescribed drug by sales value in the US (prior to 2008 it was outside the top 20 – see NHS Information Centre and drugs.com). As a result of Newcastle work, the dosing guidelines for rosuvastatin were changed to minimise the risk of dangerous co-administration of rosuvastatin with either gemfibrozil or cyclosporine. Since the effect of cyclosporine on rosuvastatin clearance was so large (leading to a 7.1 fold increase in drug exposure), it can be confidently asserted that serious morbidity, including breakdown of skeletal muscle, will have been avoided in some patients. For example, 60-80% of organ transplant

recipients suffer from abnormally high lipid levels in blood at some point, and are therefore potentially indicated for statins and cyclosporine.

The information given on drug-drug interactions in the FDA-approved rosuvastatin medication guide is mirrored in the current British National Formulary entry on the drug (Ev c).

Worldwide impact on the regulation and activity of the pharmaceutical industry

Investigation of drug-drug interactions (DDI) is becoming an increasingly important issue for the pharmaceutical industry and regulatory authorities, in part because populations are ageing and older people are much more likely to be suffering from multiple medical conditions (co-morbidities), thus taking more than one drug at the same time. The transporter OATP1B1 is a main target for investigation because it has broad substrate specificity and plays a major role in the clearance of many drugs by the liver, including the antibiotic rifampicin, the anti-cancer drug paclitaxel and some statins.

In March 2010 the International Transporter Consortium, which comprises representatives from academia, industry and regulatory authorities, published a white paper that contains a section about the investigation of transporter-mediated drug-drug interactions at OATP1B1. It refers to the rosuvastatin-cyclosporine interaction and cites R4 (Ev d). In March 2012, at the second workshop of the International Transporter Consortium, a presentation about OATP-mediated drug interactions also referred explicitly to data from R4:

"Rosuvastatin (Crestor®) as a case study to study OATP1B1 inhibition and the risk of DDI. Simonson SG *et al"* (Ev e).

Through the International Transporter Consortium's white paper, Newcastle research has influenced thinking within the FDA. In February 2012, the FDA published *Guidance for Industry Drug Interaction Studies — Study Design, Data Analysis, Implications for Dosing, and Labeling Recommendations*. It includes two decision trees that should be used during the development phase of new compounds and help determine (our underlining):

"whether an investigational drug is a substrate for OATP1B1 or OATP1B3 and when an in vivo clinical study is needed" and *"whether an investigational drug is an inhibitor of OATP1B1"* (Ev f).

As justification for this guidance, the document refers to studies, including R4, that have shown clinically significant OATP1B1-mediated drug-drug interactions (our underlining highlights the data from R4):

"For example, co-administration of cyclosporine increases the area under the plasma concentration-time curve (AUC) of pravastatin, rosuvastatin, and pitavastatin by 10-fold, 7-fold, and 5-fold, respectively… Because these statins are not significantly metabolized, the interactions appear to result from inhibition of transporters, including OATP1B1." (Ev f).

The European equivalent of the FDA, the European Medicines Agency, published its final guideline on investigating drug-drug interactions in July 2012. It states:

"[the] *inhibition of OATPs has been reported to result in marked increases in the systemic exposure of drugs subject to hepatic uptake transport … the possible involvement of OATP1B1 and 1B3 uptake transport should be investigated in vitro for drugs estimated to have ≥ 25% hepatic elimination"* (Ev g).

Since adherence to guidelines is mandatory for pharmaceutical companies for the US and European markets, Newcastle research has had worldwide impact on the pharmaceutical industry. A representative of one of the major international pharmaceutical companies who is also a member of the International Transporter Consortium has agreed:

"- that Colin's [Dr Brown's] *in vitro work showed that the rosu/cycl DDI* [rosuvastatin/cyclosporine drug-drug interaction] *was in part mediated by the OATP1B1 transporter (published with the clinical trial in the Simonson paper);*

- that this in vitro data was used to inform the development of regulatory guidelines and influential papers (e.g. ITC white paper) on transporter-mediated DDIs;

- that these guidelines have influenced the operations of the pharmaceutical industry (for example,

testing for DDIs if NCE's [new chemical entities] *are likely to be cleared hepatically* [by the liver] *and be OATP1B1 substrates)."* (Ev h)

Commercial impact

Several companies, such as Solvo Biotechnology (Ev i) and Qualyst Transporter Solutions (Ev j), now offer products and services that have been designed for the study of transporter-mediated drug-drug interactions. Qualyst Transporter Solutions is a global provider of hepatic drug transporter products and contract research services, and it has sold kits or undertaken research projects (typical contracts range in value from $65 to $250k) for major pharmaceutical companies including Pfizer, Novartis, Merck and Eli Lilly. One of their products – B-CLEAR – has been designed in response to 2012 FDA guidance on drug transporter interactions. The company has stated:

"One of the transporters that are [sic] *extremely important for drug interactions and required by both the FDA and EMA for any drug being developed is OATP1B1. Colin Brown's group did work, for the first time, showing OATP1B1 drug-drug interactions between rosuvastatin and cyclosporine, as well as rosuvastatin and gemfibrozil. We can confirm that in the last few years, particularly since the initial International Transporter Consortium report in 2010, this work has directly led to a significant impact on regulation and activity of the pharmaceutical industry. Indeed this is now a routine part of every transporter drug interaction study Qualyst performs for its pharmaceutical clients"* (Ev j).

5. Sources to corroborate the impact

Ev a. Senior Project Director, AstraZeneca. (At the time of the research, Assoc. Director Drug Metabolism and Pharmacokinetics)

Ev b. Crestor [rosuvastatin] prescribing information sheet (December 2012). http://www.crestor.com/c/explore-crestor/side-effects.aspx [Click full prescribing information] (Quotations from the Drug Interactions and Clinical Pharmacology sections.)

Ev c. Entry on rosuvastatin in the British National Formulary. http://www.medicinescomplete.com/mc/bnf/current/bnf_int974-rosuvastatin.htm

Ev d. International Transporter Consortium, Giacomini KM et al. (2010). Membrane transporters in drug development. *Nature Reviews Drug Discovery.* 9(3):215-36. (See Table 3: Selected transporter-mediated clinical drug–drug interactions.)

Ev e. Presentation at the International Transporter Consortium Workshop Two (2012). Membrane transporters in drug development - Best practices and future directions. Tools to study drug transporters. http://www.ascpt.org/2012AnnualMeeting/2012SpeakerPresentations/InternationalTransporterConsortiumWorkshopTwo/tabid/12596/Default.aspx (Quotation from slide 47.)

Ev f. Guidance for industry: drug interaction studies — Study design, data analysis, implications for dosing, and labeling recommendations. US Food and Drug Administration (February 2012). http://www.fda.gov/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/ucm064982.htm (Decision tree, page 67; quotation on cyclosporine-rosuvastatin DDI, page 10.)

Ev g. Investigation of drug interactions. European Medicines Agency (July 2012). http://www.ema.europa.eu/ema/index.jsp?curl=pages/regulation/general/general_content_000370.jsp&mid=WC0b01ac0580032ec5 (Quotation from page 13.)

Ev h. Statement from the Worldwide Director of [Company information removed for publication].

Ev i. Solvo Biotechnology. Transporter services catalogue (May 2013). http://www.solvobiotech.com/documents/SOLVO-Product-and-Service-catalog-short-version.pdf

Ev j. Statement from the CEO of Qualyst Transporter Solutions, USA.