Slide 1	# Variability Due to Genetic Differences Nick Holford Dept Pharmacology & Clinical Pharmacology University of Auckland	

Slide 2	# Objectives
	
	- Understand how between individual variation may contribute to :
	  » drug effectiveness
	  » adverse reactions
	  » drug interactions
	
	- Understand how identification of genes that contribute to disease pathophysiology can be used to:
	  » identify new targets for drug therapy and the
	  » potential to "customise" pharmacotherapy	

Slide 3	# Pharmacogenomics and Pharmacogenetics
	
	ICH-E15 Guideline : "Terminology and Definition in Pharmacogenomics", Step-4, 2007
	
	- **Pharmacogenomics** (PGx) is defined as the investigation of variations of DNA and RNA characteristics as related to drug response (genome science)
	
	- **Pharmacogenetics** (PGt) is a subset of PGx and is defined as the influence of variations in DNA sequence on drug response (clinical relevance)
	
	  » PGx and PGt are applicable to activities such as drug discovery, drug development, and clinical practice.
	  » Drug response includes drug disposition (pharmacokinetics) and drug effect (pharmacodynamics).
	
		ICH. DEFINITIONS FOR GENOMIC BIOMARKERS, PHARMACOGENOMICS, PHARMACOGENETICS, GENOMIC DATA AND SAMPLE CODING CATEGORIES E15 http://www.ich.org/products/guidelines/efficacy/efficacy-single/article/definitions-for-genomic-biomarkers-pharmacogenomics-pharmacogenetics-genomic-data-and-sample-cod.html

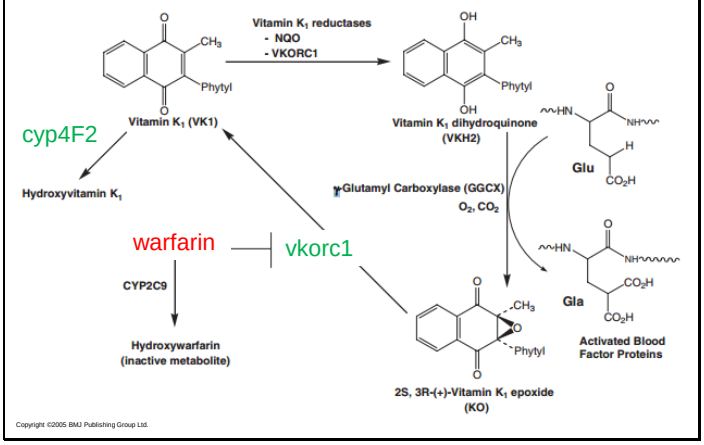
Slide 4	**Variability**
	
	- "Predictable"
	  » 50% of total
	  » Genetic
	  eg fast acetylator
	  » Environmental
	  eg renal function
	
	- "Unpredictable"
	  » 50% of total	

Slide 5	**Predictable Variability**
	
	- Pharmacokinetics (Clearance)
	  » Cytochrome P450
	  – CYP1A1
	  – CYP2C9,19
	  – CYP2D6
	  – CYP2E1
	  – CYP3A4, CYP3A5
	  » Non-Cytochrome Metabolism
	  – Acetylation
	  – Glucuronidation
	  – Purine breakdown
	  – Alcohol/Acetaldehyde Dehydrogenase
	  » Transporter
	  – P-Glyco-Protein
	  – Organic Anion
	
	- Pharmacodynamics (C50)
	  » Vitamin K Epoxide Reductase VKORC1
	  » Vitamin K1 Oxidase CYP4F2	

Slide 6	**CYP3A5**
	
	- **Marker Drug:** Tacrolimus
	- Clinically Relevant Drugs
	  » Tacrolimus
	  » Used as an immunosuppressant e.g. kidney transplant
	- Clearance by CYP3A5
	  » *1/1 genotype "expressors" have 2 fold increased CL
	  » *3/*3 "non-expressors"
	- Genotype guided dosing leads to better achievement of tacrolimus target concentration
	  » No difference in clinical outcome (study too small?) (Thervet 2010)
	- Ethnicity:
	  » 80% Caucasians but only 25% of African descent are *3/*3
	
		Thervet E, Loriot MA, Barbier S, Buchler M, Ficheux M, Choukroun G, et al. Optimization of initial tacrolimus dose using pharmacogenetic testing. Clin Pharmacol Ther. 2010;87(6):721-6.

Slide 7	**N-acetyl-transferase Acetylation** ▯ Marker Drug ***procainamide*** ▯ Clinically Relevant Drugs » anti-arrhythmic (procainamide) » anti-tuberculous (isoniazid) ▯ NAT genotype ▯ Ethnicity (% with low clearance) » Caucasian 50% » African 30% » Asian 10% 	
Slide 8	**UDP-Glucuronosyl-transferase Glucuronidation** ▯ Marker Drug ***bilirubin?*** » Hereditary hyperbilirubinaemia – Gilbert syndrome (3-10% of population) ▯ Clinically Relevant Drugs » Anti-cancer drug (Irinotecan) ▯ Active metabolite (SN-38) is eliminated by glucuronidation » Severe neutropenia and diarrhoea in 25% of patients » UGT1A1*28 predicts those who will get toxicity ▯ Ethnicity (UGT1A1*28) » African-American 40% » Caucasian 35% » Asian 5% (also UGT1A1*6 variant) 	UGT1A1*28 and UGT1A1*6 ethnic frequencies: http://www.wjgnet.com/1948-9366/full/v2/i1/14.htm
Slide 9	**Thio-Purine-Methyl-Transferase Purine Metabolism** ▯ Marker Drug: **6-mercaptopurine** ▯ Clinically Relevant Drugs » 6-mercaptopurine Childhood acute lymphocytic leukaemia » Azathioprine (prodrug for 6-MP) Crohn's disease, rheumatoid arthritis ▯ Severe toxicity (diarrhoea and neutropenia) » Thio-Purine-Methyl-Transferase (TPMT) genotype – Homozygote deficiency 1 in 300 – Heterozygote partial deficiency 1 in 10 – 10 fold dose reduction required in homozygotes ▯ Ethnicity? 	No clear benefit of using TPMT genotype or 6-thiguanine-nucleotide phenotype in irritable bowel disease in children. Konidari A, Anagnostopoulos A, Bonnett LJ, Pirmohamed M, El-Matary W. Thiopurine monitoring in children with inflammatory bowel disease: a systematic review. Br J Clin Pharmacol. 2014;78(3):467-76.

Slide 10	**Alcohol/Acetaldehyde Dehydrogenase** • Marker Drug ***ethanol*** • Clinically Relevant Drugs » Recreational drug (ethanol) • Alcohol Dehydrogenase » ADH1B*1/*1 is normal ethanol metabolism » ADH1B*2 increases ethanol metabolism • Aldehyde Dehydrogenase » ALDH2*1/*1 is normal acetaldehyde metabolism » ALDH2*1/*2 decreases acetaldehyde metabolism » ADH1B*2 + ALDH2*1/*2 -> very low ethanol/very high acetaldehyde » ADH1B*2 + ALDH2*1/*1 -> lower ethanol/normal acetaldehyde • Ethnicity » 40% of Asians have excessive flushing with ethanol because of increased acetaldehyde formation » Others can tolerate larger quantities of ethanol 	Wall TL, Peterson CM, Peterson KP, Johnson ML, Thomasson HR, Cole M, et al. Alcohol metabolism in Asian-American men with genetic polymorphisms of aldehyde dehydrogenase. Ann Intern Med. 1997;127(5):376-9. http://themedicalbiochemistrypage.org/ethanol-metabolism.php Yokoyama A, Tsutsumi E, Imazeki H, Suwa Y, Nakamura C, Yokoyama T. Polymorphisms of alcohol dehydrogenase-1B and aldehyde dehydrogenase-2 and the blood and salivary ethanol and acetaldehyde concentrations of Japanese alcoholic men. Alcohol Clin Exp Res. 2010;34(7):1246-56.
Slide 11	**P-Glyco-Protein Transporter** • Multi-drug resistance to treatment » Cancer » Rheumatoid Arthritis » Inflammatory Bowel Disease » Epilepsy • Common Mechanism » Increased activity of PGP transporter » Drug absorption decreased » Transport out of brain & tumour increased » PGP activity associated with ABCB1 polymorphism? ABC=ATP Binding Cassette • Ethnicity? 	
Slide 12	**Organic Anion Transporter** • Statins are among the most widely used and effective medicines in New Zealand • Myopathy (rhabdomyolysis, myositis) occur 1 in 10,000 • *SLCO1B1* gene encodes the organic anion–transporting polypeptide OATP1B1, which has been shown to regulate the hepatic uptake of statins • Transporter deficiency leads to lower statin clearance and higher statin concentrations • 16 x higher risk of myopathy with *SLCO1B1* mutation 'CC' 	The SCG. SLCO1B1 Variants and Statin-Induced Myopathy -- A Genomewide Study. N Engl J Med. 2008:NEJMoa0801936.

Slide 13	**Warfarin VKORC1 and CYP4F2**  	McDonald MG, Rieder MJ, Nakano M, Hsia CK, Rettie AE. CYP4F2 Is a Vitamin K1 Oxidase: An Explanation for Altered Warfarin Dose in Carriers of the V433M Variant. Mol Pharmacol. 2009;75(6):1337-46.
Slide 14	**Warfarin** **Sensitivity and Resistance** ▯ Genetic differences in VKOR » "Normal Daily Dose" – 5 mg/day » Sensitivity Vitamin K epOxide Reductase – Decreased VKORC1 activity – 80% of Asians – 3 mg/day » Resistance Vitamin K1 Oxidase – Decreased CYP4F2 activity – 60% Caucasians – Perhaps also mutant enzyme does not bind warfarin well – 6 mg/day (but some need much higher doses) 	There are two kinds of VKOR genes: Sensitivity: These patients have a VKORC1 mutation with reduced recycling of inactive vitamin K epoxide to active vitamin K1. This makes them more sensitive to the action of warfarin. For this reason they need a lower dose. Resistance: These patients include those who have been previously called familial warfarin resistance. They appear to have reduced CYP4F2 activity which is an elimination pathway for vitamin K1. It is also possible that some VKOR mutants do not bind warfarin efficiently and thus higher concs (and thus doses) are need to give the required degree of inhibition. See http://en.wikipedia.org/wiki/Warfarin and McDonald MG, Rieder MJ, Nakano M, Hsia CK, Rettie AE. CYP4F2 Is a Vitamin K1 Oxidase: An Explanation for Altered Warfarin Dose in Carriers of the V433M Variant. Mol Pharmacol. 2009;75(6):1337-46.

Slide 15	# HLA-B - Phenytoin is contraindicated in individuals with the HLA-B*1502 variant allele because of an increased risk of adverse skin reactions (Stevens Johnson syndrome and toxic epidermal necrolysis) https://www.pharmgkb.org/guideline/PA166122806. - This can be considered a form of pharmacodynamic source of phenytoin variability. - Other drugs have also been associated with HLA-B genotype variants (*1501 carbamazepine, *5801 allopurinol) and increased risk of these skin adverse effects. 		
Slide 16	# Body size, renal function and post-menstrual age are the most important determinants of drug dose - In comparison to these factors other covariates such as **genotype often pale into insignificance**. - Quantitative pharmacology can help put the role of using covariates to predict drug dose into **a realistic perspective**. 		
Slide 17	# Pharmacogenetics Personal View