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Pharmacokinetic and pharmacodynamic modelling of PR-104: dissecting the "bystander effect" of hypoxia-activated metabolites

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Introduction. Hypoxia is a unique feature in solid tumours and is therefore a potentially exploitable therapeutic target. PR-104, currently in clinical trial, is a phosphate ester that is rapidly converted to its alcohol prodrug, PR-104A. This prodrug is activated by reduction to hydroxylamine (PR-104H) and amine (PR-104M) metabolites, both in hypoxic cells and independent of hypoxia by aldo-keto reductase (AKR) 1C3. However, it is unknown whether the high single-agent activity of PR-104 in some human xenografts reflects a "bystander effect", due to metabolite diffusion from hypoxic zones, or metabolism by AKR1C3.
Aims. To understand the contributions of hypoxia-activated bystander killing and oxygen-independent metabolism in the anti-tumour activity of PR-104.
Methods. Initially, a population model was developed to describe the plasma pharmacokinetics (PK) of PR-104 and PR-104A in rodents, dogs and humans. The pharmacodynamics (PD) of PR-104A and PR-104H was then measured in tissue culture studies, and their extravascular transport investigated using an *in vitro* multicellular layer (MCL) model. A spatially resolved (SR) model was then developed to predict the PK and PD (cell kill) at each position in a three-dimensional tumour microvascular network.
Results. Population PK analysis estimated rapid conversion of PR-104 to PR-104A, with a faster clearance of PR-104A in dogs and humans than in rodents. In SiHa cervical carcinoma cells, the Area under the concentration-time curve (AUC) was identified as the key exposure variable that correlates with clonogenic cell killing. The diffusion coefficient of PR-104H was similar to that for PR-104A, but its lower metabolic stability gave a calculated diffusion half distance of 50 μm in tissue. SR-PK/PD simulations showed that the monotherapy activity of PR-104 occurs via 3 pathways: hypoxia-activated bystander killing, hypoxia-independent activation and circulating metabolites.
Discussion. While the current SR-PK/PD model under-predicted measured activity in SiHa xenografts, an improved model is under development to further our investigation of the relative importance of bystander cell killing.

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Metabolic drug activation in rheumatoid arthritis and drug toxicity

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Introduction. Conventional Disease Modifying Anti Rheumatic Drugs (DMARDs) are the cornerstone of treatment for early rheumatoid arthritis. These drugs tend to have multiple mechanisms of action with respect to both efficacy and toxicity, and metabolic activation is often an important step in achieving both therapeutic and toxic effects.
Aims. To provide an overview of the role that metabolic drug activation plays in the toxic effects of sulfasalazine and leflunomide, including identification of factors that may be used to identify patients that are more likely to cease these drugs due to toxicity.
Methods. Patients who were included in the Royal Adelaide Hospital Early Arthritis inception cohort were included in this retrospective analysis. Patients were treated according to a structured treatment algorithm with a 'treat to target' approach. Information regarding patients' demographic, pathological and genetic variables was collated, and the effect of genetic markers on the rate of cessation due to toxicity with sulfasalazine and leflunomide was determined by a Cox proportional Hazard model.
Results. Cessation due to toxicity was more likely amongst users of leflunomide who were CYP2C19 poor metabolisers (i.e. carried one or two *CYP2C19* loss-of-function alleles and no gain-of function alleles). Likewise, NAT2 slow metabolisers were more likely to cease sulfasalazine due to toxicity. *ABCG2* genotype did not appear to influence cessation of sulfasalazine or leflunomide due to toxicity.
Discussion. Genetic variables in pathways involved in metabolic drug activation may be able to predict individuals who are more likely to cease sulfasalazine and leflunomide due to toxicity, but further work must be conducted so that this information can be used to personalise treatment with conventional DMARDs and improve outcomes for patients with rheumatoid arthritis.

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Structure activity studies and therapeutic potential of venom peptides that target acid-sensing ion channels.

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INTRODUCTION: A marked decrease in pH, or acidosis, is often associated with painful pathological conditions such as inflammation, ischemia, trauma, infection, and malignant tumours (Deval et al, 2011). Furthermore, central nervous system acidosis is a key mechanism of neurodegeneration (in stroke and multiple sclerosis) and seizures (in epilepsy). Acid-sensing ion channels (ASICs) are activated by the drops in pH reached during acidosis (~pH 6-6.5). They are the primary neuronal proton sensors in mammals and play central roles in pain perception and in mediating neurodegeneration that follows acidosis. Inhibition of ASICs has been shown to be analgesic in rodents (1a and 3) and humans (1a)(Deval et al, 2011), and neuroprotective in models of stroke (Pignataro et al, 2007) and multiple sclerosis (Vergo et al, 2011) with minimal side effects. Therefore potent and selective ASIC modulators are very attractive as potential broad range and safe therapeutic leads for pain and neurodegeneration. AIM: To give an overview of the therapeutic potential of peptidic ASIC modulators and the work we are doing to understand their binding sites and the molecular basis of their interaction with these sites and their mechanism of action. METHODS: We used a combination of chemical synthesis and recombinant expression in *E. coli*, NMR, two-electrode voltage-clamping of *Xenopus* oocytes and *in silico* approaches such as restraint based docking using HADDOCK. RESULTS/DISCUSSION: We have successfully determined new structures and the pharmacophores of two ASIC modulating peptides, PcTx1 (ASIC1a) and APETx2 (ASIC3) both of which are in pre-clinical development. Furthermore, we have discovered the most potent blocker of ASIC1a to date and are carrying out work on its structure, mechanism of action and therapeutic potential.

Deval E et al (2011) Pharmacol Ther 128:549-558
Pignataro G et al (2007) Brain 130:151-158
Vergo S et al (2011) Brain 134:571-584

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Consequences of human TRPV4 mutations: implications for drug targeting and disease

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Introduction. The TRPV4 cation channel, is widely expressed in mammalian tissues and is the subject of intense study to determine if it has therapeutic potential in a range of diseases. TRP ion channels have diverse roles in sensing environmental stimuli either directly (e.g. TRPV1 and TRPA1), or indirectly like *Drosphila* TRP, which responds to phospholipase C signalling from light-activated rhodopsin. TRPV4 is activated by osmotic swelling, shear stress, temperature and by agonists, but the physiological mechanisms of its activation are not well understood. Recently, we and others identified point mutations of TRPV4 in humans which produce 3 distinct phenotypes in affected individuals affecting nerves, bone or joint development (see Lamandé et al 2011 for details). We may be able to exploit distinct tissue-specific TRPV4 modulation for new therapies.
Aims. Test activation of TRPV4 by activating endogenous GPCRs or by applying hypotonic solutions in HEK293 cells. Use signalling inhibitors and activators on wildtype and mutant TRPV4 constructs to investigate signalling mechanisms involved.
Methods. TRPV4 activation was assayed by measuring intracellular calcium levels with FURA2 Fluorescence in HEK 293 cells.
Results. Human mutations in the N-terminus reduced TRPV4 responsiveness to hypotonic solutions whereas other human mutations reportedly increase TRPV4 responsiveness. We identified a common signalling pathway for hypotonicity and GPCR activation of TRPV4 and have partially characterised it
Discussion. TRPV4 mutations have tissue-specific effects suggesting that cell-specific interactions modulate it. activity. TRPV4 appears to be a receptor-operated channel that is activated by intracellular signalling resulting from cell swelling or activation of specific GPCRs. Targeting tissue-specific activation pathways of TRPV4 may be a fruitful area for therapeutics with reduced side-effect potential.

Lamandé, S. et al (20011) Nature Genetics 43 (11), 1142-1146.

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$GABA_A$ receptors and flavonoids: Achieving subtype selectivity anxiolytics devoid of sedative effects

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Recent genetic and pharmacological studies have demonstrated that targeting α_2- and α_4-containing $GABA_A$ receptors may mediate the anxiolytic effects of benzodiazepines and other agents without inducing sedation. Flavonoids are a class of polyphenols found in plants and have a range of pharmacological actions including anti-anxiety effects. In our laboratory, we have developed a number of synthetic flavonoids including 2'methoxy-6-methylflavone (2'MeO6MF) and 3-hydroxy-2'methoxy-6-methylflavone (3-OH-2'MeO6MF) that mediate their action specifically via α_2- and α_4-containing $GABA_A$ receptors when evaluated on over 40 human recombinant $GABA_A$ receptors expressed in *Xenopus* oocytes using 2-electrode voltage clamp methods. Intraperitoneal injection of 1-100 mg/kg of either compound to Balb-C mice showed a significant ($p^*<0.05$, $p^{**}< 0.01$ One way ANOVA, n=8) dose dependent increase in both the number of open arm entries and the time spent in open arms in the elevated plus maze (EPM) and an increase in the time spent in the light compartment along with the number of transitions in the light dark tests. Sedative effects were only observed with 2'MeO6MF at the higher doses. No myorelaxant effects were observed with any agent in the horizontal wire test. The anxiolytic effects of both agents were not reversed by the benzodiazepine antagonist flumazenil in the EPM showing that the effects are mediated via nonbenzodiazepine allosteric sites of $GABA_A$ receptors. This study highlights the fact that targeting non-benzodiazepine allosteric sites on $GABA_A$ receptors can lead to new anxioselective agents with fewer side-effects.

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TRPM7 in cardiac hypertrophy: more than just a cation channel

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Introduction. The transient receptor potential melastatin 7 (TRPM7) channel is a ubiquitously expressed cation channel that is permeable to Mg^{2+} and Ca^{2+}. Unusually for a cation channel, TRPM7 also contains a kinase domain, making this a dual function protein that can both transport cations and activate intracellular signalling cascades. TRPM7 is essential for cell growth and embryonic development, however its role in cardiac cells remains poorly understood.

Aims. This study aimed to determine whether TRPM7 could mediate growth and remodelling of cardiac cells.

Methods. Ventricular cardiomyocytes were isolated from neonatal rats. Cells were then treated with short hairpin RNAs to downregulate TRPM7 expression, or with a recently described pharmacological inhibitor of TRPM7 (NS8593, 10 μM) before being exposed to either angiotensin II (AngII) or neuregulin (NRG) to induce hypertrophy. Hypertrophic signalling was measured using promoter-driven reporter assays for classical 'pro-hypertrophic' genes, and reorganization of the actin cytoskeleton examined with phalloidin staining.

Results. Both AngII and NRG caused activation of myosin light chain 2v, atrial natriuretic peptide and cyclin D (indicating an increase in hypertrophic signalling). Downregulation of TRPM7 using three independent shRNAs significantly inhibited the AngII-induced activation of these hypertrophic reporters. Activation of hypertrophic signalling was also reduced by pharmacological inhibition of TRPM7 with NS8593. AngII and NRG both cause sarcomeric re-organization and cellular hypertrophy in cardiomyocytes; this could be prevented by either pharmacological inhibition or downregulation of TRPM7.

Discussion. The TRPM7 cation channel contributes to hypertrophic signalling, cellular growth and reorganization in cardiomyocytes. Further investigations are required to determine whether this is due to Mg^{2+}/Ca^{2+} transport, or activity of the TRPM7 kinase domain.

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Systematic Reviews of Adverse Effects – Why Bother?

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While systematic reviews and meta-analyses are top of the evidence hierarchy, most of the work has concentrated on evaluation of treatment benefit. This talks aims to present a structured framework and provide practical relevant examples that illustrate how adverse effects can be evaluated in a comprehensive, unbiased manner. There are unique methodological challenges stemming from the diversity of adverse outcomes ranging from common, mild symptoms to rare, fatal events, thus making it almost impossible to design a single study that addresses all facets. Retrieval of the most appropriate studies should usually be specifically tailored to fit the nature of the adverse effects, according to the primary objective and study question. This depends on whether the main aim is towards scoping/ hypothesis-generation, or to statistically calculate magnitude of risk (with hypothesis testing), or clarifying characteristics and risk factors of the adverse effect.

Selection of appropriate data sources depends on characteristics of the adverse effect (e.g. background incidence and effect size of the drug, pharmacological predictability, clinical presentation, time of onset after drug exposure). Reviewers should bear in mind possibility of Type II errors (a particular problem when evaluating rare adverse effects) that lull us into a false sense of security (e.g. wrongly concluding that there was no significant difference in harm between drug and control, with the drug erroneously judged as safe). Hence, it is important to retrieve particular study designs that are most likely to yield robust data on the adverse effects of interest, rather than rely on studies that are poor at measuring certain types of adverse effects, thus leading to 'false negatives'. Reviewers and readers should also be aware of methodological limitations or controversies in the conduct of meta-analyses that can lead to conflicting or differing interpretations of the dataset.

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Public Health: drug safety epidemiology

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The findings that rofecoxib and rosiglitazone increased the risk of myocardial infarction drew attention to the limitations of spontaneous adverse reaction reporting programs which have been the mainstay of pharmacovigilance since they were established following the thalidomide disaster. Regulators have now moved to a far greater emphasis on active investigation and less emphasis on the passive surveillance of spontaneous reporting. This change includes a requirement that companies submitting an application for a new chemical entity provide a Pharmacovigilance Plan which describes how the company will investigate gaps in knowledge about the safety of their product.

Spontaneous reporting programs are not able to determine that a drug increases the risk of an event which has a significant background incidence in the population of individuals taking the drug, and which has no specific features which lead to an association with the drug being made. They also are poor at identifying events with a long time to onset, such as malignancies.

Thus the association between rofecoxib and myocardial infarction was made in a randomised controlled trial (RCT), and meta-analysis of RCTs was required for the association with rosiglitazone. Typically adverse reactions, at least the serious ones, are rare, and even meta-analysis may not provide a sufficiently sizable cohort to make an association. Case-control designs are the epidemiological approach to investigating a rare occurrence in the context of a defined exposure. This methodology can be used to investigate associations and also risk factors. Our group has used cases reported to the spontaneous reporting program to investigate risk factors for a number of adverse reactions, including hepatitis with flucloxacillin, cystitis with tiaprofenic acid and myocarditis with clozapine. For the last of these three we plan to conduct a pharmacogenetic analysis shortly.

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Pharmacogenomics and drug safety

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The last decade has seen elucidation of many genes related to drug metabolism, drug transporters and pharmacodynamic factors of specific drugs that have the propensity to drive dose-dependent and pharmacologically predictable adverse drug reactions and drug interactions. Associations between immune response genes such those in the major histocompatibility complex (HLA) have shed light on the immunopathogenesis of serious adverse drug reactions such as drug hypersensitivity syndromes (DRESS/DIHS), Stevens-Johnson Syndrome and toxic epidermal necrolysis (SJS/TEN). The association between HLA-B*5701 and abacavir has been a notable discovery which has acted as a roadmap for T1→ T4 translation from discovery of an association between a gene and a drug toxicity through to clinical and laboratory translation, through to implementation of a screening test which has now been routinely used as part of guideline-based HIV clinical practice in the developed world to successfully essentially eliminate abacavir hypersensitivity. Whether a test will translate is driven by characteristics of the drug and the availability of therapeutic alternatives, attributes of the test itself, nature of the drug toxicity, having an environment or individual to champion the test, the ability to generate high levels of evidence to support the clinical utility and cost-effectiveness of the test, the development of appropriate laboratory support, infrastructure and quality assurance, and the design and implementation of appropriate clinical systems. More recent research has provided important structural and functional insights as to how drugs like abacavir and carbamazepine may specifically interact with HLA-B*5701 and HLA-B*1502 respectively to cause hypersensitivity and SJS/TEN. It is now feasible that these approaches examining how drugs specifically interact with HLA and other genes could be applied as part of a pre-clinical pharmacogenomic screening strategy to inform the design and development of safer drugs.

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The safety of medicines in older people

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Older people are the major users of medicines. In Australia older adults take between 5-12 different prescription medicines every day and this tends to be higher in frail elderly people and those in residential care facilities. Such use of high medicine is unquestionably associated with increased risk of harms. Our prospective study of 1705 community-dwelling older men (CHAMP study) has shown that high risk prescribing such as polypharmacy, hyperpolypharmacy and a high Drug Burden Index is associated with a variety of poorer outcomes including increased risk of frailty, falls, death and institutionalization, taking into account comorbidities and other subject characteristics. The use of psychotropic medicines including antipsychotics and opioid analgesics is very prevalent in older people and the use is increasing dramatically in these age groups particularly those in residential care. Yet these medicines have been linked with significant adverse effects including an increased risk of death in older people, especially those with dementia. Such prescribing is usually undertaken in the absence of good quality evidence for the efficacy or effectiveness of these medicines in very elderly people or those with comorbidities. On the other hand, deprescribing and reduction of drug burden has been linked with beneficial outcomes or at least the absence of harm in many situations. This critical situation will only be resolved by including real life frail older people in clinical trials or at least high quality observational studies. Moreover, research is required to establish the benefits of deprescribing in older people with polypharmacy and to develop educational methods that are effective at improving the prescription to older people.

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Drug delivery to the eye: barriers and delivery system prospects

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Ocular drug delivery is practiced in most cases as eye drops, but this method is suitable only in the treatment of anterior segment disorders. For retinal drug delivery, invasive intravitreal injections must be used. This lecture illustrates the relevant barriers and constraints in ocular drug delivery to the anterior and posterior segment. In addition, the prospects of delivery systems will be highlighted, especially for the clinical benefits in the retinal drugs delivery.

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Translational pulmonary drug delivery

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Bringing a therapeutic molecule from the laboratory bench to the patient's bedside involves many translational steps, including physicochemical and biological aspects of both in vitro and in vivo studies. Pulmonary drug delivery has experienced rapid growth over the past two decades, with new inhalation products coming into the markets for treatment of diseases no longer limited to only asthma and chronic obstructive pulmonary diseases. These therapeutic compounds include deoxyribonuclease, tobramycin and mannitol for cystic fibrosis, prostacyclin for pulmonary arterial hypertension, insulin for diabetes, vaccines and antiviral drugs for influenza, as well as mannitol for measurement of airway hyperresponsiveness. While some of these compounds are new drug molecules, the rest are old drugs but repositioned for a new indication. This presentation will highlight some key considerations in translational pulmonary drug delivery, with an emphasis on the formulation and delivery aspects for drug repositioning and its effectiveness.

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Utilizing oligoarginine to enhance cellular uptake of PECA nanoparticles

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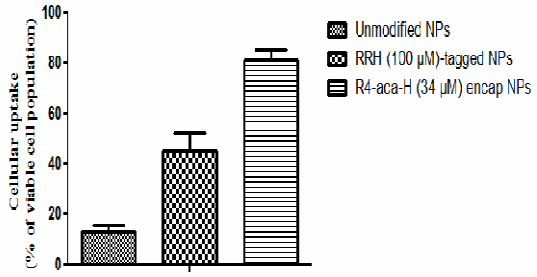

Introduction. Cell-penetrating peptides, such as oligoarginine, can be covalently bonded with bioactives to enhance their cellular uptake (Fonseca et al, 2009). However, this modifies the bioactive moiety and so potentially its bioactivity. An alternative approach is to associate arginine with nanoparticles that encapsulate the bioactive, allowing the bioactive to be delivered in its native form.

Aims. To compare covalent and non-covalent association of oligoarginine with nanoparticles for improving cellular uptake.

Methods. Poly(ethyl-cyanoacrylate) (PECA) nanoparticles were prepared using a microemulsion template. Oligoarginine of different chain lengths attached to histidine (RRH or R4-aca-H) and FITC-dextran (MW = 2,000 kDa) were added to the aqueous phase of the microemulsion prior to polymerization. Covalent binding of the oligoarginine via histidine anchoring to the histidine residue was determined using MALDI-TOF (Matrix-Assisted Laser Desorption/Ionization – Time Of Flight) spectrometry. Flow cytometry was used to quantify the cellular uptake of the different nanoparticle formulations into Caco-2 cells.

Results. Di-arginine-histidine (RRH) covalently bound to PECA nanoparticles and had a higher uptake than the unmodified PECA nanoparticles. R4-aca-H did not covalently bind to the PECA nanoparticles, however when it was encapsulated, 80% cell uptake was observed.

Discussion. Associating oligoarginine either covalently or entrapping it within PECA nanoparticles increases cellular uptake compared with nanoparticles without arginine. Encapsulated oligoarginine resulted in greater uptake compared to covalently-tagged PECA nanoparticles. Polymeric nanoparticles administered with cell-penetrating peptides may thus have potential to improve absorption of bioactives.

Fonseca S B et al (2009) Advanced Drug Delivery Reviews, 61:953-964

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Bio-mimetic prodrugs to promote lymphatic transport of immunomodulators: A balance between digestive stability and ease of re-esterification

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Introduction. The lymphatic system plays an important role in immune function, as well as serving as the main conduit for transport of dietary triglyceride (in the form of lipoproteins) from the intestine. The current study examines the use of triglyceride(TG)-mimetic prodrugs to target the delivery of an immunomodulatory drug, mycophenolic acid (MPA), to the lymphatic system via integration into triglyceride metabolic pathways. Enhanced immunomodulator exposure to sites of action within lymphoid tissue is expected to improve the treatment of immune system diseases.
Methods. Biotransformation of the prodrugs was assessed via incubation with rat digestive fluid and rat plasma, and analysis of prodrug derivatives in mesenteric lymph was facilitated by HPLC-MS. Lymphatic drug transport was examined in mesenteric lymph-cannulated rats following intraduodenal drug/prodrug administration under conditions modulated by metabolic enzyme inhibitors.
Results. All four TG-mimetics markedly increased lymphatic drug transport (2-96 fold) when compared to parent MPA where lymphatic transport was low (0.14% of dose). Prodrugs were digested rapidly (<2min) by rat digestive fluid to form the monoglyceride equivalent prodrugs (MEPs) prior to being re-esterified with fatty acids in enterocytes. Lipolysis was a prerequisite for efficient lymphatic transport as inhibition of MEP generation by orlistat (320μM) markedly attenuated lymphatic recovery (MPA-TG, 13.5% vs 1.3%, in the absence and presence of orlistat). Stabilisation of labile MEPs by steric hinderance of the ester bond showed both improved (MPA-C6-Me-es-TG, 13.5%) or reduced (MPA-C6-et-TG, 0.31%) lymphatic transport when compared to MPA-C6-es-TG (9.6%), depending on the impact of structural modification on re-esterification.
Discussion. The current mechanistic evaluation of the drivers of lymphatic transport of TG-based prodrugs revealed the need for luminal liberation of MEPs, the prevention of further breakdown of MEPs to parent drug (MPA) and derivatives, and the need for subsequent re-esterification to the TG forms in the enterocyte.

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Enhanced exposure of a chemotherapeutic agent to the lymphatic system with the use of nano-sized and PEG-capped drug vectors

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Introduction. The lymphatic system is a major pathway by which metastasizing cancers spread throughout the body. Cancers that arrest within lymph nodes are also able to seed further tumours at distal locations. Targeting chemotherapeutic agents towards the lymphatic system and lymph nodes offers an alternative treatment option to surgical lymph node removal and further provides the potential to limit the side effects commonly associated with chemotherapy.
Aims. To directly compare the lymphatic uptake of doxorubicin in a clinically available PEGylated liposomal formulation (Doxil®) and a PEGylated dendrimer containing doxorubicin covalently linked to the dendrimer surface via an acid-labile hydrazine linker after IV and SC administration to rats.
Methods. Male Sprague Dawley rats were cannulated via the right carotid artery to facilitate blood collection, the thoracic lymph duct to facilitate lymph collection, and the right jugular vein to allow intravenous dosing and replacement of fluid lost via the drainage of thoracic lymph. Rats were dosed either IV via the jugular vein cannula or SC into the left heel and blood and lymph samples were collected for 30 h in thoracic duct cannulated rats and up to 7 days in non-lymph cannulated rats. Plasma and lymph were analysed for doxorubicin concentration via HPLC.
Results. Liposomal and dendritic formulations of doxorubicin significantly increased the lymphatic exposure of doxorubicin by ~110 fold and ~490 fold respectively after SC dosing. Interestingly, IV administration of the dendritic formulation also significantly improved lymphatic exposure to doxorubicin when compared to IV administration of doxorubicin (by 330 fold) or the liposomal formulation (by >2 fold).
Discussion. This work demonstrates that the dendritic formulation was able to target the lymphatics from both an SC injection site and from the blood after systemic absorption, suggesting the potential for colloidal and polymer-based drug delivery systems to improve the exposure of lymph-resident metastases to chemotherapeutic drugs.

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Expression, regulation, deorphanisation and putative nutrient-sensing function of taste GPCRs in the heart

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Introduction. G protein-coupled receptors (GPCRs) are critical for cardiovascular physiology, yet only a small fraction of the cardiac-GPCR repertoire is therapeutically targeted. Taste receptors are functional beyond the mouth, but have not been studied in heart.
Aims. To investigate taste GPCR expression, regulation and function in human and rodent heart.
Methods. RT-qPCR taste receptor screens were performed on failing human hearts, in rodent heart, and in cultured cardiac myocytes and fibroblasts. Taste GPCR localisation was investigated using *in situ* hybridisation and *Tas1r1* gene-targeted mice (Tas1r1^{Cre}/Rosa26^{tdRFP}). Five cardiac-expressed type 2 taste receptors (*Tas2*) GPCRs were cloned from rat heart and screened against 102 natural or synthetic bitter compounds in a heterologous expression system.
Results. Discrete subsets of *TAS1/Tas1* and *TAS2/Tas2* family GPCRs were identified in human and rodent hearts, were enriched in cultured rat myocytes, and were localised in distinct myocardial cells. Following starvation, several *Tas2r*s were upregulated ~3-fold in cultured rat myocytes and in the mouse heart *in vivo*. We identified novel agonist ligands for three *Tas2* GPCRs (Tas2r108, Tas2r137 and Tas2r143). Ongoing work is focused on testing the endogenous cardiac-expressed taste GPCR responses in primary rat ventricular myocytes.
Discussion. The discovery of taste GPCRs in the heart opens an exciting new field of cardiac research. We predict that these taste receptors may function as cardiac nutrient sensors and using our identified agonist ligands, we hope to delineate the physiology of taste receptor activation in the heart.

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A reduction in random between subject variability is not mandatory when adding a new covariate

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Introduction. Population PKPD analyses include models for heterogeneity between subjects. The remaining between subject variability that cannot be explained by patients' characteristics, known as covariates, is assumed to arise from random variability. Random variability should decrease when more variability can be explained from predictable sources (e.g. weight or genetics).
Aim. To explore circumstances where random variability may not reduce even though a significant covariate is included in the analysis.
Methods. This work is based on simulation and estimation and does not include any patient data. Simulations were performed using the software MATLAB (ver 2010b) and estimation using the software NONMEM (ver 7.2). For simplicity we assumed the underlying PK was described by an *iv* bolus 1-compartment model. Five scenarios were considered with correlations between clearance (CL) and the theoretical covariate ranging from 0-100%. We considered two relationships between CL and the covariate, (1) where the relationship had a forced intercept of zero – reflecting when the covariate value (e.g. weight) equals zero then CL equals zero and (2) where we allowed for an intercept, i.e. when the covariate value equals zero (e.g. weight) then CL equals a positive value. Each simulated scenario was also estimated by a base model (i.e. no covariate) which was used for comparisons.
Results. Relationship 1 resulted in inflated random variability with correlations 0 - 75%. Models that allowed for an intercept performed well with slight inflation which was evident only at low correlation (25%).
Discussion. A moderate to strong covariate may not reduce random variability and in fact it may inflate this variability when not correctly implemented. We note here that correct implementation should not necessarily be based on what appears to be a biologically plausible relationship. This would lead to wrong conclusion that covariate was not relevant.

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Ghrelin gene-derived peptides have protective roles in the cerebral circulation and brain

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Introduction. Cerebrovascular dysfunction - deficits in nitric oxide (NO·) function and Nox2 oxidase-driven oxidative stress - occurs after ischaemic stroke leading to further neuronal damage. The ghrelin gene is expressed in the stomach where it encodes three peptides - acylated ghrelin, des-acylated ghrelin and obestatin. In addition to their neuroendocrine functions, evidence suggests that these peptides may have important protective functions in the cardiovascular and central nervous systems.
Aims. Test whether ghrelin gene-derived peptides: (1) exert protective cerebrovascular effects; and (2) protect the brain against ischaemia.
Methods. In cerebral arteries from wild-type (WT) and ghrelin knockout ($Ghr^{-/-}$) mice, NO· function was assessed in a myograph via the vasoconstrictor response to L-NAME (NO· synthase inhibitor; 100 μmol/L), and Nox2 activity was assessed by measuring phorbol 12,13-dibutyrate (PDBu; 10μmol/L)-stimulated superoxide production. Stroke was induced in WT and $Ghr^{-/-}$ mice by middle cerebral artery occlusion (MCAo) for 0.5h followed by reperfusion (23.5h). At 24h, neurological and sensorimotor function, and infarct and oedema volumes were evaluated.
Results. The magnitude of L-NAME-induced constrictions of cerebral arteries from $Ghr^{-/-}$ mice (Δ diameter = -27.4±2.1%; n=5, P<0.05) were lower by ~36% when compared to WT mice (-42.7±2.1%; n=5). PDBu-stimulated superoxide production was two-fold higher in $Ghr^{-/-}$ mice (n=7, P<0.05). In WT mice, exogenous des-acylated ghrelin or obestatin (100fmol/L-10nmol/L; n=5-8) elicited NO·-dependent cerebral vasodilatation, whereas acylated ghrelin (n=8) had no effect on tone. Furthermore, des-acylated ghrelin (1nmol/L-10nmol/L), but not acylated ghrelin or obestatin, inhibited PDBu-stimulated superoxide (n=5, P<0.05). After MCAo, $Ghr^{-/-}$ mice had worse neurological and sensorimotor impairment, and significantly larger infarct ($Ghr^{-/-}$: 37.7±3.9 vs WT: 26.3±2.7, mm^3, n=8-10, P<0.05) and oedema ($Ghr^{-/-}$: 7.8±1.7 vs WT: 4.03±0.6, mm^3, n=8-10, P<0.05) volumes.
Discussion. These data reveal a previously unrecognised protective role for ghrelin gene-derived peptides in the cerebral circulation and brain, and highlight their potential as novel approaches for stroke treatment.

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Calcium dependent regulation of multidrug resistance-associated protein 3 (MRP3/ABCC3) gene expression in a model of epithelial-mesenchymal transition (EMT)

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Introduction. Increased expression of members of the ATP binding cassette (ABC) transporter superfamily in cancer cells is linked to chemoresistance, and more recently has been correlated with malignant progression and a more aggressive cancer phenotype. Epithelial-mesenchymal transition (EMT) is a process implicated in cancer metastasis and is a feature of an aggressive tumour subtype. Aberrant Ca^{2+} signaling is a feature of tumourigenesis and tumour progression in some cancer types, and has recently been linked to EMT. The relationship between EMT, ABC transporter expression and Ca^{2+} signaling has not yet been fully assessed.
Aims. 1. To quantify specific MRP/ABCC transporter mRNA expression in a model of EMT using the MDA-MB-468 basal-like breast cancer cell line. 2. To assess the effect of intracellular free Ca^{2+} chelation prior to induction of EMT on MRP/ABCC mRNA expression.
Methods. For induction of EMT, MDA-MB-468 cells were treated with epidermal growth factor (EGF, 50 ng/mL) for 24 h. Intracellular free Ca^{2+} chelation was achieved by loading the cells with 100 μM BAPTA-AM for 1 h at 37°C prior to treatment with EGF. Quantitative real-time RT-PCR was used to assess changes in MRP1/ABCC1, MRP3/ABCC3 and MRP5/ABCC5 mRNA expression following EGF and/or BAPTA-AM treatment.
Results. EGF-induced EMT in MDA-MB-468 cells resulted in a significant increase in MRP3/ABCC3, but not MRP1/ABCC1 or MRP5/ABCC5, mRNA relative to the control. This effect was abolished in cells pre-treated with BAPTA-AM, indicating that intracellular Ca^{2+} plays an essential role in facilitating the increased expression of MRP3/ABCC3 in MDA-MB-468 cells undergoing EMT.
Discussion. MDA-MB-468 cells undergoing EGF-induced EMT demonstrate a calcium dependent increase in MRP3/ABCC3 mRNA expression. Further studies are required to elucidate the precise mechanisms underlying the regulation of MRP3/ABCC3 mRNA expression by Ca^{2+}.

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Small airway reactivity to constrictors is differentially altered by chronic and acute inflammatory stimuli

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Introduction. The distal lung is an important site of inflammation, remodelling and airway hyperresponsiveness (AHR) in asthma. These changes are evident in mouse models following chronic exposure to ovalbumin (OVA) while lipopolysaccharide (LPS) directly induces airway inflammation. Limited studies have assessed small airway reactivity *in vitro* following these stimuli.
Aim. To assess small airway reactivity in mouse lung slices following chronic OVA treatment *in vivo* or acute LPS treatment *ex vivo*.
Methods. Balb/C mice sensitised with OVA on days 0 and 14, were challenged with nebulized OVA or saline 3 times/week from day 21 to 64, and small airway fibrosis confirmed using Masson's trichrome stain. Lung slices (150μm) from saline and OVA groups were cultured in the absence or presence of IL-1α (10ng/mL) and/or TNFα (50ng/mL) for 48h. Slices from untreated mice were exposed to LPS (10μg/mL, 48h) and supernatants assayed for TNFα by ELISA. Changes in small airway lumen diameter in response to serotonin (5HT) and methacholine (MCh) were measured using phase-contrast image analysis.
Results. Following OVA challenge, small airway contractile responses to MCh were slower, with a 13-fold loss in potency and reduced maximum relative to saline controls. *In vitro* incubation with IL-1/TNFα did not alter reactivity in slices from either group. LPS treatment increased TNFα release (175.6 ± 65.9pg/mL, ND in untreated slices, n=5) and increased airway reactivity to 5HT, but not MCh.
Conclusion. Airway hyporesponsiveness following OVA challenge, even in the presence of inflammatory cytokines, was unexpected and may reflect the influence of fibrosis to oppose small airway narrowing. Increased cytokine release and reactivity with LPS provides conditions to assess the clinical potential of novel anti-inflammatory and bronchodilator therapies targeting the small airways.

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Characterisation of the hybrid orthosteric/allosteric bitopic nature of TBPB at the M_1 muscarinic ACh receptor

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Introduction. Therapeutics targeting the M_1 muscarinic ACh receptor (mAChR) hold promise in the treatment of cognitive dysfunctions (Langmead *et al*., 2008). Various novel selective agonists (NSAs) have been identified for the M_1 mAChR and are purported to solely engage the receptor via an allosteric site, rather than the classical ACh (orthosteric) binding site. However, this may not be the case for some NSAs (Valant et al., 2009). We hypothesised that the recently identified NSA TBPB [1-(1'-2-methylbenzyl)-1,4'-bipiperidin-4-yl)-1*H* benzo[*d*]imidazol-2(3*H*)-one], may interact simultaneously with both the orthosteric binding site and an allosteric site in a bitopic mode of action (Jacobson *et al*., 2010; Lebois *et al*., 2010).
Aims. To characterise the bitopic mechanism of action of TBPB at the M_1 mAChR.
Methods. Truncated fragment molecules of TBPB were synthesised and pharmacologically characterised via radioligand binding and cellular signalling assays using cells recombinantly expressing the M_1, M_2, M_3 and M_4 mAChRs, as well as mutant variants of the M_1 subtype.
Results. Two fragment compounds representing opposite ends of TBPB, VCP794 and VCP813, were identified with pharmacological behaviours consistent with that of orthosteric ligands. Functional studies indicated a molecular moiety of TBPB required for agonism (VCP794), whilst radioligand dissociation kinetics studies revealed a structural moiety required for interaction with an allosteric site (VCP813). Subsequent studies in M_1 mutant receptors identified amino acid residues that differentially affect the binding and function of TBPB and its fragments compared to classical orthosteric ligands.
Discussion. These findings provide evidence of a novel bitopic mode of interaction with the M_1 mAChR by TBPB and highlight the potential existence of more bitopic ligands and their consideration in drug discovery programs.

Jacobson et al (2010) Mol Pharmacol 78(4):648-657
Langmead et al (2008) Pharmacol Ther 117:232-243
Lebois et al (2010) ACS Chem Neurosci 1:104-121
Valant et al (2009) Mol Interv 9(3):125-135

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***CYP2B6*6* mutation significantly reduces *in vitro* N-demethylation of ketamine enantiomers.**

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Introduction. Ketamine is metabolised to the active metabolite norketamine (NK) mainly by the polymorphic CYP2B6 and CYP3A4 enzymes.

Aims. To examine the relative contribution of CYP2B6 and CYP3A4, and the impact of *CYP2B6*6* mutation on *in vitro* NK formation from ketamine enantiomers in human liver microsomes (HLMs) and expressed enzymes.

CYP2B6 genotype	High affinity enzyme (CYP2B6)		Low affinity enzyme (CYP3A4)	
	K_m (μM)	Cl_{int} (ml/min/p mol CYP)	K_m (μM)	Cl_{int} (ml/min/p mol CYP)
**1/*1* (n=4)	28±15	200±101	747±620	51±32
**1/*6* (n=4)	93±33	85±29	373±132	38±10
**6/*6* (n=3)	78±6.7	39±16	847±182	21±5.5

Methods. Kinetics of S- and R-ketamine metabolism were determined using cDNA-expressed CYP3A4, CYP2B6 with coexpression of Cyt b5, cDNA-expressed CYP2B6.1 and CYP2B6.6 variant, and HLMs genotyped for *CYP2B6*6*. Effects of CYP-selective inhibitors on NK formation were also studied.

Results. Two-enzyme Michaelis-Menten equation best fitted the HLM kinetic data (Table). Expressed enzymes showed that the high and low affinity enzymes were CYP2B6 and CYP3A4, respectively. K_m and CL_{int} values for the CYP2B6.6 variant were 200% and 29% that for the CYP2B6.1 protein, respectively. CYP2B6- but not CYP3A-selective inhibitors reduced NK formation from (S)-ketamine with the degree of inhibition significantly different among three genotypes (gene-dose $p<0.05$). Results for R-ketamine were similar.

Discussion. These results indicate a major role of CYP2B6 and the *CYP2B6*6* mutation on liver NK formation at therapeutic concentrations, which support our preliminary finding that the *CYP2B6*6* allele reduces ketamine metabolic clearance to norketamine clinically.

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Simultaneous deletion of the α_{1A}-adrenoceptor and $P2X_1$purinoceptor generates male mice which are infertile

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Introduction. Research into male contraceptives has focused on disrupting the production of sperm by altering hormone levels. Such approaches require factors such as the reversibility of such treatments and the efficiency of sperm reduction to be met to be truly suitable targets for a male contraceptive. Furthermore loss of normal hormone signaling frequently results in side effects and impairs sexual function. We have generated α_{1A}-adrenoceptor / $P2X_1$-purinoceptor knockout (DAPKO) mice and observed that males are completely infertile. α_{1A}-adrenoceptors and $P2X_1$-puinoceptors are G protein-coupled receptors and ligand-gated ion channels respectively and have numerous physiological functions. In blood vessels and the genitourinary tract these receptors are primarily involved in smooth muscle contraction.

Aims. To investigate the mechanism of infertility caused by dual α_{1A}-adrenoceptor / $P2X_1$-purinoceptor knockout (DAPKO) in male mice.

Methods. Sexual behaviour and breeding was observed by behavioural video analysis. Sperm motility and viability was investigated by microscopy and intracytoplasmic sperm injection (ICSI). Contractile responses to electrical field stimulation and exogenously applied agonists of genitourinary tissues from DAPKO mice was observed using functional isolated organ bath studies.

Results. Crossing of male DAPKO mice with non DAPKO female mice (n=16) did not result in pregnancy despite copulation occurring with normal vigour and libido. Sperm taken from the epididymis of DAPKO mice were motile (n=6) and could readily fertilise oocytes following ICSI (n=3). Testicular weight was not changed in DAPKO mice however the contractile response of the vas deferens was markedly impaired by α_{1A}-adrenoceptor / $P2X_1$-purinoceptor deletion (n=6, $p<0.01$).

Discussion. These data show that infertility observed in male DAPKO mice is due to a loss of contractile function of the mouse vas deferens which prevents the transport of sperm from the epididymis to the urethra. Selective dual pharmacological blockade of these receptors may be a fast acting, safe and effective non-hormonal male contraceptive.

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Safe to crush? Pilot study into solid dosage form modification in aged care.

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Introduction. Dosage form modification such as tablet crushing is common practice in ACFs for residents who have difficulty swallowing. (Stubbs et al, 2008) Crushing solid dosage forms can alter their efficacy and safety parameters with clinically significant consequences. (Haw et al, 2010)
Aims. To observe solid dosage form modifications in participating ACFs within the ACT, identify commonly modified medications and the methods employed, and determine aged care staff levels of self-perceived medication knowledge, and the types and quality of resources currently available to staff regarding dosage form modification.
Methods. Medication rounds in a convenience sample of ACT ACFs were observed by MN. Nursing staff knowledge of dosage form modification and available resources was assessed by written anonymous questionnaire. Data were collated to assess the range of modifications and knowledge level.
Results. From 160 medication observations, 29 residents had a total of 75 medications modified by nursing staff prior to administration, with 32% of these medications modified inappropriately. The methods used for crushing and administration resulted in drug mixing, spillage, and incomplete dosing. Staff reported adequate resources; however a lack of knowledge on how to locate and use these resources was found.
Discussion. Mixing all medications together is not recommended, but was observed in all cases. Incomplete or no cleaning of equipment between residents was observed and has the potential to cause adverse reactions. No staff were observed to use available resources for doseform modification. These results show medication crushing is common practice in ACF and many of these modifications are inappropriate.

Stubbs J et al (2008) International Psychogeriatrics 20: 616-27.
Haw C et al (2010) International Psychogeriatrics 22: 409-16.

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Can software assist the home medicines review process by identifying clinically relevant drug-related problems?

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Introduction. Home medicines reviews (HMRs) are conducted by accredited pharmacists to detect and address drug-related problems (DRPs), to improve the quality use of medication. Identification of DRPs involves an assessment of existing drug treatment in conjunction with other factors. Australian decision support software, Medscope Medication Review Mentor (MRM), has been developed to assist the identification and resolution of DRPs.
Aim. This study assessed the ability of MRM to identify clinically relevant DRPs and to provide suitable recommendations for DRP resolution.
Methods. HMR information and pharmacist-identified DRPs and recommendations were obtained from a database of almost 700 Australian HMRs collected for another project. The HMR information was entered into MRM and findings were recorded. A random sample of 20 HMRs with DRPs found by pharmacists (N=73) and MRM (N=125), were independently assessed by a panel of 12 clinical pharmacology and pharmacy experts. Experts were blinded to each source of DRPs and provided Likert scale responses regarding clinical relevance and recommendation appropriateness.
Results. Experts agreed that the pharmacists (645 of 876 opinions; 74%) and MRM (1092 of 1500; 73%) identified clinically relevant DRPs. There was no significant difference between pharmacists and MRM regarding clinical relevance (Wilcoxon Rank Sum Test, $W = 674591$, $p = 0.212$). There was a significant difference regarding recommendation appropriateness ($W = 568346$, $p < 0.001$) reflecting greater support for MRM's recommendations.
Discussion. MRM identified more DRPs than pharmacists but not at a cost of irrelevance. The software's recommendations compared favourably with those of the pharmacists. MRM appears capable of identifying clinically relevant DRPs and providing appropriate recommendations for DRP resolution.

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An observational study of pharmacists' interventions to minimise medication misadventures in paediatric inpatients

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Introduction. It has been widely documented that medication misadventures present a significant burden on the health care system. There has been extensive research to investigate medication misadventures in adults, yet data is still lacking in the paediatric population. The lack of effective strategies to address this safety issue has significant ramification in children despite being a very vulnerable patient group. However, previous studies have claimed that ward-based clinical pharmacists' intervention plays a role to reduce the incidence of medication errors in hospital.
Aims. This study aimed to document and evaluate the role of pharmacists' interventions in minimising medication misadventures in paediatric inpatients at a paediatric hospital in Perth.
Methods. Clinical interventions performed by ward-based clinical pharmacists in two general medical wards during 35-day data collection period were observed and documented by the primary investigator. The pharmacists' intervention data were analysed to identify the occurrence of medication misadventures.
Results. It has been revealed that a total of 298 interventions were performed for 1215 patients. The rates of intervention were 5.2/100 medication orders and 24.5/100 patients. Approximately 9 interventions were performed per day. Taking medication history/counselling activities were the most common interventions (66.1%), followed by provision of drug information (11.1%) and drug therapy changes (10.4%). When categorised according to therapeutic groups, antiinfectives and analgesics accounted for the top two medications implicated in the interventions. Of all interventions, 44 interventions (14.8%) were considered active interventions. Nearly all pharmacists' active interventions identified medication errors (n=42) and the majority of errors were intercepted by pharmacists.
Discussion. Pharmacists through their interventions contributed substantially to patient safety in paediatric area by minimising the occurrence of medication misadventures in the study population.

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Prevalence and factors associated with antidepressant use in Tasmanian nursing homes

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Introduction. The rate of depression in nursing home residents is high. As depression is associated with increased morbidity and mortality, there has been an increased focus on effective treatment in this setting.
Aims. The aim of this study was to examine the prevalence and pattern of antidepressant use in nursing home residents. Analysis of commonly used antidepressants, dosages and concurrent use of other psychotropic drugs will enhance understanding of depression treatment choices and identify predisposing factors associated with the use of antidepressants in this setting.
Methods. This retrospective cross-sectional study involved the analysis of 562 de-identified Residential Medication Management Review (RMMR) case notes collected by accredited pharmacists in seven nursing homes in Hobart, Tasmania. Residents' demographics, prescribed medications and medical diagnosis details were extracted. Details on antidepressant use were recorded. Bivariate analysis examined factors associated with antidepressant prescribing. Multivariate logistic regression analysis was used to examine independent predisposing factors associated with antidepressant prescribing.
Results. The overall prevalence of antidepressant use in the nursing homes was 46%. Mean residents' age was 85.1 years, 72.8% were females. SSRIs were the most commonly prescribed class of antidepressants (20.8%), followed by SNRIs (6.9%) and TCAs (6.9%). Mirtazapine was the most commonly prescribed individual antidepressant (13%). Antidepressant prescribing was associated with concurrent benzodiazepine use and age, with the odds of receiving an antidepressant lower in residents aged > 85 years. Documented history of depression increased the likelihood of antidepressant prescribing, as did a history of chronic pain and anxiety. A history of falls/fractures was not found to be associated with the antidepressant use.
Discussion. Prevalence of antidepressant prescribing in nursing homes is high at 46%. Predisposing factors included younger age, depression diagnosis, presence of chronic pain/anxiety and concurrent benzodiazepine use. Further studies are needed to examine appropriateness of antidepressant use in this demographic.

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Prevalence of frailty and number of medicines use in elderly Australian: A comparison of four frailty measures

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Introduction: Frailty has been associated with an increased risk of adverse health outcomes; however, frailty has not been clearly defined. Several measures to identify frailty have been developed, each using different assessment criteria. Consequently, classification of patients' frailty status may vary depending on the measure used.
Aims: To compare the prevalence of frailty using four different frailty measures and to examine the number of medicines used among the frail elderly.
Methods: This study used data from the Australian Longitudinal Study of Ageing (ALSA), which is a population based cohort study of older people aged 65 years old and over, who were resident in Adelaide, South Australia. ALSA contains information on the health and wellbeing of 2087 participants. Frailty measures that were used in the analysis included two multidimensional measures (a frailty index and a prognostic frailty score) and two unidimensional measures (a frailty phenotype index and a simplified frailty phenotype index).
Results: ALSA participants were aged between 65-103 years; majority of participants lived in the community (94%). The mean age was 78 years and the median number of medicines used was 3 (range 0-9). Across four measures, prevalence of frailty varied from 2% to 49%. 35% of participants were identified as frail by at least one measure and only 0.6% were classified as frail on all four measures. Among the frail groups, 53% to 73% used 4 or more medicines compared to only 28% to 36% in the non-frail groups.
Discussion: The variation in frailty status between different measures provide challenges for researchers to evaluate the safety and effectiveness of medicines use among the frail elderly.

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Pharmacy staff perspectives on patient safety issues involved in the supply of dose administration aids

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Introduction: Dose Administration Aids (DAAs) are used to assist patients and health facilities manage medication and reduce administration errors. However, their preparation and use is not without risk. DAA quality assurance research has highlighted stability issues and identified types and rates of error that occur but has not explored broader systems and social issues affecting product quality and patient safety.
Aims: To conduct a qualitative exploration of pharmacy DAA dispensing processes and identify factors that impact quality and compromise patient safety.
Methods: Community pharmacy staff were purposively recruited from metropolitan Sydney to participate in focus groups, discussing processes in DAA service provision, associated problems and possible solutions. Transcripts were analysed to identify recurring themes.
Results: Despite variability in supply processes among pharmacies, focus groups identified a number of common, often interacting, factors that can underlie risk to patient safety; to which participants suggested some solutions. These factors align with categories identified by Phipps *et al*., 2009, including: inter-professional and patient relationships; demands on the pharmacist; workplace design and management; procedural standards and resource issues. Common elements that can affect safe practise included effective communication, workplace culture and the value pharmacy staff and managers attribute to DAA services.
Discussion: Participants suggested potential solutions to mitigate error including formalised training and plain-English guidelines; however, effective solutions need also address broader cultural, social and economic drivers of quality.

Phipps DL, Noyce PR, Parker D, Ashcroft DM.(2009) Medication safety in community pharmacy: a qualitative study of the sociotechnical context. BMC Health Serv Res. 9:158

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Key Health Professionals' views of prescribing resources for older patients

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Introduction. Prescribing resources aimed at improving the quality use of medicines in the aged are either too simple and do not address potential interactions when there are many diseases and many medications, or, when providing complex information, appear to be impractical to use during consultations.
Aims. To explore health professionals' views on currently available geriatric medication management resources and to determine what health professionals consider makes medication management resources useful.
Methods. Purposive convenience sampling was used to recruit geriatricians, GPs and accredited pharmacists for one hour, individual, semi-structured interviews. Recruitment continued until data saturation was achieved. Themes were identified using NVivo9 software.
Results. Participants felt that currently available prescribing resources did not meet their needs when managing aged patients primarily due to lack guidance on how to deal with complex issues in aged people and lack of relevance to the Australian setting. Identified barriers to providing optimal care included: lack of access to appropriate literature; issues, such as lack of time, with contextualising vast amounts of new health information; and incomplete or fragmented healthcare records. Key components which make resources useful included clear formatting, simplicity, use of peer-reviewed evidence-based recommendations and ready electronic access via an easy to use interface.
Discussion. Current resources do not meet health professionals' needs when they seek practical assistance for prescribing to older people with multiple medical problems.

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Major medication discrepancies in patients with type 2 diabetes mellitus (T2DM), referred from primary care, to a tertiary ambulatory diabetes centre.

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Introduction. Multiple medications are typically used to manage glycaemia and prevent/treat the comorbidities/complications of diabetes. Patients who transition between interfaces of care, are at risk of medication discrepancies - intended or unintended differences, between recorded medication regimens.
Aims. To identify, classify and determine predictors of medication discrepancies for T2DM patients, referred from primary care to the Royal Prince Alfred Hospital (RPAH) Diabetes Service.
Methods. A retrospective audit of a random sample of 300 adult patients, who attended the RPAH Diabetes Service between 01 January 2010, and 31 December 2011, was conducted. Rates/types of medication discrepancies were identified by comparing the medication list obtained via a structured nurse-patient interview (SNPI) with that in the GP referral letter.
Results. Based on the SNPI, patients took a total of 1328 diabetes-related (anti-hyperglycaemic, anti-hypertensives, and anti-lipid) medications, averaging 4 diabetes-related medications per patient. Discrepancies were found for 72.4% (n=962/1328) of diabetes-related medications. Comparing the GP letter with the SNPI, 40.6% (n=391/962) of medications had a dose discrepancy, 44.2% (n=425/962) were listed in the SNPI, but not in the GP letter and 9.7% (n=92/962) were listed in the GP letter but not in the SNPI. Over half of all patients (54%) had a medication discrepancy with the number of discrepancies per patient ranging from 1 (27% of patients) to 7 (0.3%). The majority of discrepancies affected anti-hyperglycaemics (56.2%) (n=541/962), then anti-hypertensives (26.9%) and anti-lipid medications (16.8%). Predictors of discrepancies were: type of GP referral (hand-written/computer-generated), practice type (group/individual), duration of diabetes, and total medication number.
Discussion. Medication discrepancies from primary to tertiary care were prevalent for patients referred to the RPAH Diabetes Service. Automated GP referral letters/inaccurate GP records may have contributed. This suggests the need for routine medication reconciliation in all transitions of care to clarify the correct regimen, ensure efficacy and patient safety.

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High-throughput assay for simultaneous quantification of the plasma concentrations of morphine, fentanyl, midazolam and their major metabolites using automated SPE coupled to LC-MS/MS

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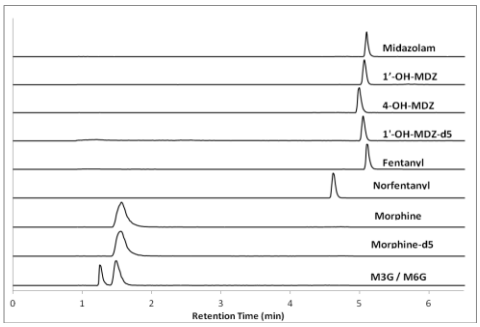

Introduction: A high throughput assay was required to measure the plasma concentration of morphine, fentanyl, midazolam, and their major metabolites; morphine-3-β-D-glucuronide (M3G), morphine-6- β-D-glucuronide (M6G), norfentanyl, 1'-hydroxymidazolam and 4-hydroxymidazolam, in plasma samples collected from critically ill patients receiving extracorporeal membrane oxygenation (ECMO) .
Aims: To develop and fully validate an LC-MS/MS method utilising on-line robotic SPE to extract and quantify the plasma concentrations of the analytes of interest in human plasma.
Methods: Aliquots (150 μl) of human plasma and of a mixture of two internal standards, morphine-d3 (200 ng/mL) and 1'-hydroxymidazolam-d5 (50 ng/mL) in 50 mM ammonium acetate buffer (pH 9.25) were mixed and loaded onto polymeric SPE cartridges which were washed using 10% methanol in 50 mM ammonium acetate buffer, pH 9.25, before elution with mobile phase comprising 0.1% formic acid in water, and acetonitrile with a flow rate of 0.6 mL/min using an 11.5 min run time. The analytes were separated on a C18 X-Terra® analytical column.
Results: The linear concentration ranges were 0.5-100 ng/mL for fentanyl, norfentanyl, and midazolam; 1-200 ng/mL for 4-hydroxymidazolam, 2.5-500 ng/mL for 1'-hydroxymidazolam and 3.5-700 ng/mL for morphine, M3G and M6G. The method showed within-run and between-run precision (RSD and accuracy <20%) for quality control samples. Moreover, analytes were stable for at least 48h in the autosampler (except for 4-hydroxymidazolam which decreased by 22% after 24h), 5 h at room temperature and after three cycles of freeze and thaw. The absolute recovery was in the range 40% (midazolam) to 110% (morphine).
Discussion: The developed assay is convenient by minimising sample volume (150 μl), replacing at least 3 separate assays, eliminating manual solvent handling, evaporation and reconstitution steps used in previously reported methods. This assay is now being successfully utilised for an international, multi-centre, clinical study investigating pharmacokinetic changes during ECMO.

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Prescribing in the elderly – cytochrome P450 (CYP) enzyme inhibitors and substrates

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Introduction. Drugs that are CYP enzyme inhibitors may increase the concentration of drugs that are CYP enzyme substrates and the risk of their adverse effects. Given that the elderly tend to be on more medications than younger patients, there is a greater chance for the occurrence of such drug interactions.
Aims. The aim was to determine whether CYP enzyme inhibitors and substrates are being co-prescribed.
Methods. 1,076 patients, aged 75 years or older, were recruited at four locations (Newcastle, Sydney, Melbourne, and Adelaide) in 2006-2009. A table of clinically relevant CYP drug interactions was used to search for the co-prescription of strong or moderate CYP inhibitors and their relevant substrates (Flockhart, 2007).
Results. There were 2 instances of strong CYP2D6 inhibitors (causes a > 80% decrease in substrate clearance - paroxetine and fluoxetine) being co-prescribed with a CYP2D6 substrate. Paroxetine (40 mg daily) was co-prescribed with flecainide (50 mg daily). Fluoxetine (20 mg daily) was co-prescribed with metoprolol (200 mg daily). Ten patients were prescribed verapamil, a moderate CYP3A4,5,7 inhibitor (causes a 50-80% decrease in substrate clearance) and the substrates for this enzyme, simvastatin (8 patients) or atorvastatin (2 patients). 29 patients were prescribed diltiazem, also a moderate CYP3A4,5,7 inhibitor and simvastatin (14 pts) or atorvastatin (15 patients). No patients on verapamil or diltiazem were co-prescribed pravastatin or rosuvastatin, which are not CYP3A4,5,7 substrates. All 14 patients were taking a higher than manufacturer recommended dose of simvastatin (10 mg) for the diltiazem/simvastatin combination and 6/8 patients for the verapamil/diltiazem combination.
Discussion. The degree of clinical relevance of these interactions will vary between patients and should be assessed on a case to case basis. This highlights the value of medication reviews.

Flockhart DA (2007) http://medicine.iupui.edu/clinpharm/ddis/table.aspx Accessed 8/10/12

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Fruit juices as perpetrators of drug interactions: The role of intestinal transporters

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Introduction. While grapefruit juice is widely recognized to cause important drug interactions via an inhibition of CYP3A4, a wider range of fruit juices, including orange and apple juice, have been shown to inhibit influx transporters in enterocytes known as organic anion transporting polypeptides (OATPs). Fruit juice coadministration significantly reduces the oral bioavailability of numerous important medicines relying on this anion transporter pathway for absorption.
Aims. We aimed to systematically review and critically analyse all studies investigating interactions between clinically used substrates of intestinal OATPs and fruit juices.
Methods. Systematic literature search (with defined terms) to identify and critically appraise relevant clinical studies.
Results. 26 studies met the inclusion criteria. Significant reductions in systemic exposure with fruit juice co-administration have been demonstrated for atenolol, talinolol, ciprofloxacin, ivermectin and etoposide, with major reductions (50-85%) in bioavailability for fexofenadine, celiprolol and aliskiren. Flavonoid constituents of fruit juices such as naringin and hesperidin, found in grapefruit and orange juice respectively, are potent inhibitors of intestinal influx transporters such as OATP1A2 and OATP2B1. The mechanism of interaction appears to be mediated via an inhibition of OATP uptake transport; differences in juice volume consumed between studies and high variability in flavonoid concentrations between juice products complicate the assessment of OATP-mediated fruit juice–drug interactions.
Discussion. OATP-mediated fruit juice–drug interactions are a clinically important, rapidly developing area of research that is helping to inform the optimal use of medicines. The involvement of the more commonly consumed orange and apple juices in OATP-mediated fruit juice–drug interactions, in addition to grapefruit juice, is likely to substantially increase the prevalence and importance of fruit juice–drug interactions in clinical practice.
Dolton et al (2012) Clin Pharmacol Ther. Online ahead of print 3/10/12; doi: 10.1038/clpt.2012.159.

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Ion chromatographic separation and isolation of oligosaccharides of intact low-molecular-weight heparin for the determination of their anticoagulant and anti-inflammatory properties

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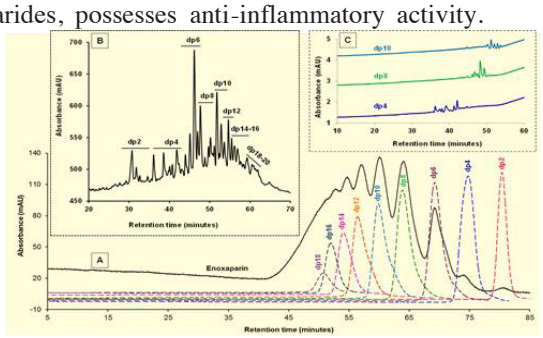
Introduction. It is well-known that enoxaparin, a widely used anticoagulant and low-molecular-weight-heparin (LMWH) containing a large number of oligosaccharides, possesses anti-inflammatory activity. While enoxaparin has shown promising results in various inflammatory disorders, some of its oligosaccharides have anti-inflammatory properties and others increase the risk of bleeding due to their anticoagulant effects.



Aims. To develop an effective ion-chromatographic (IC) technique which allows the separation, isolation and consequently the identification of different oligosaccharides of enoxaparin with or without anticoagulant activity.
Methods. Separations were performed on a semi-preparative CarboPac PA100 column (250 × 9 mm). The optimised NaCl eluent gradient was: 0-70 min: gradient from 32-74% 2 M NaCl in Milli-Q water (0.64-1.48 M NaCl). Total flow rate of 2.0 mL/min was maintained and UV detection at 232 nm was performed.
Results. The method successfully resolved enoxaparin into more than 30 different peaks. IC-derived oligosaccharides with high, moderate, low or no anticoagulant activity were identified using an anti-factor Xa assay. The anti-inflammatory activity of selected oligosaccharides was investigated using the Griess assay. Using this technique, the oligosaccharides of enoxaparin with low or no anticoagulant activity, whilst exhibiting significant anti-inflammatory activity, could be fractionated.
Discussion. In the present study, structurally complicated enoxaparin was successfully fractionated using a newly-developed IC method. An important application of this method was demonstrated by investigating the anti-inflammatory effects of its oligosaccharides. A number of studies have investigated the anti-inflammatory effects of intact enoxaparin. However, to our knowledge this is the first study demonstrating the anti-inflammatory effect of enoxaparin oligosaccharides obtained without prior chemical or enzymatic modification of the parent LMWH. This technique can provide a platform to identify the oligosaccharides which are devoid of significant anticoagulant activity and are responsible for the therapeutic effects of enoxaparin that have been observed in various inflammatory conditions.

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The Physicochemical Stability of Diluted Iron Polymaltose in Polyvinyl Chloride Infusion Bags

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Introduction. The iron polymaltose (IPM) for intravenous infusion is commonly used iron therapy for patients with iron deficiency anaemia. It is prepared by diluting commercially available IPM injection (318 mg/2 mL) with 0.9% sodium chloride. The stability of extemporaneously prepared IPM is unknown beyond 24 hours. Therefore, such preparations must be prepared on daily basis which results in substantial limitations and problems.
Aims. To investigate the physico-chemical stability of diluted IPM over a 28-day period under different storage conditions.
Methods. The IPM infusion samples (2 mg/mL) were prepared under aseptic conditions and kept in light protective bags or exposed to artificial light and stored up to 28 days at either 4 or 25°C. Aliquots were withdrawn on days 0, 1, 2, 3, 6, 7, 14, 21 and 28. Samples were analysed by size-exclusion chromatography (SEC) to measure the changes in concentration or molecular weight of IPM before and after storage. Samples were also investigated for the particle size distribution by dynamic light scattering (DLS) and for the free iron (III) content by ion chromatography (IC). Furthermore, samples were visually inspected for discoloration, clarity and precipitation, and the pH values of samples were also measured.
Results. Visual, HP-SEC, DLS and IC analyses showed that IPM (2 mg/mL) in 0.9% sodium chloride for intravenous infusion prepared under aseptic conditions remained physically and chemically stable at room temperature (with or without light exposure) or in refrigerator for at least 28 days.
Discussion. This is the first study that investigated the physicochemical stability of diluted IPM. The results indicate that assigning the 28 days stability of IPM diluted in 0.9% sodium chloride and stored at room temperature or in a refrigerator would be appropriate.

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The Physicochemical Stability of Ceftazidime or Cephazolin in Peritoneal Dialysis Fresenius Infusion Bag

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Introduction. Fresenius balance is relatively new types of infusion bags containing dialysis solution used for patients with renal failure. This solution contains low-glucose degradation products which prolongs the function of the peritoneal membrane, thereby yielding better patient outcomes. Ceftazidime or cephazolin is often used for the treatment of Gram-negative or Gram-positive peritonitis in peritoneal dialysis (PD) patients. However, the stability of these antibiotics in Fresenius balance solution has not been investigated thus far.
Aim. To investigate the stability of ceftazidime or cephazolin in Fresenius balance peritoneal dialysissolution under two different storage temperatures.
Methods. Ceftazidime (500 mg/mL) or cephazolin (500 mg/mL) was injected into Fresenius PD bag to obtain the concentration of 500 mg/L. Bags (n=3) containing either ceftazidime or cephazolin were then stored at4°Cor 25°C. Each PD bag was removed from its respective storage condition and an aliquot (approximately 2 mL) was withdrawn on days 0, 1, 2, 3, 4, 5, 7 and 14. The concentration of each antibiotic before and after storage was determined by stability indicating high-performance liquid chromatography.
Results. Ceftazidime or cephazolin lost more than 90% of its initial concentration within 24 hours when stored at 25°C. On the other hand, both the antibiotics retained more than 95% of the initial concentration when kept at 4°C.
Discussion. Ceftazidime or cephazolin antibiotic admixture stored at 25°C was found to be stable for up to 24 hours. However, at 4°C the antibiotic solution was stable for up to 14 days. Hence, ceftazidime or cephazolin admixture may be prepared in advance and stored at 4°C for at least 14 daysavoiding the necessity for frequent preparation.

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A practical synthesis of D-rhamnose building blocks for synthetic bacterial O-polysaccharide conjugate vaccines

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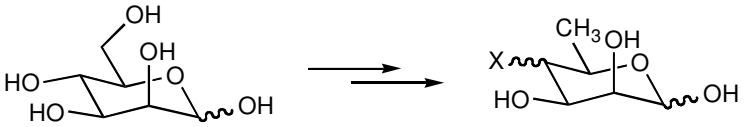
Bacterial cell surface glycans play an important role in infectious diseases. As our general knowledge of the importance of glycans in infectious diseases grows, so to do efforts directed towards understanding the role of specific components within these cell surface glycoconjugates. In this regard, 6-deoxy-D-hexoses have been receiving increased attention, most notably because of their role in infectious diseases as key components associated with pathogenic bacteria.[1]

One example of these rare sugars in bacterial pathogenesis is D-rhamnose and its 4-deoxy derivatives, which are common components of the LPS and EPS from many human and plant pathogenic species. However, importantly due to the fact that D-rhamnose is exclusively found in microorganisms and not in mammals or plants, this rare bacterial monosaccharide is a promising target for the development of new anti-infective agents, practically synthetic bacterial O-polysaccharide conjugate vaccines.[2]

Although the synthesis of D-rhamnose (and D-rhamnosides) have been reported numerous times, it has been noted by many that the synthesis of selectively functionalised D-rhamnose derivatives has been hampered by lack of ready access to D-rhamnose in large quantities [3]. This presentation will discuss our efforts towards the synthesis of selectively functionalized D-rhamnose derivatives, and will describe our successful strategy that allows for the quick and efficient synthesis of D-rhamnose derivatives. To the best of our knowledge the approach we have developed is the most efficient and high yielding synthesis of D-rhamnose reported to date.



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SPEAKER ABSTRACTS

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Glutamate efflux from brain to blood - Transport and uptake studies in a bovine *in vitro* blood-brain barrier model

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Introduction. The concentration of the excitotoxic amino acid, L-glutamate, in brain interstitial fluid is tightly regulated by uptake transporters and metabolism in astrocytes and neurons. Recent studies suggest that the blood-brain barrier may play a role in brain glutamate homeostasis.

Aims. The aim of the study was too investigate whether the blood-brain barrier play a role in brain glutamate homeostasis.

Methods. Transendothelial transport- and accumulation studies of ^{3}H-L-glutamate, ^{3}H-L-aspartate and ^{3}H-D-aspartate were performed in a tight bovine endothelial/rat astrocyte blood-brain barrier co-culture model. Results. The co-cultures displayed transendothelial resistance values of 1014 ± 70 $\Omega \cdot cm^2$, and 14C-D-mannitol permeability values of 0.88 ± 0.13 x 10^{-6} cm · s^{-1} after six days of culture. Unidirectional flux studies showed that L-aspartate and L-glutamate, but not D-aspartate, displayed polarized transport in the brain-to-blood direction, however all three amino acids accumulated in the co-cultures when applied from the abluminal side. The transcellular transport kinetics were characterized with a K_m of 69 ± 15 μM and a J_{max} of 44 ± 3.1 pmol · min^{-1} · cm^{-2} for L-aspartate and a K_m of 138 ± 49 μM and J_{max} of 28 ± 3.1 pmol · min^{-1} · cm^{-2} for L-glutamate. The EAAT inhibitor, DL- *threo*-ß-Benzyloxyaspartate, inhibited transendothelial brain-to-blood fluxes of L-glutamate and L-aspartate. Expression of EAAT-1 (*Slc1a3)*, -2 (*Slc1a2)* and -3 (*Slc1a1*) mRNA in the endothelial cells was confirmed by conventional PCR and localization of EAAT-1 and -3 in endothelial cells was shown with immunofluorescence. Abluminal uptake studies demonstrated that EAAT1 was responsible for uptake from the abluminal solution into the brain endothelial cells.

Discussion. The present study indicate that the blood-brain barrier effluxes glutamate from brain to blood via abluminal uptake via EAAT1 and luminal exit via a not yet characterized carrier system. Overall transport kinetics indicate that the efflux system may play a role under pathophysiological conditions.

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Heterogeneous vasoconstrictor responses amongst men and women

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Introduction. Women have increased in-hospital mortality following coronary artery bypass grafting (CABG)[Vaccarino et al, 2002]. Multiple mechanisms are implicated but sex-differences in vascular reactivity have not been examined.

Aims. To evaluate sex-differences in the vasoconstrictor responses of isolated internal mammary artery (IMA) from patients undergoing CABG and subcutaneous microvessels of non-cardiovascular diseased (CVD) patients.

Methods. Concentration-response curves to phenylephrine, noradrenaline, thromboxane mimetic (U46619) and serotonin were assessed by wire myography in IMA remnants and subcutaneous microvessels from CAD and non-CVD patients respectively.

Results. In the IMA, women were more sensitive to phenylephrine and serotonin (EC_{50}: 0.53μM±0.13 and 0.14μM±0.03 respectively), compared to men (EC_{50}: 1.14μM±0.23 and 0.82μM±0.32, $p<0.05$). While in the non-CAD microvessels, women were more sensitive to phenylephrine (EC_{50}: 2.92μM±0.63 Vs 8.42μM±1.55), with increased constrictions to noradranaline (E_{MAX}: 117.4±5.68 Vs 147.5±16.19) and U46619 (E_{MAX}: 161.2±18.41 Vs 124.5±4.83). Western blot analyses for total α1 and β2 adrenoreceptor abundance in IMA segments were not different between sexes, however, serotonin receptor density was higher in women than men.

Discussion. In the large arterial segments women have selective hypersensitivity to serotonin and phenylephrine, with increased serotonergic receptor density. The subcutaneous microvessels of non-CAD female patient had greater response to phenylephrine, noradrenaline and U46619. This may predispose to myocardial ischemia in women and contribute to the increased post-operative mortality.

1. Vaccarino V et al (2002) Circulation. 105:1176-81

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The role of NOX2 NADPH oxidase in macrophage polarisation

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Introduction. Advanced atherosclerotic plaques are associated with an increased ratio of pro-inflammatory M1 to anti-inflammatory M2 macrophages and increased activity of the reactive oxygen species generating enzyme, NOX2 NADPH oxidase. Given NOX2 deletion reduces atherosclerotic lesion size (Judkins et al., 2010), we hypothesized that NOX2 may contribute to the atherogenic actions of M1 macrophages and promote M1 polarisation.

Aims. To establish an in vitro model of macrophage polarisation and determine the role of NOX2 in M1 polarisation.

Methods. Phorbol-12,13-dibutyrate (PDBu, 10nM, 24hrs)-differentiated human monocytes (THP-1) were polarised toward either a M1 (lipopolysaccharide 10ng/ml + interferon-gamma 5ng/ml; LPS-IFN- , 72hrs) or a M2 (interleukin-4 25ng/ml; IL-4, 72hrs) phenotype. Quantitative real-time PCR was used to measure expression of M1 (CCR7, CXCL11) and M2 (MRC1) markers, and NADPH oxidase subunits. L012-enhanced chemiluminescence was used to measure PDBu (NOX2-derived) and ionomycin (NOX5-derived)-stimulated superoxide production.

Results. LPS/IFN- treatment selectively increased expression of M1 markers CXCL11 (50-fold, n=6; $P<0.01$) and CCR7 (250-fold, n=4; $P<0.01$) in THP-1 cells, whereas IL-4 treatment increased MRC1 expression (4-fold, n=6; $P<0.05$). M1 polarisation was associated with upregulation of NOX2 (4-fold, n=6; $P=0.05$) and its regulatory subunit, p47phox (7-fold, n=6; $P<0.01$), as well as an increase in PDBu-stimulated superoxide production. NOX2 expression was not elevated in M2-polarised cells, but expression of another isoform, NOX5 (3-fold, $P<0.05$) was upregulated. Consistent with this, ionomycin-stimulated superoxide production was elevated in M2 macrophages. Finally, chronic treatment with the NOX2 inhibitor, apocynin, did not prevent M1 macrophage polarisation.

Discussion. M1 and M2 macrophages are associated with increased NOX2 and NOX5 activity, respectively. While NOX2 inhibition may be a viable therapeutic strategy to limit the damaging effects of M1 macrophages, it may not lead to a change in polarisation state.

Judkins CP et al (2010) Am J Physiol Heart Circ Physiol 298:H24-H32

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An investigation of the anti-inflammatory role of nitroxyl (HNO) on the endothelium.

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Introduction. Nitric oxide ($NO^{\bullet}$) interferes with key events in plaque development, including endothelial-leukocyte adhesion. Unfortunately, the use of the $NO^{\bullet}$ donor, glyceryl trinitrate (GTN) as a treatment for cardiovascular disease is limited due to its susceptibility to tolerance and clearance by superoxide. The reduced congener of $NO^{\bullet}$, nitroxyl (HNO) is resistant to tolerance, yet the anti-inflammatory potential of HNO is unknown.
Aims. To assess the effects of the HNO donor, Angeli's Salt (AS) compared to GTN, on endothelial inflammation.
Methods & Results. Both AS and GTN attenuated endothelial-leukocyte adhesion of monocytes (THP-1) to TNFα-activated HUVECs in a concentration-dependent manner (TNFα (10ng/mL); 100±0%, TNFα+AS (10μM); 36±4%, TNFα+GTN (10μM); 46±8%; n=3-4; $P<0.001$). The effects of AS and GTN were diminished with the soluble guanylate cyclase (sGC) inhibitor, ODQ (10μM; n=3; $P<0.01$), and GTN, but not AS, with the $NO^{\bullet}$ scavenger, hydroxocobalamin (100μM; n=3; $P<0.05$). Flow cytometry analysis revealed that the protein expression of intercellular adhesion molecule-1 (ICAM-1) on TNFα-stimulated HUVECs was significantly reduced by AS (10μM; n=3; $P<0.001$), but not by GTN ($P>0.05$). In proof of concept experiments using mouse aorta stimulated with TNFα, both AS and GTN also attenuated adhesion of leukocytes from human blood compared to TNFα alone-treated vessels after 10 mins (TNFα; 18±2, TNFα+AS (10μM); 4±1, TNFα+GTN (10μM); 5±1 leukocytes/field; n=5-10; $P<0.001$). The effects of AS were abolished by the HNO scavenger, L-cysteine (3mM; n=3; $P<0.001$), and ODQ (n=3; $P<0.01$), and GTN by hydroxocobalamin (n=3; $P<0.001$). Aortic levels of ICAM-1, monocyte chemotatic protein-1 (MCP-1) and interleukin-6 (IL-6) mRNA were increased by TNFα (n=4-6; $P<0.05$) but were unaffected by either AS or GTN (n=3-6; $P>0.05$) suggesting post-transcriptional activity.
Discussion. These results demonstrate, for the first time, that AS reduces inflammation through an endothelial sGC-dependent mechanism and therefore may be a viable therapeutic agent in the treatment of cardiovascular disease.

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Differential roles of KCNQ4 and KCNQ5 potassium channels in cerebral artery reactivity

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Introduction. Voltage-dependent potassium channels encoded by KCNQ genes (Kv7) participate in the myogenic control of cerebral artery diameter but there is no information on the role of individual isoforms in either myogenic activity or receptor-mediated vasorelaxation.
Aims. This study aims to define the specific contribution of Kv7.4 and Kv7.5 channels to myogenic and calcitonin gene-related peptide (CGRP) mediated regulation of cerebral artery diameter.
Methods. Potassium channel modulators and small interfering RNA (siRNA) strategies were allied with isometric and isobaric myography to investigate the contribution of Kv7.4 and Kv7.5 subtypes to CGRP-evoked dilation and pressure-induced myogenic constriction in middle cerebral arteries (MCA).
Results. Dilations to CGRP in precontracted MCA were inhibited by the pan-Kv7 channel blocker linopirdine (E_{max}: 43.7±9.8% compared to control 113.1±12.3%, n=6, P<0.01). Similarly, arteries depleted of KCNQ4 using selective siRNA were markedly less responsive to CGRP (E_{max}: 31.4±15.3% compared to scrambled siRNA 90.8±6.7%, n=5-8, P<0.05) and the Kv7 channel activator S-1 (P<0.01). In contrast, responses to CGRP and S-1 in arteries incubated with KCNQ5 siRNA were not different from scrambled siRNA transfections. However, downregulation of Kv7.5 channels by targeted siRNA markedly increased active myogenic constriction at nearly all pressures including the physiological range (40 to 80 mmHg).
Discussion. The present study identifies differential roles for Kv7 subtypes in the control of cerebral arterial tone. While Kv7.4 channels underlie a substantial portion of the dilation produced by CGRP, Kv7.5 channels predominate in the regulation of myogenic constriction.

Zhong XZ et al (2010) J Physiol. 588:3277-93

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Hydrogen sulfide protects endothelial function under conditions of oxidative stress.

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Introduction. Hydrogen sulfide is an endogenously produced gas that is reported to have anti-oxidant effects.
Aims. The aim was to examine the capacity of the H_2S donor, NaHS to scavenge superoxide anions (O_2^-) and to examine whether this effect elicited protection of endothelial function in oxidative stress.
Methods. O_2^- were generated in Krebs' solution via the reaction of hypoxanthine (Hx 100μM) with xanthine oxidase (XO 0.01U/ml) or pyrogallol (PG 20μM). Thoracic aortae were collected from male C57Bl6/J mice and vascular reactivity and NO bioavailability were examined by myography. NADPH (100μM)-dependent lucigenin-enhanced chemiluminescence was used to examine the ability of NaHS to scavenge O_2^- with and without vascular tissue.
Results. NaHS scavenged O_2^- generated from Hx-XO in Krebs' solution in a concentration-dependent manner (maximum reduction 59±4%, IC_{50} 0.12μM, $P<0.001$). Aortic rings exposed to either Hx-XO or PG in the myograph displayed significantly attenuated vasorelaxation to the endothelium-dependent vasodilator acetylcholine (control: 90±7%, Hx-XO: 58±4%, PG: 65.3 ± 4.0%, $P<0.001$) which was completely reversed by NaHS (100μM) or superoxide dismutase (250U/ml). Similarly, NO bioavailability was attenuated by PG ($P<0.05$), but restored by NaHS (100μM). NaHS treatment (100nM-100μM) for 30min inhibited vascular O_2^- generation in a concentration-dependent manner (maximum reduction 88±3%, IC_{50} 2.4μM, $P<0.001$). This effect persisted when the aortic segments were incubated with NaHS (100nM-100μM) for 30min and the NaHS washed out before NADPH (100μM) was added (maximum reduction 58+7%, IC_{50} 0.4μM, $P<0.001$).
Discussion. These data show that H_2S scavenges O_2^- generated via Hx-XO or PG and additionally inhibits NADPH-dependent O_2^- production. These properties protect endothelial function against oxidative stress *in vitro*.

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Carvedilol induces greater control by PDE3 of β_2- than β_1-adrenoceptor-mediated inotropic effects in human failing myocardium while PDE4 has no effect

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Introduction. Phosphodiesterases PDE3 and/or PDE4 control ventricular effects of catecholamines in several species but their relative effects in failing human ventricle are unknown.
Aims. To determine whether the PDE3-selective inhibitor cilostamide or PDE4 inhibitor rolipram modify the positive inotropic and lusitropic effects of catecholamines in human failing myocardium.
Methods. Ventricular trabeculae from freshly explanted hearts of 5 non-β-blocked, 11 carvedilol-treated and 15 metoprolol-treated patients with terminal heart failure were paced to contract at 1Hz. The effects of (-)-noradrenaline, mediated through β_1-adrenoceptors (β_2-adrenoceptors blocked with ICI118551), and (-)-adrenaline, mediated through β_2-adrenoceptors (β_1-adrenoceptors blocked with CGP20712A), were assessed in the absence and presence of PDE inhibitors.
Results. The positive inotropic and lusitropic effects of (-)-noradrenaline were potentiated 2-5-fold by cilostamide in metoprolol-treated but not in non-β-blocker-treated patients. Cilostamide caused 3-5-fold and 10-35-fold potentiation of the inotropic and lusitropic effects of (-)-adrenaline in metoprolol-treated and carvedilol-treated patients respectively. Rolipram did not affect the inotropic and lusitropic potencies of (-)-noradrenaline or (-)-adrenaline.
Discussion. Treatment of heart failure patients with carvedilol, and to a lesser extent with metoprolol, facilitates PDE3-induced reductions of the inotropic and lusitropic effects mediated through β_2-adrenoceptors. PDE4 had no effect.

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Impact of the putative nitroxyl (HNO) donor 1-nitrosocyclohexyl acetate (1-NCA) on the intact rodent heart

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Introduction. HNO (a redox sibling of NO•) elicits vasodilator, antihypertrophic and superoxide-suppressing actions. HNO however is resistant to scavenging by superoxide, does not develop tolerance to its vascular actions, and is highly thiophilic. The impact of the putative mixed HNO/NO donor 1-NCA on left ventricular (LV) function however remain largely unresolved.
Aims. We tested the hypothesis that 1-NCA enhances LV function, and sought insight into its effectiveness in the context of diabetes-induced LV dysfunction.
Methods. The acute dose-response curve to 1-NCA (10^{-7}-10^{-1}mol) following U46619 preconstriction was determined in isolated adult male Sprague-Dawley rat hearts subjected to Langendorff perfusion *in vitro*, on coronary flow and LV function. In parallel studies, 1-NCA (83mg/kg/daily i.p. for 4wks) was administered to both normal and streptozotocin-diabetic adult male FVB/N mice (commencing after 4wks diabetes), with LV function determined on both echocardiography and LV catheterisation.
Results. Bolus 1-NCA doses significantly increased coronary flow (by 3.9±0.9ml/min, n=3 at the highest dose studied) in the normal rat heart *in vitro*. Although modest trends for enhanced LV function were evident, these were not significant. The HNO scavenger L-cysteine (4mM, n=7), but not the NO• scavenger hydroxocobalamin (50μM, n=3), significantly blunted 1-NCA coronary vasodilatation. Chronic treatment with 1-NCA significantly reduced LV superoxide generation by 28±4% ($P<0.0005$, dihydroethidium fluorescence), but did not affect other measures of LV systolic or diastolic function in non-diabetic mice. In contrast, diabetes-induced impairments in LV diastolic function were significantly ameliorated by chronic 1-NCA, on both peak atrial (A) wave velocity and the ratio of peak early (E):A wave velocities, descriptors of LV filling (both $P<0.05$, n=4-7).
Discussion. In conclusion, 1-NCA elicits HNO-dependent coronary vasodilatation, accompanied by superoxide suppression. Its ability to enhance LV function in the intact heart may be selective for settings where diastolic function is impaired.

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Compound 21, a synthetic AT_2 receptor agonist, evokes neuroprotection in a conscious rat model of ischaemic stroke. Lachlan J Facey[1], Jennifer K Callaway[2], Robert E Widdop[1] & Claudia A McCarthy[2]. [1]Dept of Pharmacol, Monash Univ, Clayton, VIC. [2]Dept of Pharmacol, Univ of Melbourne, Parkville, VIC

Introduction: Central AT_2 receptor stimulation using the peptide agonist CGP42112 has been shown to be neuroprotective (McCarthy *et al* 2009). The current study has used the non-peptide, Compound 21, as an AT_2R agonist.
Aim: We hypothesised that the peripheral administration of Compound 21 would evoke neuroprotection in an ischaemic model of stroke.
Methods: Animals were anaesthetised with ketamine (75mg/kg; i.p) & xylazine (10mg/kg; i.p) for stereotaxic insertion of a guide cannula. Ischemia was induced in conscious rats by administering endothelin-1, through the guide cannula, to the right middle cerebral artery. Compound 21 (1 mg/kg/day), candesartan (0.5 mg/kg/day), or vehicle, was administered for 4 days via an osmotic mini-pump (i.p), starting 24 hours before stroke induction. For osmotic pump implantation, animals were re-anaesthetised 24 hours prior to stroke using isoflurane inhalation. Motor and neurological deficit was assessed at 24 and 72 hours post stroke. Infarct volume was measured and immunohistochemistry performed post mortem. Neurite outgrowth was assessed in PC12W cells incubated with either compound 21 (10^{-7}M) or Ang II (10^{-7}M) alone and in combination with PD123319 (10^{-5}M).
Results: When administered prior to stroke, Compound 21 and candesartan reduced cortical infarct volume compared to vehicle by approximately 58% and 54%, respectively ($P<0.05$ vs. vehicle), independent of any changes in blood pressure. Moreover, both treatments significantly improved behavioural deficit 24 hours after stroke. When Compound 21 was administered six hours after stroke (3 mg/kg, i.p. bolus), and continued for 3 days (0.3 mg/kg/day), it caused less neuroprotection than seen with pre-treatment. Compound 21 significantly increased neurite outgrowth (50.5% of cells expressing neurites, $p<0.001$ vs. vehicle), in PC12W cells compared to vehicle (13.54%), and this was abolished with the co-administration of PD123319.
Discussion: The current study has shown, for the first time that systemic administration of the novel AT_2 receptor agonist, Compound 21, is as protective as AT_1R blockade in a physiologically relevant conscious model of stroke.

McCarthy CA et al (2009) *Stroke*, 40, 1482-1489

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Much ado about NSAIDs

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Although in clinical practice since 1899 the chameleon nature of nonsteroidal anti-inflammatory drugs (NSAIDs) continues to provide challenges. NSAIDs are a homogeneous group in terms of inhibition of the cyclooxygenase enzymes COX-1 and COX-2 but a chemically heterogenous group that includes derivatives of acetic, enolic, fenamic, propionic and salicylic acid and the diaryl heterocyclic COX-2 inhibitors. Metabolism is primarily hepatic variably involving functionalisation and conjugation reactions. Early studies established that the hepatic metabolism of the R(-) enantiomers of the 2-arylpropionates involved formation of a coenzyme A (CoA) thioester intermediate, which following racemisation and subsequent hydrolysis, yielded the pharmacologically active S-enantiomer. Formation of the CoA conjugate was catalysed by a hepatic microsomal ATP dependent long-chain acyl CoA synthetase. Chiral inversion is the major determinant of (R)-ibuprofen clearance in humans and we established that gender and hormonal factors did not influence metabolism via this pathway. Subsequent studies characterising NSAID glucuronidation established the presence of enzymes of the UDP-glucuronosyltransferases superfamily UGT1A and UGT2B7 in the human nephron. We established that fatty acids were potent inhibitors of glucuronidation and that inclusion of albumin in microsomal incubations improves the accuracy of in vitro-in vivo extrapolation for substrates of UGT1A9 and UGT2B7. The common involvement of UGT2B7 in the metabolism of NSAIDs and aldosterone led us to screen NSAIDs as inhibitors of aldosterone glucuronidation. Determination of the inhibition constant of five NSAIDs established a rank order of potency of inhibition of aldosterone glucuronidation as mefenamic acid>diclofenac>naproxen> indomethacin>S-ibuprofen. Further we established that >3months use of diclofenac was associated with greater arterial dysfunction in comparison to naproxen, indomethacin and ibuprofen thus alluding to a role in the cardiovascular toxicity of NSAIDs. Over the course of 113 years NSAIDs have been identified as gastrotoxic, nephrotoxic and now cardiovascular toxic drugs, is there more to come?

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The best of both worlds: creating an inclusive learning environment for experimental and clinical pharmacology

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Changes in teaching methods (e.g. from traditional to integrated teaching), the diversity of degree programs into which pharmacology is taught and the expanding knowledge base of the discipline have all contributed to the challenges of teaching pharmacology. In 2008 - 2009, a national audit of pharmacology curricula in Australia was undertaken (Hinton et al., 2010). Five degree programs (science, medicine, pharmacy, nursing and allied-health) were surveyed for course content, teaching methods and assessment. Similar subject areas were found to be taught across the five degree programs supporting the view that a generic pharmacology curriculum (or core curriculum) can be defined for science and health-related degree programs. Differences between degree programs, however, were found with respect to the breadth and depth of teaching within a given subject area and the pedagogical approach used for teaching and learning. In order to harness these differences, whilst retaining common core content, it is proposed that specific learning outcomes and levels of attainment expected for each degree program are articulated alongside curriculum content to create a flexible curriculum framework. Such a proposal could provide a national pharmacology curriculum that can be tailored to specific degree requirements. It offers several advantages. Defining the broad knowledge base of pharmacology preserves discipline identity and assists in curriculum mapping; articulating learning outcomes and standards ensures that the requirements of each degree program will be met; and creating an inclusive learning environment facilitates the sharing of valuable expertise and innovative teaching practices to enhance the standard of teaching and learning across experimental and clinical pharmacology.

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The devil is in the detail

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Teaching Pharmacology across different courses presents some issues in setting the level of difficulty, depth and breadth. It also raises the idea of having different tasks of varying levels of cognitive complexity with matched activities across any one course; with low level activities relying on recall and recognition to high activities tasks that engage students in reasoning and analysis. While the low cognitive complexity activities are likely to be shared across courses, the high cognitive complexity activities are not. Moreover, the time devoted to each of these "depth of knowledge" activities could vary across different courses. Do we intrinsically "assign" different levels of depth of knowledge and parse the level of complexity across different courses? The level of recall and the basic application of pharmacological knowledge for medical students would be similar to science / biomedical science students; but, there would be divergence when it comes to extended thinking requiring the differentiation between a family of clinically used compounds to the detailed investigation of the pharmacodynamics characteristics of agents targeting a receptor system. Highly complex tasks for science / biomedical science students tend to construct models for research, while in clinical related courses such as medicine; students tend to solve a problem based on several alternatives finding the best outcome.

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Teaching the 'new' prescribers: The challenges of knowledge, attitudes and skills

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The extension of prescribing rights to multiple groups of health care professionals other than medical practitioners started in the USA in 1965 when Nurse Practitioners emerged in response to a shortage of physicians in rural and disadvantaged communities. In the UK 'nurse prescribing' was adopted nationally in 1998 and all nurses with a district nursing or health visiting qualification were eligible for training to prescribe from the Nurse Prescribers Formulary. Nurse independent prescribing was introduced in the UK 2002 and this was followed shortly after by nurse and pharmacist supplementary prescribing (2003); radiographers, podiatrists, chiropodists, physiotherapists and optometrists supplementary prescribing (2005); pharmacist independent prescribing (2006) and optometrists independent prescribing (2008). In New Zealand midwives were granted prescribing rights in 1998 while in Australia the first Nurse Practitioner was appointed in NSW in 2001. The other Australian States followed with the authorisation of prescribing rights to Nurse Practitioners in Victoria in 2000 and South Australia in 2002. Since that time it has been identified that the greatest education challenges facing higher education providers involved in the education of the Nurse Practitioners include meeting the needs of diverse groups of students with varied interests, differing levels of prior knowledge and varied clinical backgrounds. Development of pharmacology curricula, assessment methods, determination of prescribing skills and competencies and the incorporation of flexible learning environments is all impacted by an overarching background of National Competency Standards for the individual health care professions, the National Prescribing Service "Prescribing Competencies Framework", HealthWorkforce Australia's "Health Professionals Prescribing Pathway Project" and TEQSA. The educational challenges for the Discipline of Pharmacology are enormous and teaching and learning is but one aspect of the challenge.

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Myocardial stress and ischemic injury - sex and sex steroid influences

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The occurrence of myocardial infarction in women and men is associated with differential clinical outcomes relating to sudden cardiac death and progression to failure. For women there is also evidence that coincident diabetes and underlying cardiac hypertrophy selectively and negatively modulate the severity of response to a clinical event. An understanding of the mechanistic bases for these differences is currently lacking. Experimentally at the level of the single cardiomyocyte, we have identified fundamental differences in the contractile performance and in the handling of activator Ca^{2+}. We have also characterized sex differences in the ex vivo responses of isolated hearts to ischemic stress (Bell et al, 2008). These investigations suggest increased mechanical reserve in female hearts in association with upregulation of signalling through PI3K activation of Akt and also PKC activation of ERK1/2. When ischemic challenge is assessed in hearts which exhibit genetically-determined hypertrophy, these sex-specific response differences are blunted. Furthermore, when energy stress is induced to perturbs PI3K myocardial signalling, sex differences in the energy mobilization responses are observed. It is postulated that sex steroids play a role in determining/modulating sex-specific aspects of function, signalling and injury vulnerability. Recently we have reported that myocardial tissue expresses the enzyme aromatase - indicative of the capacity for local cardiac androgen-to-estrogen conversion. Our experiments with genetically manipulated rodents, where tissue aromatase expression has been suppressed or enhanced, provide evidence that altered sex-steroid conversion impacts on cardiac injury responses - including signalling, inotropy and arrhythmogenesis (Bell et al 2011). Further exploration of these sex-specific responses to myocardial stress will yield clinically relevant sex-targeted therapies for the ischemic heart.

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Y chromosome dependent blood pressure regulation in the SHRSP is mediated, in part, by the renal renin angiotensin system.

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Introduction. The Y chromosome influences blood pressure regulation with studies demonstrating introgression of the Y chromosome from a normotensive into a hypertensive rat strain reduces blood pressure by 10-15mmHg. One candidate gene implicated to contribute to the Y-chromosome dependent regulation of blood pressure is the Sry3 gene. It is located exclusively on the Y chromosome from the spontaneously hypertensive rat (SHR) and has been shown *in vitro* to interact with the renin angiotensin system (RAS); upregulating angiotensinogen, renin and ACE gene promoter activity. However, the functional consequence(s) of this interaction on renal function and renal RAS responses *in vivo* remains unknown.

Aims. We aimed to investigate the functional consequences of the interaction of the Y chromosome with the renal RAS *in vivo* using 16 week old normotensive (WKY), hypertensive (SHRSP) and 2 consomic strains; one in which the WKY Y chromosome was introgressed into the SHRSP background (SP.WKY_{Gla}Y) and vice versa (WKY.SP_{Gla}Y).

Results. Systolic blood pressure, measured by radiotelemetry, was lower in the SP.WKY_{Gla}Y vs SHRSP (195±5mmHg vs 227±8mmHg, n=8, $P<0.03$) and was higher in the WKY.SP_{Gla}Y vs WKY (157±3mmHg vs 148±3mmHg, n=8, $P<0.05$). The ratio of plasma Ang(1-7):Ang II was higher in the SHRSP when compared to all other strains ($n\geq5$, $P<0.01$). In addition, SHRSP had greater renal AT1R, AT2R and MasR mRNA gene expression ($P<0.005$ compared to WKY, $n>6$) which was not present in the SP.WKY_{Gla}Y. Renal blood flow responses to graded intrarenal bolus doses (1, 3, 10, 30, 100ng/kg) of Ang I and Ang(1-7) were blunted in the SHRSP when compared to both the SP.WKY_{Gla}Y and WKY ($P_{STRAIN}=0.0007$ and $P_{STRAIN}=0.0008$, respectively). Renal responses to intrarenal Ang II (1,3,10,30,100 ng/kg) were similar in all strains.

Discussion. This study provides novel evidence that the Y chromosome enhances the vasodilatory components of the RAS in the SHRSP which is restored following introgression of the WKY Y chromosome.

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Sex differences in the role of the renin-angiotensin system in the regulation of arterial pressure and renal function.

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Introduction: Women are protected from cardiovascular and renal disease relative to men, prior to menopause. The mechanisms are poorly understood, but evidence demonstrates that estrogen plays a protective role against cardiovascular disease in women. There are major sex-differences in the expression levels of components of the renin-angiotensin system (RAS) and also differences in the way males and females respond to stimulation and inhibition of the RAS under physiological and pathophysiological circumstances.

Our studies: We have demonstrated that the depressor RAS pathways are enhanced in females and that the angiotensin type 2 receptor (AT_2R) has a vasodilatory role in the response to chronic angiotensin II (AngII) infusion in female but not male rats. This action of AngII to decrease arterial pressure in females was mediated via an AT_2R, estrogen dependent, mechanism. Thus, the AT_2R plays a role in countering the pressor actions of AngII at the angiotensin type 1 receptor in females. Importantly, no differences in acute pressor responses to AngII were observed between the sexes, rather the differences become apparent during chronic RAS activation in females. However, we have shown acute sex-differences in the contribution of the depressor arm of the RAS to the renal mechanisms that control extracellular fluid homeostasis (pressure-natriuresis; tubulo-glomerular feedback) and hence arterial pressure. Thus, RAS depressor mechanisms in the kidney, which promote salt and water excretion, confer protection from increases in arterial pressure in females.

Future perspectives: Our work now focuses upon the role of the RAS during pregnancy and in ageing, to determine if the RAS depressor pathways are potential therapeutic targets for the treatment of hypertension and renal disease.

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Gene-environment interactions and sexual dimorphism in mouse models of brain disorders

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Huntington's disease (HD) is a tandem repeat (CAG) expansion disorder involving a triad of psychiatric, cognitive and motor symptoms. In a transgenic mouse model of HD we have demonstrated that environmental enrichment (which enhances sensory stimulation, cognitive activity and physical exercise) can delay onset of the affective, cognitive and motor endophenotypes. Detailed investigations of these HD mice have also revealed sexually dimorphic depression-like behaviours which precede cognitive and motor deficits. The female HD mice demonstrate early affective abnormalities which can be rescued by administration of clinically effective antidepressant drugs as well as increased physical exercise. This is consistent with the clinical sexual dimorphism in the incidence of depression. We have thus been able to investigate these mice as a model of depression and HD and have discovered various molecular abnormalities, including specific deficits in neurotrophin, serotonergic and dopaminergic signalling pathways. A selective subset of these molecular changes have been found to be sexually dimorphic and, along with a possible role of sex hormones, may help explain the depression-like behaviours in these female HD mice.

These findings have been extended to additional environmental factors (e.g. stress), neuroendocrine modulators (e.g. sex hormones) and animal models of other brain disorders. For example, we have characterized behavioural and molecular changes in knock-out and knock-in mice modelling autism spectrum disorder. These models involve X chromosome gene mutations and thus they exhibit strong sexual dimorphism, reflecting the high clinical incidence of autism in boys.

Together with epidemiological studies and clinical trials, our findings are informing mechanisms of pathogenesis and the subsequent design of future intervention studies for these sexually dimorphic brain disorders. The models of gene-environment interactions can also be used to identify novel molecular targets for 'enviromimetics', drugs which mimic or enhance the beneficial effects of environmental stimulation.

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Association of the human Y chromosome and cardiovascular disease risk

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There is a sexual dimorphism in the incidence and prevalence of coronary artery disease (CAD) - men are more commonly affected than age-matched women. We explored the role of the exclusively male Y chromosome in CAD in the context of this sexual inequity.We genotyped 2978 biologically unrelated white men from the cross-sectional BHF - Family Heart Study and the prospective West of Scotland Coronary Prevention Study for 11 markers of the male-specific region of the Y chromosome. Based on this information each Y chromosome was then tracked back into one of 13 ancient lineages defined as haplogroups. We then examined for associations between common Y chromosome haplogroups and the risk of CAD in both populations. This was followed by functional analysis of Y chromosome effects on monocyte and macrophage transcriptome in 255 British men from Cardiogenics Study.
Of nine observed haplogroups, two (R1b1b2 and I) accounted for approximately 90% of variation in the Y chromosome. We found that carriers of haplogroup I had an approximately 50% higher age-adjusted risk of CAD than men with other Y in both populations [OR: 1·56 (95% CI: 1·24-1·97), P=0·0002]. The association between haplogroup I and increased risk of CAD was independent of traditional cardiovascular risk factors. Analysis of macrophage transcriptome revealed that 19 molecular pathways showing strong differential expression between men with haplogroup I and other lineages of the Y chromosome were interconnected by common genes related to inflammation and immunity, and that some of them have a strong relevance to atherosclerosis.
Our findings indicate that the Y chromosome determines not only maleness but also the risk of cardiovascular disease, possibly through effects on immunity. Our observations may also explain some of the "male disadvantage" in susceptibility to CAD.

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Understanding idiosyncratic adverse drug reactions through integrative systems approaches

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Idiosyncratic adverse drug reactions (ADRs) remain intractable problems that continue to challenge clinicians and the pharmaceutical industry. The severest manifestations can cause irreversible morbidity in individuals and death; often with little warning. The inherently rare nature of these events and the absence of reliable animal models or in-vitro methods to study the mechanisms behind these ADRs render their occurrence largely unpredictable. While a number of theories may surround many idiosyncratic ADRs, they often involve consideration of a restricted part of the broader biological network of interactions (e.g. the role of hERG potassium channel inhibition in Torsade de Pointes (TdP)). Such reductionist approaches have generated valuable information; however they often fall short in accurately predicting vulnerability in patients or risks from specific medicines. This paper explores principles of off-target drug action within biological networks - complex adaptive systems. The features of these systems can explain the limitations of reductionism in explaining the underlying mechanisms for these rare toxicities and in accurately identifying "at-risk" individuals. Integrative systems approaches seek to identify important features and interactions in biological networks that together may explain vulnerability to idiosyncratic ADRs. These approaches rely on the wealth of knowledge that resides in disparate and growing data repositories. The growth in power of computational and information technologies and the development of tools and processes that can link information across these datasets can identify previously-unrecognised patterns of interaction among drugs and components of biological systems at a number of levels and may reveal the roles of specific targets, biological pathways and risk factors involved in the development of drug toxicity. The combination of adverse reaction and drug (chemistry, pharmacokinetic and pharmacodynamic) information with knowledge of variability among genes, proteins and biological metabolites presents opportunities for understanding the factors that conspire to produce toxicity.
Examples of integrative approaches to understanding drug toxicity will be presented together with our recent work that has linked human population pharmacovigilance data with computational chemistry data and ligand-protein interaction information to identify structural and biological components and potential biological pathways implicated in drug-induced liver injury, parasomnias and cardiac arrhythmia (TdP).

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Care track as a methodology and relevance for drug safety

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The landmark CareTrack study revealed that in 2009-10 Australians received appropriate care for 22 common conditions accounting for 40% of the burden of illness 57% of the time (95% CI, 54%–60%) of 35,573 eligible health care encounters). The range across conditions was 13% (alcohol dependence) to 90% (coronary artery disease) and for individual practitioners with more than 300 encounters, 32% to 86%. These data indicate room for improvement. Many hypothesise that feedback of quality of care data to individual clinicians in respect of their own patients will lead to improvements in achieving minimal standards of care.

Many of the conditions such as heart failure and type II diabetes are chronic, heavily reliant on multiple medications and more often affect the elderly where adverse drug reactions and interactions and medication errors are more likely.

Automatic data retrieval and checking against indicators of care using the general approach of the CareTrack study is possible with the advent of the electronic health record. Critical to the effort is a more systemised approach to developing and owning standards of care and associated indicators. These can then be used as tools that can be used to monitor and feedback quality of care to individual practitioners. Wider access to the standards of care not only to clinicians but also to their patients will drive improvement.

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Injectable and oral contraceptive use and cancers of the breast, cervix, ovary, and endometrium in black South African women: case–control study

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Background: Oral contraceptives are known to influence the risk of cancers of the female reproductive system. Evidence regarding the relationship between injectable contraceptives and these cancers is limited, especially in black South Africans, among whom injectable contraceptives are used more commonly than oral contraceptives.

Methods and Findings: We analysed data from a South African hospital-based case–control study of black females aged 18–79 y, comparing self-reported contraceptive use in patients with breast (n = 1,664), cervical (n = 2,182), ovarian (n = 182), and endometrial (n = 182) cancer, with self-reported contraceptive use in 1,492 control patients diagnosed with cancers with no known relationship to hormonal contraceptive use. We adjusted for potential confounding factors, including age, calendar year of diagnosis, education, smoking, alcohol, parity/age at first birth, and number of sexual partners. Among controls, 26% had used injectable and 20% had used oral contraceptives. For current and more recent users versus never users of oral or injectable contraceptives, the odds ratios (ORs) for breast cancer were significantly increased in users of oral and/or injectable contraceptives (OR 1.66, 95% CI 1.28–2.16, p,0.001) and separately among those exclusively using oral (1.57, 1.03–2.40, p = 0.04) and exclusively using injectable (OR 1.83, 1.31–2.55, p,0.001) contraceptives; corresponding ORs for cervical cancer were 1.38 (1.08–1.77, p = 0.01), 1.01 (0.66–1.56, p = 0.96), and 1.58 (1.16–2.15, p = 0.004). There was no significant increase in breast or cervical cancer risk among women ceasing hormonal contraceptive use ≥10 y previously (p = 0.3 and p = 0.9, respectively). For durations of use ≥5 y versus never use, the ORs of ovarian cancer were 0.60 (0.36–0.99, p = 0.04) for oral and/or injectable contraceptive use and 0.07 (0.01–0.49, p = 0.008) for injectable use exclusively; corresponding ORs for endometrial cancer were 0.44 (0.22–0.86, p = 0.02) and 0.36 (0.11–1.26, p = 0.1).

Conclusions: In this study, use of oral and of injectable hormonal contraceptives was associated with a transiently increased risk of breast and cervical cancer and, for long durations of use, with a reduced risk of ovarian and endometrial cancer. The observed effects of injectable and of oral contraceptives on cancer risk in this study did not appear to differ substantially.

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Pharmacometrics methods to individualise dose

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Pharmacometrics has been defined as "the science of developing and applying mathematical and statistical methods to: characterize, understand, and predict a drug's pharmacokinetic and pharmacodynamic behaviour; quantify uncertainty of information about that behaviour, and rationalize data-driven decision making in the drug development process and pharmacotherapy."[1] The approach has been shown to be useful over the years to not only guide decision making in drug development, but more so in 'getting the dose right' in the clinical setting. The research presented here has mainly focused on the later and will showcase how the pharmacometrics approach can help find the right dose for therapeutic subgroups such as children and patients with cystic fibrosis[2-4]. At the same time it will also be shown how using a model-based approach can enhance study design. In particular these topics have been looked at for designing studies with sparse sampling, under clinical restrictions, for therapeutic drug monitoring purposes or for drugs with a narrow therapeutic index[4-6]. Lastly, I will present research into improving and enhancing the understanding of the currently used methods and clinical trial designs[7-10]. Further efforts have been made in the last years to support the application of this methodology through teaching and facilitating of user-friendly software[11].

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Preventing resistance of bacterial "superbugs" by synergistic combinations of available antibiotics

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Introduction. Resistant bacterial 'superbugs' present one of the three most serious threats to global health. As this severe problem is exacerbated by a long-term lack of new antibiotics, novel strategies to rationally optimise combinations of available antibiotics are very promising to combat this global health crisis. *Pseudomonas aeruginosa* is one of the most problematic Gram-negative 'superbugs' and has an exceptional capacity to become resistant during therapy.
Aims. To develop innovative strategies how to identify, prospectively optimise, and rationally translate synergistic antibiotic combinations that maximise bacterial killing and prevent resistance *via* latest experimental and mechanism-based mathematical modelling approaches.
Methods. *In vitro* time-kill studies assessed combinations of two β-lactam antibiotics or of a β-lactam and an aminoglycoside antibiotic with different receptor occupancy patterns against wild-type and hypermutating *P. aeruginosa* strains. Viable counts of susceptible and 'resistant' bacteria were quantified in static time-kill studies over 48 h. Antibiotic dosage regimens were prospectively optimised in dynamic hollow fibre *in vitro* infection models over 10-days. Antibiotic concentrations were determined by LC-MS/MS. Novel, mechanism-based mathematical models accounted for specific receptor occupancy patterns and resistance mechanisms.
Results. Combinations of β-lactam antibiotics binding penicillin-binding proteins (PBPs) 1 to 4 achieved substantial and synergistic killing against a high inoculum of *P. aeruginosa*, whereas monotherapy with all tested anti-pseudomonal cephalosporins achieved limited or no killing over 48 h. Optimal combinations of β-lactams with different resistance mechanisms achieved synergistic killing without resistance in 10-day hollow fibre models. Imipenem binds all PBPs in *P. aeruginosa* and led to rapid killing, but also to non-replicating persisters which could be eradicated without resistance by double β-lactam combinations or tobramycin.
Discussion. Synergistic combinations of available and safe antibiotics were prospectively optimised to maximise bacterial killing and prevent resistance via latest experimental and mechanism-based mathematical models. This approach holds excellent promise to combat resistant bacterial 'superbugs'.

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Positive and negative outcomes from medicines in older adults

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Introduction. In older adults, evidence on medicines exposure and risks is commonly obtained from post-marketing studies. Aims. To discuss the role of pharmaco-epidemiological studies in determining positive and negative medicine-related outcomes in older adults. Methods. Pharmacoepidemiology employs epidemiological methodologies to study the utilisation and effects (beneficial or adverse) of medicines in large populations. Observational studies commonly utilise data from large cohort studies, healthcare and clinical databases, and drug and disease registries. Results. In older populations, pharmaco-epidemiological studies are essential to assess the medicine-related adverse outcomes, and to evaluate efficacy of medicines in real-world settings. This is because the representation and representativeness of older people in published randomised clinical trials is generally poor. However, quantification of causality requires judicious interpretation of observational data. For instance, pharmaco-epidemiological studies can be influenced by the study population's characteristics, and their country or region's system of health care. Therefore, testing the hypotheses across different countries with different health systems is essential to achieve external validity and generalisability. Discussion. Observational studies are critical in quantifying medication-related outcomes in older adults. Where possible, findings of observational studies should be tested with pragmatic real-world interventional trials specifically designed for older adults.

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Allosteric modulation of G protein-coupled receptors

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It is now well established that virtually all G protein-coupled receptors (GPCRs) possess topographically distinct allosteric binding sites that can be targeted to modulate the activity of orthosteric ligands. Accordingly, recent years have seen a dramatic increase in the discovery of allosteric GPCR modulators. A key challenge to the field is the means to optimally describe allosteric effects in a manner that can capture experimentally observed observations and facilitate enriched structure-activity studies and/or inform drug candidate selection matrices. One approach to this challenge is to assign numbers to allostery using operational modeling. Such models describe GPCR allosterism minimalistically in terms of modulator affinity (K_B) for the free receptor, modulation of the binding (α) and/or signaling (β) of the orthosteric ligand, and intrinsic agonism (τ_B) of the allosteric modulator itself; it is apparent that most allosteric ligands are likely to display mixtures of these properties in a cell-dependent manner. Other recent paradigms that have emerged from the study of GPCR allostery are the concepts of allosteric ligand signaling bias (functional selectivity) and probe-dependence, with both having major consequences for novel drug discovery programs. Additionally, amongst the growing categories of ligands for GPCRs, a novel type of ligand has emerged, the bitopic ligand, i.e., compounds composed of distinct orthosteric and allosteric pharmacophores joined by an appropriately chosen linker. An advantage of such ligands is the ability to ensure receptor activation/inactivation through an appropriately chosen orthosteric moiety, while inducing either subtype and/or functional selectivity through the allosteric moiety. With the recent solution of multiple Family A GPCR crystal structures, the possibility of more rational exploitation of novel binding pockets either above or below the orthosteric site promises to facilitate true structure-based drug discovery for allosteric GPCR ligands.

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Effect of an educational workshop on pharmacists' knowledge, attitudes and beliefs towards low back pain (LBP).

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Introduction. Pharmacists are among the most commonly sought health care providers among people with Low Back Pain (LBP) and are well positioned to provide appropriate management advice to these people.
Aims. This study aimed to investigate pharmacists' knowledge, attitudes and beliefs towards LBP and whether participation in an educational workshop can influence these.
Methods. Pharmacists were invited to attend an evidence-based 2 h educational workshop on LBP management. Knowledge, attitudes and beliefs towards LBP were evaluated before (pre-) and after (post-) the educational workshop using the "Pharmacists' Back Beliefs Questionnaire" (PBBQ) with items from two validated back beliefs questionnaires (Buchbinder et al, 2009; Symonds 1996). Participants indicated their agreement with statements about LBP on a 5-point Likert scale of 1 "Strongly Disagree to 5 "Strongly Agree". Preferred responses were based on current guidelines for the evidence-based management of LBP.
Results. Responses from 204 pharmacists participating in the educational workshop and who completed the pre- and post- PBBQ showed that the educational workshop led to significant changes to misconceptions regarding bed rest (median pre- and post- scores respectively (IQR): 3 (2-4) vs 1 (1-1) n=204; $p<0.001$) and the need for imaging in non-specific low back pain (median pre- and post- scores respectively: 3 (3-4) vs 1 (1-2) n=204; $p<0.001$).
Discussion. The provision of an educational workshop on the evidence-based management of LBP can significantly influence pharmacists' knowledge, attitudes and beliefs towards LBP so that it more closely aligns with current evidence-based guidelines. Given the positive results, the next challenge is to develop strategies to reach a larger number of pharmacists and to expand such interventions to allied health care providers.

Buchbinder R et al (2009) Spine 11:1218-1226.
Symonds TL (1996) Occup Med (Oxf) 1:25-32.

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Online support for pharmacology practical teaching

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Introduction. Typical UK BSc Pharmacology courses now deliver far fewer hours of practical teaching than a decade ago. Many university departments have, in part, replaced practical classes with computer simulations developed by third party organisations and thus have little control of the content of those simulations or what is being delivered to their students.
Aim. To develop a database of quality-assured traces from a variety of tissue/whole animal preparations typically used in undergraduate education which teachers can access, download and incorporate into their own teaching materials. Other resources such as textual descriptions, animations and video-recordings will also be made available together with a laboratory manual in the form of an e-book.
Methods: Design and build an online searchable repository and populate with quality-assured resources. Develop a laboratory manual in e-book format describing a range of exemplar student activities which can be developed using the resources available in the database.
Results: the structure and functionality of the database will be demonstrated and exemplar sections of the e-book will be described. As a result of this development pharmacology students will have access to accurate data from experiments investigating the effects of various drugs/drug combinations on a number of *in vitro* and *in vivo* pharmacological preparations (the start point will be the >1000 traces currently owned by the author). The development will support research-informed student learning by providing students with access to primary data sources, via teacher designed learning activities, which will support acquisition of knowledge and a variety of skills such as data handling (measurement, presentation, interpretation), experimental design and communication. The online laboratory manual will provide exemplars of learning activities.
Discussion. Making resources of this type freely available and accessible to teachers of pharmacology will benefit student learning by supporting a variety small group teaching sessions, lectures and independent learning activities.

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The development of an experiential and learning programme for undergraduate pharmacy students at Alcohol and Drug Services

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Introduction. Community pharmacists offer pharmacotherapy programs and are expected to detect, monitor and advise patients with substance abuse disorders. However, undergraduate students have only limited awareness of these disorders and their management by the time they graduate.
Aims. To develop, institute and evaluate a problem-based drug and alcohol educational program for students, the main component of which involves a 3-hr experiential placement at the Alcohol and Drug Service.
Methods. The alcohol and substance abuse educational and experiential program consisted of three lectures and a three-hour placement for 72 third year students. To assess impact the brief Substance Abuse Attitude Survey (BSAAS) and an alcohol and drugs knowledge-based questionnaire was completed by all participants. The difference between the pre and post scores was statistically compared. Students were also asked to complete a qualitative questionnaire about the placement.
Results. A total of 62 students completed the baseline surveys, with 42 of these surveys matched up to post-surveys, giving an overall response rate of 58%. Seventy-seven percent of respondents were female, with ages ranging from 20-36 years (M = 22.80, SD = 3.24). No significant differences were found between the pre and post BSAAS. However; the knowledge-based questionnaire had a significant increase in correct answers, ($p<0.005$), and decrease in 'don't know' responses, ($p< 0.005$). Forty-eight students (66%) completed the qualitative questionnaire. Sixty-three percent said that their attitude towards people with substance abuse issues had changed as a result of the placement. Common themes were that students attained greater appreciation of the challenge of fighting substance abuse and that negative stigma was unhelpful. All students recommended the placement for future pharmacy undergraduates.
Discussion. Despite limited quantitative evidence of increasing positive attitudes, students' knowledge about substance abuse significantly increased as of the program. Qualitative analysis suggests that the placement should be continued in future.

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An international survey of health literacy education within schools of pharmacy

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Introduction. Health literacy is defined as the ability to access, understand, appraise and communicate health information. Initiatives to improve doctors' awareness of health literacy challenges have been reported internationally, but the concept is relatively new to pharmacy. The incorporation of health literacy in academic pharmacy curricula has not been examined. Taking stock of current health literacy training will help to develop an understanding of the needs of the pharmacy profession in health literacy.
Aims. To examine methods for teaching health literacy in schools of pharmacy internationally.
Methods. An anonymous online questionnaire was developed to examine health literacy education in pharmacy curricula, with reference to key themes identified by our group's literature research. These included how health literacy is defined, if and when in a course it is taught, the expertise of those teaching it, resources used to enhance teaching, and perspectives on the importance of teaching health literacy. We targeted academics who taught within pharmacy degree courses from countries where English is the main language, identified through academic networks.
Results. Twenty-three academics participated from 21 schools of pharmacy in seven countries. Of these, 21 stated that health literacy was taught within their pharmacy degree, in four as a stand-alone topic. Drivers were predominantly professional practice standards and the scope of pharmacy practice in their country. The majority of respondents (16) stated that health literacy was taught later in the degree (third or fourth year). Small-group tutorials and lectures were the most commonly reported forms of teaching.
Discussion. Of our limited sample, the majority of schools of pharmacy reported teaching health literacy using a variety of teaching methods. The results will inform the content and structure for a health literacy educational package for pharmacists and pharmacy assistants in Australia that could be adopted internationally.

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Development of a national medication safety online training course: because safety is no accident

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Introduction. The need for a broad-reaching training program on medication safety was identified by national organisations.
Aim. To develop a modular online medication safety training course suitable for doctors, nurses and pharmacists working in the acute care setting and students looking towards hospital practice.
Methods: Existing medication safety material, including a previously developed online program, was reviewed by medication safety experts who identified priority themes for the new training program. Examples of medication errors from Australian practice were collated. Content for the program was written then reviewed by medication safety experts prior to building into a website. Once built, the program was tested by education and training experts and end-users from each professional group.
Results. Seven learning modules and two case studies were developed. The themes of the modules are: understanding medication safety; types and causes of medication errors; wrong drug errors; wrong route errors; intravenous errors, formulation errors and communication. The modules aim to ensure participants understand why errors occur, how to avert them in their own workplace and what systems are in place to assist safe medication management. The modules include activities and real examples of errors to engage the user and encourage reflection. The interactive case studies illustrate the safety points made in the modules. Users are provided with a certificate at completion of each module and educators are able to monitor students' progress.
Discussion. An online medication safety training course has been developed that provides a broad overview of safety issues for all professionals involved in medicines management in acute care. The interdependency of healthcare professionals when managing medicines is a key message. The next steps regarding the use of this educational tool include widespread implementation across Australian healthcare, evaluation of the tool and, upon proven success, development of further modules and case studies.

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Experimental Design and Statistical Analysis in Intermediate Medical Sciences Curricula – A Pilot Study

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Introduction. Graduates of pharmacology and other medical science disciplines are expected to have sufficient skills to understand experimental design, collect and analyse data, and draw conclusions from experimental results. To achieve these graduate outcomes, students must learn the tools for quantitative literacy and research enquiry in science. However there is often a gap between when students learn experimental design and statistical analysis (EDSA) - often in junior units of study - and when they use them later in their studies, work or research (Gordon and Nicholas, 2010).
Aims. To develop, implement and evaluate a pilot module for EDSA, integrated in a discipline-specific context in an intermediate level medical science curriculum.
Methods. Pharmacology and physiology unit coordinators were surveyed on their expectations of graduates in EDSA skills. Responses were used to help develop two lectures and two tutorials which were delivered across semesters 1 and 2, to convey the basics of EDSA, along with discipline-specific experimental scenarios and data. A brief survey of student confidence in EDSA was conducted in semester 1, and a formative quiz (7 questions) to assess statistical understanding before and after the lecture and tutorial was carried out in semester 2.
Results. Survey results indicated that only 59% of the cohort had undertaken training in EDSA prior to their intermediate year, and of those, 25% were only fairly confident and 66% only a little confident in applying their knowledge. A statistically significant increase in the average score on the formative quiz was demonstrated after the EDSA lecture and tutorial compared with before.
Discussion. The EDSA pilot module was effective in improving student skills and confidence in using EDSA in medical sciences. Further integration of EDSA into discipline-specific practicals, and evaluation of student skills and confidence, is required.

Gordon, S. & Nicholas, J. (2010). Int. J. Innov. Sci. Math. Ed., 18:14-25.

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Does evidence-based education on complementary medicines change students' attitudes and likelihood of recommending them in a pharmacy setting?

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Introduction. Curtin University introduced a core evidence-based complementary medicine (CM) unit into the 3rd year pharmacy syllabus in 2010, consistent with Australian Pharmacy Council accreditation standards (2009) for pharmacy curriculum content. The unit was designed to provide students with the knowledge and ability to incorporate CMs into their pharmacy practice, thus meeting increased consumer demands for CMs and in keeping with pharmacists' professional responsibilities.
Aims. Research was conducted to evaluate the impact of the evidence-based CM unit.
Methods. Pre- and post-unit surveys were administered over two consecutive years to assess changes in students' attitudes, knowledge and likelihood of recommending CMs following evidence-based CM education.
Results. CM education resulted in a positive change in pharmacy students' perceptions towards CMs and increased their willingness towards recommending CMs within a pharmacy setting. Survey results show that completion of the unit led to a statistically significant positive change towards a belief that there is evidence for a number of CMs for the treatment of several conditions, diseases or for symptom management [2010, p=0.002; 2011, p=0.027] and that there is scientific evidence to support efficacy of many CMs, beyond a placebo effect [2010, p=0.002]. There was also a statistically significant shift in students' personal confidence in making recommendations to customers as part of a pharmaceutical care plan [2010, p=0.000; 2011; p=0.000].
Discussion. The evidence-based CM unit at Curtin University proved valuable in equipping pharmacy students with the ability and willingness to make appropriate patient-specific CM recommendations as part of their professional pharmacy practice, taking evidence on efficacy, safety and place in therapy into consideration, and in line with pharmaceutical care educational guidelines for pharmacy students. This educational initiative is a valuable step forward in bridging the gap between the identified need for greater evidence-based CM instruction and the reality of current pharmacy educational practices.

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Use of an audience response system and collaborative learning increases engagement in post graduate pharmacy.

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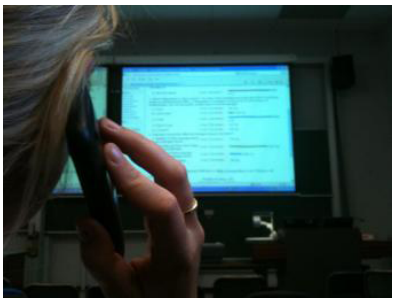

Introduction. Over the last few years one of the most successful implementations of technology in the learning environment has been the use of audience response systems (Cain, Black, & Rohr, 2009). These systems are shown to increase student engagement in a variety of learning environments. In my post graduate pharmacy course I detected a need for an intervention with the ability to engage students while listening to peer presentations.
Aims. I aimed to increase the benefit students derived from student presentations in terms of student engagement.
Methods. A series of clinical-style cases were devised as motivational examples to increase immediacy and highlight the relevance of the material to students. Student groups were required to present a case in turn each week. Each student within a group had to set an MCQ on an important point from the case. The whole class was tested at the end of each presentation using an audience response system (Votapedia, www.urvoting.com).
Results. Student participation (quiz responses) averaged 72% of the class over semester (range 53 – 90%, SD = 12%). In structured interviews students revealed they found setting questions with Votapedia increased the meaning they attributed to their presentation and that collaborative learning provided opportunities for increasing their own understanding.
Discussion. The participation rate and student feedback indicate how students valued the activity. This strategy provides an opportunity to demonstrate the benefit of collaborative learning as well as the use of technology to increase student engagement with the curriculum.

Cain J, Black EP, Rohr J (2009). An audience response system strategy to improve student motivation, attention, and feedback. Am J Pharm Educ 73(2): 21.

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Safety before efficacy? Australian pharmacists' attitude to homeopathic products in pharmacy

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Introduction. Despite Cochrane reviews concluding there is no evidence for the efficacy of homeopathic preparations, they are sold in most community pharmacies in Australia. This availability may put professional credibility at risk.
Aims. This study explored factors that influence pharmacists' preparedness to recommend, or at least make available, homeopathic products.
Methods. A mixed method approach was used incorporating two focus groups of South Australian pharmacists with a variety of backgrounds (n=13); telephone interviews with pharmacists from each state and territory (n=18), and a cross-sectional survey for community pharmacists (n=185). Non-parametric tests were used to identify significant patterns in attitudes toward OTC and homeopathic products.
Results. More pharmacists ranked efficacy above safety in OTC products and were more likely to discourage homeopathic product use, than those who ranked safety before efficacy. Almost all (96%) indicated that they would like the public and health colleagues to think of them as a source of information about all medicine and health products. Pharmacists believe that they will only sell a product if it will be useful for a patient, with homeopathic products sometimes regarded as a "safe" option for consumers. Some pharmacists see a place for these products if consumers seek them, although few would personally recommend them. Those who did not support their provision were concerned with the reputation of the profession. The impact of homeopathic products on business profitability was found to be minimal.
Discussion. Pharmacists want to be seen as caring for the health of consumers and as medication experts. Pharmacists want access to reliable information sources, education, and desired that undergraduate education should include skills to critically evaluate evidence for a range of health products. There is a strong argument for removing homeopathic products from pharmacy based on the lack of evidence, lack of commercial return and risk to credibility.

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Exploring community pharmacists' views on medication-safety management

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Introduction: Preventable harm from medicine use in the community continues to occur despite the best intentions of healthcare professionals and strategies to support quality use of medicines. Incident Reporting Programs (IRPs) can identify system weaknesses and generate strategies to prevent medication-safety incident (MSI) recurrence. IRPs depend on healthcare professional engagement. An IRP was offered to pharmacists alongside a medication-safety campaign in 2011; however, few pharmacists engaged. This may stem from pharmacists' perceptions of IRPs and the profession's role in reporting. Beliefs of pharmacists towards current MSI management have not been explored.
Aims: To gain insight into (1) community pharmacists' perspectives on management and response to MSIs and (2) barriers and facilitators to IRP participation.
Methods: Semi-structured interviews involved presentation of 1-2 MSI vignettes followed by discussion of contributing factors. Workplace responses to these were explored. The interviewer provided education on how system failures contribute to MSIs and the potential for IRPs to inform systems change, followed by reflection on beliefs of MSI management and the role of IRPs. Barriers and facilitators to IRP participation were discussed.
Results: Interim analysis of 11 interviews reveals blame culture as a factor in typical workplace responses. Lack of time, competing priorities and fear of punishment for MSI disclosure were reported as barriers to IRP participation. Facilitators included education on the systems approach to MSIs, as well as incentives (e.g. continuing professional development points).
Discussion: Our findings suggest pharmacists possess a broad range of beliefs and approaches towards MSI management depending on individual workplace culture. Future medication-safety initiatives will need to consider pharmacists' beliefs and needs, including education and additional resources and incentives for reporting.

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Improving consumer access to medicines: innovative medicines reclassification in New Zealand (NZ)

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Introduction. Prescription to non-prescription medicines reclassification is generally regarded as a positive development to increase consumer access to medicines and relieve limited health resources, although reclassification rates have slowed in many developed countries. Despite a small population and negative pharmaceutical industry environment, reclassification is progressing in NZ. Recent changes include influenza vaccination, and first-in-world reclassifications such as calcipotriol and trimethoprim.
Aims. To ascertain why NZ is one of the most progressive developed countries regarding medicines reclassification.
Methods. Two main sources of data were used. Interviews were conducted with 12 purposively selected key informants on NZ medicines reclassification, exploring barriers and enablers for reclassification. Secondly, analysis of minutes of the Medicines Classification Committee (1990-2011), and related documents provided reclassification rates and insight into deliberations around each case. The experience of the first author in reclassification in NZ was also used. A heuristic approach was used to thematically analyse the content of the transcribed interviews, which were then compared with 54 similar interviews from other countries including Australia, the United Kingdom, the United States and Japan.
Results. Multiple enablers to reclassification exist in NZ, with the key themes arising from the interviews including: the pharmacist-only medicine category; a 'can do' approach; the smallness of the NZ market; openness to change; trust in pharmacy and consumers; harmonisation with Australia; and facilitation by key individuals. Mandatory training, and non-sponsor reclassification (by a pharmacy retail group) enable reclassification. The pharmacy retail group has driven four reclassifications, with another pending.
Discussion. Innovation, as has occurred in NZ, may provide useful insights to move reclassification forward internationally. Research is required to ascertain the effect of non-prescription provision of these medicines on patient outcomes.

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Community-acquired pneumonia: why aren't national antibiotic guidelines followed?

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Aims: To assess adherence to the Australian Therapeutic Guidelines (TG14) for the empirical management of community acquired pneumonia (CAP), and explore the potential barriers affecting adherence to these guidelines.
Methods: Medical records were reviewed for all patients who were diagnosed with pneumonia within 24 hours of presentation (either admitted or not) at the Royal Hobart Hospital (RHH) from June 2010 to March 2011. A survey to identify potential barriers affecting adherence with TG14 was distributed to prescribers in the RHH's emergency department and medical units.
Main measuring outcomes: The adherence rate to TG14 for the management of CAP in terms of selected antibiotic, route of administration and dose; and the extent of doctors' agreement with the statements in the survey regarding the potential barriers.
Results: A total of 193 patients were assessed. The overall adherence to TG14 for the empirical antibiotic management of CAP was 23.8% (25.5% and 22.6% for patients with severe and non-severe CAP, respectively).Twenty-nine different antibiotic regimens was utilised during the audit period. Ceftriaxone-based therapy was prescribed to 57% and 30% of patients with severe and non-severe CAP, respectively. The response rate to the barriers survey was 50.9%; of those who responded 46.4% thought the influence of senior doctors could be a factor affecting junior doctors' adherence to the guidelines. Other barriers noted were a lack of guideline awareness (39.3%), the requirement to calculate to assess the severity of CAP (35.7%), and the existence of other guidelines that conflict with TG14 (28.6%).
Conclusion: Adherence with TG14 was poor for the treatment of CAP. Efforts to improve this should consider the potential barriers that hinder adherence.

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Implications of the Personally-Controlled Electronic Health Record for community pharmacy

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Introduction. International developments embrace the concept of electronic health records for consumers, aiming to reduce fragmentation in health data. The Australian Government has driven the national implementation of Personally-Controlled Electronic Health Records (PCEHRs) from 1st July 2012. Despite national and international data addressing the impact of e-health records on logistical and professional aspects of practice from doctors' and nurses' perspectives, data regarding the impact on community pharmacy is almost non-existent.

Aims. To determine, in the months prior to the launch, community pharmacists' perceptions about the developments and how they might integrate PCEHRs into pharmacy practice.

Methods. Semi-structured interviews of 20-25 minutes' duration were undertaken during March-April 2012 with 25 pharmacy owners and managers from 24 community pharmacies in Perth, Western Australia. Independently-owned and 'banner group' pharmacies were included from a range of suburbs. Interviewees were briefed about the PCEHR, before exploratory questions regarding the potential integration, benefits and challenges of the system in pharmacy practice. Data were recorded, transcribed and thematically analysed.

Results. Most pharmacists perceived benefits in enhanced access to patient data, and expected system flexibility to record clinical activities and health services. Patients' control over their data management was a concern, potentially resulting in incomplete and untrustworthy data, with potential for litigation of health professionals for decisions made on incomplete data. Concerns were also raised about workload, technical upgrades and work flow. The pharmacists called for remuneration, medico-legal guidelines and boundaries, and clarification of roles and responsibilities. The pharmacists universally voiced a need for PCEHR training.

Discussion. Awareness of the perceived benefits and challenges with the PCEHR will advise Australian practice guidelines, training priorities and policies to assist with adoption and optimal use of PCEHRs by community pharmacists. Training priorities and practice guidelines should address ethical data management and optimal use of electronic health records for clinical services.

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Challenges and safety concerns for community pharmacy personnel in the provision of services to young people

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Introduction. Young people aged 12-25 are a population with specific health needs distinct from those of either children or adults. Research indicates that these additional considerations can present challenges to primary healthcare providers. This study explored provision of services to youth from the perspective of community pharmacy personnel.

Aims. To explore challenges faced by community pharmacists and pharmacy staff in the provision of services to young people.

Methods. Questionnaires were distributed to pharmacy personnel at 500 randomly selected pharmacies across New Zealand. In addition to quantitative data collected from youth-health related questionnaire items, an open question section collected qualitative data on i) participants' comments and experiences, ii) participants' suggestions for development of services for youth. These data were analysed in NVivo using a general inductive approach to identify barriers, facilitators, suggestions and training needs. This presentation will describe themes relating to safety concerns and challenges.

Results. Three mail shots yielded response rates of 50.5% for pharmacists and 37.1% for pharmacy staff. Just over a third of participants (n = 171) answered the open questions. Common concerns relating to safety included beliefs that young people are less health literate, and seek information from unreliable sources such as peers or the internet. Young people were also reported to be less adherent, and more likely to require referral. Participants were concerned about broad safety and wellbeing issues for young people, particularly with regards to provision of sexual health services, and medicines with potential for misuse such as weight management products.

Discussion. Many challenges faced by pharmacists and pharmacy staff providing services to young people relate to concerns about the safety or appropriateness of medications and services for this vulnerable population. Clarification of guidelines and legislation regarding this age group is necessary to support pharmacy personnel and improve quality of care for youth.

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End-users' perceptions of the electronic medication repository, MedView.

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Introduction. MedView, developed as part of the Australian government's personally controlled electronic health record initiative, is an electronic medication repository that allows access to patients' medication histories across hospital and community settings. It was trialed in early 2012 in the Barwon region, Victoria.
Aims. To evaluate end-users' perceptions of MedView at "Pre-" and "Post-" implementation.
Methods. "Pre-MedView" evaluation involved semi-structured interviews and surveys with hospital doctors, general practitioners, and hospital and community pharmacists. "Post-MedView" implementation involved semi-structured interviews of healthcare practitioners and consumers. Interviews were transcribed verbatim and thematically analysed. Data was triangulated.
Results. "Pre-MedView": Health professionals (n = 38) were interviewed, and 14% (123/875) of surveys were returned. General practitioners (78%), hospital doctors (80%), hospital pharmacists (74%) and community pharmacists (52%) had difficulties accessing accurate medication information at least some of the time in current practice. MedView's perceived benefits included facilitating continuity of care between different healthcare settings, reducing medication misadventure and improving work efficiencies. The opt-in consent process in community was thought to negatively influence work efficiencies. Hospital pharmacists were likely to be the most frequent MedView users (58% likely to use for ≥ 60% of all patients). Perceived barriers included incomplete medication information in MedView. Concerns about possible misinterpretation of information and increase rate of medication errors were raised. "Post-MedView": Health professionals (n = 24) and 38 consumers participated. Most viewed MedView as user-friendly and easily accessible. Most consumers (84%) consented to sharing their medication information on MedView.
Discussion. MedView was perceived to have the potential to provide benefits, in terms of improving medication use and safety across the continuum of care. MedView was user-friendly. Refinement in roll-out and education strategies will be required for wider roll-out.

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Management of high blood pressure in pregnant women attending an Australian maternity hospital

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Background: Hypertension complicates approximately 10% of pregnancies in Australia. While the importance of managing severe hypertension is undisputed, management of mild-moderate hypertension remains contentious.
Aim: To review antenatal management of women with pre-pregnancy or pregnancy-induced hypertension (PIH), and compare outcomes.
Methods: An electronic search identified women, who gave birth at Mercy Hospital for Women in 2010, with an ICD code corresponding to any hypertensive disorder of pregnancy. A manual record review of eligible patients was performed to compare pregnancy and blood pressure(BP)management, perinatal and obstetric outcomes according to diagnosis of PIH and pre-pregnancy hypertension, and between women receiving treatment and their untreated counterparts.
Results: 513 women (9.1%) were identified as having hypertension – pre-pregnancy hypertension (n=59, Group1) or PIH (n=454). Among women with PIH, 76 (16.7%) received treatment (Group 2) and 378 did not (Group 3). The women in Group 1 were significantly older than those in Group 3 ($p < 0.001$). Group 2 had the most frequent development of pre-eclampsia (any form)(64%), shortest gestation (35 weeks and 2 days± 4 weeks and 3 days) and highest number of babies with fetal growth restriction(26%) in comparison with the other two groups; one baby in this group was stillborn. Group 3 had the only two incidences of placental abruption and one separate incidence of fetal death. The BP reading at which hypertension was diagnosed in Group 2 was significantly higher than in Group 3(152/94mmHg vs. 142/90mmHg;p=0.001). The diagnosis of hypertension was also significantly earlier in Group 2 (31 weeks and 1 day± 4weeks and 4 days vs. 35 weeks and 2 days ± 4 weeks;p=0.001
Conclusion: The majority of women who develop PIH are managed by close monitoring without antihypertensive medication. There is potential for improvement of BP management in both the untreated and treated groups.