Institution: King's College London

Unit of Assessment: 3B - Pharmacy and Nutritional Sciences

Title of case study: Novel class of medicine to treat lung diseases

1. Summary of the impact

The discovery of a novel, inhaled dual phosphodiesterase 3 and 4 inhibitor, RPL554 – first developed in the Sackler Institute of Pulmonary Pharmacology, King's College London – led to the creation of a SME, Verona Pharma plc, which then successfully demonstrated clinical benefit in Phase II clinical trials. This is a major breakthrough as a "first in class" drug with both bronchodilator and anti-inflammatory activity in a single medicine for the treatment of important respiratory diseases, asthma and chronic obstructive pulmonary disease.

2. Underpinning research

Both asthma, which globally affects an estimated 300 million individuals, and chronic obstructive pulmonary disease (COPD), currently the sixth leading cause of death worldwide, are inflammatory diseases that involve narrowing of the airways. As such, treatment involves both anti-inflammatory measures and those that aid bronchodilation. Gold standard treatment is combination long-acting beta 2 agonist (LABA) and glucocorticosteroid inhalers; however, LABAs are under scrutiny for safety and there is concern about an increased pneumonia risk with glucocorticosteroids for COPD patients. Research at King's College London (KCL) by Prof Clive Page (1996-present, Professor of Pharmacology) and Dr Domenico Spina (2000-present, Reader in Pharmacology) produced a new class of therapeutic agent for these diseases – inhaled phosphodiesterase 3/4 inhibitors – that have combined bronchodilator and anti-inflammatory activity and an excellent safety event profile.

To aid in the development of a suitable anti-inflammatory treatment for asthma and COPD, the Sackler Institute of Pulmonary Pharmacology at KCL has carried out 20 years of pioneering work demonstrating the anti-inflammatory activities of PDE inhibitors. Phosphodiesterase 4 (PDE4) is the predominant PDE isoenzyme in inflammatory cells and a 1995 KCL study with ovalbumin (OVA)-immunized guinea pigs found that chronic administration of a low dose of either a type 3/4 or PDE4 inhibitor produced significant inhibition of eosinophil accumulation normally seen following aerosolized OVA administration (1). KCL researchers also showed airway anti-inflammatory effects of low doses of the non-selective PDE inhibitor theophylline via a double-blind, placebo-controlled study involving 19 atopic asthmatic subjects receiving 6 weeks treatment (2). A further study, in collaboration with Pfizer Ltd, found that either theophylline or the PDE4 inhibitor CDP840 inhibited phytohaemagglutinin-induced proliferation of mononuclear cells in a concentration-dependent manner in either asthmatic or healthy groups (3).

This work led to a number of collaborations with UK and overseas pharmaceutical companies that allowed KCL researchers to explore the potential of targeting such enzymes to treat airway inflammatory diseases. While PDE4 inhibitors have beneficial effects in asthma, side-effects, especially nausea, have limited their use. However, in 1997 KCL researchers found that either a single dose or 9.5 days of the selective PDE4 isoenzyme inhibitor CDP840 given to a total of 54 patients in a double-blind fashion was well-tolerated with no reports of nausea. CDP840 significantly attenuated the late asthmatic response to allergen challenge in the absence of any bronchodilatory or histamine antagonist effect, suggesting CDP840 may exert its effects via an anti-inflammatory mechanism (4). A 2002 double-blind study of a single dose of the novel PDE4 inhibitor V11294A in eight healthy males resulted in plasma concentrations adequate to inhibit activation of inflammatory cells *ex vivo* without any adverse reactions (5). Further studies confirmed that V11294 is an orally active PDE4 inhibitor that exhibits anti-inflammatory activity *in vitro* (with human cells) and *in vivo* (in animal models) at doses that are not emetogenic (6).

Through this work, alongside other KCL studies, it became clear that PDE4 inhibitors were anti-inflammatory and PDE3 inhibitors were effective bronchodilators. It should therefore be possible to develop an inhaled, dual-acting drug inhibiting both enzymes, while not having the unwanted, dose-limiting side effects observed when such drugs were administered systemically. Working in collaboration with Sir David Jack, on behalf of Vanguard Medica (now Vernalis), KCL researchers identified and characterised a novel class of inhaled mixed PDE3/4 inhibitors having both bronchodilator and anti-inflammatory activities in a single molecule and with a long duration of action. They first observed that the non-selective PDE isoenzyme inhibitor trequinsin induced

reversal of induced contraction of guinea pig superfused trachea and demonstrated long lasting bronchodilator responses (7). Following this, *in vitro* experiments showed the trequinsin-like RPL554 could significantly inhibit induced contraction of guinea pig superfused trachea for up to 12 hours after administration. KCL researchers also showed RPL554 could inhibit lipopolysaccharide-induced tumor necrosis factor alpha release from human monocytes and proliferation of human mononuclear cells to phytohemagglutinin. *In vivo*, orally administered RPL554 significantly inhibited eosinophil recruitment following antigen challenge in OVA-sensitized guinea pigs. Further, inhalation of a dry powder containing RPL554 by guinea pigs 1.5 hours before antigen exposure significantly inhibited the recruitment of eosinophils to the airways and histamine-induced plasma protein extravasation in the trachea and histamine-induced bronchoconstriction over a 5.5 hour period (8). These experiments paved the way for the development of human trials of RPL554.

3. References to the research (indicative maximum of six references)

1. Banner K, Page CP. Acute versus chronic administration of phosphodiesterase inhibitors on allergen-induced pulmonary cell influx in sensitized guinea pigs.Br J Pharmacol 1995;114:93-99. http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1510161/ (39 Scopus citations)
2. Sullivan P, Bekir S, Jaffar Z, Page C, Jeffery P, Costello J. Anti-inflammatory effects of low dose oral theophylline in atopic asthma. Lancet 1994;343:1006-8. Doi: 10.1016/S0140-6736(94)90127-9 (297 Scopus citations)
3. Landells LJ, Szilagy CM, Jones NA, Banner KH, Allen JM, Doherty A, O'Connor BJ, Spina D, Page CP. Identification and quantification of phosphodiesterase 4 subtypes in CD4 and CD8 lymphocytes from healthy and asthmatic subjects. Br J Pharmacology 2001;133:722-9. Doi:10.1038/sj.bjp.0704120 (30 Scopus citations)
4. Harbinson PL, MacLeod D, Hawksworth R, O'Toole S, Sullivan PJ, Heath P, Kilfeather S, Page CP, Costello J, Holgate ST, Lee TH. The effect of a novel orally active selective PDE4 isoenzyme inhibitor (CDP840) on allergen-induced responses in asthmatic subjects. Eur Respir J 1997;10:1008-14. Doi: 10.1183/09031936.97.10051008 (103 Scopus citations)
5. Gale DD, Landells LJ, Spina D, Miller AJ, Smith K, Nichols T, Rotshteyn Y, Tonelli A, Lacouture P, Burch RM, Page CP, O'Connor BJ. Pharmacokinetic and pharmacodynamic profile following oral administration of the phosphodiesterase (PDE)4 inhibitor V11294A in healthy volunteers. Br J Clin Pharmacol 2002;54:478-84. Doi: 10.1046/j.1365-2125.2002.01682.x (15 Scopus citations)
6. Gale DD, Hofer P, Spina D, Seeds EA, Banner KH, Harrison S, Douglas G, Matsumoto T, Page CP, Wong RH, Jordan S, Smith F, Banik N, Halushka PV, Cavalla D, Rotshteyn Y, Kyle DJ, Burch RM, Chasin M. Pharmacology of a new cyclic nucleotide phosphodiesterase type 4 inhibitor, V11294. Pulm Pharmacol Ther 2003;16:97-104. Doi:10.1016/S1094-5539(02)00175-X (10 Scopus citations)
7. Spina D, Ferlenga P, Biasini I, Moriggi E, Marchini F, Semeraro C, Page CP. Effect duration of selective phosphodiesterase inhibitors in the guinea pig. Life Sciences 1995;62:953-65. Doi: 10.1016/S0024-3205(98)00015-0 (8 Scopus citations)
8. Boswell-Smith V, Spina D, Oxford AW, Comer MB, Seeds EA, Page CP. The pharmacology of two novel long-acting phosphodiesterase 3/4 inhibitors, RPL554 [9,10-dimethoxy-2-(2,4,6-trimethylphenylimino)-3-(N-carbamoyl-2-aminoethyl)-3,4,6,7-tetrahydro-2H-pyrimido-[6,1-alisoquinolin-4-one] and RPL565 [6,7-dihydro-2-(2,6-disopropylphenoxy)-9,10-dimethoxy-4H-pyrimido[6,1-a] isoquinolin-4-one]. J Pharmacol Exper Ther 2006;318:840-8. Doi:10.1124/jpet.105.099192 (23 Scopus citations)

Research Grants

- C Page. Investigating the pharmacology of PDE inhibitors. Napp Pharmaceuticals. 1991-3: £215,000
- C Page. Phosphodiesterase and inflammation. Purdue Frederick Inc. 1990-4: £350,000; 1994-5: £174,512; 1995-6: £162,512; 1996-7: £178,000; 1997-8: £150,000.
- C Page. Development of a novel PDE inhibitor. Vanguard Pharmaceutics Ltd. 1998-9, £77,000; 1999-2000, £57,000
- C Page. The role of PDE isoenzymes in the regulation of inflammatory cells. Institute Jouveinal/Park Davies (now Pfizer). 1998-2000: £108,000, 2004-7: £128,000
- C Page. PDE inhibitors and inflammation. UCB/Celltech. 2003-6: £180,000
- C Page. PDE inhibitors and airway diseases. Altana. 2004: £35,000; 2006-8: £88,000
- C Page, D Spina. Investigating the duration of action of PDE inhibitors in vitro. Zambon S.p.A.

1993-6: £45,000
- D Spina. Investigation of anti-inflammatory molecules. Veronapharma plc. 2006-12: £508,646

4. Details of the impact

The KCL work described above directly led to the development of the novel mixed phosphodiesterase (PDE) 3/4 inhibitor RPL554. This drug has a long duration of action and clinically possesses both anti-inflammatory and bronchodilator activity in a single molecule against these two enzymes known to be of importance in the development and progression of immunological respiratory diseases. This dual activity of RPL554 makes it unique amongst drugs being developed for the treatment of respiratory diseases. Asthma and COPD affect millions of people worldwide; however current gold standard treatment for these conditions is under scrutiny by the FDA for their adverse event profiles. Additionally, its ability to be administered in a broad range of doses with few adverse events puts it ahead of other drugs for these conditions that have been investigated but not developed or rarely used due to their especially pro-emetic effects.

KCL researchers form Verona Pharma plc

The discovery of RPL554 led to the formation of Verona Pharma plc in 2006 by KCL's Prof Clive Page. Verona Pharma plc is an AIM listed Company that employs four people. Prof Page has helped raise more than £11 million to allow Verona Pharma to develop a suitable inhaled formulation of RPL554 for the necessary regulatory inhaled toxicology and early clinical studies to be undertaken (1a,b). The publication list provided by Verona Pharma plc includes several of the papers discussed above alongside reference to a number of other KCL-led studies (1c).

RPL554 makes it to Phase II clinical trials

The vast majority of drugs tested for efficacy in animal models do not make it into human clinical trials. Verona Pharma plc has therefore made a huge impact by successfully completing double-blind, placebo-controlled, randomised clinical studies of RPL554 in people with mild to moderate asthma and mild to moderate COPD to show the clinical effectiveness of RPL554. Following completion in May 2008 of toxicological studies of RPL554, the Company commenced a Phase I/IIa clinical trial at the Centre for Human Drug Research at Leiden in the Netherlands. This demonstrated that RPL554 has a good safety profile, with no evidence of cardiovascular or gastrointestinal side effects. It also showed that RPL554 has beneficial effects in terms of bronchodilation and bronchoprotection in asthmatics and a reduction in the numbers of inflammatory cells in the nasal passages of allergic rhinitis patients with mild asthma. Further studies demonstrated the safety and bronchodilator effectiveness of the drug administered at higher doses (2a). RPL554 has subsequently been successfully tested in four Phase II clinical trials in approaching 100 patients with asthma or mild to moderate COPD and shown to be an excellent bronchodilator drug with bronchodilation maintained over a period of 6 days with daily dosing of RPL554. In March 2013, the Company demonstrated positive airway anti-inflammatory activity with respect to COPD at a clinical trial carried out at the Medicines Evaluation Unit in Manchester (2b-e) demonstrating in 26 healthy subjects exposed to an inflammatory stimulus that RPL554 can inhibit inflammatory cell recruitment to the lung (2f).

Further interest in RPL554 and mixed PDE3/4 inhibitors

The discovery of RPL554 has increased interest in the development of mixed PDE3/4 inhibitors. This is evidenced by submission of research by other pharmaceutical companies to exploit this new mechanism for COPD, which mention the discovery of RPL554, and from articles written by key opinion leaders and by major pharmaceutical companies. For instance, both Kyorin Pharmaceutical Co., Ltd. in Japan (3a) and the Swiss company Novatis (3b) have published overview papers on how they are investigating mixed PDE3/4 inhibitors. The announcement of trials for RPL554 also generated considerable media interest including on the BBC webpage (3d) and in the Telegraph (3e) and Daily Mail (3f).

5. Sources to corroborate the impact (indicative maximum of 10 references)

1. Verona Pharma plc

a) Website: http://www.veronapharma.com/joomla/index.php/products/lead-drug-rpl554
b) Professional letter of support on file from the CEO of Verona Pharma
c) References: http://www.veronapharma.com/joomla/index.php/products/publications

2. Clinical Trials

a) Phase I/IIa trial:
   - A Combined Clinical Phase I/IIa Study of the Safety and Efficacy of Nebulised RPL554 in Healthy Subjects, Allergic Asthmatics, and Allergic Rhinitics: http://apps.who.int/trialsearch/Trial.aspx?TrialID=EUCTR2008-005048-17-NL
   - Results: http://www.veronapharma.com/joomla/index.php/news/project-news/2009/163-a-single-dose-of-rpl554-is-safe-and-effective-in-patients-with-asthma
   - Franciosi L, et al. A combined phase I/IIa study of the safety, bronchodilator and bronchoprotective effects of nebulized RPL554, a dual PDE3/4-inhibitor, in healthy subjects and asthmatics. Clin Translat Allergy 2013 3(Suppl 1):O13. doi:10.1186/2045-7022-3-S1-O13: http://link.springer.com/content/pdf/10.1186%2F2045-7022-3-S1-O13.pdf
   - Letter of professional support on file from the Dept Respiratory Diseases & Allergeology Skate University, Groningen, The Netherlands.

b) Evaluation of the efficacy and safety of 6 repeated daily doses of nebulised RPL554 0.018 mg/kg (6X) in allergic asthmatics:
   - Trial: http://apps.who.int/trialsearch/Trial.aspx?TrialID=EUCTR2011-001698-22-NL
   - Results: http://www.veronapharma.com/joomla/images/Documents/News/2011-08-17_NR.pdf

c) Randomised, Double-Blind, Placebo-Controlled Evaluation of the Safety and Duration of Action of 2 Single Inhaled Doses, 0.036 mg/kg (12X) and 0.072 mg/kg (24X), of RPL554, a Dual PDE 3/4 Inhibitor
   - Trial: http://apps.who.int/trialsearch/trial.aspx?trialid=EUCTR2010-021349-36-NL
   - Results: http://www.veronapharma.com/joomla/images/Documents/News/2011-02-22_NRa.pdf

d) COPD trial
   - Cazzola M, et al. Safety and bronchodilator effects of nebulized RPL554, a novel dual PDE3/4 inhibitor In COPD. American Thoracic Society International Conference 2013. Chapter DOI: 10.1164/ajrccm-conference.2013.187.1_MeetingAbstracts.A1497: http://www.atsjournals.org/doi/abs/10.1164/ajrccm-conference.2013.187.1_MeetingAbstracts.A1497
   - COPD trial: http://www.veronapharma.com/joomla/images/Documents/News/2012-09-04_NR.pdf

e) Franciosi L, et al. Efficacy and safety of RPL554, a dual PDE3 and PDE4 inhibitor, in healthy volunteers and in patients with asthma or chronic obstructive pulmonary disease: findings from four clinical trials. Lancet 2013 [epub ahead of print] Doi: http://dx.doi.org/10.1016/S2213-2600(13)70187-5

f) Verona press release: http://www.veronapharma.com/joomla/index.php/news/project-news/235-rpl554-demonstrates-positive-airway-anti-inflammatory-activity-in-clinical-trial

3. Further interest in RPL554 and mixed PDE3/4 inhibitors

a) Ochiai K, et al. Phosphodiesterase inhibitors. Part 5: hybrid PDE3/4 inhibitors as dual bronchorelaxant/anti-inflammatory agent for inhaled administration. Biourg Med Chem Lett 2013;23:375-81/. Doi: 10.1016/j.bmcl.2012.08.121

b) Compton C, et al. The Novartis view on emerging drugs and novel targets for the treatment of chronic obstructive pulmonary disease. Pulm Pharmacol Ther 2013 Jun 4. pii: S1094-5539(13)00129-6. Doi: 10.1016/j.pupt.2013.05.009. [Epub ahead of print]

c) BBC webpage. Asthma and Hayfever drug tested. 10 Sept 2008. http:news.bbc.co.uk/1/hi/health/7607665.stm

d) Telegraph. Side-effect free asthma and hayfever drug for sale 'within three years.' 9.Sep.2008: www.telegraph.co.uk/health/2712351/Side-effect-free-asthma-and-hayfever-drug-for-sale-within-three-years.html

e) The Mail Online: Breakthrough drug could cure asthma and hay fever symptoms. 10.Sep.2008. www.dailymail.co.uk/health/article-1053968/Breakthrough-drug-cure-asthma-hay-fever-symptoms.html