Pharmacogenetics and cardiovascular disease management

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Patients differ in their response to drugs. Part of this variability may reflect genetically-determined characteristics of target genes or metabolizing enzymes. A knowledge of an individual's genetic makeup could allow drug therapy to be targeted at those most likely to benefit.

Cadman and O'Connor (2003) stated: 'The central dogma on which pharmacogenomics is founded is that the patient's response to any pharmaceutical is influenced by variations in proteins encoded by the genome.'

INTRODUCTION

In the era of evidence-based medicine, clinicians are guided by the results of large randomized trials. A 34% relative risk reduction of cardiac events as a result of treatment with simvastatin, in groups of patients with coronary disease, mandates the use of statins in all such patients. A failure to demonstrate significant differences in outcomes between various types of antihypertensive drugs, in groups of hypertensive patients, supports the indiscriminate use of drugs to control blood pressure, with cost playing a major role.

From a public health and epidemiological perspective, such clinical trials allow an estimate of benefit and risk; the problem becomes one of implementing such treatment across the whole population. However, even those clinicians who wholeheartedly have embraced this approach recognize that within the population who could benefit there will be some who fail to respond as expected. Hidden within the Kaplan–Meier curves are individuals with a greater-than average response, some with a lesser-than average response, and some who are harmed.

TABLE 1.
Suggested reasons for individual variability in response to drugs

Severity of underlying illness
Disorders of metabolism as a result of renal or hepatic impairment
Compliance/concordance with treatment
Interaction with other substances
External (lifestyle) influences
Genetic differences

VARIABILITY OF RESPONSE

The reasons for variability in response to a drug are numerous (*Table 1*). Adverse drug reactions may be predictable, but are often idiosyncratic. Recent British data suggests that adverse drug reactions account for 1 in 16 hospital admissions and 5700 deaths within hospitals each year (Pirmohamed et al, 2004); 28% were judged to be 'unavoidable'. Clinicians attempt to alleviate such problems by, for example, adjusting drug doses in cases of renal impairment, using weight-adjusted doses or avoiding recognized drug interactions. Genetic influences are also taken into account, albeit indirectly. The advice to use diuretics and calcium channel blockers, rather than angiotensin receptor blockers (ARBs) and angiotensin-converting enzyme (ACE) inhibitors, in hypertensives of Afro-Caribbean origin, or to avoid thiazide diuretics in those prone to gout, are examples of treatment choices that reflect factors under genetic control, with regards to ethnic group or urate metabolism.

'Pharmacogenetics' seeks to use knowledge of genetic variability to target drugs towards those in whom they will be effective and away from those in whom they will cause side-effects. Using a broader exploration of the genome is described as 'pharmacogenomics'. This tailoring of therapy allows 'the right drug in the right dose to the right patient at the right time' (Roden, 2003).

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VARIABILITY IN THE GENOME

Humans share at least 99% of our DNA sequence. Variations include deletions, duplications and variable number tandem repeats. Rare DNA variations or mutations may result in recognized diseases. When a variation affects more than 1% of the population it is termed a polymorphism. The most frequently occurring polymorphism affects single nucleotides – single nucleotide polymorphism (SNP) – of which up to 3 million have been detected (Roden and Brown, 2001). Such SNPs may not be associated with an overt phenotype, but may lie in a gene-coding region, with resultant alteration of amino acid sequence, protein configuration and function, or in a non-coding region, with consequent alteration in protein expression. If the protein is the target for a drug, or is involved with its metabolism, important pharmacodynamic or pharmacokinetic consequences may result.

Genomic DNA can be extracted from blood cells or buccal smears, and polymerase chain reactions (PCR) allow prompt genotyping. Microarrays of panels of assays using fluorescent markers can be used to detect multiple SNPs, for example 74 SNPs have been found in 25 genes implicated in blood pressure regulation (Liljedahl et al, 2003). Newly developed techniques, such as high throughput genotyping, allow interrogation of the entire human genome. Commercial exploitation of these techniques will undoubtedly follow.

PRACTICAL EXAMPLES OF PHARMACOGENETICS

Metabolism

Genetically-determined variation in drug metabolism can be predicted to have significant affects on pharmacokinetics and drug effects (*Table 2*).

The cytochrome P450 (CYP) family of liver enzymes is responsible for the metabolism of many drugs. The isoform of CYP involved in the metabolic inactivation of active enantiomer S-warfarin is CYP2C9. Although a number of polymorphisms of the enzyme have been identified, two are relatively common – CYP2C9*2, an SNP at position 144 of exon 3 associated with 30% reduction in enzyme activity, and CYP2C9*3, an SNP at position 359 on exon 7 associated with 80% reduction in enzyme activity. A retrospective observational study of anticoagulation clinics confirmed that the doses used to maintain a therapeutic international normalized ratio (INR) correlated with genotype (*Table 3*). More importantly, compared with the 'wild type' (most common variant) genotype, it found that those with polymorphisms took a median 95 days longer to achieve a maintenance dose and were more likely to experience bleeding complications during initiation and during follow-up (10.92 per 100 patient years *vs* 4.89 per 100 patient years) (Higashi et al, 2002).

This association of CYP2C9 genotype with the maintenance dose of warfarin was confirmed by Gage et al (2004), who developed a predictive tool that included pharmacogenetic, demographic (body size, age, ethnic group) and clinical (amiodarone and statin use) details to guide dosing. In their view this tool was capable of halving overdosage compared with standard practice.

Similarly, a variety of polymorphisms of the CYP2D6 gene are described, with corresponding variability in enzyme activity, from poor metabolizers through to ultra-rapid metabolizers. This particular cytochrome enzyme is implicated in the metabolism of many drugs in cardiovascular therapeutics, including flecainide, propafenone, metoprolol and perhexiline. Compared with patients with two fully functioning alleles, those with genetically deficient enzyme activity have higher plasma levels of metoprolol for a given oral dose (Rau et al, 2002), exhibit greater beta-blocker effects and CNS side effects of propafenone (Siddoway et al, 1987), and have impaired metabolism of perhexiline, with corre-

TABLE 2.
Some consequences of polymorphisms affecting drug metabolism

Extended pharmacological effect
Increased effective dose
Lack of pro-drug activation
Metabolism by alternative, deleterious pathway
Adverse drug reactions
Exacerbated drug–drug interaction

TABLE 3.
Median dose of warfarin to achieve INR 2.0–3.0 with respect to genotype of CYP2C9

Genotype	Prevalence	Median warfarin dose
*1/*1	68.6%	5.27 mg
*1/*2	15.1%	4.64 mg
*1/*3	9.7%	2.92 mg
*2/*2	2.2%	3.86 mg
*2/*3	1.6%	2.32 mg
*3/*3	2.7%	1.60 mg

From Higashi et al (2002). INR = international normalized ratio

sponding increased risk of hepatotoxicity and peripheral neuropathy (Barclay et al, 2003).

Drug target genes

Knowledge of drug effects dependent upon polymorphisms of drug receptor and effector protein genes is perhaps a little less developed, and results of studies are less consistent. However, it would appear self-evident that inter-individual variability in sensitivity to a drug, if not the likelihood of an adverse reaction, may be determined by genetic variations of such genes. In fact, a number of the polymorphisms that have been shown to affect drug responsiveness have also been shown to predict the severity of disease states (Roden and Brown, 2001; Bristow, 2003).

Hypertension: Adducin is a cytoskeletal protein that links cell surface receptors with cytosolic actin filaments, and is involved with renal epithelial-cell ion transport. In a case-control study of treated hypertensives, a polymorphism of the α-adducin gene was detected in about 35% of individuals. In these hypertensives, the odds ratio of stroke or myocardial infarction was significantly lower in those treated with diuretics compared with those receiving other antihypertensive drugs; no such difference between drug classes were seen in those with the more common 'wild-type' α-adducin gene (Psaty et al, 2002). Screening for this variant among hypertensive patients could allow selection of appropriate therapy.

Heart failure: A common polymorphism exists in intron 16 of the ACE gene, whereby each allele may or may not contain a 287-base period insertion (I = insertion, D = deletion – leading to II, ID and DD genotypes). The D allele is associated with higher ACE activity and angiotensin II levels and the magnitude and duration of ACE inhibitor drug effects are governed by the ACE I/D genotype (Ueda et al, 1998). A prospective study of patients with severe left ventricular dysfunction revealed a significantly poorer transplant-free survival rate in those with the D allele. The widespread use of ACE inhibitors or ARBs (95% of patients) failed to eliminate the adverse prognostic effect of the D allele, yet this genotypic risk was not evident in those treated with β-blockers. Only those with DD had significant survival benefit from β-blockers (McNamara et al, 2001). The mechanism by which the β-blocker effect is altered by ACE genotype is uncertain.

The effectiveness of β-blockers may also be affected by polymorphisms of the β-receptor. For example, in patients with congestive heart failure the improvement in ejection fraction following carvedilol therapy was affected by the presence of polymorphisms of the β2 receptor (Kaye et al, 2003).

Hyperlipidaemia: Genetic polymorphisms have been reported to be responsible for variation in response to lipid-lowering treatments and dietary interventions. For example, the ability of pravastatin to delay the angiographic progression of coronary disease is blunted in those with the ACE DD genotype; progression of coronary atheroma following lipid lowering with a combination of lovastatin and colestipol or niacin and colestipol is influenced by polymorphisms of the transcription-start site of the hepatic lipase gene (to such an extent that those with the genotype associated with the least hepatic lipase activity (TT) show progression of coronary disease while other genotypes show improvement); polymorphisms of the first intron of the cholesterol ester transfer protein (CETP) affects disease progression in response to treatment with pravastatin (Maitland-van der Zee et al, 2002). Many of these polymorphisms are not necessarily those affecting drug target receptors, but clearly genes that are involved in disease pathogenesis are able to modify drug effects.

Apolipoprotein E (apoE) is a component of chylomicrons, very-low-density lipoprotein (VLDL) and intermediate-density lipoproteins, and is involved in mediating the uptake of remnants of these particles into the liver and their clearance from the plasma. The gene for apoE resides on chromosome 19, and three isoforms exist – apoE2, apoE3 and apoE4 – leading to six genotypes resulting from combinations of two alleles. Variation in apoE genotype is associated with variability of lipid responses to fish oil ingestion (Minihane et al, 2000). A number of studies have shown interactions between apoE genotype and lipid response to statin therapy. Survivors of myocardial infarction are at greater risk of subsequent death if they carry the apoE4 allele. This excess risk is abolished by simvastatin. In fact, in those with low lipoprotein(a) and without an apoE4 allele there appeared to be no obvious mortality benefit of statin therapy (*Table 4*) (Gerdes et al, 2000).

Smoking cessation: Craving for cigarettes in response to stress is affected by genetic polymorphisms of both the dopamine receptor gene (DRD2) and dopamine transporter gene (Erblich et al, 2004), and individuals who are homozygous for the 'Asn40Asp' variant of the μ-opioid receptor (OPRM1) are more likely to be abstinent with less mood disturbance or weight gain following cessation programmes using transdermal nicotine (Lerman et al, 2004). The CYP2B6 is implicated in both bupropion and nicotine

metabolism. In a placebo-controlled trial of bupropion for smoking cessation, those with genetically-determined reduction of enzyme activity were more likely to crave cigarettes and to relapse after cessation, although the effects of the genotype were attenuated by bupropion in females (Lerman et al, 2002).

THE BENEFITS

By implementing a prescribing policy based upon pharmacogenetics, treatment becomes tailored to the individual rather than simply being dependent upon the medical condition. Following a genetic test, performed only once, drugs that are likely to be ineffective or to cause adverse effects can be avoided. This is advantageous both to the patient and to the health service. Polypharmacy can be reduced, and those receiving treatment can be more certain of obtaining benefit without harm. Put simply, the number needed to treat decreases and the number needed to harm increases.

The pharmaceutical industry may initially baulk at the prospect of smaller markets for particular drugs (although perhaps purchasers could be persuaded to pay more for a bespoke service), but may be encouraged to re-explore effective drugs that have been previously withdrawn from use following a small number of adverse events. Moreover there may be the opportunity to exploit new markets by designing medicines that target specific populations. Drug interactions could be more accurately predicted and the success rate of phase III trials might increase.

WILL IT HAPPEN?

There are significant ethical issues involved with any technique that involves exploration and use of the human genome, and these are yet to be fully addressed. The economics of a pharmacogenetic approach have not been adequately described. While models suggest that the approach is financially viable (Maitland-van der Zee et al, 2004), it may still prove cheaper to pursue the present policy of general prescribing, with monitoring for later adverse and beneficial effects. More importantly, there are concerns that the complexity of the association of disease status, drug metabolism and human genetics will make a pharmacogenetic approach impractical for the foreseeable future.

In most cases, the effects of drugs depend upon polygenic and non-genetic influences rather than single SNPs, and the examples presented above have been chosen to simplify the concept. Even in the case of warfarin, where the metabolism of the drug is almost exclusively mediated by CYP2C9, the predictive tool based upon pharmacogenetics explained only 39% of the variance in the maintenance dose (Gage et al, 2004). Other studies have shown the importance of genetic variations of the code for the P-glycoprotein that controls efflux of warfarin into target cells in predicting warfarin dosage, while failing to show a corresponding association with bleeding risk (Wadelius et al, 2004). Some drugs, for example flecainide, may be excreted unchanged in the urine, so genetic influences of CYP-mediated metabolism only manifest when renal function is significantly impaired. Most drugs are metabolized by more than one route, and clinically important effects may require the coincident presence of two or more polymorphisms.

Further, the overall effect of a drug will depend upon the net effect on polymorphisms within its metabolic pathway and genetic variations within target cells. Multiple effects of a single drug may, therefore, be determined by different genetic variants. For example, haplotypes in the CETP gene predict the increase in high-density lipoprotein cholesterol and decrease in triglycerides (Winkelmann et al, 2003), while SNPs of the 3-hydroxy-3-methyl-glutaryl coenzyme A (HMG-CoA) reductase gene predict the reduction in low density lipoprotein (LDL) cholesterol, following statin therapy (Chasman et al, 2004). Finally, if pharmacogenetic drug targeting is to achieve all that is expected of it, evidence of clinically important outcomes needs to be demonstrated.

CONCLUSIONS

Pharmacogenetic testing has the potential to maximize the benefit of drug treatment within a population while minimizing the likelihood of adverse effects in individual patients. Whether this promise will be realized is yet to be determined. HM

TABLE 4.
Adjusted relative mortality hazard ratios as a result of treatment with simvastatin: role of apoE and lipoprotein(a)

Patient group		Hazard ratio as a result of treatment
All		0.49 (0.31–0.79)
without apoE4		0.66 (0.35–1.24)
with apoE4		0.33 (0.16–0.69)
with apoE4	High lipoprotein(a) (≥30mg/dl)	
no	no	0.87 (0.36–2.08)
yes no	no yes	0.48 (0.25–0.93)
yes	yes	0.22 (0.06–0.77)

From Gerdes et al (2000)

Conflict of interest: none.

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KEY POINTS

- Individuals vary in their response to drugs.
- Part of this variability is genetically determined.
- Genetic variants may affect drug metabolism and pharmacodynamics.
- Knowledge of pharmacogenetics may allow 'tailored' drug treatment.