Cardiff School of Pharmacy & Pharmaceutical Sciences Research Abstracts

19th Edition
2019

Editors: R Thomas, D Routledge
& J Jenkins
School of Pharmacy & Pharmaceutical Sciences
Cardiff University

Published by STS Publishing, Redwood Building, Cardiff CF10 3NB, Wales, United Kingdom

Published 1st October 2019

ISBN: 978094897579

British Library Cataloguing-in-Publication Data.
A catalogue record for this book is available from the British Library.

Edited by: Rhys Thomas [thomas115@cardiff.ac.uk], D Routledge [routledged1@cardiff.ac.uk] and Justine Jenkins [jenkinsj@cardiff.ac.uk]

CONTENTS

FOREWORD

The School of Pharmacy and Pharmaceutical Sciences, Cardiff University, is the only school of pharmacy in Wales and is one of the top schools of pharmacy in the UK. Our students graduate well-prepared and satisfied, as seen by the consistently high pass rate in the pharmacist registration examination and high ranking in the National Student Survey respectively.

In addition to supporting individual pharmacists in their initial and ongoing education and development, the School is active in research that has been independently judged to be predominantly of international standing, more than half of which is recognised as world-leading or internationally excellent and with many interdisciplinary and external collaborators. Research at the School encompasses medicinal chemistry, drug delivery and microbiology, pharmacology and physiology, and pharmacy practice and clinical pharmacy, and it impacts on healthcare and pharmaceutical sciences throughout the UK and the world. Further information on the School's research activities and degree programmes, along with contact details for academic staff can be found at http://www.cardiff.ac.uk/phrmy.

At the Cardiff School of Pharmacy and Pharmaceutical Sciences, the combination of these strengths allows us to successfully deliver research-led learning and teaching. All of our MPharm students undertake a significant, independent Masters level research project in the final year of the four-year degree, and present and defend their research. The high numbers of well-qualified UK, EU and international students that we attract to our postgraduate diplomas and degrees also contribute significantly to our research output.

This is the 18th year in which we have published the abstracts of our students' research. Within this publication the student is the first named author, and collaborators and supervisors follow. An alphabetical list of authors appears in the index.

Many thanks to our colleagues for their assistance in collating this book.

Rhys Thomas, Dean Routledge & Justine Jenkins
July 2019

Evaluation of a Community Pharmacy-based Asthma Care Plan

Bushra Abdirizaq and WR Ford
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Asthma is a common chronic condition associated with inflammation and obstruction of the airways[1]. Despite the availability of asthma research data and effective treatments on the NHS, approximately three people die every day in the UK as a result of their symptoms[2]. This study aimed to investigate effectiveness of asthma management in primary care by assessing asthma control, inhaler technique and medication adherence in a community pharmacy.

Convenience sampling was used to recruit 63 asthma patients in South Wales for this study. Participants completed an Asthma Control Test (ACT) and the Test of the Adherence to Inhalers (TAI). The ACT was used to determine patient's levels of asthma control and the TAI gave an indication of patient's adherence levels. Inhaler technique was assessed by an Aerosol Inhalation Monitor (AIM)[3]. The Kruskal Wallis and Mann Whitney tests were used to identify statistical significance of the results.

The findings of this study revealed that the majority of participants (60%) had uncontrolled asthma and the same proportion of patients were hospitalised or had taken oral steroids to control their symptoms. Less than a third (28%) of those who completed the TAI (n=57) were fully adherent with their medications. Inhaler technique was performed better ($p<0.05$) with dry powder inhalers (DPI) in comparison with metered dose inhalers (MDI) and the use of a spacer with an MDI was significantly better than using an MDI alone ($p<0.05$). Overall, inhaler technique was poor amongst the participants and only 18% of inhaler techniques (n=80) were correct.

It is evident that the asthma guidelines are not being followed appropriately[4]. Therefore, a different approach is required to improve the management of asthma in primary care. Healthcare professionals need to place greater emphasis on exploring medication concerns and educating patients on correct inhaler use to maximise the benefits of therapy.

1. NHS. Asthma - Overview. 2018 [accessed 5 Jan 2019]. Available from: https://www.nhs.uk/conditions/asthma/symptoms/
2. Asthma UK. Asthma facts and statistics. 2016 [accessed 7 Jan 2019]. Available from: https://www.asthma.org.uk/about/media/facts-and-statistics/
3. Vitalograph. User Training Manual. 2018 [accessed 30 Dec 2018]. Available from: https://vitalograph.co.uk/downloads/view/41
4. Barnes PJ. Asthma guidelines: recommendations versus reality. Respiratory Medicine. 2004;98(S1):S1-S7 doi: https://doi.org/10.1016/j.rmed.2004.02.002

A Retrospective Secondary Analysis of Prescribing Data for Parkinson's Disease throughout Wales

Aymen Abdullah, K Orayj and EL Lane
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Parkinson's disease (PD) is a complex neurodegenerative condition that affects motor and non-motor function.[1] There is no cure yet, though a wide range of symptomatic treatment options are available. However, guidelines on these are not universally agreed upon and have been debated without resolution[2] – as such the field of PD is in a climate of unceasing research. This study looks at how PD treatment varies throughout Wales.

GP data extracts were downloaded from NHS Wales.[3] These contained information on all medicines prescribed in Wales and dispensed in the communities of Wales or England. PD medicines from these were singled out by their BNF codes, then grouped accordingly and assigned to their health board (HB) locations of where they were prescribed and dispensed using statistical analysis software. Prevalence rates (PRs) were calculated for data display.

Results indicate variation in PD management. Average PRs of medicines vary across Wales, and there are also some notable differences in the percentage use of these medicines between the HBs. Inconsistencies in which medicines are preferred e.g. anticholinergics vs. non-ergot-derived dopamine agonists are also evident.

The average annual number of PD medicines per patient is 31.1, but Cardiff & Vale UHB and Cwm Taf UHB are outliers, being unusually high (64.0) and low[2,3] respectively.

General preference of medicines is consistent with the guidelines, the evidence for levodopa is the strongest and this is clearly the drug of choice. Variations in PR and annual number of PD medicines per patient could be due to ongoing uncertainties with the absolute clinical nature of PD medicines, but also departmental activity such as staff numbers and the fact not all medicines are PD-exclusive, especially anticholinergics.[4] These highlight limitations with this study being at population level, and opens the door to further research at patient level.

1. Jankovic J. Parkinson's disease: clinical features and diagnosis. J Neurol Neurosurg Psychiatry. 2008;79(4):368-76. doi: 10.1136/jnnp.2007.131045
2. Fahn S, Oakes D, Shoulson I, Kieburtz K, Rudolph A, Lang A, et al. Levodopa and the progression of Parkinson's disease. N Engl J Med. 2004;351(24):2498-508. doi: 10.1056/NEJMoa033447
3. NHS Wales. General practice prescribing data extract. 2018 [accessed 15 Oct]. Available from: http://www.primarycareservices.wales.nhs.uk/general-practice-prescribing-data-extrac
4. Joint Formulary Committee. British national formulary. 76th ed. London: BMJ Group and Pharmaceutical Press; 2018.

Continuous DOPA Synthesis by Adeno-Associated Virus Using Gene Therapy in Parkinson's Disease

Fatimah Abu Shamat, K Fears, M Lelos, EL Lane
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Parkinson's disease (PD) is a progressive neurodegenerative disease.[1] Current medications used to alleviate motor symptoms become less effective and cause dyskinesias in late stages of the disease.[1] Gene therapy using viral vectors could increase the effectiveness of therapy and reduce side effects.[2] Current PD clinical trials using gene therapy produced a modest efficacy.[3] This study integrates viral linkers into the viral sequence to increase genes co-expression involved in dopamine synthesis and thus increase treatments efficacy.[3,4] Our aim is to test for the efficacy of newly designed viral vectors using the unilateral 6-hydroxydopamine lesioned rat model of PD. The objectives of the study are to confirm the validity of the model and determine the expression profile of different vectors using behavioural and immunohistochemical techniques.

Fifty-two rats were unilaterally injected with 6-hydroxydopamine into the medial forebrain bundle. Four weeks post lesion, behavioural tests were performed. Forty-six rats then received stereotaxic intrastriatal injections of different viruses. Behavioural testing was performed 4 weeks post vector administration. The brains were then sliced and histological analysis was performed to stain for tyrosine hydroxylase (TH) and truncated TH. TH cells in the substantia nigra were quantified and images of striatal sections were captured to measure their optical density.

The results show little expression of truncated TH in the lesioned striatum. The titre used may have been subthreshold (too low to be detected), future studies should aim to have the titre at a threshold level. Moreover, the immunohistochemical stain was weak and hard to conclusively interpret. Optimising the staining may reveal low genes expression levels. However, behavioural tests still demonstrated limited improvement post virus injection.

The limitations in the experiment have hampered our ability to adequately evaluate the hypothesis. However, there is evidence supporting its theory, hence it should be tested for in the future.

1. Lees AJ, Hardy J, Revesz T. Parkinson's disease. Lancet.2009; 373 (9680) : 2055–2066. doi: 10.1016/S0140-6736(09)60492-X.
2. Lundstrom K. Gene Therapy applications of viral vectors. Technol Cancer Res Treat. 2004 ;3(5):467-477. doi:10.1177/153303460400300508
3. Björklund T, Kirik D. Scientific rationale for the development of gene therapy strategies for Parkinson's disease. BBA. 2009; 1792 (7): 703-713. doi: https://doi.org/10.1016/j.bbadis.2009.02.009
4. Furler S, Paterna JC, Weibel M, Büeler H. Recombinant AAV vectors containing the foot and mouth disease virus 2A sequence confer efficient bicistronic gene expression in cultured cells and rat substantia nigra neurons.Gene Ther. 2001;8(11):864-873. doi: 10.1038/sj.gt.3301469

Help or hindrance: An evaluation of the practical application of thestandards for medicines management

Tahmid Ahmed, A Ilyas, ML Hughes, LK James and L Williams
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Medication safety incidents have become a global health concern, and statistics show medication errors cost around 42billion USD annually.[2] One third of the medication errors occur particularly during medication administration in hospitals where nurses are mainly involved.[1] The Nursing and Midwifery Council's (NMC) and the All Wales Medicine Strategy Group's (AWMSG) medicine management standards and policies are used by nurses to provide safe and consistent care. Therefore, these policies have been targeted as a means to reduce these errors through identifying the causes and barriers to the use of these guidelines.

Both qualitative and quantitative methods were utilised to explore deviations from standards for medicine management by nurses. Following ethics approval, uninterrupted observations of practice were carried out using an observation schedule adapted from the guidelines in 3 different wards over 2 hospitals. A focus group was then conducted with nurses using a semi-structured schedule, which was transcribed ad verbatim and analysed thematically. All data in this study was kept confidential and non-attributable to any individual and was done with the consent of the nursing participants.

A total of 321 medication administrations were observed and one focus group of four nurses was conducted. Emerging themes involved: distractions, time, staff, convenience and familiarity of patients as potential barriers to the adherence of these standards. For example, the double checking of preparation and administration processes was not observed for any high-risk medicines.

Based on these barriers, recommendations included; single checks for administration and preparation of high-risk medicines; checking of patient details/ allergies once a shift, and the acceptance of human error playing a role in deviations from standards. Although the study was conducted on a small scale, the research findings will remain useful in the future research and development of guidelines for medicines management.

1. Leape, L. L. et al. 1995. Systems Analysis of Adverse Drug Events. JAMA 274(1), pp. 35-43. doi: 10.1001/jama.1995.03530010049034
2. World Health Organisation. 2010. The third WHO Global Patient Safety Challenge: Medication Without Harm. Available at: https://www.who.int/patientsafety/medication-safety/en/ [Accessed 10 October 2018]

An exploration and evaluation of the opinions of undergraduate MPharm 2 and 4 students on the prospect of prescribing after the pre-registration year

Fahad Ali, T Gandhi and M Weiss
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK. School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Current regulations regarding Pharmacist Independent Prescribing came about in 2006 allowing pharmacists to prescribe within their clinical competence excluding controlled drugs for the purpose of treating addiction. The requirements include; two years of appropriate clinical based practice (in the UK) either in a community or hospital pharmacy and the completion of a GPhC accredited programmed which typically entails six months of both face to face learning and self-directed study.[1] Previous work has tended to focus on the limitations and barriers of integrating Independent Pharmacist Prescribers into a secondary care environment, but no research has chosen to focus on the undergraduate perspective.

The objective of this study was to explore the views of MPharm 2 and 4 students around the prospect of prescribing immediately following their pre-registration year. Four focus groups were conducted with a semi-structured approach using a topic guide to guide questioning. The discussions were audio recorded and transcribed ad verbatim with the intent of extracting any overarching themes, sub-themes and patterns found in the data.

The findings of the study show, that there is a consensus amongst both MPharm 2 and 4 students that prescribing immediately after their pre-registration was beyond their capability. Whilst many felt it was the direction that the profession is heading towards, the current MPharm programme failed to reflect this ongoing

change with its course content and teaching. Several suggestions were made to adopt a more clinical and practical skills-based approach to teaching.

The findings of this study hope to create a foundation for future studies to widen the conversation around solutions to the shortage of healthcare workforce within the UK. It is vital to see where pharmacy teaching can be improved and optimised in order to enable well trained pharmacists to help meet healthcare demands and ensure high quality patient care.

1. General Pharmaceutical Council. Pharmacist Independent Prescriber. General Pharmaceutical Council. 2018. (Last Accessed 13th November 2018) Available at: https://www.pharmacyregulation.org/education/pharmacist-independent-prescriber

---

Dissolution of microfluidic droplets for the development of 3D printed skin model

Ameer Alshukri and CP Thomas
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Bioprinting technology is the placement of cells and tissue matrix components in three dimensions while maintaining adequate spatial control.[1] Current bioprinting systems include laser, inkjet and piezoelectric actuators. All of these approaches are complex, expensive and inaccessible, requiring a lot of technical background knowledge.[2] An affordable and accessible microfluidic based 3D bioprinter has been developed using LEGO Mindstorms® kit which utilises a bioink to encapsulate cells into microscope droplets. The bioink is biocompatible and able to mimic the extracellular matrix, but it has reduced cellular proliferation and differentiation, and cells cannot degrade the surrounding matrix which may limit cell-cell interactions [(3)]. Dissolving the bioink will allow for the positioning of pockets of other cell types and the potential reconstruction of whole epidermis.

Chelating agents were used to dissolve the bioink because other methods such as exposure to high temperature or high pH cause cellular death. Cell-free droplets were deposited into a monolayer of a circular pattern, and fixed volumes of different chelator solutions and concentrations of either EDTA or sodium citrate were added. The average dissolution time of the constructs was recorded. The biocompatibility was investigated by exposing HaCaT cells to mixtures of the dissolved bioink-chelator solutions for a various amount of times, and cellular viability was measured using resazurin reduction assay.

It was found that with increasing chelator concentrations the average dissolution time is decreased. However, lower concentrations showed more controlled dissolution. Cells metabolic capacity and thus viability was reduced with increasing exposure time to the chelator solutions.

General biocompatibility with other cell types such as fibroblasts is yet to be explored. Further research is required to optimise the method used for dissolving the droplets. Investigation of the impact of chelator solutions on subsequent cellular proliferation, differentiation and specific protein expression is still to be carried out.

1. Lee V, Singh G, Trasatti JP, Bjornsson C, Xu X, Tran TN, et al. Design and Fabrication of Human Skin by Three-Dimensional Bioprinting. Tissue Eng Part C Methods. 2014;20(6):473–84. doi:10.1089/ten.tec.2013.0335
2. Derby B. Bioprinting: inkjet printing proteins and hybrid cell-containing materials and structures. J Mater Chem. 2008;18(47):5717. doi:10.1088/1758-5090/8/3/032001

Design and synthesis of novel compounds as potential AspRS inhibitors for *Pseudomonas aeruginosa* therapy

Jordan Altimimi and C Simons
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Pseudomonas aeruginosa is a Gram-negative bacterium which is opportunistic and commonly causes chronic infections.[1] These infections are often associated with cystic fibrosis patients, the immunosuppressed, diabetics, newborns and the elderly.[1] The World Health Organization has outlined antimicrobial resistance as a major issue, emphasising the need for the development of new antimicrobial agents.[2] This is true for *P. aeruginsoa* infections; whereby multidrug resistant strain rates have increased globally.[3]

Aspartyl-tRNA synthetase (AspRS) is a crucial enzyme in the biological functioning and survival of *P. aeruginosa*. This makes it an ideal target for novel antimicrobial drugs.[4] The inhibition and abnormal functioning

of this enzyme will result in taking away the ability for *P. aeruginsoa* to make the necessary amino acid, aspartic acid and will subsequently lead to cell death.[4] This information validates AspRS as an antimicrobial target. Furthermore, this project will focus on the design, synthesis and development of novel *P. aeruginosa* AspRS inhibitors.

Two main pharmacophores are used within this project; thiazole and piperidine moieties. The conjugation of these moieties via either a sulfonamide or carboxamide linker group will provide a final monosubstituted compound. Synthesis of the sulfonamides is a four or five step process (depending on which synthetic route is successful): firstly, a mesylation reaction to transform the initial 'Boc' compound, then either a nucleophilic substitution or a Mitsunobu reaction, to follow is the Boc deprotection phase, a cyclisation reaction and finally the nucleophilic substitution reaction with the thiazole derivative. Carboxamide synthesis has been designed via two different pathways to anticipate reaction failure. Both pathways involve two nucleophilic substitution reaction steps utilizing trichloroethyl chloroformate, monosubstituted piperidines and 2-aminothiazole.

Molecular modelling experiments were used to determine the potential of the designed AspRS inhibitors. This allowed us to view any present binding interactions between the novel compounds and a homology model of *P. aeruginosa* AspRS along with how the compounds fitted within the active site.[5] These experiments were performed via MOE's 'Dock' software. The application also allowed for the visualisation of results from the docking study.

Although synthesis was unsuccessful and no novel AspRS inhibitors were obtained, the results acquired during this project are still very much of value to the wider field of research. This project was also undertaken by a colleague of mine who, attempted synthesis with alternative moieties to the thiazole one that I attempted synthesis with. These derivatives were delegated randomly meaning we each contributed to test the potential of the different derivatives. Furthermore, the designed compounds showed promising results during the molecular modelling phase with many derivatives exhibiting key binding interactions with AspRS and fitting well in the active site. With refinement it is hoped the final reaction stages which failed will become successful and final novel compounds can be synthesized. Alternative reaction methods, reactants, conditions and substituents are able to be researched and utilised to optimize these final reaction stages.

In conclusion, this project has provided a good basis in the research of novel AspRS inhibitors and provides a platform from which process optimisation can make a significant change in successful AspRS inhibitor synthesis.

1. Enoch D, Kuzhively J, Sismey, A, Grynik, A & Karas J A. Pseudomonas Aeruginosa Bacteraemia in Two UK District Hospitals. Infect Dis Rep. 2013;5(1):e4. doi:10.4081/idr.2013.e4
3. WHO. ANTIMICROBIAL RESISTANCE Global Report on Surveillance. 2014 [accessed 28 Nov 2018]. Available from: www.who.int/drugresistance/documents/surveillancereport/en/
4. Dantas R, Ferreira M, Gontijo-Filho P, Ribas R. J. Pseudomonas aeruginosa bacteraemia: independent risk factors for mortality and impact of resistance on outcome Med. Microbiol. 2014;63(12):1679-1687 doi:10.1099/jmm.0.073262-0
5. Sauter, C., Lorber, B., Cavarelli, J., Moras, D. The free yeast aspartyl-tRNA synthetase differs from the tRNA(Asp)-complexed enzyme by structural changes in the catalytic site, hinge region, and anticodon-binding domain. J.Mol.Biol. 2000;299:1313-1324. doi:10.1006/jmbi.2000.3791
6. Suzuki T, Nakamura A, Kato K, Söll D, Tanaka I, Sheppard K et al. Structure of the Pseudomonas aeruginosa transamidosome reveals unique aspects of bacterial tRNA-dependent asparagine biosynthesis. PNAS. 2015;112(2): 382-387. doi:10.1073/pnas.1423314112

Investigating the antibacterial activity of propolis and honey in combination with polyphenols against the multi-antibiotic resistant pathogens, Methicillin-Resistant *Staphylococcus aureus* (MRSA) and *Klebsiella pneumoniae*.

Naima Bakawala, R Williams, and L Baillie
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

The aim of this project was to investigate the antimicrobial activity of honeybee-derived products against methicillin-resistant *Staphylococcus aureus* (MRSA) and multi-drug resistant *Klebsiella pneumoniae* isolates. Honey samples combined with the polyphenol, epigallocatechin-gallate (EGCg)[1], were also tested for activity. Bee products such as honey and propolis are known for antimicrobial activity and have been used in wound healing.[2] Antibiotic resistance is an increasing global risk[3]; propolis and honey could present themselves as alternative antimicrobials.

An agar well diffusion assay was used to measure antimicrobial activity for honey samples alone and in combination with EGCg. The efficiency of various propolis extraction methods was also assessed for antibacterial activity. Two-tailed T-tests determined if results were statistically significant.

MRSA and the *K. pneumoniae* isolates KPC19 and KPC15 were the most sensitive to honey; honey samples 165, H251 & H291 showed activity alone and when combined with EGCg. However, EGCg was found to reduce activity of the honey samples against MRSA and varied in its impact on antibacterial activity when tested against *K. pneumoniae* isolates. Propolis extracted using either ethanol or PEG-400 were the only methods that showed activity against MRSA and KPC15. Longer extraction times increased activity against MRSA. P-values showed some results to be statistically significant.

The Welsh & South-English honey samples demonstrated antimicrobial activity against MRSA and *K. pneumoniae* isolates. EGCg was used to inhibit catalase in the honey samples to determine its effect on H_2O_2 production, which is responsible for antibacterial activity.[4] EGCg reduced activity against MRSA, but its effect against *K. pneumoniae* isolates requires further investigation. Ethanolic and PEG-400 extraction of propolis allows extraction of bioactive compounds with a range of polarities. The variety of bioactive compounds are responsible for antimicrobial activity. Further investigation to establish these components will enable the exploitation of bee products as alternative antimicrobials.

1. Eng QY, Thanikachalam PV, Ramamurthy S. Molecular understanding of Epigallocatechine gallate (EGCg) in cardiovascular and metabolic diseases. *J Ethnopharmacol*. 2018; **210**: 296-310. doi: 10.1016/j.jep.2017.08.035
2. Pasupuleti VR, Sammugam L, Ramesh N, Gan SH. Honey, Propolis, and Royal Jelly; A Comprehensive Review of Their Biological Actions and Health Benefits. *Oxid Med Cell Longev*. 2017; **2017**: 1-21. doi: 10.1155/2017/1259510
3. Gould K. Antibiotics: from prehistory to the present day. *J Antimicrob Chemother.* 2016; **71**(3): 572-575. doi: 10.1093/jac/dkv484
4. Arakawa H, Maeda M, Okubo S, Shimamura T. Role of Hydrogen Peroxide in Bactericidal Action of Catechin. *Biol Pharm Bull*. 2004; **27**(2): 277-281.

An evaluation of the repeat use of the Common Ailment Scheme

Fahmeedha Bari, KL Hodson, ML Hughes and [1]A Evans

Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [1] Welsh Government, Cathays Park, Cardiff CF10 3NQ, Wales, UK

The Common Ailment Scheme (CAS) aims to have pharmacists as the patients' first port of call for minor ailments to relieve the demand for doctors. In 2016, funding was approved to roll out the Choose Pharmacy Application (CPA) nationally, hence allowing more pharmacies to undertake CAS if commissioned.[1] This study aims to evaluate the repeat use of CAS, as part of a joint project. The objectives include identifying repeat users of CAS, establishing the most common ailments patients frequently re-use CAS for and determining how patients use the scheme.

Secondary analysis of consultation data between September 2013 and August 2018 from CAS (via Choose Pharmacy database) was conducted. The database contained ailment, duration, date undertaken, patient ID, referral and treatments and/or advice provided. There are approximately 50,000 entries. Microsoft Excel® was used to obtain a dataset of repeat users only. Datasets were exported into SPSS® to gather data on the demographic of users, frequencies of ailments, consultations, days, number of active pharmacies and referrals.

Results showed repeat use of the service for hay fever by patients aged 20-69 was most common. The majority of patients were aged ≤9 years old and used CAS for childhood related ailments. There is a 2:1 ratio of female to male users. Repeat users often come back for the identical ailment as previous consultation. The highest frequency of times a patient used CAS was 31 times.

This study showed repeat users are a positive aspect of CAS as patients are recognising the purpose of the scheme and taking advantage of this. This suggests a reduction in pressure on doctors but would need further research. Hay fever is most frequently presented ailment it may worth exploring repeat prescriptions for these patients in the future as well as promotion of self-care.

1. Henley J. Choose Pharmacy national implementation. 2016 [accessed 17 Jan 2019]. Available from: http://www.cpwales.org.uk/Services-and-commissioning/Choose-Pharmacy-IT-Platform/Contractor-News-Choose-Pharmacy/20160907-Contractor-News-Choose-Pharmacy-Roll-Out.aspx

Design of a Scalable Nanoparticle Formulation of the Topical Anaesthetic Tetracaine

Laura Baron and MO Ivory
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Application of topical anaesthetics prior to invasive procedures reduces pain experienced by patients but the time taken to achieve anaesthesia is a deterrent to their use.[1] Restricted delivery through the stratum corneum results in a prolonged onset time. Liposomes are able to encapsulate drug and increase permeation through the stratum corneum.[2] This project therefore aimed to use microfluidics to create a scalable nanoparticle formulation of tetracaine to increase drug permeation and decrease onset time.

Liposomes were formulated with soy-PC, DSPE-PEG and cholesterol in ethanol and tetracaine in PBS. Particle size, PDI and zeta potential were measured during a 15-day stability study. Ultracentrifugation was used to determine entrapment efficiency. Franz cells were used to determine permeation of formulations through Strat-M membranes with and without microneedle pre-treatment.

Liposomes formulated using 10mg/ml lipids in the mass ratio of 52:45:3 (soy-PC:cholesterol:DSPE-PEG) and 1mg/ml tetracaine were produced with a particle size of 102.9+-3.76nm and PDI of 0.061. Stability studies showed no significant increase in particle size throughout the 15-day period and no significant change to zeta potential until day 15. The entrapment efficiency studies were inconclusive. Permeation did not occur without microneedle pre-treatment due to inappropriateness of the artificial membrane. Microneedle pre-treated membranes showed no significant difference between the formulations tested in the amount of tetracaine to cross the membrane.

The liposomes produced were smaller and more consistent than tetracaine liposomes that have been produced previously. The formulations remained stable, important if used in clinical settings. Franz cell studies without microneedle pre-treatment suggest Strat-M membranes were not a good model for human skin in this study; future studies should be performed with human skin. Permeation studies with microneedle pre-treatment demonstrate that tetracaine liposomes can effectively deliver tetracaine through artificial membranes.

1. MacLean S, Obispo J, Young KD. The gap between pediatric emergency department procedural pain management treatments available and actual practice. Pediatric Emergency Care. 2007;23(2):87-93. doi: http://dx.doi.org/10.1097/PEC.0b013e31803
2. Mezei M, Gulasekharam V. Liposomes - a selective drug delivery system for the topical route of administration. I. Lotion dosage form. Life Sciences. 1980;26(18):1473-7. doi: http://dx.doi.org/10.1016/0024-3205(80)90268-4

Targeting ErbB2 endocytosis: is Herceptin the rational approach?

Lauren Beamish and AT Jones
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Breast cancer is one of the most common cancers worldwide with new cases increasing annually. Cells overexpressing ErbB2, termed ErbB2 positive, develop a more aggressive cancer type.[1] A favoured ErbB2 targeting therapy is Herceptin. However, despite successful binding, ErbB2's internalisation resistance disrupts Herceptin's ability to remove it from the membrane and downregulate.[2] Therefore, a newer formulation is required to induce internalisation. Herceptin conjugated nanoparticles are a strategy to selectively target ErbB2 and improve uptake and drug bioavailability.[3] In addition, receptor crosslinking and clustering using avidin/streptavidin linkers and Herceptin fragments induce internalisation, and enhance ErbB2's lysosomal trafficking for degradation.[2,4] The aim of this study is to explore Herceptin/ErbB2 internalisation and distribution using these formulations, and also analyse how low Herceptin internalisation translates in terms of tumour response in clinical settings.

A literature search was conducted using online databases to create two information databases. One database included 12 in-vitro studies on Herceptin based drug delivery systems, consisting of nanoparticles, avidin/streptavidin linkers and the biparatopic antibody. Herceptin/ErbB2 cellular uptake was assessed to determine efficacy. The other database included 16 clinical trials on ErbB2 positive breast cancer treatments; Herceptin, Pertuzumab, Kadcyla, Lapatinib, Herceptin with Pertuzumab and Docetaxel, Herceptin with Lapatinib. Study endpoints surrounding tumour response, disease progression and survival were analysed, comparing Herceptin based treatments to other treatments.

Higher Herceptin/ErbB2 internalisation was observed using nanoparticles, avidin/streptavidin linkers and the biparatopic antibody; with the linkers and antibody maximising ErbB2 internalisation and lysosomal trafficking. Low Herceptin internalisation corresponded to a pathological complete response (PCR) of 30.3-36%, and was higher than other monotherapies. PCR raised when Herceptin combined with Pertuzumab (36.1%) and Lapatinib (56.7%).

Low ErbB2-Herceptin internalisation correlates to poor patient response, indicating the need to improve the current Herceptin formulation. These 3 drug delivery systems are promising future strategies.

1. Rubin I, Yarden Y. The Basic Biology of HER2. *Annals of Oncology.* 2001;**12**(suppl_1):S3-S8. doi: 10.1093/annonc/12.suppl_1.S3
2. Bertelsen V, Stang E. The Mysterious Ways of ErbB2/HER2 Trafficking. *Membranes*. 2014;**4**(3):424-46. doi: 10.3390/membranes4030424
3. Panyam J, Labhasetwar V. Biodegradable nanoparticles for drug and gene delivery to cells and tissue. *Advanced Drug Delivery Reviews*. 2012;**64**:61-71. doi: https://doi.org/10.1016/j.addr.2012.09.023
4. Li JY, Perry SR, Muniz-Medina V, Wang X, Wetzel LK, Rebelatto MC, et al. A Biparatopic HER2-Targeting Antibody-Drug Conjugate Induces Tumor Regression in Primary Models Refractory to or Ineligible for HER2-Targeted Therapy. *Cancer Cell*. 2016;**29**(1):117-29. doi: 10.1016/j.ccell.2015.12.008

Inhibiting the mechanism of zinc transporter ZP10 activation in cancer cells

Sabaoun Belal Khel and KM Taylor
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Zinc, an important trace element, at high levels, has been associated with breast cancer.[1] ZIP10, a zinc transporter, has been shown to be involved in breast cancers with invasive and metastatic properties.[2] ZIP10 has been shown to form a heteromer with ZIP6[3] in order to influx zinc into cells. A previous study has shown ZIP7, also a zinc transporter, needed to be phosphorylated in order to transport zinc.[4] Therefore the current study investigated the potential for ZIP10 to be phosphorylated by different kinases as a means of activation.

Predicted phosphorylation sites of ZIP10 were researched and CK2 and CDK1 were implicated. Immunofluorescence microscopy was used to count the number of ZIP10 positive MCF-7 breast cancer cells and how that varied according to kinase inhibitor treatment. The CK2 inhibitor CX-4945 was used and also a CDK1 inhibitor was used. Results were confirmed by Western Blotting analysis.

This study confirmed an increase in ZIP10 levels on the plasma membrane by both fluorescence microscopy and western blot. Treatment of cells with the CK2 inhibitor CX-4945 reduced the number of ZIP10 positive cells on the plasma membrane, suggesting that CK2 has a role in activating ZIP10. Treatment with a CDK1 inhibitor had little effect suggesting no involvement.

The study produced results which are consistent with CK2 phosphorylation of ZIP10 as a requirement for ZIP10 mediated zinc influx into cells which may now offer a new target in cancer.

1. Mahmoud A, Al-Alem U, Dabbous F, Ali M, Batai K, Shah E et al. Zinc intake and risk of prostate cancer: case-control study and meta-analysis. *PLOS One*. 2016;**11**(11):e0165956
2. Kagara N, Tanaka N, Noguchi S, Hirano T. Zinc and its transporter ZIP10 are involved in invasive behavior of breast cancer cells. *Cancer Sci*. 2007;**98**(5):692-697.
3. Taylor K, Muraina I, Brethour D, Schmitt-Ulms G, Nimmanon T, Ziliotto S et al. Zinc transporter ZIP10 forms a heteromer with ZIP6 which regulates embryonic development and cell migration. *Biochem J*. 2016;**473**(16):2531-2544.
4. Taylor K, Hiscox S, Nicholson R, Hogstrand C, Kille P. Protein kinase CK2 triggers cytosolic zinc signaling pathways by phosphorylation of zinc channel ZIP7. *Sci Signal*. 2012;**5**(210):ra11-ra11.

Numerical Approach to Investigate Adhesion Between Dental Biomaterials and Relevant Bacteria

Chloe Bennett and P Prokopovich
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Around 10,000 dental implant procedures are carried out on the lower jaw in the UK annually.[1] Bacteria adhere to tooth and restorative material surfaces, leading to the formation of complex structures called biofilms.[2] The development of biofilms predisposes an individual to infections, requiring the prescribing of

antibiotics. With the growing global problem of antibiotic resistance, this generated the motivation for this study. A greater understanding of the factors influencing the adhesion and the material properties which encourage adhesion would allow minimisation of bacterial adhesion and therefore complications post-implantation.

The aim of this study was to determine how different material properties (surface energy, elastic modulus, Poisson ratio) affect adhesion of bacteria to the material surface. The parameter data was sourced from open literature and input into a numerical code developed by Dr Prokopovich group[3], using a multi-asperity model which was adapted from the single-asperity Johnson-Kandall-Roberts (JKR) model of adhesion.[4] This generated quantitative data on adhesive force between two surfaces, which was repeated a minimum of six times and analysed by one-way ANOVA tests to determine the statistical significance of the data ($p<0.05$).

The adhesion force data collected showed stainless steel to have the lowest average adhesive forces with all bacteria tested. Adhesion of the gram-negative bacterium *Escherichia coli* to all biomaterials was significantly lower than the other bacteria. Adhesion of *Escherichia coli* to the stainless steel surface was over 23 times lower than seen with *Streptococcus mutans*. The results suggested that a higher surface energy increases adhesion due to favourable adhesion of the bacterium to reduce the overall free energy of the system. However, other factors influence adhesion such as the reduced presence of adhesins on the gram-negative bacterial surface. Adhesion is multi-factorial so further studies need to be carried out in the future to understand more influences of adhesion.

1. BBC News. Dentists warn of risks of not looking after implants. 2015 [accessed 06 Feb 2019]. Available from: https://www.bbc.co.uk/news/health-32086134
2. Flausino JS, Soares PBF, Carvalho VF, Magalhães D, da Silva WM, Costa HL, et al. Biofilm formation on different materials for tooth restoration: analysis of surface characteristics. J Mater Sci. 2014;49(19):6820-9.
3. Prokopovich P, Perni S. Multiasperity contact adhesion model for universal asperity height and radius of curvature distributions. Langmuir. 2010;26(22):17028-36.
4. Barthel E. Adhesive elastic contacts – JKR and more. *J Phys D Appl Phys*. 2008;41:163001.

Assessing the stability of cytarabine within a Continuous Ambulatory Delivery Device (CADD) using High Performance Liquid Chromatography (HPLC)

Sandy Berriman, AG Cosslett and CP Thomas
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Cancer incidence is on the rise, which is putting a strain on the NHS and costing billions.[1] With the development of Continuous Ambulatory Delivery Devices (CADDs), chemotherapy can be given within a home setting. Cytarabine, a chemotherapeutic agent used in several treatment regimens, has been suggested to be infused using the CADD. The current information on the stability of the drug within the device is limited.[2] Therefore it was deemed necessary to assess the stability of the drug within the device to replicate its storage and use in practice.

Fifteen CADD cassettes were prepared containing 20mg/ml of cytarabine solution in Water for Injections. Ten cassettes were stored at 40°C and five were stored at 4°C for 14 days. Three samples of 5ml were taken from 2 cassettes from the 40°C and 1 cassette from the 4°C batches at 0, 4, 7, 12 and 14 days. Each sample was diluted to 0.2mg/ml and analysed using a developed High Performance Liquid Chromatography (HPLC) method. Physical tests including visual testing, pH and turbidity were also conducted on each of the samples.

The cassettes stored at 4°C maintained above 90% of the original cytarabine concentration over the 14 day period. No increase in degradation peak area was observed over the time period. The cassettes stored at 40°C showed an increase in the degradation peak area at 12 days suggesting instability. Physical testing showed no significant change over the 14 days for either temperature.

Results show that cytarabine is stable for between 7-12 days when in use within the device. A minimum of seven days' supply can be loaded within the CADD and infused to a patient within a home setting with the knowledge that the drug is stable within the device. Cassettes can also be made up in advance and stored before use.

1. Department of Health & Social Care. *2010 to 2015 government policy: cancer research and treatment*. 2015 [accessed 31 Oct 2018]. Available from: https://www.gov.uk/government/publications/2010-to-2015-government-policy-cancer-research-and-treatment/2010-to-2015-government-policy-cancer-research-and-treatment

2. Smiths Medical. *Pharmaceutical report on chemical and physical stability of drugs commonly used in CADD® infusion pumps.* [no date] [accessed 11 Nov 2018]. Available from: https://m.smiths-medical.com/~/media/M/Smiths-medical_com/Files/Import%20Files/IN193109GB-032017_LR.pdf

Investigating TEAD1, VGLL1 and VGLL3 proteins in anti-hormone resistant breast cancer cells

Sarah Bickford, L Thomas and JMW Gee
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Anti-hormones are established treatments for oestrogen receptor positive (ER+) luminal-A and B breast cancer. However acquired resistance remains a clinical issue and this can involve ER loss.[1] At Cardiff University, recent research found that loss of ER in luminal-A-derived breast cancer cells with long-term acquired anti-hormone resistance is paralleled by gain in the transcription factor TEAD1 and its coactivators VGLL1/3 at the mRNA-level.[2] This project aimed to investigate whether TEAD1/VGLL1/VGLL3 are also upregulated at the protein-level, profiling luminal-A and B-derived models with acquired resistance. It also considered if this TEAD/VGLL pathway was independent of EGFR and Src-kinase pathways, which have already been targeted (with limited success) in anti-hormone resistance. [3, 4]

Immunocytochemistry assays were optimised for TEAD1/VGLL1/VGLL3 proteins using anti-hormone resistant in vitro model coverslips and paraffin-embedded (FFPE) pellet sections, including cells treated with 1μM EGFR inhibitor (Gefitinib) or Src-kinase inhibitor (Saracatinib). H-scoring semi-quantified TEAD1/VGLL1/VGLL3 nuclear expression, with non-parametric statistical analysis versus control cells. ER-stained slides were also assessed. Finally, sections of an anti-hormone resistant xenograft were immunostained.

Nuclear TEAD1 and VGLL1 were significantly increased in luminal-A (T47D)-derived cell models with acquired Faslodex or oestrogen deprivation resistance which had complete ER loss, but not in Tamoxifen resistance which retained some ER. Nuclear TEAD1 and VGLL1 were also detected in a luminal-A-derived resistant xenograft with ER loss. In luminal-B (MDA361)-derived models, TEAD1 was absent and VGLL1 only slightly increased in Tamoxifen and Faslodex resistant cells with decreased ER. Neither TEAD1 nor VGLL1 were suppressed by EGFR or Src inhibitors. VGLL3 was mostly negative in all the models.

Upregulation of nuclear TEAD1/VGLL1 proteins in luminal A-derived, acquired anti-hormone resistant models with ER loss suggests the TEAD/VGLL pathway may be functional. Given TEAD1/VGLL1 were also EGFR/Src-independent, they may warrant further investigation to evaluate potential as a novel target in equivalentpatients.

1. Clarke, R. et al. 2015. Endocrine resistance in breast cancer- An overview and update. *Molecular and Cellular Endocrinology* **408**(2015), pp. 220-234. doi: http://dx.doi.org/10.1016/j.mce.2015.09.035.
2. 2. Davies, L. 2018. TEAD1 in Endocrine Resistant Breast Cancer and its interplay with VGLL1. MSc dissertation. Cardiff University.
3. Gutteridge, E. et al. 2009. The effects of gefitinib in tamoxifen-resistant and hormone-insensitive breast cancer: A phase II study. *International Journal of Cancer* **126**(8), pp. 1806-1816. doi: https://doi.org/10.1002/ijc.24884
4. Hiscox, S. et al. 2010. Combining Src inhibitors and aromatase inhibitors: A novel strategy for overcoming endocrine resistance and bone loss. *European Journal of Cancer* **46**(12), pp. 2187-2195.

The Development of High Strength Loperamide Suspensions

Ebubechi Osuji , CP Thomas and JC Birchall
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Loperamide hydrochloride is only commercially available and licensed in the UK as a 1mg/5ml solution and 2mg capsules. It is used at a standard dose of 6-8mg daily for antidiarrheal action.[1] However, it is also used to treat high output stoma and short bowel syndrome patients with doses of up to 64mg daily which is about 32 capsules a day.[2] Therefore, this project aims to develop higher strength loperamide suspensions at 8mg/5ml and 16mg/5ml in response to clinical requests submitted to the Saint Mary Pharmaceutical Unit.

A formulation was initially created using the loperamide active, sucralose and a vehicle which contained xanthan gum and some preservatives. This formulation however, required further optimisation hence, a surfactant; Tween 80 was initiated into the formulation. Due to an interaction between xanthan gum and Tween 80, the amount of xanthan gum used was reduced and a preferred formulation was developed. This

formulation was tested for content uniformity, stability and dissolution with quantification using a High-Pressure Liquid Chromatography (HPLC) method.

The preferred formulation had low viscosity and a high sedimentation rate which are not ideal properties of a suspension formulation. However, loperamide was proven to remain stable drug following storage under accelerated conditions. Unfortunately, we were unable to effectively demonstrate content uniformity and drug dissolution of the suspensions developed due to issues in analysing drug content via HPLC.

Our collaborators at St Mary's Pharmaceutical Unit have expressed interest in further research and optimisation of these products. Future considerations include the use of an ionic surfactant and a viscosity modifier. The content uniformity and dissolution test also needs be repeated when a more suitable formulation has been developed.

1. Joint Formulary Committee. British National Formulary. 76th ed. London: BMJ Group and Pharmaceutical Press, 2018.
2. U.K. Medicines Information. Can high dose loperamide be used to reduce stoma output? England: NHS, 2013. [accessed 01 Dec 2018] Available from: http://www.surreyandsussex.nhs.uk/wp-content/uploads/2013/04/UKMi-High-DoseLoperamide-Stoma.pdf

Influences on the uptake of technology in community pharmacies

Jennifer Bristow and MW Smith
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

The role of community pharmacists is becoming increasingly clinical, as the NHS evolves to provide more integrated care between multiple healthcare professionals to fully utilise pharmacists' skills.[1] However, changes to funding have increased emphasis on clinical services[2] whilst prescription volumes increase[3], which has put financial pressure on community pharmacies.[2] This has led to recommendations to increase technology use within community pharmacies, freeing pharmacists' time up to perform a more clinical role.[4] Technology can also bring increased integration with the wider healthcare environment and improve efficiency.[4] The aim of this project was to understand what influences uptake of technologies within community pharmacies.

Surveys were posted to 200 randomly selected independent community pharmacies in England and Wales. Multiples were excluded on the basis that they would have less autonomy about technology use, as this would be decided by management. Ethical approval was granted from Cardiff University before the surveys were disseminated. Questions included asking from which sources participants learnt about new technologies, and what the most important features of new technologies would be for them. This data was then analysed using SPSS to look for key trends.

A response rate of 22% was achieved. Respondents indicated peers were one of the most influential factors, with friends and colleagues working in pharmacies being the most likely source of information for finding out about new technologies. Their opinions and recommendations were seen as more important than leading organisations. However, social media was seen as a very uninfluential source. If respondents were going to change their PMR system the most important features were ease of use, support with implementation and cost effectiveness.

This study identified the extent of some influences on pharmacists' uptake of technology. However, it was limited by the low response rate, so future research could involve using improved methodology to increase returns.

1. The Scottish Government. Prescription for Excellence: A Vision and Action Plan for the right pharmaceutical care through integrated partnerships and innovation. 2013 [accessed 7 Oct 2018]. Available from: http://www.gov.scot/resource/0043/00434053.pdf
2. Thomas M, Anscombe J, Plimley J. The Future of Community Pharmacy in England. 2012. [accessed 12 Oct 2018]. Available from: https://www.atkearney.com/documents/20152/434294/The%2BFuture%2Bof%2BCommunity%2BPharmacy.pdf/d9c24e84-9870-1ffb-7860-7d768c43e9d4RPS.
3. Welsh Government. Community pharmacy services in Wales, 2017-18. 2018.[accessed 22 Dec 2018]. Available from: https://gov.wales/docs/statistics/2018/181031-community-pharmacy-services-2017-18-en.pdf
4. RPS. Improving Health and Patient Care through Community Pharmacy– A Call to Action Royal Pharmaceutical Society (RPS) response. [accessed 21 Oct 2018]. Available from: https://www.rpharms.com/Portals/0/Documents/Oldconsultations/consdoc140318.pdf

An evaluation of the effect of inhaler technique and adherence on asthma control

Alice Broom and WR Ford
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Asthma is an inflammatory condition of the respiratory tract resulting in airflow obstruction. Despite the availability of medications clinically proven to treat asthma, the disease was still responsible for 14,830 deaths in England from 2002-2015.[1] There are three clear factors that potentially impact asthma management: inhaler technique; adherence and efficacy of drugs to treat asthma. This study aims to investigate and identify factors that influence asthma control within the asthmatic population of South Wales.

Participants were enrolled on to the study by convenience sampling, completing an Asthma Control Test (ACT) and adherence questionnaire to determine level of asthma control and adherence to regular asthma medications within a community pharmacy setting.[2,3] Participants demonstrated inhaler technique using a Vitalograph Aerosol Inhalation Monitor (AIM) device to provide an objective measure. Qualitative data was analysed using Statistical Package for Social Sciences (SPSS). Mann-Whitney U and Kruskal-Wallis tests were conducted to compare participants with controlled and uncontrolled asthma. An ethics approval was obtained prior to study conduct.

Data from 109 participants was analysed. The highest frequency of good inhaler technique was recorded with Dry Powder Inhalers (DPI's) (39%). 67% of the inhalers reported in the study were Metered Dose Inhalers (MDI's) which had a fail rate of 69%. Levels of adherence within the study group were lower than expected, with 29% identified with good adherence. 71% of participants taking regular asthma medication reported having intermediate or poor adherence.

No statistically significant relationship was found between inhaler technique and asthma control, nor between adherence and asthma control. Device type did not affect asthma control, leaving the efficacy of drugs to treat asthma in question. Clinical trials for such medication may have overestimated their effectiveness, however further studies would need to be conducted to establish this.

1. Gupta R, Mukherjee M, Sheikh A, Strachan D. Persistent variations in national asthma mortality, hospital admissions and prevalence by socioeconomic status and region in England. *Thorax.* 2018; 73(8):706. doi:10.1136/thoraxjnl-2017-210714
2. Schatz M, Sorkness C, Li J, Marcus P, Murray J, Nathan R et al. Asthma Control Test: Reliability, validity, and responsiveness in patients not previously followed by asthma specialists. *J Allergy Clin Immunol.* 2006;117(3):549-56. doi:10.1016/j.jaci.2006.01.011
3. Plaza V, Fernandez-Rodriguez C, Melero C, Cosio B, Entrenas L, Perez de Llano L et al. Validation of the "Test of the Adherence to Inhalers" (TAI) for asthma and COPD patients. *J Aerosol Med Pulm Drug Deliv*. 2016;29(2):142-52. doi: 10.1089/jamp.2015.1212

Maximising endotoxin recovery from clinical samples using different sample preparation techniques

Chalina Chan, N Demertzis and JL Bowen
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue Cardiff CF10 3NB, Wales, UK

Sepsis is an often fatal condition arising from infection.[1] Outer cell walls of Gram-negative bacteria contain endotoxins/lipopolysaccharides (LPS) which play a big role in the development of sepsis in humans. Therefore, LPS may be a useful biomarker.[1] The Limulus Amoebocyte Lysate (LAL) Assay is a quantitative way of detecting endotoxin from pharmaceutical samples but it is prone to interference from clinical samples.[2] The aim of this project was to investigate different sample pre-treatment approaches to enable efficient recovery of endotoxin from plasma samples.

Plasma samples were spiked with different, known concentrations of LPS (5-50pg/ml). Dilution of plasma, heat and acid treatment have been reported as possible methods for recovering endotoxin from clinical samples.[3] These methods were used individually and in combination with each other prior to testing using the Charles River 'Endosafe® nexgen-PTS™. Results were compared with a standard calibration, obtained from testing different standard solutions of LPS, to determine degree of recovery.

Application of each of the sample preparation approaches individually yielded poor recoveries, with the majority of results sitting below 10%. The most promising result came from using 2.5% hydrochloric acid treatment with a dilution of sample 1:10 with LAL reagent water to recover endotoxin from a sample that was spiked with a final LPS concentration of 5pg/ml. This combined approach led to ~100% recovery, however, when higher spike concentrations were investigated it was much less effective.

Throughout the project, by exploring and using different preparation techniques, improvements were seen in recovering endotoxin from plasma samples. However, the same technique(s) used for one concentration of spiked LPS did not necessarily produce the same/similar result for a higher/lower concentration. The use of an apta-MIP sensor instead of enzyme based assays could be an alternative for detecting endotoxin in clinical samples, due to its high sensitivity and stability.[4]

1. Manthous CA, Hall JB, Samsel RW. Endotoxin in Human Disease; Part 1: Biochemistry, Assay, and Possible Role in Diverse Disease States. *Chest*. 1993; **104**:1572-81.
2. Nachum RO, Berzofsky RN. Chromogenic Limulus amoebocyte lysate assay for rapid detection of gram-negative bacteriuria. *J Clin Microbiol.* 1985; **21**(5):759-63.
3. DuBose DA, Lemaire M, Basamania K, Rowlands J. Comparison of Plasma Extraction Techniques in Preparation of Samples for Endotoxin Testing by the *Limulus* Amoebocyte Lysate Test. *J Clin Microbiol.* 1980; **11**(1):68-72.
4. Das AP, Kumar PS, Swain S. Recent advances in biosensor based endotoxin detection. *Biosens Bioelectron.* 2014; **15**(51):62-75.

Insertion and characterisation of pore forming proteins in Droplet Interface Bilayers

Marianne Collins and O Castell
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Understanding how protein pores work could be instrumental in harnessing their function to develop methods of site-specific drug delivery in the future.[1] Protein pores can be studied in artificial lipid bilayers using electrophysiology. Droplet Interface Bilayers (DIB) have recently been developed to succeed traditional bilayer techniques and provide a stable, simple method of measuring ion flux through protein pores which.[2] The aim of this research was to develop reconstitution strategies for a range of toxin pore forming proteins into DIBs and characterise their behaviour.

DIBs were made using two aqueous droplets suspended on electrodes and submerged in a lipid solution. Lipid monolayers formed around the droplets and moving them together created a lipid bilayer.[2] Protein pores present in the aqueous solution could insert in the bilayer and allowed ion flux, which was measured using electrophysiology. The current traces produced were used to characterise the pores.

Conductance of individual toxin pore insertions were typically measured at 8-10pA. Proteins with theorised structural similarity were found to have similar measured conductance. Unexpectedly, some evidence of pore-like behaviour was witnessed in control experiments with water soluble toxin protein which was not-expected to form membrane-spanning channels. However, unlike the known pore-forming proteins where activity was abolished on heating the protein, these samples continued to exhibit activity, warranting further investigation.

This research developed a generalisable strategy for reconstitution of a number of toxin proteins into DIBs. The pore-forming behaviour observed for proteins not expected to exhibit pore forming behaviour was surprising and may arise from more general membrane destabilisation mechanisms or as a result of sample contamination. The limited knowledge surrounding such toxin proteins has been increased by these studies and may be beneficial to the drug delivery field in the future.

1. St Jean, A. T. et al. 2014. Bacterial delivery of Staphylococcus aureus alpha-hemolysin causes regression and necrosis in murine tumors. *Mol Ther* 22(7), pp. 1266-1274. doi: 10.1038/mt.2014.36
2. Bayley, H. et al. 2008. Droplet interface bilayers. *Molecular bioSystems* 4(12), pp. 1191-1208. doi: 10.1039/b808893d

---

Pastoral support for pre-registration and newly qualified pharmacists: what is needed and how should it be provided?

Olivia Dalton and KL Hodson
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

NHS Wales have had a recent initiative from the Welsh Government to invest in their staff.[1] There is evidence that poor well-being can affect people's work and how a poor work life can affect people's well-being. [2,3] Therefore, Aneurin Bevan University Health Board (ABUHB) sought feedback on the pastoral support they provide to their pre-registration and newly qualified pharmacists. Thus, the aim of this study was to establish the challenges for this group of staff, how support for their well-being could be improved and how it should be delivered.

Approval was obtained from Cardiff University School of Pharmacy and Pharmaceutical Sciences Ethics Committee and Research and Development at ABUHB. All pre-registration and newly qualified pharmacists within ABUHB were invited, via email, to participate in the study. Once participants' consent was obtained, three focus groups were conducted across two hospitals. A semi-structured interview guide was used to conduct the focus groups. All groups were recorded using Philips Pocket Memo DPM6700 and transcribed verbatim. Inductive and deductive analysis was undertaken to identify themes.

A number of personal and professional challenges were identified, for example time management and feeling undervalued by staff. Feedback on current support provision was mixed and linked to the individual. It highlighted that pastoral support tended to be reactive, except at one hospital, and seemed to be linked to working relationships. Suggestions for improvement to the support provided included communicating and promoting the services available for well-being.

In conclusion to optimize staff well-being it is essential that pastoral support is individualized and that all members of staff take responsibility for building relationships and supporting each other. Support needs to be given proactively to prevent minor issues from escalating.

1. Welsh Government. 2018. *A Healthier Wales: our Plan for Health and Social Care.* Available at: https://gov.wales/docs/dhss/publications/180608healthier-wales-mainen.pdf
2. Picker Institute Europe. 2015. *Understanding staff wellbeing, its impact on patient experience and healthcare quality.* Available at: https://www.picker.org/wp-content/uploads/2015/06/2015-06-10-StaffWellbeingBriefing.pdf
3. Royal College of Physicians. 2015. *Work and wellbeing in the NHS: why staff health matters to patient care.* Available at: https://www.rcplondon.ac.uk/guidelines-policy/work-and-wellbeing-nhs-why-staff-health-matters-patient-care

---

Design and synthesis of Novel CYP51 inhibitors as Therapeutics for *Candida* Infection

Katie Davies and C Simons
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Candida infections are the most common type of fungal infections [1] and are becoming a growing concern as many *Candida* species are now resistant to the first and second line treatments- azoles and echinocandins.[2] This has led for the need to develop new azole type antifungals (CYP51 inhibitors) in order to treat the multidrug resistant *Candida.*[3] Therefore, the aim of this research project is to design and synthesise new CYP51 inhibitors as a treatment for *Candida* infections.

Firstly, Molecular Operating Environment was used to dock and compare the novel compounds with the crystal structure posoconazole. This was used to determine the similarity of the novel compounds with their interactions in the CYP51 active site. A four step experimental method was used and this produced six final novel CYP51 inhibitors. The final compounds and intermediates were analysed and verified using TLC, ^{1}H and ^{13}C NMR. This was to make sure the right compounds were produced and the purity was high enough for biological testing.

Two out of the six compounds synthesised had impurities with one of these having to be remade however all six compounds will go on to be biologically tested against two different *Candida* strains to see if they are effective treatments. With such low yields produced, the experimental method must be optimised this could be done by trying to purify the amine in the earlier stages of the experiment or see if there is another method that could produce higher yields. The four step method was successful as six novel compounds were produced however it will need to be optimised before the reactants can be scaled up to produce more compounds.

1. Vázquez-González D, Perusquía-Ortiz AM, Hundeiker M, Bonifaz A. Opportunistic yeast Infections: candidiasis, cryptococcosis, trichosporonosis and geotrichosis. J Dtsch Dermatol Ges. 2013; 11(5): 381-93. doi: https://doi.org/10.1111/ddg.12097
2. Whaley SG, Berkow EL, Rybak JM, Nishimoto AT, et al. Azole Antifungal Resistance in *Candida albicans* and Emerging Non-albicans *Candida* Species. Front Microbiol. 2017;7:2173. doi: 10.3389/fmicb.2016.02173
3. Strushkevich N, Usanov SA, Park HW. Structural basis of human CYP51 inhibition by antifungal azoles. J Mol Biol. 2010;397(4):1067-78. doi: 10.1016/j.jmb.2010.01.075

Investigation of Antibiotic Prescribing by NHS Primary Care Dentists across Wales between April 2012 and August 2018.

[1]Alice Dullehan, [2]K Haines, [2]PN Deslandes and [1]MO Ivory
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK. [2]All Wales Therapeutics and Toxicology Centre (AWTTC), University Hospital Llandough, Penlan Road, Llandough, Penarth, The Vale of Glamorgan, UK

Antimicrobial resistance is a growing threat to global health.[1] Dentists contribute to a substantial part of the antibiotic prescribing cohort, however, there was a lack of quantitative data on dental antibiotic prescribing practice at a national and health board level.[2] This study aimed to evaluate trends in prescribing by NHS primary care dentists across Wales whilst assessing adherence to guidelines and to identify gaps for further education of dental professionals at a national and health board level.

Quantitative, anonymous and monthly prescribing data on dental antibiotic prescribing between April 2012 and August 2018 was extracted and analysed using Comparative Analysis System for Prescribing Audit (CASPA). This was compared to the existing body of literature surrounding dental antibiotic prescribing to identify compliance to current guidelines.

Overall dental antibiotic prescribing decreased over the study period. Amoxicillin and metronidazole were the most prescribed dental antibiotics across Wales. Contrary to current guideline recommendations, since April 2012, erythromycin and its salts has been the third most prescribed antibiotic across Wales. The pattern of dental antibiotic prescribing appeared to peak between April 2012 to March 2013. A varied preference on the antibiotic choice by dentists was observed across the different health boards. This was demonstrated by the differences in rank of most commonly prescribed dental antibiotics between April 2017 to March 2018.

First line drugs recommended are; amoxicillin, metronidazole and penicillin V however this is not reproduced clinically, suggesting that guidelines have not been fully implemented into clinical practice. [3]The absence of All Wales dental guidelines creates difficulties in attempts to streamline prescribing practice across Wales. Further study into the reasons behind the observed trends would enable more successful antibiotic stewardship methods to be implemented into clinical practice and establish why guidelines are not being followed.

1. WHO 2015. Global action plan on antimicrobial resistance. *WHO Press*, pp. 1–28.
2. Cope, A.L. and Chestnutt, I.G. 2014. Inappropriate Prescribing of Antibiotics in Primary Dental Care: Reasons and Resolutions. *Primary Dental Journal* **3**(4), pp. 33–37.
3. SDCEP 2016. *Drug Prescribing For Dentistry.*

An investigation into the use of an additional excipient to retard the release of Gentamicin from bone cements, without affecting their mechanical properties

Daniel Ellesmere, P Hansal and JC Birchall
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward XVII Avenue, Cardiff, CF10 3NB, Wales, UK

Gentamicin is a commonly used antibiotic within polymethylmethacrylate (PMMA) bone cements, which have a very important use in orthopaedic surgery.[1,2] Yet there are fundamental problems with the release characteristics of gentamicin from the cement and concerns that this may increase the risks of antibiotic resistance.[3] The aim of this project was to investigate whether an additional excipient could modify the release of gentamicin from bone cement formulations, compared to a commercially available preparation (Palacos R+G), without adversely affecting the mechanical properties of the cement.

The effects of additive concentration and mixing technique on the formulation were explored. Scanning electron microscopy and particle size analysis helped provide an understanding of the interaction between additive and gentamicin and the aggressiveness of the mixing process. A compression test was carried out on cement cylinders to determine the impact of the addition of the additive on the mechanical strength of the formulation. A gentamicin release study using cement disks was conducted over four weeks, with samples taken at different time points and analysed using liquid chromatography – mass spectroscopy.

The different mixing methods produced varying effects on the size, structure and distribution of the particles as well as the extent of additive surface coating of gentamicin. The degree of coating was also affected by additive concentration. All the novel cement formulations significantly slowed the release rate of gentamicin compared to Palacos R+G, without any significant differences on the mechanical strength of the cement.

These results demonstrate the potential for the use of additional excipients within bone cements to alter the release of gentamicin. Further studies are required to determine the effects such additives have on the long-term release of gentamicin from the cements and whether the rate of release results in levels of gentamicin that would prevent infection without promoting antibiotic resistance.

1. 1 Chang Y, Tai C, Hsieh P, Ueng SWN. Gentamicin in bone cement: A potentially more effective prophylactic measure of infection in joint arthroplasty. Bone Jt Res. 2013;**2**(10):220–6.
2. 2 National Joint Registry. NJR Research Project Portfolio. 2017. [accessed 17 Dec 2018] Available from: http://www.njrcentre.org.uk/njrcentre/Portals/0/Documents/England/ Research/Research Portfolio Uploads/NJR Research Project Portfolio - v 15 October 2017.pdf?ver=2017-10-17-104831-383
3. 3 Nishio Ayre W, Birchall JC, Evans SL, Denyer SP. A novel liposomal drug delivery system for PMMA bone cements. J Biomed Mater Res Part B. 2015;**104**(8):1510–24.

Role of adhesive forces in the interactions between orthopaedic devices and bacteria: a numerical approach

Lara Fraser and P Prokopovich
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

A combination of complicated, time-reliant diagnosis and treatment complicated by antibiotic resistance makes orthopaedic device-related infection the most serious complication in patients with prosthesis. ODRI's negatively impact patient outcomes and increase expenditure on the NHS.[1] Studies have demonstrated the mechanisms of bacterial adhesion to biomaterials[2], thus this study aims to describe the relationship between material characteristics and bacterial adhesion.

The JKR multi-adhesion model by Dr. Prokopovich and Perni[3] formed the basis of the numerical investigation, aiming to find values for bacterial adhesion between chosen bacteria and orthopaedic material combinations. It could then be concluded which material produces the least bacterial adhesion, thus most effective in reducing ODRI.

Results showed polypropylene to be the most effective material in reducing bacterial adhesion to *S. epidermis, S. aureus, E. coli and P. aeruginosa* while alumina showed the highest mean bacterial adhesion. The additions of silver and chitosan coatings to the material showed no additional benefit in reducing bacterial adhesion, but this deviates from the results of current in vitro studies.[4]

In conclusion, the study showed positive relationships between reducing surface energy and surface roughness and increasing material hydrophobicity and material elasticity with decreased bacterial adhesion. The results are in correlation with previous studies in the research field, showing them as a promising basis for further investigation. However, the study was restricted by time, with all findings entirely theoretical and not wholly representative of the complex process of bacterial adhesion within the body.

1. 1- Moriarty TF, Kuehl R, Coenye T, Metsemakers WJ, Morgenstern M, Schwarz EM, Riool M, Zaat SA, Khana N, Kates SL, Richards RG. Orthopaedic device-related infection: current and future interventions for improved prevention and treatment. *EFORT open reviews*. 2016;**1**(4):89-99.
2. 2- Katsikogianni M, Missirlis YF. Concise review of mechanisms of bacterial adhesion to biomaterials and of techniques used in estimating bacteria-material interactions. *Eur Cell Mater*. 2004;**8**(3):37-57.
3. 3- Prokopovich P, Perni S. Multiasperity contact adhesion model for universal asperity height and radius of curvature distributions. *Langmuir*. 2010;**26**(22):17028-36.
4. 4- Bumgardner JD, Wiser R, Gerard PD, Bergin P, Chestnutt B, Marini M, Ramsey V, Elder SH, Gilbert JA. Chitosan: potential use as a bioactive coating for orthopaedic and craniofacial/dental implants. *Journal of Biomaterials Science, Polymer Edition*. 2003;**14**(5):423-38.

Evaluation of the Common Ailment Service (CAS) in Wales over the past five years

Daniel Freeman, KL Hodson, ML Hughes and [1]A Evans
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, Redwood Building, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK and [1]Welsh Government, Cathays Park, Cardiff, CF10 3NQ, Wales, UK

A common ailment is an acute health condition that can be treated by patients themselves with simple actions.[1] Reports show in a 12-month period, 34% of patients visited a general practitioner (GP) for self-treatable minor ailments and 12% went directly to an accident and emergency department.[2] Therefore, the Community Pharmacy CAS was introduced to Wales during October 2013.[3] It is a free service allowing patients to seek advice and/or treatment for 26 common conditions.[3] The overall aims of the CAS are to allow pharmacists to

utilise their skills and treat patients without them having to book a GP appointment, thus freeing up GP time.[3] This study aims to evaluate the CAS between 2013-2018.

Secondary data analysis was used. This dataset, containing details of every patient consultation under the CAS from 20/09/2013-30/09/2018, was acquired from NHS Wales Informatics Service. This quantitative data was analysed using IBM® SPSS and Microsoft® Excel. Ethical approval was not required as the data received was anonymous secondary data.

A total of 49,985 consultations were recorded over the five years, rising from 2,646 in 04/14-03/15 to 16,099 in 04/17-03/18. These consultations were for 38,164 patients and undertaken within 630 community pharmacies. The most common condition was hay fever with 11,264 (23%) consultations contrasting with the least common, colic with 4 (0.01%) consultations. The cumulative percentage for the top 10 most common conditions accounted for 86% of all consultations. In contrast, the bottom 10 conditions accounted for just over 4%.

The CAS has been increasing in number of consultations and pharmacies implementing the service since its introduction in 2013. These increasing numbers are encouraging in helping to meet the Welsh Government 'Choose Well' campaign which promotes self-care.[4] However, a review of the CAS conditions is recommended due to many of these conditions being underutilised and therefore unnecessary.

1. Welle-Nilsen LK, Morken T, Hunskaar S, Granas AG. Minor ailments in out-of-hours primary care: an observational study. *Scand J Prim Health Care.* 2011;**29**(1):39-44. doi:10.3109/02813432.2010.545209
2. Proprietary Association of Great Britain. Self care nation - self care attitudes and behaviours in the UK. 2016 [accessed 15 Dec 2018]. Available from: https://www.pagb.co.uk/content/uploads/2016/11/PAGB-SELF-CARE-NATION-REPORT-NOVEMBER-2016-1.pdf
3. Welsh Government. Evaluation of the choose pharmacy common ailments service: final report. 2015 [accessed 16 Dec 2018]. Available from: https://gov.wales/docs/caecd/research/2015/150730-evaluation-choose-pharmacy-common-ailments-service-en.pdf
4. Welsh Government. Choose well. 2016 [accessed 19 Sep 2018]. Available from: https://gov.wales/topics/health/nhswales/healthservice/choosewell/?lang=en

---

To explore the knowledge and perceptions of pharmacy students regarding the possibility of prescribing post pre-registration exams.

Tanisha Gandhi, F Ali and M Weiss
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Pharmacists have outstanding prescribing practice, bringing vast benefits to patients, healthcare professionals and an opportunity for career progression. [1,2] The NHS is struggling, [3] yet only 7% of qualified pharmacists are independent prescribers. [4] Hence, healthcare initiatives to increase efficiency could be attainable by allowing pharmacists to have the ability to prescribe within their competency, immediately after pre-registration. This study therefore aims to explore undergraduate students' perspectives on this proposed change.

Prior to conducting focus groups ethical approval, to protect participants and researcher alike, was attained. Fourth-year and second-year Cardiff University students participated via purposive sampling. A topic guide was created to aid the semi-structured approach of conducting the focus groups. Sessions were audio recorded and transcribed verbatim, allowing thematic analysis.

Twenty-three students participated between four focus groups, leading to the identification of three themes with several sub-themes. Participant views of prescribing; barriers associated with independent prescribing; and future changes required to enable pharmacist independent prescribing (PIP) immediately after pre-registration, were the main themes. Major barriers regarding students' reluctance towards PIP included insufficient clinical knowledge and placements to consolidate theoretical knowledge in the undergraduate degree. This also contributed to the lack of confidence to potentially prescribe medications, further heightened by limited access to patient medical records.

For pharmacy to progress from a primarily dispensing role, which is often the public's perception, changes to the undergraduate course appear to be the most viable option. For example, tailoring the course to integrate the PIP certification, providing a higher standard of work placements, and removing some significant science content was necessary. Changes to improve accessibility to the PIP course could also encourage a larger uptake by qualified pharmacists. However, more research is required to understand the views of pharmacy students across the UK, for appropriate changes to be made by those in a position of leadership.

1. Avery A J, Rodgers S, Cantrill J A, Armstrong S, Cresswell K, Eden M, et al. A pharmacist-led information technology intervention for medication errors (PINCER): a multicentre, cluster randomised, controlled trial and cost-effectiveness analysis. The Lancet. 2012;379(9823):1310-1319. doi: 10.1016/S0140-6736(11)61817-5
2. Pharmaceutical Services Negotiating Committee. Case study: Community pharmacist independent prescribers can alleviate GP supply-demand issues; 2018 [accessed: 26 Dec 2018]. Available from: https://psnc.org.uk/our-news/services-case-study-community-pharmacist-independent-prescribers-can-alleviate-gp-supply-demand-issues/
3. Williams H and Stone M. The benefits of employing pharmacists in GP practices. England: Medeconomics; 2014 [accessed 23 Dec 2018]. Available from: https://www.medeconomics.co.uk/article/1286630/benefits-employing-pharmacists-gp-practices
4. Robinson S. Why this NHS hospital trust asks all its clinical pharmacists to become independent prescribers. Pharm J. 2015;294:7853 [accessed 18 Oct 2018]. Available from: https://www.pharmaceutical-journal.com/your-rps/why-this-nhs-hospital-trust-asks-all-its-clinical-pharmacists-to-become-independent-prescribers/20068078.article?firstPass=false

Validity of colour spot tests on MDMA and its analogues

Emmanuel Ghunney and AD Westwell
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

It was reported in 2016 that there were 320 deaths related to any amphetamine drug[1] which highlights an issue. The MDMA reagent test kit claims that their kit is useful in reducing harm prevention from MDMA as it states it can accurately detect MDMA in its different forms, its analogues and adulterated forms[2]. The aim of my project is to see whether this test will be able to do as it states as well test substances like 2C-B as it can be confused with MDMA.

Before the test was started a control using a white crystalline powder was set up. This was to see if the reagent test kit would actually work and this was repeated five times and the initial colour change was documented. The test was then split into different sections: the colour section, its concentration, MDMA mixed with other drugs and its analogues as well as 2C-B. A tiny amount of every drug tested was placed into a ceramic spot dish test and each test was repeated three time with a droplet of each reagent.

It was found that the reagent test kit was able to detect MDMA regardless of colour but it could not determine the different concentrations. The kit also could not distinguish between adulterated forms of MDMA. Although the kit could tell the difference between analogues which are very closely related it cannot. The kit could also determine 2C-B.

The MDMA reagent test kit is not an effective way for harm prevention because although it is able to identify MDMA effectively it cannot determine many things like its concentration and adulterated forms and closely related analogues.

1. Office of National Statistics. Deaths related to drug poisoning in England and Wales: 2016 registrations. 2017 [Accessed at 16 January 2019]. Available from: https://www.ons.gov.uk/peoplepopulationandcommunity/birthsdeathsandmarriages/deaths/bulletins/deathsrelatedtodrugpoisoninginenglandandwales/2016registrations#deaths-involving-selected-substances
2. Reagent Tests UK. MDMA Reagent Test Kit. United Kingdom: Reagent Tests UK; 2015 [Accessed at 16 January 2019]. Available from: https://www.reagent-tests.uk/product/mdma-reagent-test-kit/

Investigating the influence of material and surface properties of various biomaterials on adhesion, to aid the wound healing process

Kate Griffiths and P Prokopovich
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Due to an aging population, the prevalence of wounds in the UK is likely to increase.[1] Consequently, the demand for advanced dressings is also likely to rise. In many medical applications, including advanced dressings, controlling the force of adhesion between contact materials is necessary to maintain functionality.[2] Evidence has demonstrated that a moist wound healing interface accelerates the wound healing process.[3] Consequently, in order to allow dressings to maintain this optimum environment, the fundamental aspect is adhesion to the skin. This study aims to investigate the effect of material and surface properties of biomaterials on adhesion and how this can be improved at the wound-dressing interface.

Elastic modulus (E), surface energy ($\Delta\gamma$) and poisson ratio (v) values for both the biomaterials and skin were obtained from open literature. Following the input of these parameters, a computer programme ('Force 2')

estimated adhesion forces between each chosen biomaterial and the skin. The influence of these material and surface properties on adhesion was then investigated.

To test the validity of the results, data was analysed using SPSS. A one-way ANOVA test demonstrated an overall statistical significance between the results (p=0.000). Therefore, a post-hoc tukey test was performed to identify where these differences lay. 2% alginate hydrogel, and gellan gum films with Manuka honey showed differences against all other biomaterials. This supported the results obtained.

This study has provided a greater insight into how certain parameters can be altered to enhance the forces of adhesion in dressings. The results identified that a material with a lower elastic modulus, combined with a high surface energy and high contact area, confers a greater adhesive force. It therefore shows promising potential in allowing the ideal moist wound healing environment to be maintained, by overcoming the problem of adherence between human skin and dressings.

1. [1] Guest J, Ayoub N, McIlwraith T. *Health economic burden that wounds impose on the National Health Service in the UK*. BMJ Open. 2015;5(12):5. doi: 10.1136/bmjopen-2015-009283
2. [2] Prokopovich P, Perni S. *Comparison of JKR- and DMT-based multi-asperity adhesion model: Theory and experiment.* Colloids Surf. A. 2011;383(1-3):95-101. doi: 10.1016/j/colsurfa.2011.01.011
3. [3] Benbow M. *Exploring the concept of moist wound healing and its application in practice.* Br J Nurs. 2008;17(15). Available from: https://www-magonlinelibrary-com.abc.cardiff.ac.uk/doi/pdf/10.12968/bjon.2008.17.Sup6.30705

An Electrophysiological Investigation of the Behaviour of Pore Forming Toxin in Droplet-Interface Bilayers

Aashish Grover & O Castell
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Anthrax toxin is composed of the pore-forming protective antigen (PA) and the enzymatically active lethal factor (LF).[1] They are separate moieties, unlike some other binary toxins, allowing investigation of the pore with or without LF, which aids in characterising their behaviours, which are under contention. The objective of this project was to reconstitute and examine PA samples using electrophysiology in droplet-interface bilayers (DIBs) and compare the data to a literature review to help develop better understanding of this pore.

The literature review identified 31 current traces from 12 papers pertaining to PA were identified as potentially relevant to contextualise experimental results in DIBs. DIBs were formed by placing two aqueous droplets onto electrode tips in a well containing a lipid-oil mixture. The lipid formed a monolayer around each droplet, which when brought into contact formed a lipid bilayer.[2] Only one droplet contained a PA sample. A PA-Only sample and a PA-LF_N-Lipid sample were investigated. The pHs used within the droplets were pH7.4 and pH5, and the applied voltage was varied from 20mV to 80mV positively and negatively. Analysis of the current traces produced allowed pore insertion and blocking events to be identified through changes in current.

Reconstitution of both PA samples into DIBs was achieved. The PA-Only sample exhibited short transitions between an open and closed state under both pH7.4 and pH5. The PA-LF_N-Lipid sample showed more significant blocking events, possibly indicating translocation of LF_N.[3] Rapid downward current deflections were produced by pores from the PA-LF_N-Lipid sample (pH5), consistent with the literature.

Several initial steps were achieved and observed behaviours of PA in the samples were investigated. This has furthered developing understanding of PA and LF, and has indicated areas warranting further investigation.

1. Friebe S, van der Goot FG, Bürgi J. The Ins and Outs of Anthrax Toxin. *Toxins (Basel)*. 2016;**8**(3) doi: 10.3390/toxins8030069
2. Leptihn S, Castell OK, Cronin B, Lee EH, Gross LC, Marshall DP, et al. Constructing droplet interface bilayers from the contact of aqueous droplets in oil. *Nat Protoc.* 2013;**8**(6):1048-57. doi: 10.1038/nprot.2013.061
3. Fischer A, Holden MA, Pentelute BL, Collier RJ. Ultrasensitive detection of protein translocated through toxin pores in droplet-interface bilayers. *Proc Natl Acad Sci U S A*. 2011;**108**(40):16577-81. doi: 10.1073/pnas.1113074108

Characterisation of novel anti Carcinoembryonic antigen-related cell adhesion molecule (CEACAM5) antibody fragments produced via phage display technology in comparison with commercial anti CEACAM5 antibodies

Subhaan Hafeez, J Whitehead and AT Jones
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Antibodies are components of immune cells that are capable of binding and detecting one specific antigen,[1] for this reason there is considerable interest for antibodies to be used diagnostically or therapeutically. Cardiff school of pharmacy and pharmaceutical sciences are interested in developing novel antibody fragments through phage display, a new technology that has not been previously used by this school. These novel antibody fragments will be characterised alongside other commercial antibodies that were produced through alternative methods.

New antibody fragments against Carcinoembryonic antigen (CEA) were produced, this is an antigen that is overexpressed on many cancer cells.[2] Three new antibody fragments were produced, in Germany at TU Braunschewig University in collaboration with Professor Hust and Cardiff University; named Single Chain Variable fragments (SCFV) A, D and C. The SCFV's were characterised alongside three other commercial Anti CEA antibodies; COL-1, CB30 and anti-CEA Polyclonal sheep antibody. Ability to be bind to the antigen was assessed, via the well established SDS-PAGE and Western Blotting technique. All reagents were tested against: pure CEA, endogenous CEA within lysates and lastly against cell lysates that have been supplemented with 500ng of pure CEA.

None of the tested subjects showed an ability to bind to endogenous CEA within cell lysates. COL-1 was only capable of binding to pure CEA while the polyclonal sheep antibody and CB30 was unsuccessful in showing any CEA binding. However, SCFV A, D and C were capable of binding to both pure CEA as well as lysates that have been supplemented with 500ng of CEA whereas the commercial antibodies were unable to so.

More research needs to be conducted into all SCFV's to determine if any can be used therapeutically or diagnostically. This study has shown there is greater potential for the novel antibody fragments produced via phage display compared to the commercially purchased antibodies. CB30 and anti CEA sheep polyclonal antibodies are unsuitable for any further testing as they were unable to bind to pure CEA.

1. Pacific Immunology. What is an Antibody - Pacific Immunology. 2017. [accessed 6 Jan 2019] Available from: https://www.pacificimmunology.com/resources/antibody-introduction/what-is-an-antibody/
2. Laura J. What is the CEA Test for Cancer? 2019. [accessed 9 Dec 2018] Available from: https://www.webmd.com/cancer/cea-tests#1

Investigating the performance testing methods currently used to evaluate height and mechanical strength of microneedle devices

Josana Hayles, M Dul, JC Birchall, SA Coulman
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Although there is increasing interest in microneedle (MN) technology, there are currently no MN products in routine clinical use for drug delivery. One major obstacle to commercial development is the lack of defined quality standards, therefore there is currently a need to develop standardised tests for MN critical quality attributes (CQAs).[1] The aims of this project are to evaluate the different methods that could be used to develop standardised tests for two CQAs: height and mechanical strength.

A literature review was used to compare the methods currently used to test height and mechanical strength. PubMed was used to search for specific keywords from 2005 onwards, and for 'microneedl*' in 2018. Laboratory studies evaluated the appropriateness of the tests used to determine MN heights. The heights of seven different MN devices were analysed using both scanning electron microscopy (SEM) and optical microscopy. The influence of the equipment and human error were evaluated.

The findings from 96 published papers were collated; 78 measured MN heights, using either optical microscopy (53%), SEM (40%) or both (6%). Seventy papers measured mechanical strength, with 26% measuring height reduction and 39% measuring failure force. In laboratory tests, MN devices measured from 168-629μm in

height, with differences between optical microscopy and SEM ranging from 0.1-55.6%. Different researchers produced significantly different measurements (p=0.011), with a variability of approximately 1.1% in heights.

A large range of methods have been used by MN developers to test both height and mechanical strength; exemplifying the need for standardised testing methods. This was confirmed by the laboratory studies, in which significant differences in heights were noted when using different equipment and methods. Very little data is available concerning the methods currently used by manufacturers, so future research will involve interviews to amalgamate the views of both manufacturers and regulators.

1. Lutton, REM. et al. 2015. Microneedle characterisation: the need for universal acceptance criteria and GMP specifications when moving towards commercialisation. *Drug Delivery and Translational Research.* **5,** pp. 313-331. Doi: 10.1007/s13346-015-0237-z

---

An Investigation into Non-Medical Independent Prescribing leading up to and following the Implementation of Primary Care Clusters in Wales

Elliott Hulme, RE Deslandes and [1]PN Deslandes

Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK and [1]All Wales Therapeutics and Toxicology Centre, University Hospital Llandough, Penlan Road, Llandough, Penarth, The Vale of Glamorgan CF64 2XX, Wales, UK

Welsh Government has prioritised funding towards training existing healthcare professionals in advanced practice such as prescribing, resulting in a significant increase in the number of non-medical independent prescribers (NMIPs) in recent years. In February 2015, Welsh Government published a national plan for primary care services in Wales in which local health boards (LHBs) were urged to prioritise the development of primary care clusters in their area.[1] As well as improve access to services, it was hoped that the implementation of clusters would act as a driver for non-medical independent prescribing within primary care. This study investigated the volume of prescribing by medical and NMIPs in primary care prior to and following the implementation of primary care clusters in Wales, referred to as the intervention.

Prescribing data was obtained from the Comparative Analysis System for Prescribing Audit (CASPA) and used to conduct a series of Autoregressive Integrated Moving Average (ARIMA) Interrupted Time Series (ITS) analyses comparing changes in trend and level of prescribing by NMIPs and medical prescribers pre and post-intervention in each LHB and across Wales as a whole.[2]

For NMIPs, statistically significant differences in pre and post-intervention prescribing trends were seen in five out of the seven LHBs and across Wales as a whole. Significant step changes in prescribing by NMIPs were also seen in five out of seven LHBs but were inconsistent within the given time frame. For medical prescribers, the difference in pre and post-intervention prescribing trend was significant and a step change in prescribing was seen 24 months post-intervention.

The intervention appears to have been a driver for the uptake of prescribing by NMIPs in primary care across the majority of LHBs in Wales. However, changes in level and trend of prescribing cannot be attributed to the intervention due to the retrospective, quasi-experimental design.[3,4]

1. Welsh Government. 2015a. *Our plan for a primary care service for Wales up to March 2018*. Available at: https://gov.wales/docs/dhss/publications/150218primaryen.pdf [Accessed: 25/10/2018]
2. NHS Wales Prescribing Services. 2013. Comparative Analysis System for Prescribing Audit (CASPA). 2013 [Accessed: 03/11/2018]
3. Penfold, R. B. and Zhang, F. 2013. Use of Interrupted Time Series Analysis in Evaluating Health Care Quality Improvements. *Academic Pediatrics* **13**(6), pp. S38-44. doi: 10.1016/j.acap.2013.08.002
4. Kontopantelis, E. et al. 2015. Regression based quasi-experimental approach when randomisation is not an option: interrupted time series analysis. *BMJ* **350**: h2750. doi: 10.1136/bmj.h2750

Help or hindrance: an evaluation of the practical application of the standards for medicines management.

Ashfa Ilyas, T Ahmed, ML Hughes, LK James and L Williams
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Errors present a huge problem for the NHS with up to 4% of inpatients experiencing some form of medication error, of which the most common type is a medication administration error (Choo et al. 2010). The Nurse and Midwifery Council provide Standards for Medicines Management, in an attempt to reduce these (NMC 2010). This guidance is currently under review. This research will explore deviations from the guidance to inform future guidance.

Following ethics approval, medicines administration was observed for one week on each of three wards within one Health Board using a checklist to record deviations from policy. Data were entered into SPSS® and statistically analysed. Qualitative data was collected through a focus group with nurses, and thematic analysis was carried out. This presented an opportunity to discuss barriers to using the current guidance, and ways in which they could be improved.

630 observations were made. Deviations across all clinical areas were observed. Some processes had a high percentage of adherence, such as confirming details of the prescribed medication against the drug chart, whilst some processes were never observed. The reasons behind these deviations were explored further in the focus group (n=4 participants) and suggestions were made for changes in future guidelines.

The results from this audit support lots of existing research including findings from a similar study (Kim and Bates 2013). However, this audit was vey small, and more research is needed to truly understand how nurses use the current guidelines as well as what they would want included in the future.

1. Choo, J. et al. 2010. Nurses' role in medication safety. *Journal of Nursing Management* **18**(7), pp. 853-861. doi: 10.1111/j.1365-2834.2010.01164.x
2. Kim, J. and Bates, D. W. 2013. Medication administration errors by nurses: Adherence to guidelines. *Journal of Clinical Nursing* **22**(3-4), pp. 590-598. doi: 10.1111/j.1365-2702.2012.04344.x
3. NMC. (2010). Standards for Medicines Management. Available at: https://www.nmc.org.uk/globalassets/sitedocuments/standards/nmc-standards-for-medicines-management.pdf [Accessed 17 January 2019]

Isolation and characterisation of unknown natural products derived from Narcissus pseudonarcissus

Polina Ingulova, AW White and B Dummer
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Amaryllidaceae alkaloids have been studied intensively over the last decades due to their broad range of biological activities.[1,2] This has led to discovery of galanthamine, a drug marketed for Alzheimer's disease (AD), which is currently harvested from Narcissus pseudonarcissus commonly known as daffodils.[3,4] Agroceutical Products obtains larger quantities of this naturally derived drug in the Black Mountains, Wales. Apart from being a supplier of galanthamine, this company is interested in identifying other natural products present in the harvested daffodils.4 The aim of this project is to isolate and characterise unknown natural products derived from the daffodil juice provided by Agroceutical Products.

Two orthogonal separation and purification methods were selected to isolate the unknown molecules. Extracted organic contents of the juice have first undergone size-exclusion separation on a Sephadex chromatographic column. The fractions were then combined according to the results of analytical thin layer chromatography (TLC) and further separated on preparative TLC (prep-TLC) plates. The success of separation and molecular structure characterisation was justified by mass spectrometry (MS) of medium and high resolutions, and proton nuclear magnetic resonance (1H NMR).

Results confirmed a complex mixture of natural products other than galanthamine present in the juice. MS and 1H NMR analysis suggest successful isolation and identification of masonine, an alkaloid known to be produced by Narcissus pseudonarcissus. Other findings were inconclusive for confirmation of specific molecules being present.

This data can be used by Agroceutical Products to carry out more extensive large-scale research in order to create a portfolio of natural products produced by daffodils grown in the Black Mountains. Masonine can become a starting point for testing biological activity and identifying potential applications.

1. Evidente A, Kireev AS, Jenkins AR, Romero AE, Steelant WF, Van Slambrouck S, Kornienko A. Biological evaluation of structurally diverse amaryllidaceae alkaloids and their synthetic derivatives: discovery of novel leads for anticancer drug design. *Planta Med.* 2009;**75**(5):501-507.
2. Havelek R, Seifrtova M, Kralovec K, Bruckova L, Cahlikova L, Dalecka M, Vavrova J, Rezacova M, Opletal L, Bilkova Z. The effect of Amaryllidaceae alkaloids haemanthamine and haemanthidine on cell cycle progression and apoptosis in p53-negative human leukemic Jurkat cells. *Phytomedicine.* 2014;**21**(4):479-490.
3. Hager K, Baseman AS, Nye JS, Brashear HR, Han J, Sano M, Davis B, Richards HM. Effects of galantamine in a 2-year, randomized, placebo-controlled study in Alzheimer's disease. *Neuropsychiatr Dis Treat.* 2014;**10**:391–401.
4. Agroceutical products. *About Us*. 2018 [accessed 3 Jan 2019]. Available from: http://www.agroceutical.com/aboutus.html

---

Evaluation and optimisation of a Lego based fluid delivery system for microfluidic droplet generation

Danielle Jarvis and O Castell
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB Wales, UK

Droplet microfluidics unpins a range of research and commercial applications in the life sciences, from the production of artificial cells to cell encapsulation for bio-engineering applications such as tissue engineering and repair. However, current technology used for fluid delivery is expensive and complex, and so limits accessibility. To overcome these limitations, we designed and constructed a fluid delivery system from two Lego syringe drivers and a microfluidic T-junction. This project aimed to design and charecterise these Lego syringe pumps and demonstrate successful droplet printing using more cost-effective materials.

The reliability of a commercially used syringe drivers that have been used for microfluidic droplet production were tested and flow rates 10-100ml/hr were evaluated. Lego syringe drivers were then charecterised to determine the flow rates that were produced by programmable speeds. The Lego mindstorms EV3 brick used to power the syringe was programmed with speeds 10-100(arbitrary units). Further optimization of the design took place before two Lego pumps were constructed and connected with a microfluidic T-junction. Droplet formation was tested using an established gelling method.[1]

Speeds 10-100 have been charecterised regarding the flow rates they deliver for three different Lego pump designs, one of which was characterised using deionised water, oil and alginate solution. Successful droplet generation has been demonstrated with reasonable reliability and predictability.

To the best of our knowledge, a successful Lego syringe driver used for microfluidic droplet production is a first and enables lower costs, improved accesibility of droplet microfluidics. Further characterisation and optmisation is recommended. Further work could utilise this technology in droplet microlfuidic applications, such as cell encapsulation and increase the complexity of biological models that can be printed.

1. Morgan AJ, Hidalgo San Jose L, Jamieson WD, Wymant JM, Song B, Stephens P, et al. Simple and versatile 3D-printed microfluidics using fused filament fabrication. *PLoS One*. 2016;**11**(4):e0152023. doi: 10.1371/journal.pone.0152023

Design and synthesis of novel Aspartyl tRNA synthetase (AspRS) inhibitors as potential antipseudomonal agents

Divya John and C Simons
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Pseudomonas aeruginosa (P. aeruginosa) is an opportunistic, gram-negative, rod-shaped bacterium found ubiquitously in the environment.[1] *P. aeruginosa* infects immunocompromised individuals and is a major cause of life-threatening nosocomial infections.[1, 2] Due to its intrinsic antibiotic resistance and the emergence of multi-drug resistance, infections are increasingly challenging to treat.[2] Thus, there is a clinical need for new antipseudomonals with a novel mode of action. Aspartyl-tRNA synthetase (AspRS), an enzyme essential for protein biosynthesis,[3] is an attractive target as inhibition of AspRS would halt bacterial growth. Therefore, the aim of this project was to design and synthesise novel dual site inhibitors of *P. aeruginosa* AspRS.

Novel sulphonamide and amide compounds were designed based on the structure of aspartyl-adenylate; the natural substrate of AspRS. Molecular modelling software, MOE, was used to evaluate the design of the inhibitors by identifying the binding interactions of the proposed compounds to the AspRS active site. Synthesis of the sulphonamides involved mesylation, nucleophilic substitution, Boc deprotection, cyclisation and trans-sulfonylation reactions. Synthesis of the amides involved a Mitsunobu reaction, Boc deprotection, carbamylation and nucleophilic substitution reactions. TLC analysis and column chromatography were used for identification and purification of compounds. Melting points, ^{1}H NMR and ^{13}C NMR were used to characterise products.

Molecular docking experiments demonstrated potential for the compounds as AspRS inhibitors, with interactions between the isoxazole moiety and Ser196 residues, and thiadiazole moiety with Glu438 and Arg220 residues. Two final isoxazole-amide compounds were successfully synthesised and characterised. No final sulphonamide and thiadiazole-amide compounds were synthesised. Nevertheless, all intermediate products were synthesised in good yield with confirmation by ^{1}H NMR.

The two isoxazole-amide novel compounds are pending biological evaluation after further assessment of purity. The synthesis for sulphonamides and thiadiazole-amides proved challenging. Therefore, further work including optimisation of the final synthesis steps and modification of reaction conditions could provide the desired compounds.

1. Gellatly SL, Hancock RE. Pseudomonas aeruginosa: new insights into pathogenesis and host defenses. *Pathog Dis*. 2013;**67**(3):159-73. doi: 10.1111/2049-632X.12033.
2. Giske CG, Monnet DL, Cars O, Carmeli Y. Clinical and economic impact of common multidrug-resistant gram-negative bacilli. *Antimicrob Agents Chemother*. 2008;**52**(3):813-21. doi: 10.1128/AAC.01169-07
3. Hurdle JG, O'Neill AJ, Chopra I. Prospects for aminoacyl-tRNA synthetase inhibitors as new antimicrobial agents. *Antimicrob Agents Chemother*. 2005;**49**(12):4821-33. doi: 10.1128/AAC.49.12.4821-4833.2005

Investigation into the factors that influence the formation of calcium phosphate precipitation in an aqueous parenteral nutrition admixture containing either Vamin 18 EF® or Aminoven 25®

Ellen C Jones and AG Cosslett
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Parenteral Nutrition (PN) is an indispensable therapy in treating and preventing malnourishment in patients unable to use their gut for digestion however, there are many safety implications associated with its use.[1] One such complication is the formation of calcium phosphate precipitation which is of particular concern when calcium and phosphates are required in high concentrations e.g. critically ill fluid restricted patients or neonates.[2] The presence of particulates within PN can have life-threatening consequences if administered into the circulation[3], therefore a thorough understanding of the likelihood of calcium phosphate precipitation within given admixtures is sought in order to guide clinical use.

Aqueous admixtures containing an amino acid solution (Aminoven 25® or Vamin 18 EF®), calcium chloride 14.7% w/v, Water For Injections and a variable concentration of phosphates (Sodium Glycerophosphate 21.6% w/v, Potassium Dihydrogen Phosphate 13.6% w/v or Addiphos®) were formulated. The admixtures were stored at either room temperature or 40°C for 72 hours and subject to visible inspection, turbidity testing and pH monitoring every 24 hours.

The results showed that formation of calcium phosphate precipitation was reduced in admixtures containing Vamin 18 EF® however, the results were erratic and unpredictable. In contrast, although precipitation formation was often faster and to a greater extent in the Aminoven 25® admixtures, a definite pattern of behaviour was distinguishable.

On the surface, due to the lower level of precipitation formed it appears that use of Vamin 18 EF® would be preferable in an admixture containing calcium and phosphate. However, this would be a naïve interpretation because the degree of predictability of an admixture being used in a clinical setting where rigorous testing is not available cannot be undervalued. To this end, it would be prudent to continue this line of investigation using real-life conditions to ensure reliability of results.

1. Harwood SJ, Cosslett AG. Parenteral Nutrition. In: Whittlesea C, Hodson K, editors. *Clinical pharmacy and therapeutics*. 6th ed. United Kingdom: Elsevier; 2019. p105-24

2. BAPEN. *Complication management*. 2016 [accessed: 10 Oct 2018]. Available from: https://www.bapen.org.uk/nutritionsupport/parenteral-nutrition/complication-management
3. Hill SE, Heldman LS, Goo EDH, Whippo PE, Perkinson JC. Fatal microvascular pulmonary emboli from precipitation of a total nutrient admixture solution. *JPEN J Parenter Enteral Nutr*. 1996; 20(1):81-7. doi: https://doi-org.abc.cardiff.ac.uk/10.1177/014860719602000181

The views and opinions of primary care pharmacists regarding the use of the Welsh language in professional practice

Rhianwen Jones, W Davies, L Doyle and E Jones
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Good communication is essential to effective healthcare and linguistic barriers can have detrimental effects to patient care.[1] Welsh speaking patients often face barriers when accessing healthcare services.[2] Studies show that pharmacists are aware of the difficulties minority language speakers face.[1] However, with a recent report highlighting the dissatisfaction of provision within primary care[3], and with a developing clinical role for pharmacists, further research in this area is required. The aim of this project therefore was to explore the opinions of primary care pharmacists on the use of the Welsh language.

Four semi-structured, one-to-one interviews were conducted. They were transcribed ad verbatim and thematically analysed. Four main themes were identified.

Enablers to Welsh provision, advantage to the patient, barriers to Welsh provision, and future considerations were the themes identified. Pharmacists positively referred to the improved patient outcomes seen when Welsh is used. However, due to the minority status of Welsh, participants believed that patients don't have expectations to receive Welsh services, stating that it is the patient's responsibility to ask for them. Barriers identified include limited resources and a lack of Welsh speaking healthcare professionals.

Primary care pharmacists are aware of the benefits to patients, but don't believe it's realistic for all healthcare in Wales to be offered in Welsh. It is evident that Welsh language provision within primary care needs improving.

1. 1. Welsh Language Commissioner. My language, my health: inquiry into the Welsh language in primary care. 2014 [accessed 14 Oct 2018]. Available from: http://www.comisiynyddygymraeg.cymru/English/Publications%20List/Health%20inquiry%20executive%20summary.pdf
2. 2. Madoc-Jones I, Dubberley S. Language and the provision of health and social care in Wales. *Divers Equal Health Care.* 2005; **2**:127-34.
3. 3. Hughes L, Brookes C, Cocker C, John DN, Jones AT, Longley M. *The role of the Welsh language in community pharmacy service provision in Wales.* Great Britain: Pharmacy Practice Research Trust; 2008

Exploring the ability of germinants to potentiate the antimicrobial activity of clay leachates against *Bacillus anthracis and Bacillus cereus* spores

Timothy Kellett, E Ahortor and L Baillie
Welsh School of Pharmacy and Pharmaceutical Science, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Bacterial spores are resistant to a variety of biocides. Hence, decontamination often uses harsh, environmentally damaging chemicals.[2] We have combined two novel approaches, the sporicidal activity of natural clay with the potentiating effects of germination on biocides[1] to develop a less environmentally damaging antibacterial against *Bacillus anthracis and Bacillus cereus s*pores.

Firstly, the ability of germinants to potentiate biocides was assessed. *B. anthracis* (Sterne) and *B. cereus* (6a1) spores were incubated in 100ppm Peracetic Acid (PAA) with and without germinants (500mM^{-L} L-alanine, 25mM^{-L} inosine). Secondly prospective clay leachates (0.05g/L) and a known antimicrobial clay leachate were tested for biocidal activity against the same spores with and without germinants (500mM^{-L} L-alanine, 25mM^{-L} inosine) for 24hrs (±1hr).

Germinants increased biocidal activity of PAA an average of 81% (p=<0.0003) against *B. anthracis* and 82% (p=0.005) against *B. cereus.* French green clay alone showed significant antimicrobial activity against *B. anthracis* (reduction factor = 1.87 Log_{10}, p=0.005) *and B. cereus s*pores (reduction factor = 1.98, p =0.0001) as was also seen by Moore, 2018.[3] However, all antimicrobial activity was lost in the presence of germinants.

Unexpectedly, germinants alone had an antibacterial effect which was most marked in *B. cereus* (reduction factor = 1.60, p= 0.0001).

Our results demonstrated an interaction between germinants and antimicrobial agents in the clay. Adding germinants raised the pH of antibacterial French green clay. Low pH is key to antibacterial activity[4] which could explain this phenomenon. One approach to avoid this interaction would be to deliver germinants prior to clay leachate.

Further investigations into the nature of this interaction and strategies to avoid it are needed. Future work may yield insightful progress toward a new generation of clay based environmentally friendly biocides.

1. Celebi, O et al. (2016). The Use of Germinants to Potentiate the Sensitivity of Bacillus anthracis Spores to Peracetic Acid. *Frontiers in Microbiology.* DOI: 10.3389/fmicb.2016.00018
2. Manchee, R. J., Broster, M. G., Stagg, A. J., and Hibbs, S. E. (1994). Formaldehyde solution effectively inactivates spores of Bacillus anthracis on the Scottish Island of Gruinard. Appl. Environ. Microbiol. 6, 4167–4171 Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC201953/
3. Moore, A (2018). Evaluation of the Oxidative Capacity of Clay Particulate Matter as Novel Anthrax Decontamination Solutions. Unpublished Phd thesis. Cardiff University
4. Williams, L. B., Metge, D. W., Eberl, D. D., Harvey, R. W., Turner, A. G., Prapaipong, P., & Poret-Peterson, A. T. (2011). *What makes a natural clay antibacterial?* Environmental science & technology, 45(8), 3768-73. DOI: 10.1021/es1040688

Molecular modelling of the DENV NS3/NS5 interface as a potential target for the development of novel antivirals

Hassaan Khan, A Brancale and G Nannetti
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII avenue, Cardiff CF10 3NB, Wales, UK

The Dengue virus (DENV), a mosquito-borne Flavivirus divided into four distinct serotypes (DENV 1-4), is responsible for ~390million infections per year. There are no current antiviral therapies for Dengue, and significant issues associated with the CYD-TDV vaccine has necessitated the development of novel antivirals for Dengue. NS3 and NS5 proteins encompass enzymatic activities crucial for viral RNA replication and have been found to interact in the replication complex. Mutagenesis studies have fine-mapped the interactive regions of NS3 and NS5, as well as proposed this interaction as critical for viral replication. The aim of this project was to identify potential allosteric inhibitors of NS3 and NS5 using their respective interactive regions as the template for structure-based virtual screening. A further objective comprised molecular dynamic (MD) investigations of potentially relevant protein-protein docked NS3/NS5 conformational models.

Using homology models NS3 and NS5 of DENV-2, a receptor grid was generated in each model for a series of docking and rescoring techniques including HTVS, SP, XP, PLANTS and FlexX. Following this, MD simulations were performed on two protein-protein docked NS3/NS5 conformations, using the Desmond package.

Visual inspection of the ligand poses allowed for selection of 40 potential allosteric inhibitors for each NS3 and NS5 target site. RMSD analyses of the MD simulations revealed protein-protein docked model 16 as a more stable NS3/NS5 conformation relative to model 9. H-bond analyses revealed H-bonds between the NS5 critical residue Lys-330 and residues of or close to the NS3 interactive region, for majority of the model 16 simulation.

Potentially inhibiting compounds for each NS3 and NS5 protein, as well as protein-protein docked models forming potentially key H-bonds were achieved. Further work will involve ELISA assays of the selected compounds for each protein, and possibly further molecular modelling studies to search for novel druggable pockets of model 9 and 16.

A Five Year Evaluation of the Community Pharmacy Common Ailments Service: September 2013 to September 2018

Niall Kinsella, ML Hughes, KL Hodson and [1]A Evans
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue,Cardiff CF10 3NB, Wales, UK and [1]Welsh Government Offices, Crown Buildings, Cathays Park, Cardiff CF10 3NQ, Wales, UK

The Common Ailments Service (CAS) is an enhanced pharmacy service initiated in Wales in 2013.[1] The service enables patients to seek advice from pharmacists and receive treatment, free of charge for a defined

list of 26 conditions. Common ailments, are self-treatable and usually self-limiting, requiring little or no medication.[2] The aim of CAS is to encourage patients to self-care, thus reducing workload of general practitioners.[2] This study will conduct a five year evaluation of CAS, utilising reimbursement records from Welsh Government.

An anonymised quantitative dataset containing a wide range of variables, with information from every CAS consultation conducted throughout the five year period, was obtained from the Chief Pharmaceutical Officer for Wales. Secondary data analysis was implemented to assess the pre-existing dataset. This type of methodology is advantageous in that no data collection is required and allows for extensive longitudinal research of large databases.[3] The variables were analysed using Microsoft Excel and IBM SPSS. There were no ethical considerations for this study as the sample was anonymised.

Since introduction, 38,164 patients have used CAS, culminating in 49,985 consultations, provided by 630 pharmacies. Of the pharmacies that provided CAS, the majority were in the most deprived areas and assigned a deprivation quintile 1 or 2, accounting for 63% of total consultations. The service has predominantly been used by females (64.6% of consultations) and by patients aged 0-9 (33.2 consultations per 1000 population). The ailment with which patients presented with most frequently was Hay Fever (22.5%), which showed seasonal trends.

CAS continues to be a prominent NHS service offered through community pharmacies in Wales. Results show usage is growing annually, compared to England's Minor Ailment Scheme.[4] Instalment of Choose Pharmacy software in all pharmacies will enable more patients across Wales to access this valuable community pharmacy service for free advice and treatments.

1. Welsh Government. Community pharmacy services in Wales, 2017-18. 2018 [accessed 1 Nov 2018]. Available from: https://gov.wales/docs/statistics/2018/181031-community-pharmacy-services-2017-18-en.pdf
2. Welsh Government. Community pharmacy common ailments service: service specification (Draft) [accessed 3 Oct 2018]. Available from: http://www.cpwales.org.uk/getattachment/Contract-support-and-IT/Contract-Services/IT/Choose-Pharmacy-IT-Platform/Common-Ailments-Service-(1)/Common-Ailments-Service-Specification-(draft).pdf.aspx?lang=en-GB
3. Bryman A. *Social research methods*. 4th ed. Oxford: Oxford University Press; 2012.
4. Royal Pharmaceutical Society. No plans to commission national minor ailment scheme in England, says pharmacy minister. *Pharm J*. 2018. doi: 10.1211/PJ.2018.20204855

Statins as anti-cancer agents in Triple Negative Breast Cancer

Georgina Leason and SE Hiscox
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Statins are a class of drugs used to treat hypercholesteremia through competitive inhibition of HMG-CoA reductase to reduce plasma cholesterol levels and are widely used to reduce the risk of coronary disease.[1] Recent evidence points to a role for statins in suppression of tumour cell growth, including that of Triple Negative Breast Cancer (TNBC)[2], a breast cancer subtype that is particularly hard to treat in that it lacks a well-defined therapeutic target.[1,3] This project aimed to further explore the role of statins agents in the suppression of TNBC growth and migration.

WST and Ki-67 assays were used to determine the effects of atorvastatin (0-50nM) on the growth of MCF7 (luminal A, ER+) and MDA-MB-231 (TNBC) cell models). Changes in cellular migration in response to atorvastatin were measured using an in vitro cell wounding assay. Stain-induced changes to the cell cytoskeleton were investigated using immunofluorescence microscopy with phalloidin and vinculin, for visualisation of actin and focal adhesions respectively.

WST data revealed that atorvastatin exerted a dose-dependent inhibition of growth in both cell lines although MDA-MB-231 cells were much more sensitive versus MCF7 cells (629.3nM to 2.311 respectively). These observations were further confirmed through staining of the Ki-67 proliferation antigen. Atorvastatin also prevented wound closure in the wound assay. Treatment of MDA-MB-231 cells with atorvastatin also induced a reduction in filopodia like projections and a change in colony morphology to one with a tightly-packed appearance.

In conclusion this project has confirmed the anti-tumour action of a statin drug in breast cancer and that TNBC cells appears differentially sensitive to its effects versus ER+ breast cancer. These data may further suggest that statin agents might represent a useful therapeutic tool for a particularly hard-to-treat form of breast cancer.

1. Ahern TP, Lash TL, Damkier P, Christiansen PM, Cronin-Fenton DP. *Statins and breast cancer prognosis: evidence and opportunities.* The lancet oncology. 2014;**15(**10):e461-e8.
2. Shaitelman SF, Stauder MC, Allen P, Reddy S, Lakoski S, Atkinson B, et al. *Impact of Statin Use on Outcomes in Triple Negative Breast Cancer.* Journal of Cancer. 2017;**8**(11):2026-32. doi: 10.7150/jca.18743
3. Mandal CC, Ghosh-Choudhury N Fau – Yoneda T, Yoneda T Fau – Choudhury GG, Choudhury Gg Fau – Ghosh-Choudhury N, Ghosh- Choudhury N. *Simvastatin prevents skeletal metastasis of breast cancer by an antagonistic interplay between p53 and CD44.* (1083-351X)

Computational studies on immune-checkpoint PD-1/PD-L1: identification of new potential anticancer small molecules

Su Ying Lei and S Ferla
School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Programmed cell death-1 (PD-1), also known as immune inhibitory receptor, is found on activated T-cells.[1] Programmed cell death-ligand 1 (PD-L1), the ligand of PD-1, is widely expressed on normal tissues and cancer cells. Under normal conditions, PD-1/PD-L1 interaction will inhibit the activation of T-cells toward self-antigens, protecting against autoimmune diseases.[2] However, overexpression of PD-L1 in certain cancer malignancies promotes the PD-1/PD-L1 interaction and inhibits the function of T-lymphocytes in cancer cells eradication, hence, causing uncontrolled tumour proliferation.[3] Anti-PD-L1 and anti-PD-1 monoclonal antibodies used to treat various types of advanced cancer have unfavourable pharmacokinetics profile.[4] Therefore, this project aims to identify novel small molecules that could inhibit PD-1/PD-L1 interaction.

In a ligand-based virtual screening, known inhibitors of PD-L1 (Bristol-Myers Squibb (BMS) compounds), were used to generate a shape-based query model. Databases of commercially available compounds were screened against the query model and docked on PD-L1 crystal structures. The results obtained were rescored using Glide XP, Protein-Ligand ANT system (Plants) and FlexX. Consensus scoring method was employed combining all rescoring results and compounds that lie in the top best 25% of all 3 scoring functions were selected to undergo final visual inspection step. In a structure-based virtual screening, databases of commercially available compounds were docked on PD-1 crystal structure. Rescoring, consensus scoring, and visual inspection were then applied.

The adopted virtual screening methods produced diverse sets of final compounds that were selected as potential inhibitors of the PD-L1/PD-1 interaction by either binding to PD-L1 or PD-1. These compounds have shown theoretical interaction with either PD-1 or PD-L1.

Structural insight also revealed that compounds selected have promising ability in blocking PD-1/PD-L1 interaction. These diverse sets of compounds will be evaluated in different biochemical assays and any potential hit compounds will be used as a starting point to develop more potent PD-1/PD-L1 inhibitors.

1. Ahmadzadeh M, Johnson L, Heemskerk B, Wunderlich J, Dudley M, White D, et al. Tumor antigen-specific CD8 T cells infiltrating the tumor express high levels of PD-1 and are functionally impaired. *Blood.* 2009;**114**(8):1537-1544. doi:10.1182/blood-2008-12-195792.
2. Riella L.V., Paterson A.M., Sharpe A.H., and Chandraker A. Role of the PD-1 pathway in the immune response. *Am J Transplant*. 2012;**12**(10):2575-87. doi:10.1111/j.1600-6143.2012.04224.x
3. Freeman G, Long A, Iwai Y, Bourque K, Chernova T, Nishimura H, et al. Engagement of the Pd-1 immunoinhibitory receptor by a novel B7 family member leads to negative regulation of lymphocyte activation. *J Exp Med*. 2000;**192**(7):1027–1034.
4. Alsaab H, Sau S, Alzhrani R, Tatiparti K, Bhise K, Sushil K. et al. PD-1 and PD-L1 checkpoint signaling inhibition for cancer immunotherapy: mechanism, combinations, and clinical outcome. *Front. Pharmacol.* 2017;**8**:561. doi:https://doi.org/10.3389/fphar.2017.00561

Design & synthesis of Fingolimod Phosphate Prodrugs as Potential Niemann Pick Type C Treatment

LM Lucas and F Pertusati
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [1]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Niemann-Pick Type C (NPC) disease is a fatal, neurodegenerative disease caused by mutations in NPC 1 and 2 genes.[1] These mutations result in an inability of the body to properly process lipid molecules, namely cholesterol and glycosphingolipids. Redistribution of lipid molecules within the brain and liver is responsible for the cognitive and liver symptoms associated with the disease.[2] With only one drug, Miglustat, clinically

approved to delay the disease progression, there has been huge interest in developing new therapies with the potential to treat this devastating disease.[3]

Histone deacetylase inhibitors (HDACI's) have received interest for their ability to treat NPC disease.[1] Fingolimod is an immunomodulating drug used in the treatment of multiple sclerosis, it is phosphorylated by nuclear sphingosine kinase 2 to form fingolimod phosphate/FTY720-P, the pharmacologically active species. FTY720-P is a proven class I HDAC inhibitor. A recent trial found that Fingolimod Phosphate "dramatically increases the amount of NPC 1 and 2 proteins in human fibroblast cells".[4]

In this project we synthesized a family of fingolimod phosphate prodrugs with the aim of designing a potential drug capable of improving the delivery of FTY720-P, thereby with the potential to treat NPC disease. Phosphoramidate prodrug approach has been successfully used in nucleoside drugs to deliver the respective monophosphate. We envisaged that applying this strategy to fingolimod would be a good strategy to improve the biological activity of FTY720-P. We coupled FTY720 with several phosphorochloridate compounds, each with variable amino acid, ester and aryl moieties.

In conclusion four phosphorochloridates were produced using various amino acids, and were coupled with fingolimod to produce the final prodrugs. Four prodrugs were produced in adequate yields and a fifth prodrug was produced in trace amounts. The study is awaiting biological data for the prodrugs produced.

1. Maceyka M, Milstien S, Spiegel S. The potential of histone deacetylase inhibitors in Niemann-Pick type C disease. FEBS J. 2013;280(24):6367-6372. doi: 10.1111/febs.12505
2. NORD (National Organization for Rare Disorders). Niemann-Pick disease type C - NORD (National Organization for Rare Disorders). 2018. [accessed 26 Nov 2018]. Available from: https://rarediseases.org/rare-diseases/niemann-pick-disease-type-c/
3. Lyseng-Williamson K. Miglustat: A review of its use in Niemann-Pick disease type C. Drugs. 2013;74(1):61-74. doi: 10.1007/s40265-013-0164-6
4. Newton J, Hait N, Maceyka M, Colaco A, Maczis M, Wassif C et al. FTY720/fingolimod increases NPC1 and NPC2 expression and reduces cholesterol and sphingolipid accumulation in Niemann-Pick type C mutant fibroblasts. FASEB J. 2017;31(4):1719-1730. doi: 10.1096/fj.201601041r

---

Determining the impact of Polyethylene glycol (PEG) on the mechanical performance of hard shell gelatin capsule formulations for use in dry powder inhalers

Mishel Mathew, S Parker and SA Coulman
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Dry powder inhalers (DPIs) have been used to treat pulmonary conditions since the 1960.[1] Hard-shell capsules are traditionally manufactured from gelatin[2], which contains water as a plasticiser. Exposure to low humidity conditions therefore results in a reduction in the moisture content of gelatin capsules and brittleness (2). Polyethylene glycol (PEG) can act as an alternative plasticiser in gelatin capsules.[3] This study aims to determine how PEG influences the mechanical performance of gelatin capsules at moisture contents below the "normal" specification range.

A materials testing machine was used to conduct both puncture and compression tests on empty gelatin and PEG-gelatin (8.5% PEG) capsules conditioned at different relative humidities (RHs). A lactose powder blend formulation was produced and filled into gelatin and PEG-gelatin capsules conditioned at different RHs and loaded into a Plastiape DPI. A simulated aerosolization event was conducted to examine powder retention in the capsules.

Findings showed that the force required to puncture Quali-G at 34% RH was 4.23N which increased to 4.97N for Quali-G capsules at 11% RH. For Quali-G PEG the puncture force increased from 3.66N at 34% RH to 3.95N at 11% RH. A similar trend was observed with compression testing. This indicates that PEG-gelatin may provide some resistance to capsule brittleness at moisture contents below the normal specification. Aerosolization testing did not find any gross differences in the mass of powder released from the different capsule formulations.

This study found that PEG has the potential to overcome the problems of brittleness and improve the mechanical performance of gelatin capsules. Further work is needed to determine if the addition of PEG and low storage humidities affects aerosolization of a powder blend containing an active pharmaceutical ingredient (API). Nevertheless, the findings of this study encourage the development of PEG-gelatin capsules for low moisture content formulations.

1. Bell JH, Hartley PS, Cox JSG. Dry powder aerosols I: A new powder inhalation device. *Journal of Pharmaceutical Sciences.* 1971;**60**(10):1559-64. doi: 10.1002/jps.2600601028
2. Birchall JC, Jones BE, Morrissey A. A Comparison of the Puncturing Properties of Gelatin and Hypromellose Capsules for Use in Dry Powder Inhalers. *Drug Development and Industrial Pharmacy*. 2008;**34**(8):870-6. doi: 10.1080/03639040801928903
3. Lodha A, Patel A, Chaudhuri J, Jadia P, Joshi T, Dalal J. Formulation and evaluation of transparent ibuprofen soft gelatin capsule. *Journal of pharmacy & bioallied sciences*. 2012;**4**(Suppl 1):S95-S7. doi: 10.4103/0975-7406.94154

Welsh language service provision in a community pharmacy setting and its impact on patient care: A pharmacist's perspective

Mared McNeil, E Jones, W Davies and L Doyle
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

The Welsh Assembly Government (WAG) has passed legislation and developed initiatives to encourage parity between Welsh and English language use[1]. A survey of fluent Welsh speakers showed that only 6% were offered primary care services in Welsh[2]. Within community pharmacy, research has highlighted the benefits to patients of consulting in their preferred language but access to Welsh language services is variable[3]. The aim of this project is to investigate the views of community pharmacists on the impact of using Welsh language services on patient care, the barriers to providing them and suggestions for optimising their use.

An exploratory, qualitative approach was used, developing an interview schedule to perform in-depth, one-to-one, semi-structured interviews. A non-probability, purposive and convenience sampling method identified four pharmacists to participate. Ethical approval was granted. The interviews were audio recorded and transcribed verbatim with identifiable information being anonymised. Thematic analysis was used with an inductive approach to identify themes.

Consulting in the patients preferred language of Welsh improved the quality of consultations, particularly with the elderly in rural communities. Increasing level of comfort, improving rapport and enhancing medication compliance. Providing these services was dependent on the pharmacists' confidence in their Welsh language ability. There was a lack of awareness of Welsh language initiatives and resources with little advertising of services available in Welsh. There was also a lack of training and support in using the Welsh language at both undergraduate and postgraduate levels.

Better communication between WAG and community pharmacists regarding initiatives and resources for Welsh language services needs to occur. Welsh workforce fluency was a major barrier to service provision with funding for training and support possibly being beneficial. This has been further hampered by the introduction of the ORIEL pre-registration recruitment process. Providing these services could also be helped if they attracted a financial incentive.

1. 1 Welsh Language Act 1993. c. 38
2. 2 Welsh Government and Welsh Language Commissioner. Welsh language use in Wales, 2013-2015; 2015 [accessed 10 Jan 2018]. Available from: http://www.comisiynyddygymraeg.cymru/English/Publications%20List/Adroddiad%20-%20Y%20defnydd%20o%27r%20Gymraeg%20yng%20Nghymru%2c%202013-15%20-%20Saesneg.pdf
3. 3 Hughes L et al. *The role of the Welsh language in community pharmacy service provision in Wales*. London. The pharmacy practice research trust; 2008.

Developing a rapid diagnostic test that can determine the antibiotic susceptibility of *Staphylococcus aureus*

Nia Morgan, L Baillie and E Ahortor
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Methicillin-resistant *Staphylococcus aureus* (MRSA) has become a rising danger, and with patients 64% more likely to die from its infection compared to methicillin-sensitive *S. aureus* it is a current issue requiring an urgent solution.[1] Point-of-care testing (POCT) devices could be one possibility, allowing more rapid detection (minutes) compared to the current method of culture and sensitivity testing which can take up to 18 hours.[2] Resistance is normally due to the presence of the *MecA* gene which results in an altered penicillin-binding protein.[3] The aim of this study was to detect *S. aureus* and determine its antibiotic susceptibility by using *S. aureus* and *MecA* DNA probes.

Clinical isolates of MRSA were tested using DNA probes developed at Cardiff University in a DNA-based enzyme-linked immunosorbent assay (ELISA). This differs from the majority of rapid diagnostic assays as it does not involve the polymerase chain reaction method which can be inhibited by patient derived factors (e.g. blood).[4] DNA was extracted from the bacteria either by using microwaves or a commercial-kit method prior to testing.

100% (n=10) of samples were correctly identified by the *MecA* probe and correctly distinguished from *S. epidermidis* by the *S. aureus* probe. There was no significant difference between the probes ability to detect DNA obtained following both extraction techniques (P>0.05). All controls were also correctly identified, with just an error bar crossover in one assay from one no template control.

With this data building on to data from a previous study, these are promising results which could support the development of this technique into a rapid POCT device capable of detecting and determining the antibiotic susceptibility of *S. aureus*. However, further research is needed, particularly involving more MRSA samples, inclusion of more clinical control isolates, and the addition of *MecA* negative MRSA samples to further assess the selectivity of the *MecA* probe.

1. World Health Organisation (WHO). Antimicrobial Resistance. 2018 [Accessed: 6th December 2018]. Available at: https://www.who.int/en/news-room/fact-sheets/detail/antimicrobial-resistance
2. Nathwani D, Morgan M, Masterton RG, Dryden M, Cookson BD, French G et al. Guidelines for UK practice for the diagnosis and management of methicillin-resistant **Staphylococcus aureus** (MRSA) infections presenting in the community. *J Antimicrob Chemother.* 2008;**61**(5):976-994. doi: 10.1093/jac/dkn096
3. Brown DFJ, Edwards DI, Hawkey PM, Morrison D, Ridgway GL, Towner KJ et al. Guidelines for the laboratory diagnosis and susceptibility testing of methicillin-resistant *Staphylococcus aureus* (MRSA). *J Antimicrob Chemother.* 2005;**56**(1):1000-1018. doi: 10.1093/jac/dki372
4. van Belcum A and Rochas O. Laboratory-based point-of-care testing for MSSA/MRSA detection in the age of whole genome sequencing. *Front Microbiol.* 2018;**9**(1437). doi: 10.3389%2Ffmicb.2018.01437

Communication within community pharmacy, and the technological developments needed to improve it.

Bethan Morris and MW Smith
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Community pharmacy has developed in recent years, with pharmacists undertaking more clinical, patient facing roles and providing an increasing number of services to patients.[1]However, the technology used to support pharmacists and their services has not advanced accordingly, and is thought to be insufficient, particularly in terms of supporting communication. [2] At present, communication inadequacies are said to cause a number of issues which detrimentally impact upon patient care. [3] Technological improvements have the potential to address these inadequacies. [4] This study aimed to explore current communication within community pharmacy, and the technology required to improve it.

Questionnaires were disseminated to 200 independent community pharmacies in England and Wales. These pharmacies were randomly selected from a database obtained from NHS England and Shared Services Wales. The data collected from returned questionnaires was entered into an SPSS® database and descriptively analysed.

A response rate of 17% was achieved, with responses from pharmacies with varying demographics received. Being a communicator was found to be one of the principal roles of a pharmacist, and respondents reported communicating most commonly via telephone. Respondents highlighted a number of barriers to communication, including poor accessibility of other healthcare professionals (97% of respondents) and excessive workload (94%). 79% of respondents thought that access to patients' medical records is essential for communication with prescribers, and 85% stated that integrating GP and PMR systems would benefit communication. The prospect of sharing patient records between community pharmacies was also favourably thought upon. Many respondents (85%) indicated that being able to communicate with GPs via PMR systems would be advantageous, while few (26%) thought that initiating video link systems with GPs would be beneficial.

Alongside further wider-scale research, it is hoped that the findings of this study will be used to determine the technology required to enhance communication in community pharmacy.

1. General Pharmaceutical Council. What does a pharmacist do? 2018 [accessed 18 Oct 2018]. Available from: https://www.pharmacyregulation.org/raising-concerns/raising-concerns-about-pharmacy-professional/what-expect-your-pharmacy/what-does-0
2. Westerling AM, Haikala V, Airakstinen M. *The role of information technology in the development of community pharmacy services: Visions and strategic views of international experts.* Res Social Adm Pharm. 2011;**7**(4):430-7. doi: https://doi.org/10.1016/j.sapharm.2010.09.004
3. Astrom K, Duggan L, Bates I. *Developing a way to improve communication between healthcare professionals in secondary care.* Pharmacy Education. 2007;**7**(3):279-85
4. Mercer K, Burns C, Guirguis L, Chin J, Dogba MJ, Dolovich L et al. *Physician and Pharmacist Medication Decision-Making in the Time of Electronic Health Records: Mixed-Methods Study.* JMIR Hum Factors. 2018;**5**(3). doi: 10.2196/humanfactors.9891

An exploration of the technology used in community pharmacy to support pharmacists in the provision of health-related services

Noor Nawai and MW Smith
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Community pharmacists have transitioned from their traditional role of dispensing medication, to a more patient focused clinical role, thus helping to reduce strain on the NHS.[1] They are now delivering a range of services, including administering flu vaccinations and chlamydia screening.[2] However, technology within community pharmacy has not caught up with the progression of the profession[3], as many activities are performed manually which can be time consuming. Technology is thought to aid pharmacists in their new clinical roles by making the medication supply process safer, improving workflow, and reducing the time pharmacists spend in the dispensary. This allows more time for patients.[3,4] This study aims to explore the challenges pharmacy staff face due to their changing roles, along with their opinions and current use of technology within community pharmacy. Thus, enabling the identification of technological needs that could aid the pharmacy team in their future roles.

A mixed method research approach was employed where 200 randomly chosen independent community pharmacies were sent a survey. Responses were inputted into SPSS where quantitative analysis was carried out to produce descriptive statistical data.

A 15% response rate was achieved. Results highlighted the following challenges within community pharmacy; managing stock levels, dispensing repeat prescriptions, and delivering professional services. Utilisation of technology was varied, although access to patient records was found to be very popular. Most participants did not use robots for dispensing, nor did they use many features on their PMR system that could aid with these challenges.

In conclusion, the findings of this study could be used by technological system providers to address the challenges identified by community pharmacy staff. Thus, enabling an improved work environment, and improving patient care, which will have an overall positive impact on the NHS.

1. National Health Service. The NHS Long Term Plan. 2019 [accessed 8 Jan 2019]. Available from: https://www.longtermplan.nhs.uk/publication/nhs-long-term-plan/
2. Pharmaceutical Services Negotiating Committee. Services and commissioning. [No date] [Accessed 1 Jan 2019] Available from: https://psnc.org.uk/services-commissioning/
3. Liddell A, Adshead S, Burgess E. Technology in the NHS: Transforming the patient's experience of care. 2008 [Accessed 1 Jan 2019]. Available from: https://www.kingsfund.org.uk/sites/default/files/Technology-in-the-NHS-Transforming-patients-experience-of-care-Liddell-Adshead-and-Burgess-Kings-Fund-October-2008_0.pdf
4. Goundrey-Smith S. *Information Technology in Pharmacy: An Integrated Approach*. London: Springer; 2013.

Src kinase inhibitors as anti-cancer agents in triple negative breast cancer

Mohammed Nawaz and SE Hiscox
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Breast cancer is the most common cancer in the UK and accounts for 15% of all new cases each day.[1] This study aimed to investigate the effect of Src inhibitors on specific subtypes of breast cancer (TNBC and ER+), particularly TNBC due to its limited treatment options and poor prognosis compared to ER+ and HER2+ breast cancer making it a clinical priority.[2,3] To investigate the effect of Src inhibitors on ER+ and TNBC breast cancer cells, MCF7 and MDA231 cells were treated with saracatinib (AZD0530).

The effects of saracatinib and saracatinib/EGFR inhibitor combination on cell proliferation and cell migration were investigated using WST and wound assays, respectively. The Ki-67 assay was used to confirm saracatinib's effect on proliferation reflected in WST assay data. Immunofluorescence was used to image the effects of saracatinib on the cell cytoskeleton and FAK activity/location in the cells.

Saracatinib showed inhibition of TNBC and ER+ breast cancer cell proliferation, with TNBC cells showing increased sensitivity to saracatinib. Furthermore, saracatinib showed a significant decrease in migration of TNBC and ER+ breast cancer cells compared to untreated controls. Saracatinib decreased MDA231 cell migration by approximately 30% compared to untreated controls and cell proliferation decreased by approximately 60%, thereby suggesting that both migration and proliferation are dependent on Src signalling.

WST and wound assays performed, provide additional evidence for the role of Src kinase in triple-negative breast cancer subtypes, promoting processes, namely migration and proliferation, thereby indicating its therapeutic significance, particularly in TNBC. This study's evidence, the limited treatment options for TNBC and previous work on Src kinase support the need for further investigation of Src inhibition in, *in vivo* models and clinical trials as it represents a novel target for patients suffering from TNBC.

1. Cancer Research UK. 2018. *Breast cancer statistics.* [Accessed: 03 Nov 2018]. Available from: https://www.cancerresearchuk.org/health-professional/cancer-statistics/statistics-by-cancer-type/breast-cancer#heading-One
2. National Institute for Health and Care Excellence. Managing advanced breast cancer. CG81. London: National Institute for Health and Care Excellence; 2018 [Accessed: 01 Dec 2018]. Available from: https://pathways.nice.org.uk/pathways/advanced-breast-cancer#path=view%3A/pathways/advanced-breast-cancer/managing-advanced-breast-cancer.xml&content=view-index
3. Finn R.S., Dering J, Ginther C, Wilson C.A, Glaspy P, Tchekmedyian N. Dasatinib, an orally active small molecule inhibitor of both the Src and abl kinases, selectively inhibits growth of basal-type/ "triple-negative" breast cancer cell lines growing in vitro. *Breast Cancer Res Treat.* 2007;**105**(3):319-26. doi: https://doi.org/10.1007/s10549-006-9463-x

Formulation and In-vitro Evaluation of Dissolvable Mucoadhesive Thin films Containing Pomegranate Rind Extract and Zinc(II)

Jee Ji Ong and CM Heard
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Gingivitis, an inflammation of gums potentially advances to severe periodontitis if left untreated.[1] Pomegranate rind extract (PRE) is known for its bactericidal properties and synergistic virucidal effect when co-administered with zinc sulphate ($ZnSO_4$).[2,3] This study aimed to (A) formulate dissolvanble thin polymer-based films incorporating PRE and $ZnSO_4$ (B) determine simultaneous delivery of punicalagin and Zn(II) into and across porcine buccal membrane and epidermis, as comparator (C) preliminary evaluation of anti-biofilm activity.

Polymer-based films were prepared using different PEG400 concentration containing PRE and a combination of PRE-Zn(II). Punicalagin release was determined using diffusional release testing. Porcine buccal membrane and epidermis were dosed with film samples in in-vitro uptake and permeation studies. Quantitation of punicalagin and Zn(II) was by HPLC and ICPMS respectively. The films were then evaluated for activity against *Staphylococcus aureus* in a single-specie biofilm assay (in collaboration with Dr Lau, Newcastle University).

Without the presence of Zn(II), the film containing the highest PEG400 concentration (88.8%w/v) dissolved most rapidly, within 30 minutes with >50% of punicalagin released. The punicalagin and Zn(II) uptake across buccal membrane was significantly greater than epidermis. Similar results were found for in-vitro permeation studies, except there was higher punicalagin permeation across epidermis from the PRE-Zn film. Overall, Zn(II) uptake and permeation was higher than punicalagin in part reflecting the relative loading levels. A $1\log_{10}$ reduction in biofilm cell viability was seen with PRE-Zn(II) film compared to $0.029\log_{10}$ for PRE alone.

Film dissolvability can be optimized using PEG400 as a solubilising agent. Zn(II) limits punicalagin release through interaction with carbomer; however, the anti-biofilm activity of PRE against *S. aureus* was enhanced by Zn(II). Overall, polymer-based films can deliver punicalagin and Zn(II) into and across buccal membrane and epidermis and this study supports further investigations into synergistic bactericidal and anti-biofilm effect of PRE and Zn(II) in treating periodontal infections.

1. Philstrom, B. et al. Periodontal diseases. *The Lancet*. 2005;366:1809-1820.
2. Houston, D.M.J. et al. Potentiated virucidal activity of pomegranate rind extract (pre) and punicalagin against Herpes Simplex virus (HSV) when co-administered with zinc (II) ions, and antiviral activity of pre against HSV and acyclovir-resistant HSV. *PLoS ONE*. 2017;12:e0188609.
3. Yehia, H.M. et al. Antimicrobial activity of pomegranate rind peel extracts. *African Journal of Microbiology Research*. 2011;4:3664-3668.

Design and synthesis of a novel RNA dependent RNA polymerase inhibitors against human norovirus

Deml Oni , A Brancale and G Giancotti
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [1]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Noroviruses are positive sense RNA viruses and members of the *Calciviridae* family.[1] Affecting over 20 million individuals in the US and more than 200,000 children deaths in developing countries, norovirus-induced gastroenteritis persists as a major global burden.[2] Previous drug development studies have proved unsuccessful, thus a norovirus therapeutic is yet to be available. RNA-dependent RNA polymerase presents as an attractive drug target; implicated in viral replication.[3] This project proposes the design and synthesis of a novel series of RdRp inhibitors. Structural modifications of hit compounds **1-**3, previously demonstrating inhibitory activity at Welsh School of Pharmacy, were conducted.[4]

Ration design of seven novel derivative compounds were performed using computational modelling programs. Docking studies performed includes the generation of 137 predicted binding modes within the 4LQ3 (PPNDS) crystallised RdRp pocket; enabling inspection of key residue interactions.

Preparation of designed compounds followed one general synthetic route (4 schematic steps) including: acylation of nitrogen nucleophiles, catalytic reduction, acyl chloride peptide coupling and esterification. Alternative methods for synthesis of compound **7** were followed to improve yield. 4 novel compounds were synthesised, purified by column chromatography and collected at variable yields of 0.5-65%. ^{1}H and ^{13}C NMR used as confirmation.

Preliminary biological evaluation of compounds **1** and **6-8** were conducted in PICO-green assay at fixed concentration of 20µM. Little reduction in polymerase activity (around 10%) demonstrated by compounds **6-8**. Further testing using gel-shift assays will be considered in future work.

1. Zheng D, Ando T, Fankhauser R, Beard R, Glass R, Monroe S. Norovirus classification and proposed strain nomenclature. J Virol. 2006;346(2):312-323. doi: 10.1016/j.virol.2005.11.015
2. Nguyen G, Phan K, Teng I, Pu J, Watanabe T. A systematic review and meta-analysis of the prevalence of norovirus in cases of gastroenteritis in developing countries. J Med. 2017;96(40):e8139. doi: 10.1097/md.0000000000008139
3. Kim Y, Galasiti Kankanamalage A, Chang K, Groutas W. Recent Advances in the Discovery of Norovirus Therapeutics. J Med Chem. 2015;58(24):9438-9450. doi: 10.1021/acs.jmedchem.5b00762
4. Ferla S, Netzler N, Ferla S, Veronese S, Tuipulotu D, Guccione S et al. In silico screening for human norovirus antivirals reveals a novel non-nucleoside inhibitor of the viral polymerase. Sci Rep. 2018;8(1). doi: 10.1038/s41598-018-22303-y

---

Microneedles: Exploring the perspectives of potential UK users on an innovative contraceptive delivery device

Bolanle Opawole, JC Birchall, ML Hughes and SA Coulman
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Many low-middle income countries (LMICs) have a high unmet need for family planning[1]. A novel contraceptive microneedle delivery system that could increase the range of contraceptive options and partially satisfy this need is currently in development, however, its clinical utility will rely on its usability and acceptability to stakeholders. This study aimed to explore the acceptability of a novel contraceptive microneedle delivery system to UK women and to identify their preferred features of such a device.

After attaining ethical approval from Cardiff School of Pharmacy and Pharmaceutical Sciences, 23 women between 18-35 years old were recruited into 5 focus groups through a mixture of convenience, snowball and purposive sampling. A semi-structured topic guide was used to direct group discussions around the perceived advantages and disadvantages of the concept of a contraceptive microneedle and gather the opinions of participants on five prototype dummy contraceptive microneedle applicator devices. Focus groups were audio recorded, transcribed verbatim, anonymised, then thematically analysed to uncover core themes.

Identified benefits of contraceptive microneedles included minimally invasive application, painless administration, self-application, and increased discretion. Favourable comparisons were made of contraceptive microneedles over currently available long-acting contraceptive methods. Concerns focused on how to confirm microneedle administration and the efficacy, side effect profile, duration of action and safety of the method. Several positive and negative features were recognised of prototype dummy contraceptive

microneedle applicator devices and were perceived to affect usability. Important design features included an indicator, one-step actuation method and post-actuation microneedle retraction.

This exploratory study gathered the perceptions of potential users on microneedle use in contraception. Overall, users favourably perceived the proposed contraceptive microneedle device, despite having some concerns. Suggestions to address these concerns have been made and should be considered in the development of this novel contraceptive method in order to ease its translation into clinical use.

1. Sedgh, G et al. 2016. Unmet need for contraception in developing countries: examining women's reasons for not using a method. Available at: https://www.guttmacher.org/report/unmet-need-for-contraception-in-developing-countries [Accessed 7 December 2018]

Characterising the mechanical performance of hypromellose capsules, for use in dry powder inhalers, stored in ultra-low humidities

Clara O'Reilly and SA Coulman
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Hard-shell capsules have been used in dry powder inhalers (DPIs) to treat pulmonary disorders since the 1960s.[1] They are formulated using either gelatin or hypromellose (HPMC) as the major component. There is a desire to reduce the moisture content of inhalation grade HPMC capsules to significantly below the current specification, as existing and emerging active pharmaceutical ingredients are moisture labile. At a moisture content just below the 'normal' specification the mechanical properties of HPMC capsules remain relatively unchanged.[2-4] This study aims to determine the mechanical performance of Quali-V®-I HPMC capsules at moisture contents significantly below the specification range.

Capsules conditioned at 34%, 11% and 0.5% relative humidities (RH) were punctured or compressed using bespoke tests on a materials testing machine. Capsules conditioned at these different RHs were also filled with a lactose blend and inserted into a Plastiape DPI to undergo a simulated inhalation event. This event was used to assess capsule powder retention.

The force required to puncture capsules stored at 34% and 11% remained relatively unaltered, supporting previously published data. The force required to puncture capsules stored at 0.5% was slightly lower, although possible capsule shrinkage is thought to have created an erroneous result. Compression forces emained relatively unchanged at the three storage RHs and there were no gross changes in the mass of powder released from capsules at the different RHs.

This study indicates that from a mechanical and aerosolization perspective, inhalation grade HPMC capsules could perform at extremely low moisture contents. Further work is needed to confirm this and to to determine if the release of API within a lactose blend is effected by the low RH storage condition.

1. Jones, B. The evolution of DPI capsules. *Inhalation.* 2008;2(6):20-3.
2. Birchall JC, Jones BE, Morrissey A. A Comparison of the puncturing properties of gelatin and hypromellose capsules for use in dry powder inhalers. *Drug Dev Ind Pharm.* 2008;34(8):870-76. doi:10.1080/03639040801928903
3. Torrisi BM, Birchall JC, Jones BE, Diez F, Coulman SA. The development of a sensitive methodology to characterise hard shell capsule puncture by dry powder inhaler pins. *Int J Pharm.* 2013;456(2):545-52. doi:10.1016/j.ijpharm.2013.08.011
4. Chong RHE, Jones BE, Diez F, Birchall JC, Coulman SA. Evaluating the sensitivity, reproducibility and flexibility of a method to test hard shell capsules intended for use in dry powder inhalers. *Int J Pharm.* 2016;500(1):316-25. doi:**10.1016/j.ijpharm.2016.01.034**

A review of primary dental prescribing of analgesics and mouthwashes in Wales

[1]Elin Palmer [2]K Haines, [2]PN Deslandes and [1]MO Ivory
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK. [2]All Wales Therapeutics and Toxicology Centre, Academic Building, University Hospital Llandough, Penlan Road, Penarth, Vale of Glamorgan, CF64 2XX, Wales, UK

The British Pharmacological Society states that prescribing should be effective, safe and cost-effective and where appropriate, prescribers should adhere to national and local formularies.[1] National Institute for Health and Care Excellence and Scottish Dental Clinical Effectiveness Programme guidelines are used in practice in Wales to guide dental prescribing. On a dental NHS script, dentists may only prescribe from the dental

practitioner's formulary (DPF).[2] The aim of this project was to review primary dental prescribing of analgesics and mouthwashes to determine whether it aligns with guidelines.

A literature review was conducted using Medline, Embase and Cochrane library to review available data satisfying inclusion criteria. Prescribing data was collected for the usage of analgesics and mouthwashes in Wales from Comparative Analysis for Prescribing Audit software. Statistical analysis was undertaken to assess the effects of reclassification of diclofenac and an MHRA warning on hypersensitivity in chlorhexidine using ARIMA analysis.

The effect of diclofenac reclassification reduced the rate at which prescribing was decreasing ($p<0.0001$) whereas the MHRA chlorhexidine warning did not affect prescribing trends ($p=0.413$). Diclofenac sodium, dihydrocodeine, ibuprofen, paracetamol and sodium fluoride mouthwash prescribing were all found to have decreased over the data collection period whereas benzydamine mouthwash prescribing remained consistent and aspirin, sodium chloride and hydrogen peroxide mouthwash prescribing was sporadic. Medications available over-the-counter in this project cost NHS Wales £132,176.84. Inappropriate prescribing of diclofenac prescribing was highlighted (May 2017).

Cost savings would be available to NHS Wales if they were to implement a ban on prescribing of these medications like NHS England.[3] Despite evidence against its use,[4] dihydrocodeine is still being prescribed and thus its place on the DPF must be questioned. The project highlighted areas of inappropriate prescribing and thus, where future research could be conducted. Limitations of the data meant it was difficult to draw conclusions from this project.

1. British Pharmacological Society. Ten Principles of Good Prescribing. [accessed 18/12/2018]. Available from: https://www.bps.ac.uk/education-engagement/teaching-pharmacology/ten-principles-of-good-prescribing
2. National Institute for Health and Care Excellence. Dental Practitioners' Formulary. [accessed 18/12/2018]. Available from:https://bnf.nice.org.uk/dental-practitioners-formulary/
3. NHS England. Prescribing of over the counter medication is changing: patient leaflets. 2018 [accessed 29/12/2018]. Available from: https://www.england.nhs.uk/wp-content/uploads/2018/08/1a-over-the-counter-leaflet-v1.pdf
4. National Institute for Health and Care Excellence. Analgesics. [accessed 18/12/2018]. Available from: https://bnf.nice.org.uk/treatment-summary/analgesics.html

---

Characterising the antibacterial activity of Namibian honey

Amisha Patel and L Baillie
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

A huge global concern is that bacteria are becoming resistant to all known antibiotics, which is resulting in increased mortality and more complications with treating infections.[1] Honey represents a valuable alternative due to its known therapeutic properties and efficacy against resistant bacteria such as *MRSA* and *K.pneumoniae*.[2] The aim of this project was to try to identify a novel antibacterial compound present in Namibian honey to help tackle resistance.

Fourteen honey samples from Namibia, 1 from Cardiff and 1 from New Zealand underwent screening for antibacterial activity using an agar diffusion method against *MRSA*, *E.coli*, *K.pneumoniae* and *MSSA*.[3] Promising honeys were then treated with catalase to neutralise the hydrogen peroxide activity, and honeys that retained activity underwent solvent extraction with methanol and hexane. The extracts were then separated by a thin layer chromatography (TLC) method. A bioautographic method[4] was applied to identify which bands on the TLC had antibacterial activity.

Seven honeys showed good antibacterial activity after initial screening, subsequently 3 of these honeys: Clabratant, Neudam FS and Windhoek, continued to show activity after catalase treatment against *MRSA*. This suggests some activity is due to a non-peroxide substance. The bioautographic method revealed an antibacterial spot with an Rf value of 0.2 in the hexane extracts. To identify the compound, liquid chromatography–mass spectrometry was carried out on all of the hexane extracts. A prominent peak was found with a mass-to-charge ratio of 263.

Using the information gathered, databases were searched and it was concluded that the compound could not be identified without further analysis. This indicates the possibility of a novel compound being found, however further research would need to be done to confirm this and it's antibacterial nature.

1. 1. Jim O'Neill. The Review on Antimicrobial Resistance. 2014 [accessed 8 Dec 2018]. Available from: https://amrreview.org/sites/default/files/AMR%20Review%20Paper%20%20Tackling%20a%20crisis%20for%20the%20health%20and%20wealth%20of%20nations_1.pdf

2. 2. Molan, PC. The antibacterial activity of honey: 1.The nature of the antibacterial activity. Bee World, 1992;73(1):5-28.
3. 3.Bauer AW, Kirby WM, Sherris JC, Turck M. Antibiotic susceptibility testing by a standardized single disk method. Am J Clin Pathol. 1966; 45(4): 493-496.
4. 4.Blaxland J. The antibacterial activity of Humulus lupulus against mycobacteria [PhD Thesis]. Cardiff: Cardiff University; 2015.

Investigating melanoma cell and keratinocyte crosstalk by developing a 2D cell culture model

Emily Postins and CP Thomas
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

The skin consists of three distinct layers, the epidermis, dermis and subcutaneous tissue, of which many cell types are present.[1] There is a strong functional interrelationship between keratinocytes and melanocytes which are present within the epidermis.[2] Melanoma can form as a result of this moderating interaction breaking down however the underlying mechanisms are not fully understood.[3,4] The aim of this project was to investigate melanoma cell and keratinocyte crosstalk at different stages of differentiation to establish a two-dimensional (2D) *in vitro* model.

HaCaT cells were cultured in three varying calcium concentrations, very low calcium (0.03 (millimolar (mM)), standard calcium (1.8mM) and high calcium (2.8mM). HaCaT cells were cultured in the specific calcium concentration for 24 hours and the conditioned medium was used for the analysis of melanoma. A scratch wound assay was conducted to assess the effect of conditioned medium and fresh DMEM medium on the migration of melanoma cells. Static images of the scratch wound assay were used to calculate the surface area of the scratch. The scratch closure was calculated as a percentage against the initial scratch.

Very low calcium control and very low calcium conditioned media had a scratch closure of 100% at 24 hours. Standard calcium control and standard calcium conditioned media had a scratch closure of 80% and 82% respectively at 24 hours. High calcium control and high calcium conditioned media had a scratch closure of 66% and 60% respectively at 24 hours. There was no difference in the migration of melanoma cells cultured in conditioned media, however cells migrated at a faster rate in the very low calcium condition.

In this 2D model, A375 melanoma cells are sensitive to changes in calcium concentration, providing new information in the field. Secreted keratinocyte signalling mediators had no effect on the migration of melanoma cells.

1. Tobin DJ. Biochemistry of human skin—our brain on the outside. *Chem Soc Rev*. 2006;**35**(1):pp. 52-67. doi: 10.1039/b505793k
2. Chung H, Jung H, Jho E, Multhaupt H, Couchman J, Oh E. Keratinocytes negatively regulate the N-cadherin levels of melanoma cells via contact-mediated calcium regulation. *Biochem Biophys Res Commun.* 2018;**503**(2):pp. 615-620. doi: 10.1016/j.bbrc.2018.06.050
3. Haass NK, Smalley KSM, Li L, Herlyn M. Adhesion, migration and communication in melanocytes and melanoma. *Pigment cell res.* 2005;**18**(3):pp. 150-159. doi: 10.1111/j.1600-0749.2005.00235.x
4. Wang J, Kalabis MF, Herlyn M. Crosstalk in skin: melanocytes, keratinocytes, stem cells, and melanoma. *J Cell Commun Signal* .2016;**10**(3):pp. 191-196. doi: 10.1007/s12079-016-0349-3

Investigations into the stability of intravenous lipid emulsions with varying volumes and concentrations of glucose over a period of time

Alice Preece and AG Cosslett
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, Redwood Building, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Parenteral nutrition (PN) admixtures are indicated for patients who cannot obtain nutrients from digestive processes. These admixtures are composed of over 50 components that are dependent on a patient's requirement.[1 2] If not calculated accurately, it may lead to an unstable admixture. The aim of this study was to determine the effects of glucose on the stability of the lipid components.

Smoflipid® 20%, Intralipid® 20% and glucose 10%, 50% and 70% in combinations of 25ml, 50ml and 75ml were used to make 100ml test sample. N=6 sample size was used, with 3 stored at 20°C and 3 at 40°C. Testing was performed on Days 0-3, samples containing maximum volumes of each concentration tested again on Day 21. Testing methods include visual inspection, pH analysis, microscopy, and laser diffraction to determine the largest globule size.[3]

Most samples remained stable with no significant changes. The most unstable sample was Intralipid® 20% 25ml with glucose 70% 75ml when stored at 40°C. All testing methods supported the classification: observations of a yellow colour change, water layer formation, cream layer that could not be re-dispersed when shaken and an oil layer on the surface. The sample had a pH of 2.85 and the largest globule size of 15.136µm. Both lipid samples with glucose 70% and 50% 75ml were classed as unstable from Day 3 when stored at 40°C and at both storage temperatures on Day 21. Smoflipid® 20% proved more susceptible to instabilities sooner than Intralipid® 20% as unstable results were seen on Day 21 when added to glucose 10% 75ml. This was not reciprocated in Intralipid® 20%.

PN admixtures containing lipids and glucose of high concentrations should be monitored frequently and stored for no longer than 48 hours. Using a lower glucose concentration would be the most favoured approach in practice to maintain patient safety and wellbeing.

1. Joint Formulary Committee. *British National Formulary*. **76 ed**. London: BMJ Group and Pharmaceutical Press; 2018. P1038-1043.
2. Mirtallo J, Canada T, Johnson D, Kumpf V, Peterson C, Sacks G et al. *Safe Practices for Parenteral Nutrition. Journal of Parenteral and External Nutrition*. 2004; **28**(6): S43.
3. Malvern Panalytical. *Laser Diffraction*. [Accessed 3 December 2018]. Available at: https://www.malvernpanalytical.com/en/products/technology/light-scattering/laser-diffraction

Screening of novel monomers for the recognition of endotoxin

Natalie Price, N Demertzis and JL Bowen
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Sepsis is a syndrome involving physiologic, pathologic and biochemical abnormalities that arise from a dysregulated response to infection.[1] Early detection and prompt treatment are critical, hence there is a clinical need for biomarkers indicating sepsis.[2] Lipopolysaccharide (LPS) is a component of the outer membrane of Gram-negative bacteria that has a key role in the pathogenesis of sepsis and has been identified as a possible biomarker.[3] Molecularly imprinted polymers (MIPs) act as synthetic receptors and demonstrate potential as biosensors.[4] The aim of this project was to electropolymerise a series of novel monomers and screen for interaction with LPS to determine their suitability for application in a MIP.

The novel squaramide monomers were electropolymerised using various solvents in order to determine the ideal conditions for their electropolymerisation. Squaramide 1 was taken forward as a successful polymer. Two control polymers, dopamine and aniline, were also electropolymerised. Electrochemical impedance spectroscopy (EIS) was used to measure the interaction of varying concentrations of LPS with the polymers. Using an equivalent circuit model, Nyquist plots were extracted from the EIS data and were used to calculate the change in electron transfer resistance (ΔRct), which was plotted against LPS concentrations to establish binding interaction.

An aqueous-organic solvent mixture was used to successfully polymerise squaramide 1. The squaramide 1 polymer demonstrated significant interaction with LPS, showing greater ΔRct values compared to polydopamine. Polyaniline showed the greatest ΔRct values out of all the polymers, however it is likely to be unsuitable for this application due to its porous structure.

Squaramide 1 was successfully electropolymerised onto the surface of a gold electrode. The SQR 1 polymer demonstrated superior binding interaction with LPS compared to the polydopamine control. Therefore, the novel SQR 1 monomer shows potential for future application in the development of a biosensor to detect LPS.

1. Singer M, Deutschman C, Seymour C, Shankar-Hari M, Annane D, Bauer M et al. The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3). *JAMA*. 2016;**315**(8):801. doi: 10.1001/jama.2016.0287
2. National Institute for Health and Care Excellence. Sepsis: recognition, diagnosis and early management. 2017 [accessed 2 Dec 2018]. Available from: https://www.nice.org.uk/guidance/ng51/chapter/Context
3. Ulevitch R, Tobias P. Recognition of Gram-negative bacteria and endotoxin by the innate immune system. *Curr Opin Immunol*. 1999;**11**(1):19-22. doi: 10.1016/s0952-7915(99)80004-1
4. Abdin M, Altintas Z, Tothill I. In silico designed nanoMIP based optical sensor for endotoxins monitoring. *Biosens Bioelectron*. 2015;**67**:177-183. doi: 10.1016/j.bios.2014.08.009

Role of adhesion in biofilm prevention in the wound healing process. A numerical approach investigating the influence of surface and material characteristics on adhesion.

Alana Borthwick and P Prokopovich
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Bacterial adhesion onto wound surfaces is a rising concern for health due to the implications of infection, enhanced when the bacteria adapts to form a biofilm. A meta-analysis published in the Journal of Wound Care in 2017 concluded there was a prevalence of 78.2% biofilms found in chronic wounds.1 The aim of this project is to deduce the most favourable dressings for wounds to prevent biofilm formation through a numerical approach.

An adaptation to the JKR asperity model that was generated in house was used. The JKR model assumes that one surface is rough (biomaterial) and the other smooth (bacteria) and allows the generation of adhesive force between them. Parameter values for the approach including Elastic modulus, Poisson ratio and Surface energy were collected through a literature review. Four materials were run against the bacteria Staphylococcus epidermis. These materials were then run against a further three bacteria (Staphylococcus aureus, Staphylococcus salivarius and Escherichia coli).

When the simulation was run with S. epidermis, the most adhesive material was 2% Alginate hydrogel and the least adhesive was Cellulose Based Hydrogel with Glycerol and with Cellulose Based Hydrogel. With reference to contact area and adhesive force there was significant difference between the materials excluding CBH and CBHG which had no significant difference between them. Asperity height and radii values were also collected.

From the results of this project it's concluded that Cellulose Based Hydrogel with or without glycerol would be the most appropriate choice when trying to prevent biofilm formation. This was determined as it's the least adhesive dressing for all bacteria, this would make bacteria unlikely to adhere to the dressing and therefore not in the presence of the wound surface. This choice complimented by the advantages of hydrogel dressings and the wide use of these type of dressings in wounds today.2

1. Malone et al. The prevalence of biofilms in chronic wounds: a systematic review and meta-analysis of published data. *Journal of Wound Care.* 2017;**26**(1):20-25. DOI: 10.12968/jowc.2017.26.1.20.
2. M. Bennet-Marsden. How to select a wound dressing. Pharmaceutical Journal. 2010;**2**(363) Available from: https://www.pharmaceutical-journal.com/careers-and-jobs/careers-and-jobs/career-feature/how-to-select-a-wound-dressing/11039846.article [07/12/18]

Long-acting erythropoietin stimulating agent therapy for anaemia of chronic kidney disease in pre-haemodialysis patients: comparison with South West Wales snapshot data

Qurat-Ul-Ain Qamar, J Stead, M Gumbleton, J Jenkins, C Brown[1], O Brooks[1], F Cameron[1] and W Watkins[2].
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [1]Morriston Renal Unit, Heol Maes Eglwys, Morriston, Cwmrhydyceriw, Swansea SA6 6NL, Wales, UK and [2]Sir Geraint Evans Wales Heart Research Institute, Cardiff CF14 4ED, Wales, UK

The project was initiated with the Abertawe Bro Morganwg University Health Board (ABMUHB) using real-world data to establish effectiveness and the safe dose conversion ratio of Mircera (methoxy polyethylene glycerol-epoetin beta) in comparison to patients receiving Aranesp (darbepoetin alpha) at different frequencies for the treatment of anemia in pre-hemodialysis chronic kidney disease (CKD). The primary target is to keep Hb within a range of 100-120. (1,2) The project also investigates the logistical impact of the switch comparing the cost difference of district nurses' visits for the administration of ESA to the patient which drives therapeutic tendering.

The mean monthly ESA dose and Hb (±95% CI) were recorded over a period of 16 months. In the pre-switch period the patients were on Aranesp (months -3 to -1) and in the post-switch period, patients were on Mircera (months 0 to +12). Mean evaluation dose conversion ratio (EDR) of Mircera/Aranesp and linear regression analysis were done.

The dataset contained 127 patients but only 119 were included in the analysis (after applying exclusion criteria). These patients had a mean Aranesp dose of 94.4ug (±95% CI 93.1,95.7) pre-switch and 89.9ug (88.9,90.9) post-switch with an EDR of 0.95 which indicates mean Haemoglobin (Hb) stability throughout the

study. The DCR subgroup analysis of <0.8,0.8-1.2 & >1.2 showed that Hb fluctuates when EDR >1.2. The patients who were on “weekly”, “fortnightly”, “once every three weekly" or "monthly" Aranesp doses showed Hb stability when EDR is between 0.89-1.40 while Linear Regression showed the DCR to be 0.8-1.4. The <100, 100-120 & >120g/L Hb subgroup showed an EDR of 0.91 to 0.97.

The dose conversion ranged from 0.8 to 1.4 depending on the Hb status of the patient. The switch results in efficient and effective treatment that saves £7349.7 per 30 patients on weekly Aranesp dosing regime which shows a reduced cost burden on healthcare system.

1. The Renal Association. Anaemia of Chronic Kidney Disease. 2017. [accessed 18 Jan 2019]. Available at: https://renal.org/wp-content/uploads/2017/06/anaemia-of-chronic-kidney-disease5d84a231181561659443ff000014d4d8.pdf
2. NICE. Chronic kidney disease: managing anaemia. 2015. [accessed 18 Jan 2019]. Available at: https://www.nice.org.uk/guidance/ng8/resources/chronic-kidney-disease-managing-anaemia-pdf-51046844101

User Perspectives on a Proposed New Contraceptive Method

Hina Qazi and ML Hughes
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

There is an unmet need for an ideal method of contraception in Sub Saharan Africa (SSA).[1] Development of a novel, innovative and strategically important method has the potential ability to reduce this unmet need. Therefore, a discrete, irreversible and self-applicable method is being developed using microneedle technology. This study aims to gather user acceptability data on the proposed method by obtaining viewpoints of women in the UK to supplement previous user acceptability research carried out in SSA.

Due to the exploratory nature of the research, qualitative methodology was used to conduct the study. Purposive sampling was used to recruit participants. Eligible participants included any women of childbearing age of between 18-45 years. Focus group discussions were carried out using an open-questioned and prompting discussion guide. Focus groups were audio recorded with consent and transcribed verbatim. Data analysis was conducted using inductive thematic analysis to identify key themes. Ethics approval was obtained for the study.

Twenty-three participants were interviewed in five focus groups. Four major themes identified included method attributes: drivers of user preference, user perspectives on the proposed new method, reactions to prototype applicator devices and method recommendations.

The results showed that multiple method attributes and prototype characteristics contribute towards user acceptability, however recommendations towards these attributes and characteristics were also made. Further research would allow generalizable data to be gathered from across the UK. All results are intended to guide design and development of the new method.

1. Tsui, Amy O. Contraceptive Practice in Sub-Saharan Africa: *Population and development review*. Suppl. 2017;**43**:166-191.

The Role of ZIP7 in the Activation of Immunity

Jack Raddenbury and KM Taylor
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Zinc is an essential nutrient for the correct function of many proteins in the body.[1] The amount of zinc inside cells is partly determined by the ZIP7 zinc transporter.[2] Furthermore, ZIP7 has been discovered to bind to proteins important in the immune response, such as CD40.[3] It is unknown what role, if any, ZIP7 has in the activation of these proteins. The aim of this project is to determine that ZIP7 is bound to the CD40 complex, that this is dismantled on activation, that zinc treatment can dismantle this complex and that two ZIP7 mutants which have been observed clinically behave differently to the natural ZIP7.

The ZIP7 sequence was examined for potential binding sites to TRAF proteins. Western Blot and fluorescent imaging was used to examine ZIP7, CD40, TRAF2 and TRAF6 in control conditions or when stimulated with zinc. Mutant ZIP7 constructs were also examined under these conditions to observe any change in effect.

ZIP7 potentially binds to many of these immune proteins. Western Blot reveals that ZIP7, when treated with zinc, increases the amount of these immune proteins, consistent with binding in unstimulated conditions.

Furthermore, both mutant ZIP7 constructs behave differently to wild-type ZIP7, impairing function and not showing the same increase in certain immune proteins.

This data suggests that when the ZIP7 transporter is activated it dismantles from the binding to these immune molecules, allowing these proteins to carry out their function. The difference in activity with the mutant ZIP7s suggests that the change in shape has an effect on this process. However, more research is needed to solidify these claims as the results gathered were not statistically significant and had wide error bars.

1. Tapiero H, Tew K. Trace elements in human physiology and pathology: zinc and metallothioneins. Biomed Pharmacother. 2003;**57**(9):399-411.
2. Cousins R, Liuzzi J, Lichten L. Mammalian zinc transport, trafficking, and signals. Chem Bio Chem. 2006;**281**:24085-24089.
3. Ewing R, Chu P, Elisma F, Li H, Taylor P, Climie S et al. Large-scale mapping of human protein-interactions by mass spectrometry. Mol Syst Biol. 2007;**3**:89.

Identification, design and synthesis of novel opsin ligands as a strategy against Leber congenital amaurosis and retinitis pigmentosa

Aws Radef and M Bassetto
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Leber Congenital Amaurosis (LCA) and Retinitis Pigmentosa (RP) are forms of inherited retinal dystrophy that can lead to blindness.[1,2] Due to the severity of the conditions and the lack of effective treatments there is an interest in the development of a pharmacological therapy that is potent and safe. Some types of these conditions are caused by mutations in genes that lead to opsin misfolding and render it dysfunctional, impairing vision. For this reason, opsin has been proposed as a drug target that could be rescued, allowing its correct involvement in the phototransduction pathway, thus restoring vision.

This project entails the identification and design of novel opsin ligands using molecular modelling techniques such as shape-based virtual screening and molecular docking, as well as chemical synthesis of these compounds to be tested in cell-based assays.

Libraries were screened for compounds that are similar in shape to a known opsin ligand, YC-001. After docking, rescoring and consensus scoring, 38 commercial compounds were chosen for purchase and biological testing. YC-001 derivatives were docked into the binding site of opsin and their interactions were visualised. Compounds **6** and **9** were the most promising as they had more interactions with residues in the binding site. More derivatives were designed based on these results. Compounds **1-7** and **9** were successfully synthesised with yields of 12-75%. Synthesised compounds underwent biological testing to assess their activity in rescuing mutant opsin. Compound **5** demonstrated the greatest activity by appearing to rescue a large proportion of the misfolded opsin in cells. Compounds **2** and **6** also showed promise in the assays.

The next stage of the study is to assess the biological activity of the selected commercial compounds and the remaining derivatives. The compounds showing the greatest activity will be optimised to eventually develop a novel opsin ligand that could potentially be therapeutic for LCA and RP.

1. den Hollander AI, Roepman R, Koenekoop RK, Cremers FPM. Leber congenital amaurosis: Genes, proteins and disease mechanisms. Prog Retin Eye Res. 2008;27(4):391–419. doi: 10.1016/j.preteyeres.2008.05.003
2. Parmeggiani F. Clinics, Epidemiology and Genetics of Retinitis Pigmentosa. Curr Genomics. 2011;12(4):236–7.

Design and Synthesis of Novel CYP51 Inhibitors as Candida Therapeutics

Ieuan Rosser and C Simons
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Fungal infections are a global issues affecting over 150 million people worldwide annually. 750,000 of these are caused by invasive Candida infection. Mortality from these infections is estimated to be similar to Tuberculosis and three times more than Malaria.[1] Current therapies are limited with triazole antifungals forming the mainstay of treatment. Failure with these and the next line treatments have multiple side effects. As well as this the triazoles have issues with resistance and drug drug interactions. The aim is to licence a triazole based therapy targeting the CYP51 enzyme that is involved with ergosterol synthesis that is effective against Candida without the problems of existing drugs.[2-4]

To begin with potential active structures were designed using modelling software MOE. Structures were based on the licenced inhibitor posaconazole and docking studies were performed to assess the suitability of a variety of structures. Viable compounds, with similar amino acid interactions to posaconazole were synthesised using a 4 step process. The first step, a CDI coupling reaction to form an amide, a cyclisation reaction to form an oxazoline ring, the addition of a triazole group and addition of a benzoyl chloride, benzene sulfonyl chloride or phenyl isocyanate. Purification then characterisation of the structures was undertaken using column chromatography, proton NMR, carbon NMR and TLC. Pure compounds were sent to Swansea University for biological evaluation.

Docking studies produced a number of viable compounds with similar or increased amino acid interactions when compared with posaconazole. Four final compounds were synthesised with enough purity for biological evaluation. One reaction failed to reach completion due to issues with starting material that were discovered.

Purification steps were suggested at each stage of the reaction process to remove impurities and ease work ups for subsequent reactions. Biological evaluation will determine the most effective compounds *in vitro*. This data can then be used to improve structures and identify lead compounds.

1. Sardi JCO, Scorzoni L, Bernardi T, Fusco-Almeida AM, Mendes-Giannini MJS. Candida Species: Current Epidemiology, Pathogenicity, Biofilm Formation, Natural Antifungal Products and New Therapeutic Options. J Med Microbiol. 2013; 62: 10-24. DOI 10.1099/jmm.0.045054-0.
2. Ghannoum MA, Rice LB. Antifungal Agents: Mode of Action, Mechanisms of Resistance, and Correlation of these Mechanism with Bacterial Resistance. Clin Microbiol Rev. 1999; 12(4): 501-517.
3. Bruggemann JMR, Alffenaar J-WC, Blijlevens NMA, Billaud EM, Kosterink JGW, Verweij PE, Burger DM, Saravolatz LD. Clinical Relevance of the Pharmacokinetic Interactions of Azole Antifungal Drugs with Other Coadministered Agents. Clin Infect Dis. 2009; 48(10): 1441-1458. *https://doi.org/10.1086/598327*.
4. Brown GD, Denning DW, Gow NAR, Levitz, SM, Netea MG, White TC. Hidden Killers: Human Fungal Infections. Sci Transl Med. 2012; 4(165). DOI: 10.1126/scitranslmed.3004404.

Assessment of Asthma Control, Inhaler Technique and Adherence within Community Pharmacy

Alliyah Sajid, A Broom, B Abdirizaq, D Skelton and WR Ford
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Worldwide asthma control is unsatisfactory. Although medication is clinically effective, 18% of patients within Western Europe display severe symptoms.[1,2] Possible causes for this include inadequate methods of inhaler use and compliance.[3,4] This study aims to determine potential factors contributing to poor asthma control including inhaler technique and adherence.

A total of 63 patients were recruited predominately in six community pharmacies within South Wales via convenience sampling. Patients answered a demographic survey including details of their asthma clinical history after consent was obtained. The outcome measure is asthma control, evaluated using the Asthma Control Test (ACT). Inhaler technique and adherence was assessed using the Vitalograph Aerosol Inhalation Monitor (AIM) and the Test of the Adherence to Inhalers (TAI) respectively.

Key findings include 60% of patients had uncontrolled asthma. 57% were prescribed more than two types of asthma medication, indicating previous poor control. All patients prescribed 4 different types of medication were uncontrolled. 60% of patients experienced an exacerbation; 29% occurred within the last year. Most inhaler technique assessments, 54%, resulted in a fail. Although metered dose inhaler (MDI) assessments obtained the most fails at 77%, only 16% of patients used a spacer to improve this. Among healthcare professionals (HCPs), pharmacists trained a mere 3% of patients. Furthermore, 72% of patients had poor adherence. Unexpectedly, data did not associate exacerbations, incorrect technique, poor adherence, or time of previous asthma review to uncontrolled asthma.

Patient education can improve; long-term asthma management and treatment may benefit from optimisation. There is a need to identify the most effective method of training. Understanding the lack of correct technique, good adherence and spacer use is the basis of future study. As pharmacy-based interventions can improve asthma outcomes, research regarding the role of the pharmacist in the managing chronic conditions may contribute to the development of future services.

1. Rabe KF, Adachi M, Lai CK, Soriano JB, Vermeire PA, Weiss KB, et al. Worldwide severity and control of asthma in children and adults: the global asthma insights and reality surveys. *J Allergy Clin Immunol*. 2004;**114**(1):40-7. doi: 10.1016/j.jaci.2004.04.042

2. Adams NP, Bestall JC, Malouf R, Lasserson TJ, Jones P. Beclomethasone versus placebo for chronic asthma. *Cochrane Database Syst Rev*. 2005;1:CD002738. doi: 10.1002/14651858.CD002738.pub2
3. Bårnes CB, Ulrik CS. Asthma and adherence to inhaled corticosteroids: current status and future perspectives. *Respir Care*. 2015;**60**(3):455. doi: 10.4187/respcare.03200.
4. Sanchis J, Gich I, Pedersen S. Systematic review of errors in inhaler use: has patient technique improved over time? *Chest*. 2016;**150**(2):394-406. doi: 10.1016/j.chest.2016.03.041

The development of a valid HPLC method for analysis of a high-strength loperamide suspension

Gemma Saxty and CP Thomas
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

This project was created when St. Mary's Pharmaceutical Unit (SMPU) was approached by clinicians about developing a high-strength loperamide suspension to treat patients with high-output stomas.[1] Currently, patients required to use up to 64mg loperamide daily are using 2mg capsules sprinkled across food or mixed in with water.[2,3] This invariably results in inconsistent dosing and uniformity, not to mention problems with patient compliance. Therefore, if a new high-strength suspension could be produced and analysed to meet requirements, this could greatly improve treatment for patients.

To be able to determine content uniformity, dissolution and degradation qualities of the suspension created, an appropriate HPLC method needed to be developed and tested. A flow rate of 1.5ml using a 250x4.6mm Phenomenex (Kinetix) C18 column at a temperature of 30°C was used. The wavelength of the UV detector was 219nm. Sample run time was 30 minutes. The mobile phase used for isocratic elution of the samples consisted of 58:37:5 solution A: acetonitrile: tetrahydrofuran. The method passed all SMPU system suitability tests and remained with British pharmacopoeia requirements.

Content Uniformity was carried out in accordance with the British Pharmacopoeia requirements for unlicensed medicines[4], but dissolution testing was limited due to time restraints and lack of apparatus. The suspension failed both tests. Forced degradation studies were carried out for three weeks at 40°C and 60% humidity. This provided mixed results but overall suggested the suspension was stable.

In summary, a valid HPLC method was determined for the analysis of a loperamide suspension. Further development of the method or use of an alternative method to assess for any new degradation peaks within a loperamide sample is required. The high-strength loperamide suspension also needs further improvement before it could be considered a viable treatment option for the management of a high-output stoma.

1. Joint Formulary Committee. *British national formulary*. 76 ed. London: British Medical Association, Royal Pharmaceutical Society; 2018
2. UK Medicines Information Pharmacists (UKMi). 2016. *Can high dose loperamide be used to reduce stoma output?* Available from: https://www.sps.nhs.uk/articles/can-high-dose-loperamide-be-used-to-reduce-stoma-output-2/ [Accessed 09/10/2018].
3. University Hospitals of Leicester. 2005. *Guidelines for the management of adult patients with a high output stoma.* Available from: https://www.leicestershospitals.nhs.uk/EasySiteWeb/GatewayLink.aspx?alId=41610 [Accessed 8 Oct 2018].
4. British Pharmacopoeia Commission. 2017. *The British Pharmacopoeia.* 2018 ed. London: The Stationary Office.

Pharmacist-led Antimicrobial Stewardship in Primary Care – Evaluation of current prescribing practice regarding prophylactic antibiotic treatment for recurrent UTIs

Julie-Anne Scott[1], A Tucker[2], K Davies[3], P Gimson[4] and JL Bowen[1]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK. [2]Glanrhyd Hospital, Tondu Rd, Pen-y-fai, Bridgend CF31 4LN, Wales, UK. [3]Medicines Management Team, Block A, NPT Hospital, Port Talbot, SA12 7BX, Wales, UK. [4]1000 Lives Improvement, No. 2 Capital Quarter, Tyndall Street, Cardiff, CF10 4BZ, Wales, UK.

A recurrent urinary tract infection (UTI) is defined as two or more symptomatic UTIs in a six-month period or three or more symptomatic UTIs in a 12-month period.[1] The management of prophylactic antibiotics for recurrent UTIs is an area identified by the Welsh Government to reduce unnecessary use of antibiotics and optimise prescribing practice, reducing the growth of antimicrobial resistance.[2] Primary care schemes are already in place and seek to ensure that prescribing is appropriate and in line with clinical guidelines.[3] The aim of this study is to evaluate prescribing practice and adherence to guidelines in primary care. Additionally, views from community pharmacists were collected.

Two self-administered, online, questionnaires were sent out to General Practitioner (GP) surgeries and community pharmacies across the Abertawe Bro Morgannwg University Health Board. They were designed to assess knowledge of current guidelines, resistance awareness and prescribing practice. They also focused on gathering current views and recommendations for the future. Both quantitative and qualitative data was collected through closed and open questions. Ethical approval was gained prior to starting the research.

Guideline and resistance awareness amongst clinicians was successfully identified. Many GPs' first choice antibiotic was Nitrofurantoin due to knowledge of growing resistance to Trimethoprim. Conversely, Community Pharmacists lacked awareness; 4 out of 9 participants were unaware of clinical guidelines. A main issue determined was the completion of a six-month review; barriers included lack of time and resources. Participants stated that help from the pharmacy team was beneficial in supporting reviews. Continued education by pharmacists regarding appropriate antimicrobial use was established as an important future recommendation.

Existing schemes have demonstrated an improvement in primary care, however further work is needed. Antimicrobial stewardship activities in community pharmacies is recognised as an area for future development. Continued evaluation in a bigger sample size is recommended.

1. Raz R, Gennesin Y, Wasser J, Stoler Z, Rosenfeld S, Rottensterich E, et al. *Recurrent urinary tract infections in postmenopausal women*. Clinical Infectious Diseases. 2000;**30(1)**:152-6.
2. Welsh Government. Together for Health: Tackling antimicrobial resistance and improving antibiotic prescribing - A Delivery Plan for NHS Wales and its partners. Welsh Government; 2016.
3. National Institute of Health and Care Excellence. Urinary tract infections (lower) - women. London: National Institute of Health and Care Excellence; 2015 [accessed 7th December]. Available from: https://cks.nice.org.uk/urinary-tract-infection-lower-women - !scenario:2

Does the addition of a 2A or IRES linker improve the efficacy of gene therapy in a 6-OHDA rodent model of Parkinson's Disease?

Abbie Shaw, K Fears, M Lelos, EL Lane
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Gene therapy has been investigated as a potential treatment for Parkinson's disease.[1] AAV viruses can be used to deliver the genes for Tyrosine Hydroxylase(TH) and GTP cyclohydrolase-1 (GCH-1), rate limiting enzymes for the production of dopamine.[2,3] We address whether the addition of linkers (IRES and 2A), to the genetic construct, improves efficacy of transcription and leads to increased tyrosine hydroxylase and GCH-1 production, restoring the lack of dopamine seen in Parkinson's disease.

46 rats received 6-OHDA lesioning by unilateral stereotaxic surgery. Rats were split into six groups receiving either MRX001 (containing IRES linker) at 2.5e9 or 5e9, MRX002(containing 2A linker) at 5e9, reference vector at 2.5e9 or 5e9 or the control group. Immunohistochemistry for normal TH and truncated TH followed by optical density analysis was completed to assess extent of lesioning and whether vectors had efficacy. Cylinder testing was used to see if changes in behaviour would be achieved.

The 6-OHDA lesions were successful with 98.3% of dopaminergic cells depleted. Normal TH staining on the lesioned side compared to the intact side was 83.59% less (SEM= 1.62). Cylinder testing revealed contralateral forelimb dysfunction (One-way ANOVA, P=<0.0001). Post-vector there was no difference in truncated TH histological analysis between groups (One-way ANOVA, P= 0.0523) or changes in behaviour (One-way ANOVA, P=0.3190), meaning vectors did not have efficacy.

Since we used threshold doses[3] it is possible that dosages were sub-optimal or that techniques used to detect histological changes or behavioural changes are not sensitive enough. The future of this technique depends on finding methods to ensure sufficient gene expression and spread of the vector to achieve a clinical response. Linker usage remains an option to do this, but other techniques must also be explored. Translation of this technique to the clinic also remains a major barrier.[4]

1. Palfi S, Gurruchaga JM, Lepetit H, Howard K, Ralph GS, Mason S, et al. *Long-Term Follow-Up of a Phase I/II Study of ProSavin, a Lentiviral Vector Gene Therapy for Parkinson's Disease*. Hum Gene Ther Clin Dev. 2018;29(3):148-55. doi: 10.1089/humc.2018.081
2. Cederfjäll E, Sahin G, Kirik D. *Key factors determining the efficacy of gene therapy for continuous DOPA delivery in the Parkinsonian brain*. Neurobiol Dis. 2012;48(2):222-7. doi: 10.1016/j.nbd.2011.10.017

3. Cederfjäll E, Nilsson N, Sahin G, Chu Y, Nikitidou E, Björklund T, et al. *Continuous DOPA synthesis from a single AAV: Dosing and efficacy in models of Parkinson's disease*. Sci Rep. 2013;3(5): 1-12. doi: 10.1038/srep02157

What are the views of community pharmacists in Wales, regarding anticoagulation medication errors and their prevention?

Holly C Sherrad[1], P Gimson[2] and JL Bowen[1]
1Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK
21000 Lives Improvement, Public Health Wales, Number 2 Capital Quarter, Tyndall Street, Cardiff, CF10 4BZ, Wales, UK

In March 2017 The World Health Organisation set its third Global Safety Challenge – Medication Without Harm - aiming to "reduce the level of severe, avoidable harm related to medications by 50% over 5 years, globally".[1] One action involves targeting high risk medications including anticoagulants. Anticoagulants, combined with non-steroidal anti-inflammatory drugs and antiplatelets account for over a third of hospital admissions due to avoidable adverse drug reactions (ADRs), in England.[2] This project aims to gain a greater understanding of the current obstacles to improving anticoagulant medication safety in Wales to help guide future strategy development.

Views from community pharmacists across Wales were gathered via the use of an online questionnaire. The questionnaire comprised of both open and closed questions. The study was approved by the School Ethics Committee ahead of distribution.

The results highlighted areas to improve; awareness of the WHO challenge, use of a counselling checklist, completing Continuing Professional Development (CPD) related to anticoagulant medication errors, consistent reporting of errors, and widely known and followed guidelines. 'High work pressure' was the most commonly reported factor, by 58%, considered responsible for contributing to anticoagulation medication errors. Other key themes identified included patient knowledge, drug knowledge and systematic issues that need reviewing. When asked for suggestions most community pharmacists focused on increased access and sharing of information (especially concerning International Normalized Ratio (INR) results).

This study has raised many factors pertaining to anticoagulation medication errors that could be reviewed and improved, notably access to INR results and inter-sector communication.

Further research into the views of other healthcare sectors and professionals would further aid prevention of anticoagulant medication error planning. In reference to the WHO challenge other 'high-risk' classes of medication should also be reviewed and a comparison may highlight mutual areas for future development.

1. Donaldson L, Kelley E, Dhingra-Kumar N, Kieny M, Sheikh A. *Medication Without Harm: WHO's Third Global Patient Safety Challenge.* The Lancet. 2017;389(10080):1680-1681. doi: 10.1016/s0140-6736(17)31047-4
2. Elliott R, Camacho E, Campbell F, Jankovic D, St James M, Kaltenthaler E et al. Prevalence and economic burden of medication errors in the NHS in England. 2018. [accessed 18 Jan 2019] Available from: http://patientsafety.health.org.uk/sites/default/files/resources/does_improving_safety_culture_affect_outcomes.pdf

Asthma control, Adherence and Inhaler Technique, and Their Effects on the Long Term Management of Asthma

Dafydd Skelton and WR Ford
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, Cf10 3NB, Wales, UK

Despite the availability of clinically effective drugs the need for prescribing of oral steroids and the frequency of hospitalization and death rates related to asthma are still high.[1] We hypothesized that a combination of both poor adherence and a poor inhaler technique led to patients having a greater likelihood of having poor long term outcomes. In this study we aimed to measure patients' adherence to their medication regimen of inhaled corticosteroid alongside their inhaler technique for comparison with both short term and long term asthma outcomes.[1]

Using the validated Asthma Control Test, and 10 point TAI adherence test and AIMS device to measure these, alongside using a vitalograph AIMS machine to determine the quality of a patient's inhaler technique. [2, 3].

Quantitative data were analyzed using SPSS. Kruskal-Wallis and Mann-Whitney U statistical tests were used to compare patient groups.[2,3]

Patient's asthma control and adherence levels were lower than European and WHO estimates.[4] There was no correlation between asthma control or patient adherence to their medication regimen and patient need for hospitalization or use of oral steroids. Patients demonstrating technique to healthcare professional was essential for development of basic elements of inhaler technique. However, current teaching methods are not able to coach patients to master more complex elements of inhaler techniques thus highlighting the need for a Device such as AIMS to give patient instant feedback on their technique. Patients' technique using a DPI was superior to MDI. Poor DPI technique was linked with higher levels of serious exacerbation. This was not seen in MDI.[4]

There is a need for a device such as AIMS to give patients instant feedback on their technique. Patients may benefit from being prescribed a DPI rather than an MDI if taking long term inhaled corticosteroids. Preventer medication may be underprescribed.

1. Royal College of Physicians. 2014. Why asthma still kills. The National review of Asthma Deaths (NRAD) available at file:///C:/Users/User/Downloads/Why%20asthma%20still%20kills%20full%20report%20(1).pdf [accessed 11 November 2018].
2. Nathan RA, Sorkness CA, Kosinski M, Schatz M, Li JT, Marcus P, Murray JJ et al. 2002. Development of the asthma control test: A survey for assessing asthma control. The Jounal of Allergy and Clinical Immunology. 113(1): 59-65 DOI: https://doi.org/10.1016/j.jaci.2003.09.008
3. Plaza V, Fernández-Rodríguez C, Melero C, Cosío BG, Entrenas LM, Pérez de Llano L, Guitérrez-Pereyra F et al. 2016. Validation of the "test of the adherence to inhalers" (TAI) for asthma and COPD patients. Journal of Aerosol Medicine and Pulmonary Drug Delivery. 29(2) 142-152. doi: 10.1089/jamp.2015.1212
4. Cerveri I, Locatelli F, Zoia MC, Corsico A, Accordini S, and de Marco R. 1999. International variations in asthma treatment compliance: the results of the European community respiratory health survey. European respiratory journal. 1999 (14) 288-294. (no doi available) available at https://erj.ersjournals.com/content/14/2/288.long

Exploring the chemical constituents of Hydnora africana and their antibacterial properties

John Speakman and AW White
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

It has been described as the strangest plant in the world[1], Hydnora africana grabs the attention of those interested in the weird and wonderful aspects in the botany world. Growing underground as a parasitic plant, it relies solely on another plant- Euphorbia, to survive. Having been used by locals to treat conditions including diarrhoea and dysentery for years and is one of the most trusted herbal remedies in South Africa.[2] This study aims to separate components of the root of H.africana and identify their properties.

Solvent extraction was used to separate components of H.africana by polarity. These fractions were then separated further through preparative TLC and these *components* were then analysed through mass spectrometry and Nuclear Magnetic Resonance (NMR). Six bands were isolated from the H.africana sample after preparative TLC- 2 from the hexane fraction, 3 from the ethyl acetate and 1 from the methanol fraction. There were four groups of interest, which appeared in at least three of the bands each. These bands were then tested for antibacterial activity.

The four groups were of interest could be novel compounds as they were not identified in previous research done into H.africana, nor on a chemical database.[4] The antibacterial activity assay didn't work, so the activity could not be determined. The tests completed for functional groups gave some indication as to what the potential structure of these isolated compounds was, but further tests and methods are needed, such as IR spectroscopy and Carbon-NMR.

The screening for activity against diarrrhoea causing bacteria could be completed in future research successfully, indicating the activity of these non-polar compounds. This, along with exploring how these active compounds comply with Lipinski's rule of five[3], would determine the suitability of H.africana as a lead compound in drug discovery.

1. WGBH, The Strangest Plant In The World?! 2017 [accessed 16 Jan 2019]. Available from: https://www.youtube.com/watch?v=WBVd9Stp2s8
2. Wintola, O. and Afolayan, A. A SURVEY OF MEDICINAL PLANTS USED IN THE TREATMENT OF DYSENTERY IN AMATHOLE DISTRICT MUNICIPALITY, SOUTH AFRICA. Pakistan Journal of Botany. 2014; 46(5): 1685-1692
3. Lipinski, C., Lombardo, F., Dominy, B. and Feeney, P. (1997). Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Advanced Drug Delivery Reviews*, 23(1-3), pp.3-25.
4. NORMAN MassBank. European MassBank; 2018 [Accessed 16 Jan. 2019]. Available from: https://massbank.eu/MassBank/

A Longitudinal Retrospective Study of the Dose Conversion in Clinical Practice of Erythropoietin Stimulating Agents, Aranesp to Mircera: Comparison to Snapshot Data from North Wales

Jessica Stead, M Gumbleton, C Brown[1], O Brooks[1], F Cameron[1] and J Jenkins
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Ave, Cardiff, CF10 3NB, Wales, UK and [1]Morriston Hospital Department of Nephrology, Heol Maes Eglwys, Swansea, SA6 6NL, Wales, UK

Anaemia is a low haemoglobin (Hb) level in the blood and is common in chronic kidney disease (CKD). Treatments include iron therapy and erythropoietin stimulating agents (ESAs). (1) It has been previously determined that all ESAs are equally effective, (2) however this study aims to explore if the longer-acting ESA Mircera may be superior to Aranesp, due to less frequent dosing requirements, leading to reduced cost and improved compliance. It aims to assess the ability of Mircera to maintain Hb levels in routine clinical practice and identify the dose conversion ratio (DCR) required for this ESA switch.

This was a longitudinal retrospective study, analysing 119 pre-haemodialysis CKD patients who underwent a switch from Aranesp to Mircera. Data were collected monthly during three months of Aranesp treatment and thirteen months of Mircera. For each patient, Hb, estimated glomerular filtration rate/creatinine, ESA dose frequency and total monthly ESA doses were analysed. Results were verified by comparison with a snapshot of ESA treatment in North Wales.

The mean Hb during Aranesp treatment was 109g/L and during Mircera treatment was 107g/L. The mean monthly dose of Aranesp was 73.4mcg and Mircera was 72.1mcg. Patients switched at a DCR of 0.8-1.2 showed no statistically significant difference in Hb during each ESA treatment period. Statistically, patients' pre-switch mean Hb levels contributed towards the post-switch Hb levels and the Aranesp dosing frequency was significant towards the dose of Mircera required.

Mircera maintained Hb stability in non-haemodialysis patients converted from Aranesp at a 1:1 DCR. This DCR was validated by longitudinal and snapshot methodologies and is true for patient sub-groups who presented with in-range and above-range Hb, and for sub-groups who received Aranesp doses fortnightly, every three weeks and monthly. Mircera also offers logistical savings and greater convenience for patients due to its once monthly dosing regimen.

1. Brown C, Jackson D. Diagnosis and Management of Anaemia in Adults with Chronic Kidney Disease. *Pharm J*. 2018; **10**: 269-76
2. Palmer S, Saglimbene V, Mavridis D, Salanti G, Craig J, Tonelli M et al. Erythropoiesis-Stimulating Agents for Anaemia in Adults with Chronic Kidney Disease: A Network Meta-Analysis*. Cochrane Database Syst Rev*. 2014; **12**. doi: 10.1002/14651858.CD010590

Enhanced Retinal Delivery of Idebenone by Cyclodextrin Complexation and Microneedles

Charlotte Tanner and CM Heard
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales UK

Idebenone is an analogue of coenzyme Q10 which is an endogenous antioxidant and electron carrier.[1] It is used to treat Leber's Hereditary Optic Neuropathy[2] and currently delivered orally, despite its low bioavailability.[3] This study aimed to improve the poor aqueous solubility of idebenone by forming inclusion complexes with cyclodextrins. Greater aqueous solubility of idebenone could result in ocular permeation to the retina. Thus avoiding first pass metabolism that reduces idebenone's bioavailability.

Increasing concentrations of different cyclodextrins alpha-cyclodextrin (ACD), beta-cyclodextrin (BCD), 2-hydroxypropyl-alpha-cyclodextrin (2HPACD) and 2-hydroxypropyl-beta-cyclodextrin (2HPBCD) to idebenone and then HPLC was used to determine the concentration of idebenone that was solubilised. The cyclodextrin that was found to be the most solubilising was then used to investigate ocular permeation using corneal rings and microneedles, applied to porcine eyes in vitro. The concentration of idebenone in the retina and vitreous humor were then determined using HPLC

From the phase solubility testing 2HPBCD was found to be the best solubilising cyclodextrin for idebenone - it increased idebenone's solubility by up to 196,813%. When investigating ocular permeation of idebenone it was found that the removal of the epithelium had little effect on permeation to the retina. When lower 2HPBCD

concentrations were used (24mM) idebenone could still be found in the retina. When microneedles were applied beofre dosing, it was found that solutions containing 75% PEG 400 compared to 50% doubled the idebenone concentration found in the retina to a maximum concentration of 0.033mM.

2HPBCD was found to be the most appropriate cyclodextrin to use. Based on its superior ability to improve aqueous solubility, having the best stability constant, complexation efficacy, drug: cyclodextrin molar ratio and utility number. Delivery was not significantly improved usign microneedle arrays. The retina was successfully targeted; this shows that there is potential in treating LHON by ocular delivery.

1. Montenegro L, Turnaturi R, Parenti C, Pasquinucci L. Idebenone: Novel Strategies to Improve Its Systemic and Local Efficacy. *Nanomaterials*. 2018;8(2):87.
2. Scope to inform clinical evidence review of: idebenone for the treatment of Leber's hereditary optic neuropathy (LHON). Nice.org.uk. 2018 [cited 12 December 2018]. Available from: https://www.nice.org.uk/Media/Default/About/what-we-do/Commissioning-Support-Programme/ID011-Idebenone-scope.pdf
3. Kutz K, Drewe J, Vankan P. Pharmacokinetic properties and metabolism of idebenone. Journal of Neurology. 2009;256(S1):31-35.

An exploration of the pharmacy team's perception regarding the future of community pharmacy and its technological needs

Farhana Tarofdear and MW Smith
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

The role of community pharmacists is changing; the future sees them taking on greater clinical roles and becoming more integrated within the NHS.[1] This brings challenges such as increased workload, time constraints and funding pressures, all of which can implicate patient saftey.[2,3] It is recognised that technology is a valuable resource in supporting pharmacies, pharmacy staff and their patients as it allows for costs efficiencies, improves workflow and presents better clinical outcomes.[4] This study, therefore explores the pharmacy team's perception regarding the future of community pharmacy, and their thoughts surrounding a series of technological systems needed to support future roles and overcome challenges.

Survey packs containing a questionnaire, cover letter and free-post envelope were prepared and mailed out to 200 randomly chosen independent pharmacies across England and Wales. A second reminder mailing was sent out two weeks later to pharmacies that had not yet responded. Quantitative data from returned questionnaires were inputted into SPSS to generate descriptive statistics, whilst qualitative data were used to provide further insight into these responses. Ethical approval was obtained.

44 questionnaires were returned giving a 22% response rate. The majority of respondents saw the future of community pharmacy delivering more services including that of medicines management, minor ailments, diagnostic testing and health promotion. 63.6% of respondents agreed to invest in technologies as a way of increasing business efficiency in response to reducing dispensing remuneration. The most popular perceived technologies seen as absolutely essential/very beneficial by respondents were patient apps and an integrated PMR system.

In conclusion, it was recognised that for community pharmacists to adopt increased and enhanced clinical roles, pharmacy staff need to be supported by new and/or greater adoption of technologies. Future research can be conducted to evaluate the popularly perceived technologies, and this can ultimately result in an improved community pharmacy practice.

1. National Health Service. The NHS Long Term Plan. 2019 [accessed 8 Jan 2019]. Available from: https://www.longtermplan.nhs.uk/publication/nhs-long-term-plan/
2. Royal Pharmaceutical Society. Now or never: Shaping pharmacy for the future. 2013 [accessed 21 Oct 2018]. Available from:https://www.rpharms.com/Portals/0/RPS%20document%20library/Open%20access/Publications/Now%20or%20Never%20-%20Report.pdf
3. Peryansas HC, Revell N, Alotaibi AF. Evaluation of Medication Errors in Community Pharmacy Settings: A Retrospective Report. *J Pharm Technol*. 2016;**32**(2):71-74. doi: 10.1177/8755122515617199
4. Liddell A, Adshead S, Burgess E. Technology in the NHS: Transforming the patient's experience of care. 2008 [accessed 21 Oct 2018]. Available from: https://www.kingsfund.org.uk/sites/default/files/Technology-in-the-NHS-Transforming-patients-experience-of-care-Liddell-Adshead-and-Burgess-Kings-Fund-October-2008_0.pdf

Exploring VGLL1, VGLL3 & TEAD1 proteins in acquired endocrine resistant breast cancer cells and their regulation by growth factor kinase pathways.

Llywelyn Thomas, S Bickford and JMW Gee
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

While the majority of oestrogen receptor (ER) positive breast cancer patients treated with endocrine therapy have a positive outcome, relapse is an issue for some. Mechanisms that drive acquired endocrine resistance are still poorly-described[1] but growth factor kinase pathways, such as epidermal growth factor receptor (EGFR) and SRC, have been implicated[2]. However, EGFR and SRC inhibition has had limited success and so the discovery of new resistance pathways is needed. mRNA increases in the HIPPO pathways transcription factor TEAD1 and its co-activators VGLL1/3[3] have recently been detected in resistance[4]. This project aimed to investigate whether the encoded proteins of TEAD1 and VGLL1/3 are deregulated in endocrine resistant cells, independent of EGFR/SRC pathways.

Immunohistochemical assays were optimized for TEAD1, VGLL1 and VGLL3 using in-vitro and xenograft models of acquired endocrine resistance. Assays were then applied to luminal A (MCF7)-derived or luminal B (BT474)-derived cell model panels and a xenograft model of acquired resistance (using responsive parental lines as controls), and to gefitinib (EGFRi) or saracatinib (Srci) (1uM)-treated coverslips. H-scoring with non-parametric analysis identified the impact of endocrine resistance or EGFR/SRC blockade.

Luminal A-derived faslodex resistant cells (FASRLT) that lost ER expression showed significantly increased nuclear TEAD1 ($p=0.007$) and VGLL1 ($p=0.025$) expression, which were also detected in equivalent xenografts. In contrast, luminal B-derived resistant models showed losses for TEAD/VGLL proteins. Inhibiting SRC significantly increased TEAD1 ($p=0.04$) and VGLL1 ($p=0.004$) in FASRLT, and gefitinib only partially, yet significantly, inhibited TEAD1. Nuclear VGLL3 was very low with no significant changes with either pathway blockade.

These various model data implicate TEAD1/VGLL1 in faslodex resistant luminal A-derived breast cancer with ER loss. Since there was no substantial positive regulation by EGFR or SRC signalling, TEAD/VGLL might be a new target for this type of endocrine-resistant breast cancer but requires further research and investigation.

1. Zhao M, Ramaswamy B. 2014. Mechanisms and therapeutic advances in the management of endocrine-resistant breast cancer. *World Journal of Clinical Oncology.* **5(**3), pp. 248-262. doi: 10.5306/wjco.v5.i3.248.
2. Hutcheson IR, Knowlden JM, Madden TA, Barrow D, Gee JM, Wakeling AE, et al. 2003. Oestrogen receptor-mediated modulation of the EGFR/MAPK pathway in tamoxifen-resistant MCF-7 cells. *Breast Cancer Research and Treatment.* **81**(1), pp. 81-93. doi: 10.1023/A:1025484908380
3. Pobbati AV and Hong W. 2013. Emerging roles of TEAD transcription factors and its coactivators in cancer. *Cancer Biology & Therapy.* **14**(5), pp. 390-398. doi: 10.4161/cbt.23788
4. Davies, L. 2018. TEAD1 in Endocrine Resistant breast cancer and its interplay with VGLL-1. MSc Dissertation, Cardiff University.

An investigation on the opinions of Welsh speaking hospital Pharmacists on the use of the Welsh language in secondary care

Emily Walters, W Davies, L Doyle, E Jones
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Following equal rights for Welsh and English languages in 2011.[1] the Welsh Government have introduced initiatives such as the 'More than Words' framework to improve access to Welsh services.[2] There is a need to assess the opinions of Welsh speaking hospital pharmacists on its use, their training background and to evaluate the success of language initiatives. The aim was to explore opinions on language use, barriers and further training needs.

One-to-one semi structured interviews were carried out with N=4 pharmacists. A purposive sample was used. Interviews were recorded with written consent obtained. Interviews were transcribed verbatim, coded and thematically analysed.[3]

Four main themes emerged; language expectation, benefits, confidence and resources. Although patients have perceived low expectations of Welsh language provision, they benefit greatly from the service as a bond is formed with the pharmacist. Elderly patients, young patients and those with speech and language difficulties benefitted most. Welsh is used conversationally, however, pharmacists use English during patient counselling

as they lack knowledge of medical terminology in Welsh. There is a lack of bilingual resources. Additionally, pharmacists have not received Welsh language training.

As pharmacists qualified before the language measure (2011)[1] could not specifically attribute a greater emphasis on Welsh speaking to healthcare, it is difficult to assess the success of language improvement targeted at healthcare. A lack of bilingual resources signals a need for the health board to place a greater emphasis on language provision through providing appropriate resources and further training if needed. Better access to bilingual resources for pharmacists could further encourage the use of Welsh and maximise staff resources. Recommendations include additional signs and noting patient's language preference. This would allow convenient identification of Welsh speakers, reducing the burden on patients to request services in Welsh, complying with the aim of the Active Offer.[2]

1. Welsh Government. The Welsh Language (Wales) Measure 2011. 2011 [accessed 10 Jan 2019]. Available from: http://www.legislation.gov.uk/mwa/2011/1/contents/enacted
2. Welsh Government. More than Just Words, Delivering the 'Active Offer' Information Pack- Health. Cardiff: Welsh Government; 2015 [accessed 10 Nov 2018] Available from: https://gov.wales/docs/dhss/publications/150928activeofferhealthen.pdf
3. Smith F. *Health Services research methods in Pharmacy: Qualitative interviews*. Int J Pharm Pract.1998;**6**:97-108. Doi: /epdf/10.1111/j.2042-7174.1998.tb00923.x

---

Design, Synthesis and Biological Evaluation of Niclosamide Analogues as Potential Activators of the Parkinson's Disease Associated PTEN-Induced Putative Kinase 1 (PINK1)

Aoife Ward-Farrell, H Kadri and Y Mehellou
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Parkinson's disease is the second most common neurodegenerative disease; affecting 1% of the population.[1] Despite the pathology of the disease being well understood, with the presentation of cardinal symptoms; bradykinesia, postural instability, tremor and rigidity[1], the aetiology is unknown, resulting in the absence of disease modifying therapies.[2] However, amongst one cohort of suffers, those with Early-Onset Parkinson's disease, a mutation in the Phosphate and tensin homolog-induced putative kinase 1 (PINK1) has been shown to be causative.[3] PINK1 is involved in neuronal mitophagy[4],and research is being undertaken to explore potential methods to exploit this involvement as overexpression of PINK1 has been shown to have restorative effects for this cohort. One such method is the adaptation of indirect activator niclosamide, currently approved for the treatment of Helminth infections. Niclosamide has been shown to alter mitochondrial membrane potential to activate PINK1.[4] However, adaptations are required to improve the oral bioavailability and to increase penetration of the blood brain barrier to reach the neuronal target. Synthesizing analogues that achieve both of these is the aim of this project.

Three analogues of niclosamide where synthesised in this project through an amide bond formation reaction. Successful synthesis of all 9 compounds occurred with ^{1}H, ^{13}C, ^{19}F, ^{31}P NMR and mass spectrometry where appropriate. Each of the niclosamide analogues achieved a reduction in their total polar surface area.

Overall the synthesis of the analogues was a success, however, refinement of each stage, especially the last, of synthesis is required. Biological testing to see if these improvements have an effect *in vivo* is to follow shortly.

1. Burn D.J. Parkinson's disease. In: Walker R and Whittlesea C, editors. Clinical Pharmacy and Therapeutics. 5th Ed. London. Churchill Livingstone Elseveir; 2012. p507-518.
2. Narendra D, Jin S, Tanaka A, Suen D, Gautier C, Shen J et al. PINK1 Is Selectively Stabilized on Impaired Mitochondria to Activate Parkin. PLoS Biol. 2010;8(1): e1000298. doi: 10.1371/journal.pbio.1000298
3. Valente, E.M. Abou-Sleiman, P.M. Caputo, V. Murquit, M. M.K. Harvey, K. Gispert, S et al. Hereditary Early-Onset Parkinson's Disease Caused by Mutations in PINK1. Science. 2004;304(5674):1158-1160. doi: 10.1126/science.1096284
4. Lambourne O, Mehellou Y. Chemical Strategies for Activating PINK1, a Protein Kinase Mutated in Parkinson's Disease. ChemBioChem. 2018;19(23):2433-2437. doi: 10.1002/cbic.201800497

Perception of Parkinson's Disease nurses on the utility of technology to collect Patient Reported Outcome Measures (PROMs) in a Parkinson's Clinic

Rebekah White, A Khardali and EL Lane
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Parkinson's Disease (PD) comprises a range of motor and non-motor symptoms.[1] The latter appear to be underreported.[1] Clinical tools known as patient reported outcome measures (PROMs) are questionnaires used to measure the patient's perception of their own health status and identify these under recognized symptoms.[2] Traditionally PROMs were paper based but there have been opportunities to move forward with technological advances and collect PROMs digitally.[3] Success to designing and implementing a new clinical tool requires the perceived value of those involved in use. The patient perception has been explored but no study commissioned to understand the healthcare professional's opinion. The aim of this study was to gather PD nurses' perceptions on the use of technology to collect PROMs by designing and conducting a survey.

A cross sectional survey study was conducted using a sample of Parkinson's disease nurse specialists. Ethical approval had to be sought from the Cardiff School of Pharmacy. The survey responses were analysed using descriptive and thematic analysis.

There was a total response rate of 72.2% with 90.8% of participants from England and 9.2% from Wales. 76.9% of nurses had used some form of PROM in their practice but only 50.8% used the tool routinely. Believed that ePROMs would enhance data collection in the clinics by facilitating the process (70.8%) and improve coordination between healthcare professionals (58.5%). Some thought the tool would be more time consuming (20.0%) and make data collection harder (7.7%). 76.9% of nurses were interested in using ePROMs in their clinics.

Participants predominantly perceived the tool to have potential in their clinics. Concerns were identified and solutions offered. Concerns included patients being unable to independently complete the ePROM and uncertainty over security of data collected. Further interviews with nurses to understand their thoughts in greater detail should be undertaken, ensuring this field of research is saturated.

1. O'Sullivan, S.S. et al. 2008. Non motor symptoms as presenting complaints in Parkinson's disease: a clinicopathological study. *Movement Disorders* **23**(1), pp. 101-106. doi: 10.1002/mds.21813
2. Kingsley, C. and Patel, S. 2017. Patient-reported outcome measures and patient-reported experience measures. *BJA Education* **17**(4), pp. 137-144. doi: 10.1093/bjaed/mkw060
3. Schick- Makaroff, K and Molzahn, A. 2015. Strategies to use tablet computers for collection of electronic patient-reported outcomes. *Health and Quality of Life Outcomes* **13**(2). doi: 10.1186/s12955-014-0205-1

Activation of PINK1 by Kinetin Derivatives to Treat Parkinson's Disease

Isabel Wilson and Y Mehellou
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

Worldwide, Parkinson's disease (PD) is the second most common neurodegenerative disease.[1] Studies have shown links between mutations in PINK1 and early onset PD.[2] PINK1 is a protein kinase and thought to be responsible for mitochondrial quality control and elimination of damaged mitochondria.[3] In PD patients where PINK1 is mutated, elimination of damaged mitochondria diminishes leading to cellular death.[4] Discovery of a small molecule termed kinetin[4] led to interest in the development of kinetin derivatives and their prodrugs as PINK1 activators.[1] My research aims to design and synthesise a series of N^6 substituted adenine and adenosine analogues as well as their 5'-*O*-monophosphate prodrugs as ProTides and establish their ability to activate PINK1.

Synthesis of the molecules included various methods; reflux reactions were used in order to create adenine and adenosine analogues, with benzylamine, isopropylamine and methylamine substituted at the N^6 position. In the development of the adenine analogues, the PyBOP method was also utilized and synthesis of two ProTides was explored using established methods that employ either NMI or ^{t}BuMgCl as the bases.

Successful synthesis of the adenine and adenosine analogues was achieved with good yields and purity. Both synthetic methods for the adenosine analogues were successful. However, the reflux method proved superior in purity. Finally, the two methods to synthesise the ProTides produced pure products. A low yield was accumulated, as expected. Furthermore, by identifying the compounds molecular weight, tPSA and LogP, it was established the adenine analogues were the most likely to cross the blood brain barrier. Upon comparison

to previously investigated compounds, it is assumed all compounds will activate PINK1, although with varying degrees of activation.

All of the compounds were successfully synthesised and are thought to activate PINK1. Confirmation of these findings with cell testing will help to confirm activation and identify structure activity relationships.

1. Osgerby L, Lai Y, Thornton P, Amalfitano J, Le Duff C, Jabeen I, Kadri H, Miccoli A, Tucker J, Muqit M, Mehollou Y. Kinetin Riboside and Its ProTides Activate the Parkinson's Disease Associated PTEN-Induced Putative Kinase 1 (PINK1) Independent of Mitochondrial Depolarization. J. Med. Chem. 2017; 60 (8): 3518–3524. doi:10.1021/acs.jmedchem.6b01897
2. Valente EM, Abou-Sleiman PM, Caputo V, Muqit MM, Harvey K, Gispert S, Ali Z, Del Turco D, Bentivoglio AR, Healy DG, Albanese A, Nussbaum R, González-Maldonado R, Deller T, Salvi S, Cortelli P, Gilks WP, Latchman DS, Harvey RJ, Dallapiccola B, Auburger G, Wood NW. Hereditary early-onset Parkinson's disease caused by mutations in PINK1. Science. 2004; 304(5674):1158-60. doi: 10.1126/science.1096284
3. Kumar A, Tamjar J, Waddell AD, Woodroof HI, Raimi OG, Shaw AM, Peggie M, Muqit MM, van Aalten DM. Structure of PINK1 and mechanisms of Parkinson's disease-associatedmutations. Elife. 2017; 6: e29985. doi: 10.7554/eLife.29985
4. Lambourne OA, Mehellou Y. Chemical Strategies for Activating PINK1, a Protein Kinase Mutated in Parkinson's Disease. Chembiochem. 2018; 19(23):2433-2437. doi: 10.1002/cbic.201800497

The importance of the role of ZIP10 phosphorylation in cells

Muhammad Zainuddin and KM Taylor
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Zinc serves important functions in many cellular mechanisms such as transcription, growth, proliferation, differentiation and apoptosis.[1] Zinc transporters ZIP6 and ZIP10 have been discovered to form a heteromer responsible for zinc influx into cells.[2] Since ZIP7 is known to require phosphorylation before transporting zinc[3], ZIP10 was examined for the potential of phosphorylation to trigger zinc influx.

Computer analysis revealed potential phosphorylation sites for ZIP10 and also predicted the involvement of CK2 and PLK1 kinases. MCF-7 breast cancer cells were treated with relevant kinase inhibitors to confirm the potential phosphorylation sites and examined for effect on ZIP10 function. The effect of the kinase inhibitors on ZIP10 function was investigated by fluorescence microscopy.

ZIP10 transporters were shown to increase on the plasma membrane of cells, both by fluorescence microscopy and western blot. Interestingly, treatment with a CK2 inhibitor, CX-4945, was demonstrated to reduce the number of cells positive for ZIP10, suggesting a role for CK2 in activating ZIP10. Treatment with the PLK1 inhibitor did not reduce the number of ZIP10 positive cells suggesting PLK1 has no role in the activation of ZiP10.

These findings indicate the potential importance of phosphorylation of ZIP10 by CK2 in triggering ZIP10 activation which may offer a new target in cancer cells.

1. Beyersmann D, Haase H. Functions of zinc in signaling, proliferation and differentiation of mammalian cells. BioMetals. 2001;14(3/4):331-341.
2. Taylor K, Muraina I, Brethour D, Schmitt-Ulms G, Nimmanon T, Ziliotto S et al. Zinc transporter ZIP10 forms a heteromer with ZIP6 which regulates embryonic development and cell migration. Biochem J. 2016;473(16):2531-2544. .
3. Nimmanon T, Ziliotto S, Morris S, Flanagan L, Taylor KM. Phosphorylation of zinc channel ZIP7 drives MAPK, PI3K and mTOR growth and proliferation signalling. *Metallomics*. 2017;9(5):471-481.

The following abstracts have been withheld as they have been or will be published elsewhere, and/or due to intellectual property or confidentiality issues:

In vitro evaluation of buccal delivery of iron from an iron spray product

Aaya Al-Hadd and CM Heard
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales. UK

An investigation into the parallel application of multiple microfluidic droplet generators

Phoebe Crews-Rees, SA Coulman, CP Thomas and O Castell
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB Wales, UK

Understanding the risk of emerging bacterial resistance to topical antibiotics

Chelsea Evangelista and J-Y Maillard
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Assessing the long-term efficacy and protection of perineal care washcloths in Incontinence-Associated Dermatitis (IAD) in an *ex vivo* test.

India Rose Hayes and J-Y Maillard.
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Evaluation of a new Test and Treat pilot service in Wales from the perspective of pharmacists.

Ricky Hicks, EM Mantzourani and RE Deslandes
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3NB, Wales, UK

An evaluation of patients' perceptions of the pilot sore throat 'test and treat' service in community pharmacies in the Cwm Taf University health board

Laura Houldcroft and EM Mantzourani
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Evaluation of trans-buccal delivery of cannabidiol in vitro

Ella Jenkins and CM Heard
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Molecular modelling studies on novel potential pro-coagulant phospholipids

Calum Johnson and A Brancale
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Drain biofilms and their resistance to disinfection

John Juanro Lauran, K Ledwoch and J-Y Maillard.
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Design and Synthesis of Novel Potential Broad-Spectrum Inhibitors of Enteroviruses

Tahmidur Rahman, A Brancale and M Bassetto
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, Kind Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

The determination of the impact of commercial cleaning products on the transferability of *Staphylococcus aureus* dry surface biofilms

Dulcie Williams, J-Y Maillard and K Ledwoch
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Determining the views and opinions of end users and stakeholders relating to a new contraceptive patch for Sub-Saharan Africa

James Womersley and JC Birchall
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Design and synthesis of selective mitochondrial targeting drugs for use in the treatment of Parkinson's Disease

Matthew Zsibrita and Y Mehellou
Cardiff School of Sciences and Pharmaceutical Sciences, Cardiff University, King Edward XVII Avenue, Cardiff CF10 3NB, Wales, UK

Investigating the effects of statins as an anti-cancer agent in triple negative breast cancer

Shafila Alim and SE Hiscox
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

TNBC accounts for around 15–20% of all breast cancers diagnosed and despite an aggressive and toxic systemic chemotherapy, nearly all women will eventually die from this disease.

Statins have demonstrated several antitumor molecular mechanisms in experimental studies, such as inhibition of cell proliferation, promotion of apoptosis, promotion of tumour cell differentiation and modulation of the tumour microenvironment. The majority of TNBC tumours overexpress EGFR and it has been suggested that statins can interfere with the EGFR signalling pathway. The effects of Atorvastatin and Rosuvastatin were investigated on MDA-MB-231 and MCF7 cell lines. Cell proliferation was measured via a Ki67 assay and WST assay. A wounding assay investigated cell migration and cytoskeleton changes were examined via immunofluorescent staining for actin (phalloidin) and focal adhesions (vinculin). Western blot analysis allowed for preliminary investigations into the role of the EGF receptor signalling pathway. Our results indicated that the MCF7 cell line was not sensitive towards the statin agents. The assays concordantly showed that both statin agents supressed the proliferative and migratory capacity of the MDA-MB-231 cells, Atorvastatin being more effective than Rosuvastatin. There was a dose dependent reduction of MAPK and STAT3 phosphorylation in the Atorvastatin treated MDA-MB-231 cells. The EGFR proteins and MAPK became induced when treated with EGF and were then all shown to be supressed when Atorvastatin was added to the treatment. Our findings warrant further exploration into statins as a therapeutic treatment in triple negative breast cancer and suggest that the statins exert their effects by inhibiting the mevalonate pathway via a mechanism that suppresses the EGF receptor, via the MAPK and JAK/STAT signalling pathways; signifying the potential of these pathways as a much needed therapeutic target for triple negative breast cancer.

Nanoparticle Mediated Delivery of Herceptin

Joshua Bateman[1], J Wymant[1], A Amoroso[2], I Fallis[2] and AT Jones[1]
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [2]School of Chemistry, Cardiff University, Main Building, Park Place, Cardiff, CF10 3AT, Wales, UK

HER2 is a membrane bound receptor tyrosine kinase expressed in 20-30% of breast cancers. It is associated with overactivation of growth pathway signalling, leading to aberrant cell proliferation, migration, invasion and evasion of apoptosis. Numerous treatments targeted against HER2 have been developed, or are in development including the humanised monoclonal antibody, Herceptin, which is currently in clinical use. Efficacy of Herceptin has been questioned following studies showing that Herceptin only results in minimal HER2 downregulation. Recently however, a biotin/streptavidin system of crosslinking HER2 using Herceptin has shown to vastly increase HER2 internalisation through a HER2 clustering-mediated endocytosis mechanism. This project aims to take advantage of the clustering-induced endocytosis mechanism previously shown and apply it to a nanoparticle therapy system.

Superparamagnetic iron oxide nanoparticles were synthesised and loaded with cytotoxic doxorubicin and targeted to HER2 using Herceptin. Fluorescently labelled Herceptin and intrinsic doxorubicin fluorescence allowed analysis of intracellular trafficking of the nanoparticles by confocal microscopy. This confirmed an endocytosis mechanism for nanoconstruct internalisation in HER2 positive cells in vitro, whereas HER2 lacking cells were unable to internalise the nanoconstruct. Doxorubicin however was ineffective at entering the nucleus of the cell when applied in the nanoparticle system. Western blotting revealed a large decrease in HER2 expression in cells treated with the HER2-decorated nanoparticles, this was accompanied by a moderate decrease in HER3 levels and increased ERK activation. CellTiter-Blue assay however, indicated that the nanoconstructs were ineffective in inducing any significant change to cell viability.

This projected provides evidence that HER2-decorated iron oxide nanoparticles are able to be internalised by HER2 expressing breast cancer cells possibly through HER2 clustering-mediated endocytosis. Modification of the nanoconstruct structure is warranted to achieve optimum intracellular doxorubicin release and subsequent cytotoxicity and cell death.

The role of Rab5a in the expulsion anduptake of extracellular vesicles in prostate cancer cells

Joseph Butcher[1] and A Clayton[2]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [2]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Exosomes, a sub-population of extracellular vesicles (EVs), have a wide variety of roles in both normal and diseased cells. They have been implicated in disease progression in several cancers. Prostate cancer is the most common male cancer in the UK. Not much is known about the role of exosomes in the development and progression of prostate cancer, however their disease promoting effects in other cancers could indicate similar roles in prostate cancer. A recent study found that the knockdown of endocytic regulator Rab5a in Du145 prostate cancer cells lead to increased EVs, the aim of the present study was to confirm and investigate these findings.

Three variants of the Du145 cell line were used: one control, and two which had Rab5a knocked down by distinct shRNA sequences. The increase in EVs was confirmed by multiple EV detection methods. The impact of Rab5a knockdown on the endolysosomal system was examined by immunofluorescent staining of endocytic markers. Uptake of EVs was assayed by fluorescently labelling purified wild-type Du145 EVs and adding them to Rab5a knockdown and control cells, analysis was performed using flow cytometry and apotome microscopy. This study reports that there is some evidence that Rab5a deficiency leads to increased EVs, but the data shows this is not likely to be due to altered autocrine uptake of EVs. Redistribution of some late endosomal compartments to the cell periphery and restriction of endocytosed EVs to the cell periphery was seen in Rab5a knockdown cells, indicating that a disrupted EV phenotype does indeed arise following Rab5a perturbation.

Further studies of Rab5a's role in the mechanisms regulate EVs in prostate cancer could

potentially reveal novel targets for a therapeutic setting. However, these mechanisms remain very poorly understood, more studies will be necessary to clarify the impact of Rab5a on EV regulation.

Role of Caveolin-1 and Microglia in Glioblastoma Multiforme – 2D and 3D Models in Response to Temozolomide Treatment

Ka-Oi Cheung, C Neto and M Gumbleton
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Glioblastoma multiforme (GBM) is the most aggressive form of brain tumour in adults, with a median survival of 12-15 months post-diagnosis. Both the presence of microglia, brain-resident immune cells, and Caveolin-1 (Cav-1), a transmembrane scaffolding protein, correlates with worse tumour grade in GBM. Whilst microglia is thought to contribute to the highly invasive potential of GBM tumour cells through paracrine signalling, Cav-1 in GBM is thought to be tumour-suppressive through downregulation of intracellular signalling and promotes anti-tumoral phenotype in microglia. Therefore, the present study aims to evaluate how Cav-1 status effects microglial sensitivity to temozolomide (TMZ), the standard treatment for GBM. CHME3 Cav-1 NT and Cav-1 KO microglia were cultured into 2D monolayer and 3D spheroid models and exposed to increasing TMZ treatment for 72 hours. Viability assays and live/dead cell analysis were performed following treatment. Initial CHME3 growth curve showed CHME3 Cav-1 NT cells proliferated at a faster rate than Cav-1 KO cells. Viability assays using PrestoBlue reagent showed that IC50 of CHME3 Cav-1 KO cells was higher compared to Cav-1 NT cells in 2D experiments at 500μM, but there was no significant decrease in viability for 3D spheroid experiments. Flow cytometry analysis of 2D experiments and fluorescent microscopy using Calcein-AM and propidium iodide stains in 3D experiments showed no significant difference in live/dead cell percentage with increasing concentrations of TMZ, or between Cav-1 NT and KO models. Additional diameter analysis was used to determine a non-significant decrease in sphere size. 2D experiments were most sensitive to TMZ treatment but the decrease in viability to both 2D and 3D experiments may be due to cell senescence caused by TMZ. Positive Cav-1 status confers a higher sensitivity to TMZ whilst negative Cav-1 status leads to higher resistance to TMZ treatment.

TEAD1 in Endocrine Resistant Breast Cancer and its interplay with VGLL1

Lucy Davies[1], P Finlay[1], A Green[2], and JMW Gee[1]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK; [2]Nottingham Breast Cancer Research Centre, Division of Cancer and Stem Cells, School of Medicine, University of Nottingham

Anti-hormone resistance can be acquired by ER+ breast cancer resulting in patient relapse, and this is sometimes associated with ER loss. Recent research in Cardiff University detected Vestigial-Like 1 (VGLL1) mRNA upregulation in luminal A-derived breast cancer models that have acquired anti-hormone resistance and lost ER. VGLL1 is a coactivator for TEAD transcription factors (TEAD1-4) whose function is regulated by the Hippo pathway. This project aimed to shed further light on VGLL1/TEAD signalling in anti-hormone resistance by profiling TEAD1 in these resistant cell models and in clinical breast cancers.

Immunostaining assays were optimised to detect TEAD1 and VGLL1 co-expression in two of the acquired resistant models, and to profile TEAD1 in panels of luminal A and B-derived anti-hormone resistant cell pellets and a clinical breast cancer series (n=93). Staining was H-scored and for the clinical series analysed versus signalling and clinicopathological parameters. KM plotter evaluated any relationship between TEAD1 mRNA and relapse-free survival (RFS) using microarray datasets from endocrine-treated patients.

TEAD1 protein was significantly upregulated in several Luminal A acquired anti-hormone (particularly Faslodex) resistant models, associating with increased VGLL1 and ER loss. Immunocytochemistry and immunofluorescence revealed nuclear co-localisation of TEAD1 and VGLL1 in two anti-hormone resistant cell models with ER loss. TEAD1 staining was detected in tumour epithelial nuclei of many clinical breast cancers. While most prominent in small, well-differentiated tumours with functional ER, TEAD1 also significantly correlated with MAPK activity, erbB3 and erbB4 that have been implicated in endocrine resistance. Increased TEAD1 mRNA also associated with shortened RFS in luminal A endocrine-treated patients.

TEAD1's induction, interplay with VGLL1 and signalling associations provide compelling evidence for TEAD1/VGLL1 involvement in luminal A-derived anti-hormone resistance, notably where ER is lost. Further supported by its relation to clinical resistance, TEAD1 signalling could potentially provide novel therapeutic opportunities for this disease state.

Investigating zinc-mediated cell division in cancer

Ayaat Jaway and KM Taylor
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Zinc is an essential element that has several functions in cell growth, cell proliferation and many other processes. Zinc influx and zinc homeostasis is controlled by two families of zinc transporters and ZIP transporters are responsible for zinc entry into the cytoplasm from the extracellular space and from intracellular stores. ZIP10 is a zinc transporter which allows zinc to enter the cytoplasm from the extracellular environment and from intracellular stores. However, the exact mechanism of ZIP10 function is unknown. Currently, very little is known about whether ZIP10 is phosphorylated and the exact role of this phosphorylation in ZIP10 function. Using immunofluorescence and Western blotting analysis, experiments were performed to discover the involvement of any kinases in ZIP10 function. We have confirmed that ZIP10 is phosphorylated by CK2 and for the first time discovered that ZIP10 may also be phosphorylated by Src kinase, with the latter controlling ZIP10 activation. These findings suggest that ZIP10 may be relevant as a therapeutic target for patients with diseases such as breast cancer.

Characterisation of novel in-vivo models of metastatic breast cancer and the effect of Bcl-3 inhibitors on metastatic disease

Rebecca Jones[1] and R Clarkson[2]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [2]School of Biosciences, Cardiff University, The Sir Martin Evans Building, Museum Avenue, Cardiff, CF10 3AX, Wales, UK.

Bcl-3 is a proto-oncogene candidate that has been linked to several cancer types and has been shown to mediate metastasis in breast cancer. There is currently a clinical unmet need for agents that directly target Bcl-3, but the mechanism by which Bcl-3 exerts its metastatic effects has not yet been fully elucidated. There is a Bcl-3 inhibitor currently in preclinical development that has been shown to reduce the growth rate and size of some breast cancer patient-derived xenograft (PDX) tumours, but currently lacks any biomarkers for evaluating drug response. Additionally, the interactions of Bcl-3 are only recently being investigated, with potential links to cell cycle and motility genes being explored as well as the potential effect these interactions could have on cell phenotype. During this project, immunohistochemistry assays were performed on a panel of 8 PDX model tumours to evaluate the practicality of using cleaved capsase 3 (CC3) and phospho-histone H3 (PHH3) as response biomarkers of CB1 treatment, as well as to assess each individual tumour to examine CB1 response compared to DMSO-treated controls. Additionally, PCR analysis was performed on a panel of 7 Rho-GTPase family genes in Bcl-3 knockdown cell lines, to analyse how the expression level of each gene is affected by a lack of Bcl-3. Immunofluorescence assays were performed on Bcl-3 and CDC42 knockdown cell lines to analyse the effect such knockdowns would have on cell phenotype. For immunohistochemistry and immunofluorescence, results were as a whole insignificant, with biological variability too high with too small a number of replicates to be able to reveal any statistically significant results, but it has been confirmed that a Bcl-3 knockdown does affect several of the target genes significantly in several cell lines, strengthening evidence for a role for Bcl-3 in cell motility and migration, and thus cancer metastasis.

Exploring the factors that affect CRISPR/Cas9 genome editing

Vasiliki Karkalousou and S Reed
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales,and [2]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

The CRISPR/Cas9 system is a versatile tool for genome editing in human cells. Cas9 is a programmable nuclease, guided to specific locations in the genome by a sgRNA designed to complement the sequence of interest (on-target). Once bound to it, Cas9 creates chromosomal DSBs that are repaired by the host repair machinery. The outcome of this process defines the functionality of genome editing. However, the off-target cleavage events and the unpredictability of the repair outcomes constitute limitations for the clinical implementation of the system.

Here, we provided a detailed analysis of the characteristics of CRISPR/Cas9 genome editing (efficiency, specificity, repair outcomes), while exploring the factors that influence them. Four CRISPR/Cas9 systems, targeted to different sites in the genome of HEK293T cells, were constructed and delivered as either plasmids or ribonucleoprotein complexes. Targets were PCR-amplified and Sanger-sequenced. ICE analysis provided the indel mutations formed, following error-prone repair of the DNA damage. Bioinformatics analysis revealed the genomic context of the targets.

The editing efficiency was found significantly altered between the four systems and the delivery methods used. All systems induced off-target cleavages, with some off-targets being mutagenized in comparable levels with the on-targets. Indel distribution spectra differed between the on-targets, and between the on- and off-targets that were edited by the same system, indicating that repair outcomes are independent of the targeted sequence. Genomic context was profoundly altered between the on- and off-targets. Our results suggest that CRISPR/Cas9 genome editing is more efficient when the system is delivered as a ribonucleoprotein complex and when the target has a biased nucleotide composition. Specificity might be influenced by the kinetics of Cas9 binding to the off-targets, while repair outcomes may be solely dependent on the genomic context of the targets. Conclusively, the discovery of the factors that influence CRISPR/Cas9 activity could guarantee that only the most efficient and specific systems, that have predictable repair outcomes, are used safely for genome editing, especially in clinical settings.

Could oxygen -releasing biomaterial play a future role in Glioblastoma Multiforme therapy?

Kitti Kovacs and B Newland
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Glioblastoma (GBM) is the most common primary brain tumour, and the median survival rate is 15 months. Standard treatment includes surgical resection and Temozolomide chemotherapy agent, and radiotherapy. However, this implied limited prognosis is partly due to treatment resistance and high rate of reoccurrence. Hypoxia, which is in oxygen-depleted state, contributes to many aspects that causes GBM to be highly proliferative, infiltrative, and having high capacity for neovascularity. Dysfunctional blood vessels enhance the development of hypoxic areas, which is partly responsible for stem cell maintenance, leading to intratumoural molecular heterogeneity and altered cell cycle in response to damage. Radiotherapy aims to damage the cells through direct and indirect effects, and both mechanisms are affected by the amount of molecular oxygen available. Several oxygen-generating biomaterials have been developed to increase oxygenation in tumours, however, none has made it to clinical trials yet, due to either being in early phase or due to unsatisfactory results. This review summarizes the influence hypoxia has over the cell cycle, stem cells, molecular pathways, that may lead to radioresistance of GBM, and includes the potential solution of oxygen-generating materials and their effectiveness on the 'treatment' of hypoxia.

Investigating the mechanism of zinc-mediated cell division in cancer

Kelsey Maidment and KM Taylor
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Zinc is an essential trace element identified to be involved in a variety of signalling pathways that are implicated in health and diseases such as cancer. Zinc can act either as a structural component of functioning macromolecules or as a second messenger, influencing multiple downstream signalling pathways. Both excessive and deficient levels of zinc can be detrimental to cellular health, which highlights the need for tight zinc regulation. An important component of zinc homeostasis is the LIV-1 family of zinc transporters (known as SLC39A and ZIP transporters) which function to increase cytosolic free zinc. Interestingly, some zinc transporters have previously been associated with breast cancer progression, although the mechanism is unknown. This study aims to investigate the effects of inhibiting the zinc influx into cells employing the use of kinase inhibitors in MCF7 breast cancer cells and testing the effects using immunofluorescence and western blot techniques. Immunofluorescence confirmed the presence of increased zinc transporters on the plasma membrane of cells. Several potential phosphorylating protein kinases were identified by computer analysis to define which inhibitors were used. Results of this study identified the involvement of serine/threonine phosphorylation of CK2 as an important protein kinase in the activation of zinc-mediated cell division. Additionally, for the first time, evidence of involvement of tyrosine phosphorylation was discovered in the ZIP transporter mechanism of cleavage and relocation to the plasma membrane. These preliminary findings are interesting but require further investigation to identify the exact mechanisms of action and any potential role as therapeutic targets.

Detection of hypoxic exposure in prostate cancer cells and implications for the functional stratification of in vivo systems

Oliver Smart[1], M Wiltshire[2] and R Errington[2]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales,and [2]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Tumour hypoxia is associated with poorer prognosis, and prostate cancers tend to be extremely hypoxic with a mean oxygenation of only 0.32%. This project sought to investigate the capability of detecting hypoxic burden in three prostate cancer cell lines (DU145, PC3, and LNCaP) using the fluorescent probe HypoxiTrak, and to establish the effect of hypoxia on prostate cancer cell behaviour.

Cells were treated with HypoxiTrak at 30 or 100nM for 48 or 96 hours in normoxia and 1%O2 and probe uptake and activation assessed using flow cytometry. The effect of hypoxia on etoposide sensitivity was assessed by treating cells for 48 hours in normoxia and hypoxia, viability measured using DRAQ7 and cell cycle assessed with propidium iodide. Cells were maintained in hypoxia for 8 weeks, and a subpopulation transferred to normoxia after 6-weeks to assess hypoxic recovery. Doubling time was calculated every week.

Uptake and activation of HypoxiTrak varied across the three cell lines, with significant detection of hypoxia in LNCaP (1.77-fold change in fluorescence, $p<0.05$ at 96 hours), but minimal increased fluorescence with PC3s (1.50-fold change, $p>0.05$ at 96 hours). Hypoxia significantly altered the sensitivity of PC3s to etoposide by reducing the capacity to enter polyploidy (35% in normoxia vs. 13.6% in hypoxia, 1000nM, $p<0.05$). DU145 and PC3 cells cultured in hypoxia for 8 weeks were initially slower to divide (DU145: 64.8 hours vs. 25.3; PC3: 52.3 vs. 35.7) but recovered to match the rate of growth in normoxia; LNCaP doubled faster in hypoxia (42.9 hours vs. 58.2) and slowed immediately when transferred back into normoxia.

Overall, further optimisation is required to develop a hypoxia detection assay suitable for all three cell lines or in heterogenous prostate cancer environments, and our results highlight the need for regularly monitoring tumour hypoxia in vivo to properly understand the ongoing changes in tumour behaviour.

The following abstracts have been withheld as they have been or will be published elsewhere, and/or due to intellectual property or confidentiality issues:

In vivo distribution of FGFR1 and FGFR2 expression in glioblastoma and their relationship with stem-like cancer cells

Nawal Shaya Alshahrany[1] and F Siebzehnrubl[2]

1Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and 2School of Biosciences, Cardiff University, The Sir Martin Evans Building, Museum Avenue, Cardiff, CF10 3AX, Wales, UK

Mechanistic importance and therapeutic significance of EPLIN in cancer progression

Benedict Lewis[1] and A Sanders[2]

[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [2]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

The potential interplay between OPG and HGF in prostate cancer metastasis

Ieuan Luc Morgan[1], A Sanders[2] and Sioned Owen[2]

[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales,and [2]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

EXPLORING THE ROLE OF ZEB1 IN HAEMATOPOIESIS

Elis Storch[1] and N Rodrigues[2]

[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [2]European Cancer Stem Cell Research Institute, Cardiff School of Biosciences, Hadyn Ellis Building, Maindy Road, Cardiff, CF24 4HQ

A novel Gata-2 pharmacological inhibitor to therapeutically target leukemic stem cells in acute myeloid leukaemia

Isobel Westcott[1] and N Rodrigues[2]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales UK and [2]European Cancer Stem Cell Research Institute, Cardiff School of Biosciences, Hadyn Ellis Building, Maindy Road, Cardiff, CF24 4HQ

Microneedle delivery of antigen-specific immunotherapy for Type 1 diabetes

[1]Farah Arikat, [1]SA Coulman, [2]S Wong, [2]C Dayan, [2]S Hanna and [1]JC Birchall
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [2]School of Medicine, Cardiff University, UHW Main Building, Heath Park, Cardiff CF14 4XN, Wales, UK

Antigen-specific immunotherapy (ASI) involves induction of tolerance to autoantigens. An important protein in the development of type 1 diabetes (T1D) is the autoantigen, proinsulin (PI), the precursor of insulin. Microneedles (MNs) are micron-sized needles that penetrate into the upper skin layers. MNs provide advantages for autoantigen delivery including targeted delivery to the skin's dendritic cells (DCs), with minimal inflammation. The aim of this Thesis was to develop a PI-coated solid MN system and investigate the potential of this system to induce peripheral tolerance in the non-obese diabetic (NOD) mouse model of T1D. A highly concentrated PI MN coating formulation was developed containing the PI, diluent and a surfactant. The formulation enabled uniform and reproducible coating of the PI on to MNs. Delivery of PI from the MN system was investigated in mouse skin. MN application method and duration were optimised and resulted in skin puncture and reproducible delivery of PI to the skin. In vitro studies identified the insulin-reactive G9 CD8+ T cell as an appropriate biological readout for PI delivery. In vivo delivery studies indicated that MNdelivered PI was delivered to the skin and subsequently processed by DCs into PI peptides, which were cross-presented in the skin draining lymph nodes to adoptively transferred G9 CD8+ T cells. This demonstrated that the PI-coated MN system has potential for inducing peripheral tolerance in the NOD mouse. T1D development was significantly delayed in NOD SCID mice that received cells from PI-treated NOD mice and cells from diabetic NOD mice (experimental group). However, no statistically significant difference in time to T1D development was observed between the experimental group and the control NOD SCID mice that received cells from untreated NOD mice and diabetic NOD mice. Further investigation of the dosage and dosing frequency of PI using the coated MN system is, therefore, warranted.

An exploratory study of pharmacy graduate preparedness for preregistration training

Bethan Broad, SA Coulman, ML Hughes, DN John
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Major changes to the role of the pharmacist in the past decade include an increased prevalence of pharmacist independent prescribing and a shift towards multi-disciplinary working. In 2011 the GPhC accreditation criteria for schools of pharmacy also changed significantly, moving to a more outcomes based education. Little is known about the preparedness of post-2011 graduates for modern practice in both hospital and community pharmacy settings. This study aimed to explore perceptions of this. Semi-structured one-to-one interviews were conducted to determine current perceptions of graduate preparedness for pre-registration training, what schools of pharmacy do well and areas for improvement. Fourteen members of academic staff (including teacher practitioners) from the Cardiff School of Pharmacy and Pharmaceutical Sciences, twenty-five employers (individuals involved in the supervision/training of pre-registration trainees) from hospital and community pharmacies, and seventeen recent pharmacy graduates from both hospital and community pre-registration training programmes were interviewed. A range of themes and subthemes were created through thematic analysis. The time between graduation and the early weeks of pre-registration training was identified as an important period in the transition from student to healthcare professional. This transition was eased by a graduate's prior exposure to the workplace (specific training site and more generally). All three stakeholder groups were supportive of enhancing spirality in MPharm curricula such that material learnt at university may be contextualised in pharmacy experiential placements. Students' exposure to patients improved their confidence and communication skills whilst their interactions with pharmacist role models informed their expectations of practice. While stakeholders perceive graduates to have appropriate knowledge, their ability to apply this may be improved, suggesting post-2011 graduates are not as prepared for pre-registration training as they could be. The need for enhanced student exposure to practice, patients and professions as part of the undergraduate degree has been identified.

Patient and public involvement in the benefit risk assessment of medicines: developing a semi quantitative framework to incorporate patient views as key criteria in decision making

Paul I. Cross and RDE Sewell
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

During the lifecycle of a drug, evidence must demonstrate that the benefits of the product continue to outweigh the risks. Benefit-risk (B-R) assessment is therefore a vital stage of the drug approval process and is an important task for regulators. Involving patients in B-R assessment is a recent development and they may view benefits and risks very differently when compared with the views of pharmaceutical companies or regulatory assessors. The aim of this research was to investigate this topic to ultimately propose a framework for involving patients in this process. The research strategy involved three phases. In phase I, a survey was submitted to (1) pharmaceutical companies and regulatory agencies and (2) patient advocacy groups across Europe, in order to obtain their opinions on involving patients in B-R assessment. Phase I of the research identified several challenges, including: how to ensure adequate patient representation as well as an absence of any established method. It was also identified that to date, only patient advocacy groups were directly involved in B-R assessment discussions. However, some companies were developing initiatives to involve patients in this process. Based on these findings, phase II was implemented, whereby individuals from regulatory agencies, pharmaceutical companies and patient advocacy groups participated in semi-structured interviews to identify themes around patient involvement and to establish valuable data regarding the current challenges. This data was then used to inform the development of a novel framework, one element of which was tested in phase III of the research; where qualitative focus groups were conducted with patients. The framework proposed from this research, consequently involved qualitative focus groups, enabling patients to provide insight into their disease and treatment. The information obtained, when presented alongside quantitative preference elicitation data, may then be used to contribute to B-R discussions by regulators, to ultimately support their decision-making.

When 'I' is replaced by 'we', even 'illness' becomes 'wellness': Exploring pharmacists' interprofessional practice to better prepare pharmacy students for interprofessional collaborative working

Andrew Jenkins, ML Hughes, E Mantzourani and MW Smith
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

The drive to increase interprofessional teamwork in the healthcare environment has gained significant traction in recent years. This has partly been as a consequence of UK inquiries that have cited breakdowns in communication and teamwork as contributory factors leading to poor patient outcomes. One method to prepare practitioners for interprofessional teamworking is interprofessional education (IPE). The General Pharmaceutical Council specifies that IPE must be embedded within UK Master of Pharmacy (MPharm) programmes. However, there is a paucity of literature examining IPE related to pharmacy and limited knowledge of pharmacists' interprofessional interactions with healthcare professionals (HCPs). This makes it challenging for pharmacy educators to design IPE sessions that are reflective of practice. To address this, a mapping process was undertaken to identify IPE sessions that are delivered in UK MPharm programmes (17/29 schools responded). This identified significant variation in IPE sessions delivered in terms of learning outcomes addressed, topics covered, and the range of student HCPs involved. A mixed method study was then undertaken to explore pharmacists' interprofessional interactions in practice. A questionnaire was disseminated to pharmacists in Wales via community pharmacies (61.9% response) and hospital pharmacy departments (estimated 59.1% response). Analysis of returned questionnaires identified that although the extent of interprofessional collaboration varied pharmacists in both sectors most frequently interact with doctors and nurses. Semi-structured interviews were undertaken with pharmacists from both the community (n=14) and hospital (n=15) sectors to explore the nature of interactions. Using deductive and inductive thematic analysis, the nature of pharmacists' interactions with HCPs was elucidated, facilitators and barriers to interactions were determined and suggestions for IPE developed. Findings from these studies resulted in a series of recommendations for pharmacy educators and policy makers to facilitate pharmacists' interprofessional collaboration in practice and aid the development of relevant IPE that is of value to learners.

The stability of new generation intravenous lipid emulsions

Helen King[1], AG Cosslett[1], CP Thomas[1] and R Price-Davies[2]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK; [2]Simbec Research Ltd, Merthyr Tydfil Industrial Park, Merthyr Rd, Pentrebach, Merthyr Tydfil CF48 4DR.

Intravenous lipid emulsions (IVLE's) form a staple part of parenteral nutrition (PN). PN provides life sustaining support where gastrointestinal nutrition is inadequate due to disease or prematurity. Whilst the physical stability of IVLE's is relatively well known and quantified, chemical stability is an area where little testing has occurred. Lipids are susceptible to breakdown through free radical attack leading to lipid peroxidation, a cyclical process resulting in the production of primary and secondary toxic lipid peroxidation products. This thesis presents the development and validation of a method for measurement of peroxidation and triglyceride (TAG) breakdown occurring within two IVLEs. The high-performance liquid chromatography (HPLC) method developed uses in-line ultra-violet (UV) and charged aerosol detection (CAD) to monitor the six main TAGs in Intralipid® and 10 TAGs in SMOFlipid® and detects the toxic secondary peroxidation products 4-Hydroxynonenal (HNE) and Hydroxyundecenal (HUE). The assay was validated in line and employed to test the chemical stability the well established lipid emulsion (Intralipid®) and a newer lipid emulsion (SMOFlipid®).
Both lipids were subject to up to 84 days storage within 50 ml syringes, 250 ml PN bags and 50 ml glass vials at room and fridge temperatures. The effect of light exposure was tested using light protected and non-light protected samples of each lipid. Results detail the extensive levels of TAG losses observed within each container and the detection of secondary peroxidation products. Fridge temperature limited TAG loss and peroxidation in all containers, however secondary peroxidation products were detected. Both SMOFlipid® and Intralipid® gave in excess of 30 % losses in TAGs over 84 days storage. HNE, HUE and a triglyceride remnant were all recorded in SMOFlipid® and Intralipid® syringes (both temperatures) and small volume PN bags at room temperature. Light protection within this study showed no significant difference vs non-light protection. The results obtained from the work within this thesis are of vital importance when considering the safety of IVLEs for intravenous nutrition. This work provides an initial data set on the levels of peroxidation occurring within two commercially available in-use IVLEs and highlights the necessity for the stability and storage limits of these emulsions to be re-assessed.

Eicosanoid and cytokine responses to bacterial infection

Shayda Rose Maleki-Toyserkani, L Baillie, CP Thomas and EJ Kidd
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Infectious diseases remain some of the most serious health threats facing the world. The immune system is equipped to initiate a rapid and specific response to foreign invaders of the body, with its ultimate aim being to protect an organism from injury and disease. Eicosanoids, including prostaglandins and leukotrienes, are a family of lipids that play key roles in inflammation including helping leukocytes fight infection. Cells of the innate immune system including tissue macrophages, neutrophils and sentinel dendritic cells are major contributors of local eicosanoids. In mammals an inflammatory insult will result in a cytokine cascade whereby tumour necrosis factor α (TNF-α) is released, followed by interleukin-1β (IL-1β) and then IL-6. Downstream of these cytokines, others are released that serve as potent chemoattractants to induce migration of neutrophils and macrophages to the site of infection. It is known that exposure to varying bacterial components results in a different profile of lipids and cytokines, and by characterising mediator signals it may be possible to define biomarker fingerprints predictive for early bacterial infections. To analyse this, a combination of a targeted lipidomic approach and cytokine immunoassays were employed to identify neutrophil and macrophage responses to individual bacterial components and the whole organism. Work in this thesis has identified potential markers of bacterial infection, such as 12- HETE, 14-HDOHE and TNF-α, which, along with future advances, could be used to develop novel strategies for clinicians, nurses and primary care staff to analyse patients suspected of bacterial infection at the bedside. Work here provides an insight into how the eicosanoid and cytokine storms are generated alongside each other to accompany classic inflammation during specific bacterial infection. The ability to distinguish between species of bacteria causing infection could prove invaluable, reducing the time taken to establish the cause of infection, ultimately leading to better patient outcomes.

Disruption of bacterial spores using microwaves and nanoparticles

Dimitri Malyshev, [2]A Porch and [1]L Baillie
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK and [2]School of Engineering, Cardiff University, Queen's Buildings, The Parade, Cardiff, CF24 3AA, Wales, UK

This thesis shows how microwaves and nanoparticles can be used to enhance the disruption of spores in the context of a novel microwave-based bacterial detector. Infections linked to Clostridium difficile are a significant cause of suffering. In hospitals, the organism is primarily acquired through the faecal-oral route as spores excreted by infected patients contaminate the healthcare environment. Microwave-based spore disruption is the focus of this project offers a potential rapid diagnostic method to detect spores, including C. difficile spores by making them release DNA which is then detected. One of the limiting factors of this method was the power required and it was hypothesised that by using a new microwave cavity and adding nanoparticles, the DNA release from spores can be achieved with less power. First, the spore surface properties of the different isolates of C. difficile were compared. A significant variation in both spore morphology and spore hydrophobicity of clinical isolates of C. difficile was observed. In particular, the “pineapple-like” shape of strains was associated with higher hydrophobicity in spores, while the loose outer exosporium layer was associated with lower hydrophobicity. The isolates were then tested for DNA release in response to microwaving. Spores were shown to release single-stranded, but no double stranded DNA. The spores were also not visibly changed by microwave exposure, suggesting a non-destructive mechanism of disruption. To study this mechanism further, the microwave system was updated to remove overall sample heating and tested under electric fields, magnetic fields and a combination of the two. The electric field was shown to be causing the disruption of spores, field showed a positive correlation with increasing disruption. Finally, the spores with tested with microwaves and nanoparticles, where nanoparticles showed a significant improvement in two of the four tested isolates. A computer model of the spore and nanoparticle interactions was made, which offered a plausible mechanism for the nature of microwave mediated disruption and the improvement in disruption caused by nanoparticles.

Caveolin-1: a mediator of Glioblastoma cell invasion and an independent negative biomarker of Glioblastoma patient survival

[1]Chiara Moriconi, [2]G Pilkington and [1]M Gumbleton
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, U.K. and [2]Brain Tumour Research Centre, University of Portsmouth, St Michael's Building, White Swan Road, Portsmouth, PO1 2UP, U.K.

Glioblastoma multiforme (GBM) is a malignant and highly aggressive form of brain tumour, with extremely poor prognosis. One of its features is the ability of the tumour to invade through normal brain resulting in tumour relapse. Our hypothesis was that Caveolin-1 (Cav-1), a major component of the caveolae and recognized to be involved in a number of signalling pathways, has a key pro-invasive role in GBM. We pursued our hypothesis by inhibiting the expression of Cav-1 in different adult GBM cell lines using different genetic techniques (liposome shRNA, lentiviral shRNA and CRISPR). We found that Cav-1 drives clonogenicity (CHAPTER 3) and invasion in a combination of two- and three-dimensional models (CHAPTER 5). We focused our research on the invasion phenomenon and, in order to provide a robust quantification approach to study invasion in 3D spheroid assays, we developed (CHAPTER 4) a open-source semi-automated script, INSIDIA, available for all researchers in the community to use. This tool was used to quantify the impact of Cav-1 on invasive capacity. In in-vitro systems, we explored the impact of Cav-1 expression upon molecules associated with the invasion phenomenon (CHAPTER 5). We found Cav-1 to be associated with CTSB, MMP1 and UPA and receptors like UPAR and CD44, as well as AKT activation. Interrogating the “The Cancer Genome Atlas” (TCGA) database, we confirmed that Cav-1 is an independent biomarker of poor prognosis in GBM patients (CHAPTER 6). This clinical data also found association of genes that may cooperate with Cav-1, including CD44, ITGA3, VIM, CTSB, CTSL, TSP-1, TIMP1 and MT1MMP. Collectively this thesis provides strong in vitro and clinical data supporting that Cav-1 as a key molecule promoting GBM invasion, and further identifying Cav-1 as a potential drug discovery target in GBM.

Molecular mechanism of highly potent NS5A inhibitors.

Elizabeth M Navarro Garcia and A Brancale
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Hepatitis C is responsible for causing chronic infections in over 170 million people all over the world who are at a risk of developing into liver cirrhosis and hepatocellular carcinoma, locating HCV in a major public health burden. Until recently, the standard-of care treatment consisted of Interferon-alpha and ribavirin, in addition to non-structural protein 3/4 (NS3) protease inhibitors, but due to the undesired side-effects, researchers developed more efficient therapies. Nowadays, small molecules targeting non-structural viral proteins: NS3/4 protease, NS5A D1 and NS5B polymerase activities can clear the infection in 98% of the cases. These direct acting antivirals (DAAs) are widely used, however, despite advances in recently approved potent DAAs the world-wide application of these therapies remains limited due to the expensive cost and potential drug resistance. NS5A is a nonstructural multifunctional protein. Mainly composed by an amphipatic helix, which is the major membrane anchor, Domain I, which is involved in RNA binding and assembly, and Domain II and III which are intrinsically unfolded domains and are known to interact with host factors. DAA targeting NS5A DI, Daclatasvir (DCV), has a picomolar range activity and it is used in combination therapy to combat HCV infection. Given the enormous medical relevance of NS5A inhibitors, the aim of this study was to decipher the mode of action of Daclatasvir, together with more insights to the role of NS5A structural elements. In the present study, experiments showed that DCV can block the envelopment of viral particles. Furthermore, targeting the assembly of HCV particles, this fact serve as evidence of the dual mode of action of DCV. Furthermore, we investigated the role of very conserved Proline residues in the structure of NS5A, identifying key Proline residues which are critically involved in RNA replication, and have an impact in HCV infection. This fact, also suggests that the some of these Prolines might be essential for the DCV binding, as we prove that they have a direct role in keeping the binding site of DCV. Lastly, we set up a molecular model which includes the intracellular membrane giving the full picture of how DCV works in the context of an intracellular membrane and its important interactions. Together our data, prove the dual mode of action of DCV targeting HCV replication and assembly. And importantly, we constructed a molecular model that can be use in the future to study structure-function of developing NS5A inhibitors.

Preventing nano and micro wear-particle induced inflammation

Melissa Rodrigues and P Prokopovich
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Aseptic loosening, as a consequence of an extended inflammatory reaction induced by wear particles, remains the most common complication of total joint replacement (TJR), representing a major problem for the long-term success and survival of prostheses. Despite it is high incidence, in the last decade any therapeutic approach has been found to treat or avoid aseptic loosening, leaving revision as the only effective treatment for this condition. The local delivery of anti-inflammatory drugs to modulate wear-induced inflammation has been regarded as a potential therapeutic approach to avoid aseptic loosening. In this work, an anti-inflammatory drug-eluting implant model system was developed and characterised. The model system was obtained by attaching DEX to functionalised-TiO2 particles, through different synthetic routes: i) by covalently binding DEX to carboxyl-functionalised particles (amino or mercapto routes) or ii) by coating amino-functionalised particles using Layerby- Layer (LbL) technique. The chemical and physical properties of DEXloaded functionalised TiO2 particles have been determined and the release profiles investigated. Depending on the synthetic route, the DEX release period can vary from hours (amino, mercapto routes) to 3 weeks (LbL route). The model system was then tested for its cytotoxic and anti-inflammatory properties in a rapid and reproducible in vitro mouse macrophage-like cellular model, by utilizing murine RAW 264.7 cells. In this model lipopolysaccharide (LPS) was utilized to activate the Raw macrophages, resulting in the secretion of pro-inflammatory cytokines, including nitric oxide (NO) and tumour necrosis factor alpha (TNF-α), the suppression of which was utilized to investigate the anti-inflammatory effect of DEX released from functionalised-TiO2 particles. In vitro studies showed that DEX decreased LPS-induced NO and TNF-α production at non-cytotoxic concentrations, where DEX released from LbL particles showed the most effective suppression of inflammation for at least 2 weeks. Collectively, these findings show that the model system developed can be a potential therapeutic approach to avoid wear-debris induced aseptic loosening of TJR.

Understanding how targeting zinc transporters prevents the development of aggressive cancer

Silvia Zilliotto and KM Taylor
Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff University, King Edward VII Avenue, Cardiff CF10 3NB, Wales, UK

Zinc is one of the most abundant trace elements in the human body. Cellular zinc homeostasis is primarily controlled by zinc transporters, including the ZIP family of zinc importers. Since zinc homeostasis needs to be tightly controlled, dysregulation of these zinc transporters is associated with multiple diseases including cancer. ZIP7, a zinc transporter residing on the endoplasmic reticulum membrane, was discovered to be involved in driving endocrine resistant breast cancer. Findings within this project support the hypothesis that tamoxifen-resistant breast cancer cells are driven by the increased activation of ZIP7 which drives the invasive behaviour of this more aggressive breast cancer phenotype. This study confirmed the suitability of activated ZIP7 as a good biomarker of acquired resistance to anti-hormone treatment in breast cancer, a current clinical unmet need. Zinc is also important in cell cycle progression and, in particular, is essential for progression of cells through the G2 phase and mitosis. Our group have discovered a role for zinc transporters in the process of mitosis. This study expanded this discovery and demonstrated that blocking the specific zinc transporters with unique agents could inhibit mitosis. These agents were also shown to be effective at reducing the growth of different cancer cell lines. This study revealed novel targets for proliferative diseases such as cancer, which is manifested by uncontrolled growth.

INDEX OF AUTHORS