INVITED LECTURES
VABLJENA PREDAVANJA

GENETIC POLYMORPHISMS AND DRUG METABOLISM

GENETSKI POLIMORFIZMI IN PRESNOVA ZDRAVIL

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Abstract

Background *It is estimated that genetic factors account for 15–30 % of variability in drug response, however for some drugs this may be the major determinant in drug response. Pharmacogenetics aims to identify genetic sources of variability in response to drugs by studying genetic variations affecting drug metabolizing enzymes, transporters and drug targets thus causing interindividual variability in drug levels (pharmacokinetics), drug response (pharmacodynamics) and side effects. Extensive information on genetic variability in drug metabolizing enzymes, transporters and targets is available from public databases.*
Drugs are metabolized in two phases. In Phase I drug is metabolically activated to reactive electrophilic form, mostly by cytochromes P450 (CYPs), to be conjugated to some endogenous compound by Phase II enzymes: UDP-glucuronosyltransferases (UGTs), N-acetyl-transferases (NATs), glutathione S-transferases (GSTs), or others. Genetic polymorphism of many enzymes involved in this process leads to inter-individual variations in metabolism and pharmacokinetics of drugs and could therefore influence drug response. Genetic polymorphism is the occurrence of two or more alleles at a given locus of which the rare allele has a frequency of at least 1 % or more in a given population. The understanding of a patient's genotype and its corresponding effect on drug response could help distinguish between responders and non-responders of a specific drug treatment and help to choose the most effective drug and optimal dose. A large number of different methodologies have been developed for genotyping, however at present predictive genotyping for drug metabolizing enzymes does not occur routinely in the clinical practice.

Conclusions *There is increasing evidence that genotyping for polymorphic drug metabolizing enzymes, in particular CYPs has potential to improve drug therapy and achieve higher response rates and reduced adverse effects. Open questions regarding the relevance of the knowledge of pharmacogenetic information for clinical end points and the cost-benefit aspects of pharmacogenetic based dosing need to be answered by prospective randomized clinical trials.*

Key words *pharmacogenetics; polymorphism; drug metabolism; cytochromes P450; genotyping*

Izvleček

Izhodišča *Različni bolniki se na predpisani odmerek zdravila ne odzovejo enako. Na interindividualne razlike v hitrosti in učinkovitosti presnove in delovanja velikega števila zdravil pomembno vplivajo tudi genetski dejavniki. Farmakogenetika preučuje vpliv genetske variabilnosti encimov, ki presnavljajo zdravila, prenašalcev (transporterjev) in tarčnih molekul, na katere zdravila delujejo na presnovo zdravil, učinkovitost farmakoterapije in neželene učinke zdravljenja. Izmed genetskih dejavnikov je najpomembnejša genetska variabilnost encimov, ki presnavljajo zdravila, saj vpliva na učinkovitost več kot 30 % zdravil.*

Presnova zdravil v telesu poteka s pomočjo specifičnih encimskih sistemov pretežno v dveh stopnjah. V prvi stopnji (faza I) nastane bolj elektrofilen, aktiviran presnovek, ki v naslednji stopnji (faza II) vstopi v reakcije konjugacije ki jih katalizirajo UDP-glukuronoziltransferaze (UGT), N-acetil-transferaze (NAT), glutation S-transferaze (GST) in drugi encimi, s čimer se poveča vodotopnost molekule. Za presnovo zdravil so v fazi I najbolj pomembni citokromi P450 iz treh družin, CYP1, CYP2 in CYP3, ki jih večinoma kodirajo polimorfni geni. Genetski polimorfizem pomeni, da sta v populaciji prisotna najmanj dva alela nekega gena, pri čemer je frekvenca manj pogostega alela vsaj 1 %. Po nekaterih ocenah bi genotipizacija lahko zmanjšala stroške zdravljenja, prispevala k varnejšemu zdravljenju pri 15–25 % vseh klinično uporabljanih zdravil in zmanjšala pogostnost neželenih učinkov za 10–20 %. Čeprav postopki genotipizacije postajajo vse hitrejši, enostavnejši in cenejši, se farmakogenetsko testiranje v klinični praksi še ne uporablja.

Zaključki *Vedno več raziskav kaže, da genetski dejavniki pomembno vplivajo na presnovo in odmerjanje zdravil ter povečujejo tveganje za neželene učinke zdravil. Potrebne pa so prospektivne raziskave, ki bi pokazale, ali poznavanje teh dejavnikov doprinese tudi k boljši uspešnosti in ekonomičnosti zdravljenja.*

Ključne besede *farmakogenetika; polimorfizem; presnova zdravil; citokromi P450; genotipizacija*

Introduction

The aim of drug treatment is to administer the appropriate drug in the correct dose to produce the desired therapeutic effect with a minimum of toxicity. Currently the choice of the drug and dose are based on the »trial and error« basis and there is a wide range of efficacy and side effects: although in most patients the desired therapeutic effect may be achieved and they benefit from the treatment, there may be patients that do not respond to the treatment or may even suffer an adverse effect with little or no benefit from the treatment.

There is extensive interindividual variability in drug response. Factors influencing the drug response include the demographic characteristics of the patients such as age, weight, sex and ethnicity, the nature of the disease, concomitant diseases, patients' diet, alcohol consumption, cigarette smoking, co-treatment with other drugs and others. The most important are however the dose of the drug, patient's compliance to the treatment prescribed and possible genetic variation in response to the drug. It is estimated that genetic factors account for 15–30 % of variability in drug response, however for some drugs this may be the major determinant in drug response.[1]

Pharmacogenetics aims to identify genetic sources of variability in response to drugs by studying genetic variations affecting drug metabolizing enzymes, transporters and drug targets thus causing interindividual variability in drug levels (pharmacokinetics), drug response (pharmacodynamics) and side effects. Pharmacokinetics examines the fate of the drug in the body: absorption, distribution, metabolism and excretion (ADME), while pharmacodynamics studies interactions between the pharmacokinetic properties of the drugs and their action on drug targets. These interactions define drugs pharmacological effects, either desired or adverse, and may be influenced by genetic variability. As the overall pharmacological effect of drugs may be determined by interaction of several genes encoding proteins involved in multiple pathways of drug disposition, metabolism and effects, pharmacogenomics aims to identify variability relevant for drug response at the genome level. Among genetic factors the variability in drug metabolizing enzymes which affects about 30 % of all drugs has been studied the most, but also the list of genetic polymorphism of drug transporters and drug targets that have been shown to influence drug levels and drug response has grown lately.[2]

Molecular basis of genetic variability in drug metabolism

Genetic variability is the consequence of the variations in the DNA sequence of the genome. Due to DNA sequence variation genes may exist in alternative forms, which are called alleles. Specific sets of alleles forming the genome of an individual are called its genotype, and the observable characteristics such as the capacity to metabolize a drug or drug response are called the phenotype.

Since autosomal chromosomes are paired, each position (locus) on the autosomal chromosome is represented twice. If both chromosomes have the same allele, occupying the same locus, the condition is referred to as homozygous for this allele. If the alleles at the two loci are different, the individual is referred to as heterozygous for both alleles.

Genetic polymorphism is the occurrence of two or more alleles at a given locus of which the rare allele has a frequency of at least 1 % or more in a given population.

The single nucleotide polymorphisms (SNPs) differ by a single base at a given position in the genome. SNPs are the most frequent and the simplest form of DNA variation and they account for more than 90 % of total variability in the human genome. Other types of variation include insertions or deletions of nucleotides, variable number of tandem nucleotide repeats (VNTR),

microsatellites and deletions or duplications of gene regions, complete genes, or chromosomal regions.

The total number of SNPs with a minor allele frequency of at least 5 % in the human genome is estimated to be over seven million,[3] but all the SNPs are not functional.[4] SNPs within the coding region can be classified into different groups. As a given amino acid may be specified by more than one triplet of nucleotide residues (codons) on the DNA molecule due to so called degeneracy of the genetic code, some SNPs in the exons, especially the ones that change the third nucleotide within the codon, do not change the information (silent SNPs). The change of the amino acid may not alter the function of the protein (neutral SNPs), or it may lead to a protein with altered function (missense mutation). A SNP may also produce a stop codon which signals premature termination of the polypeptide chain (nonsense mutation) and result in a nonfunctional protein. Insertions and deletions may cause a frame shift which usually also results in a non-functional protein. Alternatively SNPs in promoters, splice sites and untranslated regions may alter the expression and thus the amount of a gene product by changing its transcriptional regulation,[5] or mRNA stability.[6] Changes in splice site sequences can cause splicing defects, and polymorphisms can also affect the translation efficiency.[7] Finally, the gene's product may be missing due to a gene's deletion or may be amplified due to a gene's duplication. Examples of different mechanisms through which genetic polymorphisms can affect the activity of drug metabolizing enzymes are given in Table 1.

There is extensive information on genetic variability in drug metabolizing enzymes, transporters and targets available from public databases. The National Center for Biotechnology Information (NCBI) dbSNP database contains information on SNPs from 43 different organisms, among them more than 11 million human SNPs (http://www.ncbi.nlm.nih.gov/SNP/).

Table 1. *Common polymorphic alleles affecting the activity of three major Cytochromes P450 important for drug metabolism.*

Razpr. 1. *Pogosti polimorfni aleli, ki vplivajo na encimsko aktivnost treh za presnovo zdravil najpomembnejših citokromov P450.*

	DNA DNA	Protein Beljakovina	Enzyme activity Encimska aktivnost	Predicted phenotype Predvideni fenotip	Allele frequency in Slovenian population Frekvenca alela v slovenski populaciji
CYP2C9*1	–	–	Normal Normalna	EM	0.815 [a]
CYP2C9*2	3608 C > T	R144C	Decreased Zmanjšana	PM	0.122
CYP2C9*2	42614 A > C	I359L	Decreased Zmanjšana	PM	0.063
CYP2C19*1	–	–	Normal Normalna	EM	0.837 [a]
CYP2C19*2	19154 G > A	Splicing defect Sprememba izrezovanja	Not active Neaktiven	PM	0.159
CYP2C19*3	17948 G > A	W212X	Not active Neaktiven	PM	0.004
CYP2C19*17	–806 C > T	Increased transcription Povečano prepisovanje	Increased Zvišana	UM	ND #
CYP2D6*1	–	–	Normal Normalna	EM	0.427 [b]
CYP2D6*2	2850 C > T	R296C	Normal Normalna	EM	0.242
CYP2D6*3	2549 delA	Frame shift Sprememba bralnega okvirja	Not active Neaktiven	PM	0.035
CYP2D6*4	1846 G > A	Splicing defect Sprememba izrezovanja	Not active Neaktiven	PM	0.178
CYP2D6*5	Gene deletion Delecija gena	No enzyme Ni encima	Not active Neaktiven	PM	0.022
CYP2D6*1X2	Gene duplication Duplikacija gena	More enzyme Več encima	Increased Zvišana	UM	0.007
CYP2D6*41A	2850 C > T	R296C	Decreased Zmanjšana	IM	ND $

Legend: DNA – principal nucleotide sequence change (A – adenine, C – cytosine, G – guanine, T – thymine), R – arginine, C – cysteine, I – isoleucine, L – leucine, W – tryptophan, X – stop codon, EM – extensive metabolizer, IM – intermediate metabolizer, PM – poor metabolizer, UM – ultrarapid metabolizer, [a] – healthy Slovenian population[44], [b] – Slovenian schizophrenia patients on long-term antipsychotic treatment[45], ND – not determined, # – 0.18 in Swedes,[28] $ – 0.084 in Germans.[46]

Legenda: DNA – značilna sprememba nukleotidnega zaporedja (A – adenin, C – citozin, G – gvanin, T – timin), R – arginin, C – cistein, I – izolevcin, L – levcin, W – triptofan, X – stop kodon, EM – hiter metabolizator, IM – vmesni metabolizator, PM – počasen metabolizator, UM – ultrahiter metabolizator, [a] – zdrava slovenska populacija[44], [b] – slovenski bolniki s shizofrenijo na vzdrževalnem antipsihotičnem zdravljenju[45], ND – ni določena, # – 0.18 in Swedes,[28] $ – 0.084 in Germans.[46]

Extensive information on genetic polymorphisms can also be found in the Human Genome Mutation database (http://www.hgmd.cf.ac.uk/ac/index.php) as well as many other databases. Human Cytochrome P450 (CYP) Allele Nomenclature Committee Web Site publishes peer reviewed information on the alleles of human cytochromes P450 (CYPs) regarding their nucleotide changes, their functional consequences and links to publications where the allele has been identified and characterized, making it a web site particularly interesting for researchers in the field of pharmacogenetics (http://www.cypalleles.ki.se/).[8, 9]

SNPs as pharmacogenetic markers

It is estimated that there is a SNP every 300 base pairs (bp), but the spacing between them is not equal. Individual SNPs that are grouped together in the same gene or within a genome region form haplotypes. SNPs may serve as genetic markers if their functional significance and the association with a given phenotype are proven.

Candidate gene approach

Nonsynonymous polymorphisms or known variations in regulatory elements of genes coding for drug metabolizing enzymes, drug transporters or drug targets represent most commonly used candidate loci for pharmacogenetic testing. Although this approach has traditionally focused on functional SNPs in genes important for drug metabolism and transport, it is becoming clear that usually more than one gene is implicated in drug response and that pathway approach may be needed to predict drug's pharmacokinetics and pharmacodynamics as well as disease risk.[10]
A large amount of information about the relationships among genes, drugs, diseases and pathways and phenotypes of drug response is publicly available from the internet databases such as PharmGKB (http://www.pharmgkb.org/).[11] Such databases may help to select a limited set of candidate genes involved in a specific drug's action to increase the predictive power of genotyping.
A large number of different methodologies have been developed for SNP and VNRT genotyping.[12] Traditional genotyping approaches that require optimization of polymerase chain reaction (PCR) assay conditions for each individual SNP are laborious and not amenable to autoimmunization and are not best suited for the use in the clinical practice. Therefore new genotyping approaches are being developed to enable fast, robust and cost-effective analysis of a large number of SNPs under universal assay conditions that do not require optimization. TaqMan® Genotyping Assays (Applied Biosystems) and GenomeLab™ SNPstream SYSTEM (Beckman Coulter) are two such fast and robust approaches with capacity for multiplexing and automatization.

Whole genome approach

In whole genome approach, a large number of SNPs across the entire human genome is tested for association with drug response or other given phenotype. The advantage of the genome scan over the candidate gene approach is that there are no prior assumptions about the mechanisms of efficacy or safety of the drugs under investigation, and no prior assumptions about the location of the causative alleles. It may thus be possible to identify novel genes having a significant influence on drug response. The cost of such analysis is high and there is a chance that many significant SNPs will be false positive. Although genome-wide approaches such as expression array technology have identified regions associated with drug-induced adverse effects[13] and may help to identify more candidates by discovering genes that are activated or deactivated in response to treatment, overall results of the genome-wide approaches have been largely inconsistent. In addition whole genome SNP analyses have an expected high number of false positive associations due to the high degree of multiple testing and are not suitable for the use in the everyday clinical practice.

Tagged SNP approach

Due to the linkage disequilibrium (LD) there is a considerable redundancy in the information obtained from genotyping closely-spaced SNPs. Therefore a greatly reduced number of SNPs (so called tag SNPs) chosen on the basis of haplotype information can be used in association studies to investigate common genetic variations in a region of interest[14, 15] without significant loss of power.[14, 16] Genetic data for haplotype analysis are available from the International HapMap Project database (http://www.hapmap.org/).[17]

Genetic polymorphisms important for drug metabolism

A large number of enzymes most of which are polymorphic participate in metabolism of xenobiotics such as drugs and carcinogens (drug metabolizing enzymes – DMEs).[18] In general drugs are metabolized in two phases (Figure 1). Phase I DMEs, mostly cytochromes P450 (CYPs), metabolically activate xenobiotics to reactive electrophilic forms which is then conjugates to some endogenous compound by Phase II DMEs; such as UDP-glucuronosyltransferases, (UGTs), N-acetyl-transferases (NATs), glutathione S-transferases (GSTs), or others. Genetic polymorphism of many enzymes involved in this process leads to inter-individual variations in metabolism and pharmacokinetics of drugs and could therefore influence drug response.[18]
About 40 % of phase I metabolism of clinically used drugs is affected by polymorphic enzymes. As these genetic polymorphisms alter enzyme activity they may change the rate of drug metabolism and influence drug plasma levels. By using an enzyme specific probe drugs and measuring the metabolic rate individuals can be grouped into four phenotype groups: poor metabolizers (PM) lacking functional enzyme due to the homozygosity for two defective

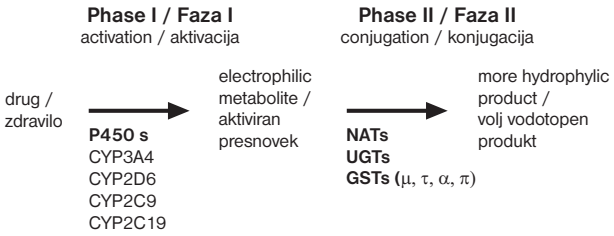

Figure 1. *Phase I and Phase II of drug metabolism. P450s – Cytochromes P450, NATs – N-acetyl-transferases, UGTs – UDP-glucuronosyltransferases, GSTs – glutathione S-transferases.*

Sl. 1. *Faza I in faza II v presnovi zdravil. P450s – Citkromi P450, NATs – N-acetil-transferaze, UGTs – UDP-glucuronoziltransferaze, GSTs – glutation S-transferaze.*

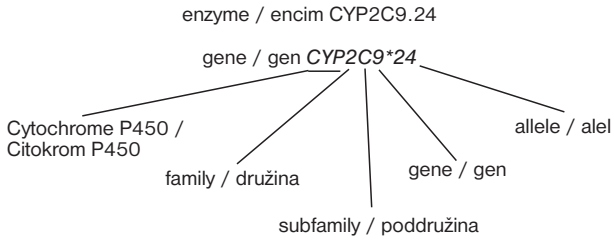

Figure 2. *Nomenclature of Cytochromes P450.*

Sl. 2. *Poimenovanje citokromov P450.*

alleles, intermediate metabolizers (IM) heterozygous for one defective allele or carrying two alleles with reduced activity, extensive metabolizers (EM) with two normal alleles and ultrarapid metabolizers (UM) carrying gene duplication or multiple gene copies. The rate of metabolism for a certain drug can differ 1000-fold between the PM and UM. Such patients may require dose adjustments as a standard population based dosing may result in a higher risk for adverse effects (ADR) due to high plasma levels in PM or unresponsiveness to treatment in UM. A meta-analysis performed in the U.S.A. revealed that serious ADRs occur in 6.7 % of all hospitalized patients, while 0.3 % of all hospitalized patients develop fatal ADR.[19] It has been shown that among the drugs that are cited in the ADR-studies 56 % are metabolized by polymorphic Phase I enzymes and that 86 % of these enzymes are actually P450s. In contrast only 20 % of drugs that are substrates for nonpolymorphic enzymes are cited in the ADR reports.[20]

Cytochromes P450

Cytochromes P450 (CYP) are a superfamily of heme-containing monooxygenases involved in metabolism of structurally very diverse endogenous substrates and xenobiotics. Following the P450 nomenclature CYP are classified according to their amino acid sequence homology. The nomenclature of P450s is well established. Abbreviation CYP indicates Cytochrome P450,while the first arabic number indicates a family characterized by more than 40 % amino acid sequence homology (CYP2 as shown in Figure 2). The letter that follows indicates a subfamily characterized by more than 55 % amino acid sequence homology (CYP2C) and the next arabic number indicates the individual enzyme (CYP2C9). Genes are written in italics and the separate alleles are designated by a star and a number that follows *(CYP2C9*24).*[8, 21]

There are 57 active CYP genes in the human genome (http://drnelson.utmem.edu/CytochromeP450.html).[22] The majority of the P450 isoforms are expressed in the liver, while some are expressed in other tissues such as central nerve system, gastrointestinal tract, lung, trachea, nasal and olfactory mucosa and adrenal gland. In particular, three families of P450s: CYP1, CYP2 and CYP3 are responsible for 75–80 % of the phase I metabolism and 65–70 % of the clearance of clinically used drugs.[23] The most important P450s for drug metabolism include CYP3A4, CYP2D6, CYP2C9 and CYP2C19. The functional effects of some of the most common *CYP2C9, CYP2C19* and *CYP2D6* alleles and their frequencies in Caucasian and Slovenian population are given in Table 1.

CYP3A subfamily accounts for more than 50 % of all CYP-dependant drug metabolism and substantial interindividual variability in CYP3A activity was observed.[24] Polymorphisms identified so far did not explain this variability as no correlation was found between the genotype and the phenotype. Induction and inhibition by drugs and some food constituents seem to be clinically more relevant, as they increase or decrease CYP3A drug metabolism.

CYP2D6 is one of the most studied P450 and the molecular basis of the interindividual variability that may lead to decreased or increased CYP2D6 activity is well understood. Because CYP2D6 is not inducible, genetic polymorphism is the major determinant of its activity and the metabolic capacity (phenotype) can be predicted by genotyping. CYP2D6 is involved in the metabolism of 20–25 % of all drugs in the clinical use, among them many antidepressants, antipsychotics, antiepileptics, antiarythmics and others.

CYP2C9 comprises 20 % of all cytochromes P450 in the liver and participates in metabolism of about 10 % to 20 % of commonly prescribed drugs such as antithrombotic and hypoglycemic agents, HMG-CoA-reductase inhibitors (statins), non-steroidal anti-inflammatory drugs, antiepileptics (phenytoin) and others. Human *CYP2C9* gene is highly polymorphic. Besides the wild-type *CYP2C9*1* allele the two most common variant alleles in Caucasian populations are *CYP2C9*2* and *CYP2C9*3* (Table 1). It was demonstrated *in vitro* and *in vivo* that these two allelic variants of *CYP2C9* gene influenced the metabolic activity of the enzyme. In accordance with other studies also our results indicate that in order to achieve the optimal therapeutic effect patients with one polymorphic *CYP2C9* allele required 44.5 % lower warfarin dose, while patients with two polymorphic *CYP2C9* alleles required 66.4 % lower warfarin doses as compared to the patients with two normal alleles.[25]

CYP2C19 is involved in metabolism of about 5 % of all drugs and is particularly important for metabolism of proton pump inhibitors and antidepressants. It has actually been shown that the cure rate of acid-related ulcers and gastroesophageal reflux disease is higher in PMs.[26, 27] Besides two common deficiency alleles *CYP2C19*2* and **3,* a novel allele *CYP2C19*17* that confers increased activity has been identified recently. This allele was reported to have a frequency of 18 % in the Swedish population and it is likely that therapeutic response to proton pump inhibitors and antidepressants will be reduced in the homozygous carriers.[28]

Also other polymorphic P450s such as CYP1A2, CYP2A6 and CYP2B6 contribute to metabolism of some clinically important drugs.

Drug transporters

Drug transporters are increasingly recognized as an important determinant of drug disposition and response.

Multidrug resistant protein *(MDR1)* gene, which codes for P-glycoprotein and functions as an ATP-dependent efflux transporter in different cells is the best characterized drug transporter. It is expressed mainly in gastrointestinal tract, liver, kidneys, blood cells and brain and plays a major role in absorption, distribution and elimination of various structurally unrelated drugs and endogenous substrates. *MDR1* gene variants were proposed as potential susceptibility factors for diseases such as inflammatory bowel diseases, Parkinson's disease and renal epithelial tumor and as determinants of treatment response to various drugs.[29–31]

There are many other transporters whose role in drug delivery and disposition needs to be determined: organic anion and cation transporters (OAT, OCT, solute carrier family SLC22A), organic anion transport proteins (OATP, solute carrier family SLCO, formerly SLC21A), and MRPs (ABCCs), also members of the ATP-binding cassette family ([32] and the references within).

Drug targets

Because genetic variability in drug target pathways alters pharmacodynamics it may affect the efficacy of drug treatment. Drug targets can be grouped into three main categories: the direct protein target of the drug, signal transduction cascades or downstream proteins, and proteins involved in disease pathogenesis.

When we recently studied genetic factors that influence warfarin requirement we have clearly shown that two common polymorphisms in *VKORC1* gene which codes for the enzyme that is directly inhibited by warfarin, influence warfarin dose requirement. The importance of accounting for variability of both pharmacokinetic and pharmacodynamic factors is evident from our observation that while *CYP2C9* polymorphisms and non-genetic factors, such as patient's age and body-weight explain 37 % of interpatient variability in warfarin dose requirement, with the addition of *VKORC1* genotyping 60.0 % of variability in warfarin dose can be explained.[33] Other studies have come to similar conclusions.[34, 35]

Examples of gene variants affecting pharmacodynamics pathways include those coding for serotonin metabolism, synthesis, transport and receptors. These gene variants may act independently, in combination with each other, and/or in combination with PK genes to affect drug response, for example to antidepressants.[36] Similarly, response to antipsychotics may be affected by polymorphic genes involved in dopamine metabolism and signaling.[37] The understanding of a patient's genotype and its corresponding effect on drug response would be useful to the practicing clinician and genetic testing could help distinguish between responders and non-responders of a specific drug treatment and help to choose the most effective drug and optimal dose.

Molecular data on disease specific proteins facilitate assessment of disease heterogeneity and genetic variability in proteins involved in disease pathogenesis proved to be very important in oncology.[38, 39]

Pharmacogenetic genotyping – the present and the future

It is becoming evident that knowledge about polymorphic drug metabolizing enzymes may provide important information for choice of drug therapy and drug dosage.[1, 23, 40] For example, a meta analysis has shown that based on the *CYP2D6* and *CYP2C19* genotype the dose adjusted would be relevant in about 50 % of clinically used drugs that are substrates for these enzymes, especially antidepressants and antipsychotics.[40]

Obviously there is a large discrepancy between the pharmacogenetic information that is available in the literature for the 174 most used drugs and the pharmacogenetic information inside the drug package label that is stated in only 13 % of drug package labels.[41]

On the other hand, the use of pharmacogenetic genotyping is not always related to the pharmacogenetic data in the literature. In the U.S.A. many psychiatrists choose to have their patients genotyped for *CYP2D6* before the start of the long term antipsychotic treatment in order to minimize the chance of occurrence of extrapyramidal side effects – in spite of the fact that studies on association between the genotype and occurrence of extrapyramidal side effects have not been conclusive.

However at present predictive genotyping for drug metabolizing enzymes does not occur routinely in the clinical practice. This could be due to the lack of awareness and knowledge about pharmacogenetic variability among health care professionals and patients, but also due to the lack of prospective studies that would clearly show that pharmacogenetic testing contributes to greater treatment efficacy.[4] Even for a drug as commonly used as warfarin there have been very few attempts to assess the benefits of pharmacogenetic testing for genetic polymorphisms involved in its metabolism.

For pharmaceutical industry pharmacogenetics and pharmacogenomics offer a way of defining variable response to drugs at an early stage of their development and a way to improve the efficiency of drug trials. The early knowledge as to whether a polymorphic pathway is involved in drug metabolism/action and of its clinical relevance will lead to a reduction of time and costs in the development of a new drug. The knowledge on genes involved in drug response has already helped to identify new targets for cancer treatment.[38, 39]

Based on the pharmacogenetic information FDA already requires testing for epidermal growth factor receptor (EGFR) before the treatment of colon cancer with cetuximab. Similarly genetic testing of HER2/neu receptor is required prior to treatment with trastuzumab (Herceptin) (http://www.fda.gov/cder/genomics/). FDA also recommends genetic testing of thiopurine methyltransferase *(TPMT)* before treatment with azathioprine and genotyping of UDP-glucuronosyltransferase 1A1 *(UGT1A1)* before treatment with irinotecan. As few as these genetics-guided recommendations may be at present, they allow for the better efficacy of the treatment.

Up to present, FDA approved only one diagnostic pharmacogenetic test that allows *CYP2D6* and *CYP2C19* genotyping.[43] As new genotyping approaches are being developed genotyping is becoming faster and more cost-effective and when applied in prospective pharmacogenetic studies the benefits of predictive genotyping could be evaluated. When the benefits of predictive genotyping are confirmed, this knowledge could be used to tailor drug choice and dosage regimens to an individual to maximize therapeutic efficacy and minimize adverse drug reactions. It has been estimated that predictive genotyping for P450s could improve clinical efficacy of 15–25 % of all drug therapy and reduce the incidence of adverse drug reactions by 10–20 %.[42]

Conclusions

There is conclusive evidence that genetic variability of drug-metabolizing enzymes, transporters or drug targets infuence drug's metabolism and disposition. There is also increasing evidence that genotyping for polymorphic drug metabolizing enzymes, in particular CYPs has potential to improve drug therapy and achieve higher response rates and reduced adverse effects. Many open questions still remain regarding the relevance of the knowledge of pharmacogenetic information for clinical end points and the cost-benefit aspects of pharmacogenetic based dosing. These questions need to be answered by prospective randomized clinical trials.

Nevertheless pharmacogenetics offers a promise of »personalized medicine« in the future. Testing for multiple common genetic polymorphisms which can modify the efficacy or the adverse effects of the treatment holds promise of the individualized treatment according to the individual's genetic background.

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