R E V I E W

Role of pharmacogenomics in the treatment of tuberculosis: a review

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Background: Tuberculosis is one of the major public health problems worldwide. Modern antituberculous treatment can cure most patients; cure rates > 95% are achieved with standard short-course chemotherapy regimens containing isoniazid, rifampicin, pyrazinamide, and ethambutol among patients with drug-susceptible strains of tuberculosis; however, a small proportion do not respond to treatment or develop serious adverse events. Pharmacogenomic studies of drugs used in the treatment of tuberculosis could help us understand intersubject variations in treatment response. In this review, we compiled pharmacogenomic data on antituberculous drugs that were available from different settings that would give a better insight into the role of pharmacogenomics in the treatment of tuberculosis, thereby enhancing the efficacy and limiting the toxicity of existing antituberculosis medications.

Methods: The PubMed database was searched from 1960 to the present using the keywords "tuberculosis", "antituberculosis treatment", "isoniazid", "rifampicin", "pyrazinamide", "ethambutol", "pharmacogenomics", and "polymorphism". Abstracts from meetings and review articles were included.

Conclusion: Studies conducted in different settings suggest that pharmacogenomics plays a significant role in isoniazid metabolism, and impacts both treatment efficacy and frequency of adverse reactions. Single nucleotide polymorphisms influencing plasma rifampicin concentrations are also reported. No data are available regarding other first-line drugs, ie, ethambutol and pyrazinamide. There is a need to incorporate pharmacogenomics into clinical trials of tuberculosis in order to understand the factors impacting therapeutic success and occurrence of adverse drug effects.

Keywords: tuberculosis, antituberculous treatment, pharmacogenomics, polymorphism, drug metabolism

Introduction

Tuberculosis continues to remain a major public health problem. One third of the world's total population are infected with *Mycobacterium tuberculosis*, the organism causative of tuberculosis. One in every 10 infected persons will break down with active tuberculosis during their lifetime. If untreated, each person with active tuberculosis is likely to infect about 10–15 persons per year, and persons infected with human immunodeficiency virus (HIV) are at higher risk.[1] Africa, followed by India and China, are among the 15 countries with the highest estimated tuberculosis incidence rates.[1] Despite the availability of highly efficacious treatment for decades, tuberculosis remains an important cause of death. In 2010, there were 8.8 (8.5–9.2) million incident tuberculosis cases, 1.1 (0.9–1.2) million tuberculosis deaths among

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HIV-negative subjects, and an additional 0.35 (0.32–0.39) million deaths due to HIV-associated tuberculosis.

Tuberculosis has been treated with combination therapy for over 50 years, following the recognition early on that *M. tuberculosis* develops resistance rapidly when single drugs are used for treatment.[2] Treatment of both pulmonary and extrapulmonary tuberculosis is usually for 6 months with isoniazid, rifampicin, pyrazinamide, and ethambutol for 2 months, followed by isoniazid and rifampicin for the following 4 months. This regimen is highly efficacious, with cure rates > 95% and relapse rates < 5% in patients with tuberculosis who are not infected by HIV and have drug-susceptible bacilli.[3] Tuberculosis is treated with combination therapy because *M. tuberculosis* develops resistance rapidly when single drugs are used for treatment. Multiple drug therapy is not only to prevent emergence of drug resistance, but also to treat different populations of tubercle bacilli.

Pharmacogenomics

Pharmacogenomics is defined as the study of the genetically determined molecular basis of drug treatment outcomes, and has evolved over the past 20 years. Treatment outcomes can be classified into two broad areas, ie, efficacy and toxicity. Interindividual differences in response to the same drugs are known to occur. These differences are higher among individuals belonging to the same population than within the same individual at different times.[4] Inheritance as a determinant of drug response can be explained by the fact that huge population variations with minimal within-subject variations exist. About 20%–95% of variations in drug pharmacokinetics and effects are estimated to be due to genetic factors,[5] although there are examples in which interindividual variations in drug response are known to occur due to sequence variants in genes coding for drug-metabolizing enzymes, drug transporters, or targets.[6–8] Nongenetic factors, such as age, organ function, concomitant therapy, nutritional status, drug interactions, and nature of the disease can also impact drug effects. However, inherited determinants remain constant throughout the lifetime of the individual.

Clinical observations of inherited differences in drug effects were first reported in the 1950s.[9–12] An important aim of pharmacogenomics is to develop a rational means to optimize drug treatment, thereby ensuring maximum efficacy with minimum toxicity.[13] The concept of "personalized medicine" holds promise in that drugs and drug combinations are optimized for the genetic makeup of the individual.[14,15] Pharmacogenomics has wide applications in several diseases, including cancer, cardiovascular disease, depression, bipolar

disorder, attention deficit disorder, HIV, tuberculosis, asthma, and diabetes.[16]

Pharmacogenomics and tuberculosis

Among the several drugs used to treat tuberculosis, pharmacogenomics is known to play an important role in the metabolism of isoniazid, a key first-line drug used to treat tuberculosis.[17] This in turn could have important clinical implications in terms of therapeutic efficacy[18] and occurrence of adverse events.[19] There are a few reports available that suggest a role with respect to rifampicin,[20,21] while not much information is available regarding genetic differences in the metabolism of other first-line and second-line antituberculous drugs.

While most of the currently available review articles have covered the relationship between pharmacogenomics and isoniazid-induced hepatotoxicity,[19,22] we have extended the scope of this review to include the effect of genetic polymorphisms on the efficacy of treatment for tuberculosis. Further, we have included available information on rifampicin and aminoglycosides.

Isoniazid

Ever since its introduction in 1952, isoniazid has been an important drug in the treatment of tuberculosis. It has all the essential properties of an ideal chemotherapeutic agent, ie, it is highly effective, quite specific in its action against the tubercle bacilli, has minimal toxicity, and is not expensive.[18]

Metabolism

Isoniazid is metabolized by acetylation, which takes place mainly in the liver and gut mucosa. This conjugation reaction is catalyzed by N-acetyl transferase (NAT2) during which an acetyl group from acetyl coenzyme A gets transferred to the acceptor amine, thereby forming an amide. Acetylation of isoniazid results in formation of acetyl isoniazid, which becomes conjugated with pyruvic acid and α-ketoglutaric acid to form hydrazones, and acetyl isoniazid becomes hydrolyzed to monoacetyl hydrazine and isonicotinic acid. Monoacetyl hydrazine undergoes acetylation to form diacetyl hydrazine, and isonicotinic acid becomes conjugated with glycine to form isonicotinyl glycine. All the metabolites of isoniazid are devoid of antituberculous activity. A schematic representation of isoniazid metabolism is shown in Figure 1.[19]
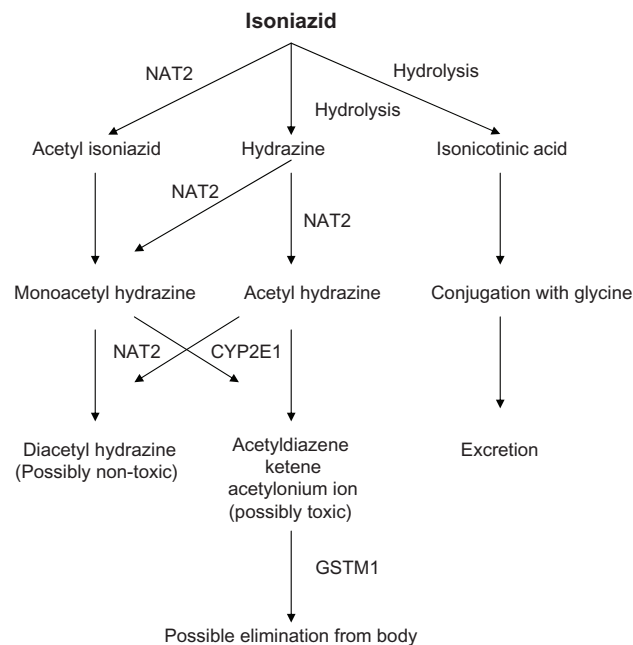
Figure 1 Suggested metabolic pathways of isoniazid and metabolites via NAT2, CYP2E1, and GSTM1. © Copyright Future Medicine. Reprinted with permission from Roy PD, Majumder M, Roy B. Pharmacogenomics of anti-TB drug-related hepatotoxicity. *Pharmacogenomics*. 2008.[19]

Acetylator genotypes and phenotypes

The acetylator status of a person is genetically determined, and the rate of acetylation of isoniazid varies from individual to individual due to differences in the concentrations of cytosolic NAT2 enzyme in the liver and gut mucosa. Many commonly prescribed drugs are detoxified by *NAT2*, which exhibits a high level of polymorphism. Variability in the expression of *NAT2* can therefore affect the therapeutic response to drugs. Individuals with reduced *NAT2* expression may have different drug levels than those with normal expression. Consequently, such persons may need a different dosage schedule of drugs that are metabolized by NAT2.

The enzyme activity is expressed at highly variable levels. Several studies over the years have shown that human subjects show a wide degree of variation in their capacity to acetylate or inactivate isoniazid to acetyl isoniazid, and they can be distinctly characterized phenotypically as being either slow or rapid inactivators (the concentration of the enzyme being higher in rapid inactivators). Figure 2 shows a bimodal distribution of plasma isoniazid concentrations in slow and rapid acetylators given the same dose of isoniazid.[23] Molecular techniques that are now available permit identification of three genotypes, ie, rapid, intermediate, and slow.

Wild-type (rapid acetylator) alleles are those in which the *NAT2*4* allele codes for a completely active enzyme. Rapid acetylators can be administered drugs that are substrates of the *NAT2* enzyme according to conventional doses.
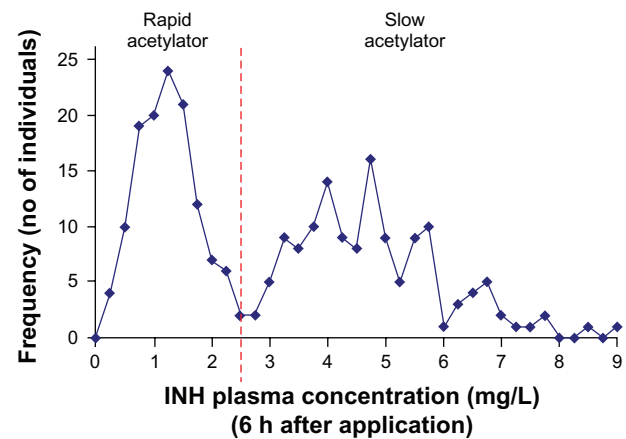

Figure 2 Bimodal variation in isoniazid exposure.[23]
Note: Plasma isoniazid concentrations were determined in subjects at six hours after isoniazid administration.
Abbreviation: INH, isoniazid.

Intermediate acetylators are those with one active and one inactive *NAT2* allele (heterozygous). They might require lower than average drug dosages for a favorable therapeutic response. Several mutations in the *NAT2* gene account for the majority of the slow acetylator genotypes in the human population (*NAT2*5A, NAT2*5B*, and *NAT2*6A*).[24] Such individuals are at greater risk of drug-induced adverse reactions due to reduced drug elimination. Acetylator gene frequency for the slow allele differs across ethnic groups and countries, being 10% in people from the mongoloid races like the Eskimos, Japanese, and Chinese, 90% in the Middle East, 60% in the Negroid, Caucasian, and south Indian populations, and 72% in the US.

A study by Kinzig-Schippers et al in 18 healthy Caucasians showed that *NAT2* genotypes accounted for 88% of variability in apparent isoniazid clearance, while isoniazid preparation and body weight accounted for 2% and 3% of variations, respectively.[24] There exists a linear relationship between isoniazid clearance and number of high-activity *NAT2* alleles (Figure 3).[25] Parkin et al observed variations in isoniazid disposition among subjects with different *NAT2* genotypes, slow acetylators having 4–6-fold higher serum isoniazid concentrations.[26] Based on this study, an individualized isoniazid dosing regimen was suggested. Similarly, a study comparing urinary isoniazid excretion between healthy volunteers and patients showed that subjects with a higher number of active *NAT2* alleles had higher levels of isoniazid acetylation.[27]

Isoniazid acetylator status and treatment efficacy

Scanty information is available on the association between isoniazid blood concentrations, efficacy, and toxicity. Mitchison
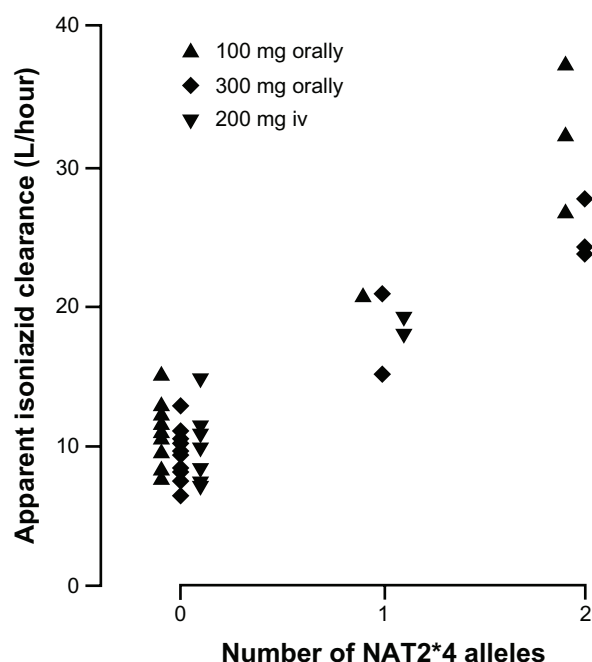
Figure 3 Individual clearance of isoniazid in relation to the number of high-activity *NAT2*4* alleles. © Copyright American Society for Microbiology. Reprinted with permission from Kinzig-Schippers M, Tomalik-Scharte D, Jetter A, et al. Should we use N-acetyl transferase type 2 genotyping to personalize isoniazid doses? *Antimicrob Agents Chemother.* 2005;49(5):1733–1738.[25]

proposed that the peak isoniazid concentration to minimum inhibitory concentration ratio could serve as a predictor of outcomes of tuberculosis treatment.[28] It has been established that plasma isoniazid levels influence the mean early bactericidal activity of isoniazid, which is lower in rapid acetylators than in slow acetylators.[29] Therefore, it is possible that low plasma isoniazid levels in rapid acetylators could be one of the causes for occasional therapeutic failure or relapse.[30] On the contrary, higher levels in slow acetylators could predispose to toxicity. Thus, *NAT2* genotyping prior to isoniazid administration would help clinicians in predicting pharmacokinetic variability, and adjusting isoniazid dose. Isoniazid dosing could be adjusted for subjects with none, one, or two rapid *NAT2* alleles to achieve similar isoniazid exposure.

Studies to understand the relationship between isoniazid acetylator status and treatment outcome in patients with pulmonary tuberculosis receiving treatment with isoniazid alone or in combination with p-amino salicylic acid were undertaken. Comparison of response to tuberculosis treatment between slow and rapid inactivators of isoniazid suggested an association between treatment response and rate of inactivation of isoniazid, and there was a difference in the rate of conversion to bacteriological negativity between slow and rapid inactivators.[31] Under trial conditions, it appears that isoniazid acetylator status has little prognostic

significance during daily, twice-weekly, or three times weekly tuberculosis treatment.[17,32,33] However, in field conditions or in cases of drug irregularity, isoniazid acetylator status may acquire some importance in daily regimens, and may have an appreciable effect on outcomes among patients on three times weekly and twice-weekly regimens.[18] Controlled clinical trials using once-weekly isoniazid-containing treatment regimens demonstrated that response to treatment was better in slow acetylators compared with rapid acetylators, the differences in cure rate being in the order of 20%–35% ($P < 0.001$, Table 1).[34] In order to elucidate the reasons for the inferior response of rapid acetylators, studies of serial serum isoniazid concentrations in slow and rapid acetylators were undertaken. Table 2 presents the peak concentration, coverage, and exposure to isoniazid attained at a dose of 15 mg/kg.[18] The peak concentration attained in slow acetylators was slightly higher than that in rapid acetylators; however, the fall-off was fast in rapid acetylators, but was more gradual in slow acetylators. Coverage, defined as the number of hours for which a bacteriostatic concentration of isoniazid (0.2 μg/mL) is maintained in the blood, was significantly greater in slow acetylators. Similarly, exposure, defined as the area under the concentration-time curve, was significantly greater in slow acetylators. Significant differences in coverage and exposure between slow and rapid acetylators suggested that failure of once-weekly isoniazid regimens was because of suboptimal coverage and exposure among rapid acetylators.[35]

Weiner et al undertook a study to understand the reasons for lower efficacy of once-weekly isoniazid-rifapentine compared with twice-weekly isoniazid-rifampicin. This study showed that poor treatment outcomes were associated with isoniazid acetylator status in patients receiving a once-weekly regimen.[30]

The isoniazid acetylator phenomenon could have important therapeutic implications in certain specific situations, such as during tuberculosis treatment in HIV-infected

Table 1 Prognostic significance of isoniazid acetylator status during once-weekly chemotherapy[30]

Once-weekly phase	Total patients		Favorable response at one year		
	Slow	Rapid	Slow	Rapid	Difference
SH	38	39	82%	60%	22%
SHZ	67	34	87%	53%	34%
SH	109	67	94%	73%	21%
SPH	98	72	95%	76%	19%
EH	57	51	91%	57%	34%

Abbreviations: S, streptomycin; H, isoniazid; P, sodium para-aminosalicylic acid; E, ethambutol; Z, pyrazinamide.

Table 2 Peak, coverage, and exposure following a dose of isoniazid 15 mg/kg body weight among patients with pulmonary tuberculosis[18]

Isoniazid inactivation rate	Peak concentration (µg/mL)	Coverage (h)	Exposure (µg/mL · h)
Slow	13.8	30	85
Rapid	10.8	14	36

individuals and in patients with chronic renal failure. A study undertaken by our group to understand the influence of HIV infection on the pharmacokinetics of antituberculous drugs, showed that differences in peak concentration and exposure of isoniazid between HIV-infected and HIV-noninfected patients with tuberculosis were more significant in rapid acetylators than in slow acetylators. Hence, among HIV-infected patients with tuberculosis, where bioavailability of antituberculous drugs has been shown to be suboptimal, rapid acetylators are at a further disadvantage.[36,37]

Determination of isoniazid acetylator status could be particularly important in patients with tuberculosis with chronic renal failure. The pharmacokinetics of isoniazid was compared between patients with chronic renal failure and healthy volunteers. While plasma isoniazid concentrations were similar in rapid acetylators and healthy subjects, peak isoniazid concentrations, exposure, and half-life were significantly higher in slow acetylators with renal failure than in healthy subjects ($P < 0.01$). This study suggested that it was necessary to reduce the dose of isoniazid in slow acetylators to reduce the incidence of adverse events.[38]

Donald et al studied the impact of isoniazid dose and *NAT2* genotype and phenotype on the pharmacokinetics and pharmacodynamics of isoniazid in adult pulmonary patients with tuberculosis. This study showed that fast acetylators of isoniazid receiving a daily dose of 6 mg/kg had similar exposure to that of slow acetylators who received a daily dose of 3 mg/kg. It was suggested that reducing the isoniazid dose below 6 mg/kg would be disadvantageous for fast acetylators, but 3 mg/kg was sufficient for slow acetylators to achieve optimal isoniazid exposure.[39]

In a population pharmacokinetic study of isoniazid in South African patients with tuberculosis by Wilkins et al, a model was developed to understand the pharmacokinetic variability of isoniazid. This study observed that the existing treatment guidelines could cause suboptimal exposure in rapid acetylators of isoniazid.[40]

A pharmacokinetic study by Jeena et al in children with tuberculosis suggested that isoniazid doses may have to be individualized based on age, acetylator status, and disease process.[41] Other studies have also shown that decreased isoniazid exposure was likely to impact tuberculosis treatment outcomes in children who were rapid acetylators compared with slow acetylators.[42–44] A recent pharmacokinetic study of antituberculous drugs undertaken in children at our center showed that isoniazid acetylator status significantly influenced drug levels and tuberculosis treatment outcome (unpublished data).

Isoniazid-induced toxicity

Isoniazid is quite nontoxic in conventional doses and is known to cause only two well recognized adverse reactions. Hepatotoxicity is the most common and serious adverse reaction that could occur during treatment of tuberculosis. The incidence of hepatotoxicity during antituberculous treatment is reported to vary from 2% to 28%.[22] The other adverse event is peripheral neuropathy, which is common with high doses, particularly among slow acetylators.[45] It is reversible and can be prevented by concomitant pyridoxine administration.[46] These toxicities have been shown to be related to the slow clearance of acetyl hydrazine, a toxic metabolite of isoniazid in affected patients, who were identified as slow acetylators.[19]

Hepatotoxicity is a major toxic effect of isoniazid. This has caused concern because isoniazid is widely used in preventive therapy programs, apart from treating tuberculosis. Isoniazid undergoes metabolism by human enzymes to produce toxic chemicals that are likely to cause liver toxicity. Isoniazid-induced hepatotoxicity has been reported to be influenced by NAT2 acetylation, oxidation by cytochrome P450 oxidase (CYP) 2EI, and detoxification by glutathione S-transferase (GST, Figure 1).[19]

NAT2 polymorphisms

A few studies have attempted to establish the molecular and chemical basis for the occurrence of isoniazid-induced hepatotoxicity. Mitchell et al have proposed that the incidence of hepatotoxicity due to isoniazid could be greater in rapid acetylators than in slow acetylators. This is due to the fact that rapid acetylators produce monoacetyl hydrazine from isoniazid faster than slow acetylators; monoacetyl hydrazine subsequently gets converted by the CYP-dependent hepatic microsomal enzyme system to a potent acetylating agent which could cause hepatic necrosis.[47] However, this hypothesis was questioned because of the fact that formation of monoacetyl hydrazine was similar in the two phenotypes, the more rapid formation for monoacetyl hydrazine being

offset by its more rapid elimination via diacetyl hydrazine in rapid acetylators.[48] Research reported by Sarma et al has shown that hydrazine formed from isoniazid is responsible for the hepatotoxic action of isoniazid metabolites. This reaction is modified by metabolic induction by rifampicin, which induces isoniazid metabolism by inducing isoniazid hydrolase, thereby producing isonicotinic acid and hydrazine.[49] This direct pathway of isoniazid metabolism induced by rifampicin was shown to be more pronounced in slow acetylators than in rapid acetylators.

A 28-fold higher risk of isoniazid-induced hepatotoxicity in slow acetylators compared with rapid acetylators has been reported in Japanese and Taiwanese populations.[50,51] Another study in a Korean population observed that slow *NAT2* acetylators had a 3–8-fold risk of developing isoniazid-induced hepatotoxicity ($P = 0.005$). This study suggested that *NAT2* genotyping could serve as a useful tool to predict antituberculous drug-induced hepatotoxicity.[52] Slow acetylator status of *NAT2* as a risk factor for isoniazid-induced hepatotoxicity was observed in a study conducted in Tunisian patients with tuberculosis, and the *NAT2*53/5B*, *NAT2*6A/6A*, *481T/T*, and *590A/A* diplotypes could serve as useful biomarkers for prediction of antituberculous drug-induced toxicity.[53] A retrospective study in 102 Japanese patients with tuberculosis showed that the occurrence of side effects was greater in slow acetylators than in rapid and intermediate acetylators.[54] A recent meta-analysis by Wang et al of 14 studies, comprising 474 cases and 1446 controls, showed a significant association between slow *NAT2* acetylators and risk of antituberculous drug-induced liver toxicity.[55] The odds ratio (OR) for slow *NAT2* acetylators compared with rapid acetylators was 4.7 (95% confidence interval [CI] 3.3–6.7, $P < 0.001$). Subgroup analysis indicated that both Asian and non-Asian slow acetylators develop liver damage more frequently. On comparing intermediate *NAT2* acetylators with rapid *NAT2* acetylators, the OR was 1.3 (95% CI 0.93–1.71, $P < 0.14$). Another subgroup analysis for the different treatment regimens showed that the combined OR for isoniazid + rifampicin, isoniazid + rifampicin + pyrazinamide + ethambutol, and isoniazid were, respectively, 34.3 (95% CI 10.4–113.0, $P < 0.001$), 4.1 (95% CI 2.8–6.0, $P < 0.001$), and 2.4 (95% CI 0.52–10.73, $P < 0.27$) on comparing slow *NAT2* acetylators with rapid acetylators. Identifying genetic *NAT2* polymorphisms in patients could help clinicians to prevent hepatotoxicity and achieve better patient management.[55]

Contrary to these findings, a case-control study undertaken in patients of Caucasian origin with tuberculosis did not find any association between increased risk of antituberculous drug-induced liver injury and the presence of slow *NAT2* polymorphisms.[56] Genotyping studies in an Indian population[57] and in a heterogenous population consisting of Hispanics, Africans, Caucasians, South Americans, and Asians[58] has reported a lack of association between isoniazid-induced hepatotoxicity and *NAT2* acetylator status.

CYP2E1 polymorphisms

CYP2E1 is one of the enzymes of the hepatic microsomal enzyme system, and brings about conversion of acetyl hydrazine to hepatotoxins, eg, acetyl diazene, ketene, and the acetylonium ion.[59] It has been proposed that isoniazid or its metabolite, hydrazine, could induce *CYP2E1* activity in the rat, and that this activity is driven by blood isoniazid levels.[60] On the other hand, isoniazid could also inhibit the activity of *CYP2E1,* this being more prominent in individuals with the variant *CYP2E1* genotype.[61] Hence, *CYP2E1* activity is likely to be increased in individuals with a common genotype at *CYP2E1* compared with those with the variant genotype. The risk of hepatotoxicity could increase due to enhanced *CYP2E1* activity via increased production of hepatotoxins.

Lee et al have reported similar findings, and also observed that *CYP2E1* was related to the severity of antituberculous drug-induced hepatotoxicity.[62] A study from Taiwan showed that the common *1A/*IA* genotype of *CYP2E1* enhanced the odds of isoniazid-induced liver toxicity by 2.5 times in adult patients with tuberculosis.[61] Further, a combination of the *1A/*1A* genotype and slow *NAT2* acetylation status increased the risk of hepatotoxicity (OR 7.43, 95% CI 2.4–22.8) compared with the presence of any one polymorphism. Another study in Indian pediatric patients observed that the risk of liver toxicity was enhanced by the variant *CYP2E1*6* allele and *IA*6*ID* haplotype at *CYP2E1* (OR 11.0, 95% CI 1.02–11.0 and OR 4.6, 95% CI 1.3–16.3, respectively).[63] The common *IA* allele at *CYP2E1* increasing the levels of liver enzymes was observed in a heterogenous group of Asians, Africans, Caucasians, South Americans, Hispanics, and Africans.[56] However, a study done in a Korean population did not find an association between liver toxicity and *CYP2E1* polymorphisms.[52]

GSTM1 and GSTT1 polymorphisms

The GSTs are a group of enzymes involved in the detoxification of carcinogens, toxic chemicals, and medications.[64,65] GSTs bring about conjugation of glutathione with the substrate, for solubilization and elimination from the body. The GST enzymes are coded by at least five distinct loci. Of these, *GSTM1* and *GSTT1* are reported to be associated

with hepatotoxicity.[52,66] GSTs are reported to play a key role in the metabolism of isoniazid. Glutathione serves as a scavenger of free radicals intracellularly by conjugating with toxic metabolites of isoniazid. Inadequacy of GST activity due to homozygous deletion at *GSTM1* and *GSTT1* loci could influence susceptibility to isoniazid-induced liver toxicity. The risk of isoniazid-induced hepatotoxicity has been shown to be increased (OR 2.13, 95% CI 1.25–3.10) in Indian patients with homozygous *GSTM1* mutation.[52] While Taiwanese patients with homozygous *GSTM1* deletion were found to have twice the risk of isoniazid-induced hepatotoxicity in a Taiwanese population, this did not occur in those with homozygous *GSTT1* deletion.[52,66] Similar studies with reasonable sample sizes now have to be done in different ethnic populations. Identifying patients with homozygous *GSTM1* deletion may help in the prevention and better management of isoniazid-induced hepatotoxicity.

Manganese superoxide dismutase

It is known that hepatic damage can be caused by reactive oxygen species formed from various reactions. Manganese superoxide dismutase brings about a decrease in the load of reactive oxygen species in the mitochondria. A study from Taiwan showed that genotypes with the variant C allele (T > C polymorphism at codon 47; alanine > valine) enhanced the risk of antituberculous drug-induced liver toxicity.[66] This is probably because manganese superoxide dismutase with the variant amino acid, valine, at codon 47 augmented formation of toxic hydrogen peroxide, which can lead to hepatotoxicity.

Human leucocyte antigen alleles

The lack of HLADQA1*0102 and existence of DQB1*0201 alleles have been reported to be independently associated with an enhanced risk of antituberculous-induced hepatotoxicity in Indian patients.[67]

Rifampicin

Rifampicin is an important first-line drug used to treat tuberculosis. It has shown concentration-dependent activity against *M. tuberculosis* under in vitro and in vivo conditions.[68–70] Organic anion transporter peptides play an important role in the transport and disposition of drugs in the human body. Rifampicin is a substrate of drug transporters such as P-glycoprotein and OATP1B1. Nuclear receptors, ie, pregnane X receptor and constitutive androstane receptor, transcriptionally regulate the drug transporters. Weiner et al undertook a pharmacokinetic study to explore the reasons for the huge interindividual variations in rifampicin levels seen in patients

receiving standard treatment dosages. The pharmacokinetics of rifampicin was compared between patients with tuberculosis and healthy subjects, between regions (Africa versus non-Africa), and between races (black versus other races). The relationship between rifampicin pharmacokinetics and polymorphisms of drug influx and efflux transporter genes (anion transporting polypeptides [*SLCO1B1* 463C > A and 521T > C and *SLCO1B3*] and P-glycoprotein [*ABCB1*]) was also studied.[20] The study findings in 72 adult pulmonary patients with tuberculosis from the US, Africa, and Spain showed that polymorphisms in the *SLCO1B1* gene had a significant influence on rifampicin exposure, this being 36% lower among *SLCO1B1* 463 CA genotypes than CC genotypes (29.8 versus 46.7 μg $\cdot$ h/mL; $P = 0.001$, Figure 4).[20] Further, *SLCO1B1* gene polymorphisms related to reduced rifampicin exposure were more pronounced in black subjects. This study was the first to report an association between rifampicin exposure and *SLCO1B1* 463 C > A gene polymorphism. Marked interindividual variations in rifampicin exposure were probably due to this polymorphism.

Chigutsa et al undertook a study in South African patients with tuberculosis to examine the impact of *ABCB1*, *SLCO1B1*, *PXR*, and *CAR* polymorphisms on the pharmacokinetics of rifampicin.[21] Patients heterozygous and homozygous for the variant allele of *SLCO1B1* rs 4149032 polymorphism had decreases in rifampicin bioavailability of 20% and 28%, respectively. This polymorphism explained 21% of the between-subject variability in drug clearance.
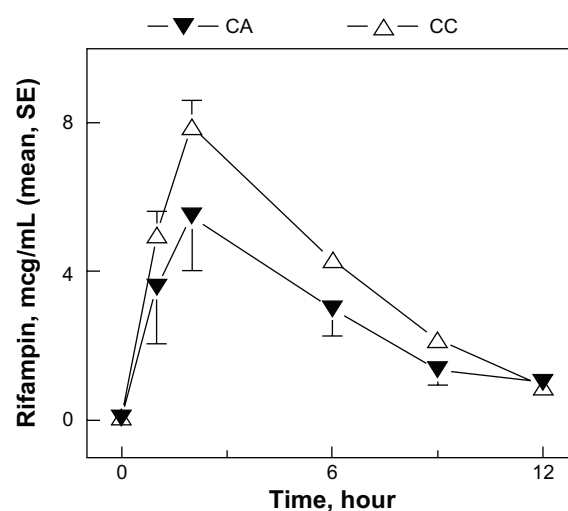

Figure 4 Plasma rifampin concentrations (μg/mL) versus time (hours) in patients grouped by *SLCO1B1* c.463CA polymorphisms (▼) versus *SLCO1B1* c.463CC genotypes (Δ). © Copyright American Society for Microbiology. Reprinted with permission from Weiner M, Peloquin CA, Burman W, et al. Effects of tuberculosis, race, and human gene SLCO1B1 polymorphisms on rifampin concentrations. Antimicrob Agents Chemother. 2010;54(10):4192–4200.[20]
Note: Arithmetic mean and standard error shown with vertical bars.

Simulations revealed that an increase in rifampicin dose of approximately 30% in patients harboring the polymorphism resulted in plasma rifampicin levels similar to those for the wild-type allele, with a typical peak concentration above the recommended minimum of 8 µg/mL. However, the other polymorphisms, ie, for *ABCB1*, *PXR*, and *CAR*, did not have any significant impact on the pharmacokinetics of rifampicin. This study suggests that an increase in rifampicin dose would be desirable for carriers of the *SLCOB1B1* polymorphism. More studies in different populations are required to understand the role of *SLCOB1B1* polymorphisms in influencing plasma rifampicin concentrations and their role in impacting tuberculosis treatment outcomes.

Aminoglycosides

Aminoglycosides, such as streptomycin, kanamycin, and amikacin, are used in the treatment of tuberculosis. Ototoxicity is an adverse event that can arise following the use of aminoglycosides. Zhao et al undertook a molecular characterization study in a Chinese family to understand the association between aminoglycoside-induced ototoxicity and mitochondrial mutations. It was observed that a C to T transition at position 1494 in the mitochondrial 12S rRNA gene was responsible in individuals genetically predisposed to ototoxicity.[71] It was likely that the deafness phenotype linked to the C > T 1494 12S rRNA gene polymorphism could be induced or worsened by aminoglycosides.

Translating pharmacogenomics to the clinic

Although genetic polymorphisms are known to impact drug effects, individualized drug therapy is seldom used in clinical practice. This could be because it is difficult to carry out well designed clinical pharmacogenomic studies to prove that individualization of drug treatment on a genetic basis helps in improving clinical outcomes. The multigenic nature of most drug effects and practical problems in controlling for nongenetic confounders, eg, drug interactions, diet, and smoking, are some of the limitations in conducting such studies. Prospective clinical trials on a large scale are necessary to quantitate drug response objectively. In order to achieve this, it is important to obtain genomic DNA from all patients recruited into clinical trials, and obtain consent to carry out pharmacogenetic studies. It would be ideal to couple these trials with preclinical experimental models that reinforce clinical associations between genotypes and phenotypes. Inclusion of pharmacogenetic testing in clinical trials is necessary for clinical progress. Because of

marked population heterogeneity, there is a need to validate pharmacogenomic relationships for every therapeutic indication and in various ethnic/racial groups. This would help in reducing the occurrence of adverse drug effects and enhancing treatment success, and in the long run will probably decrease the cost of health care. Pharmacogenomic studies have the potential to achieve this by translating genome variability into improved therapeutics.

Conclusion

Among the antituberculous drugs, isoniazid metabolism and disposition is known to be impacted by genetic factors. The relationship between ethnicity, genetic background, and response to tuberculosis treatment has not been well studied. It has been reported that no single gene variant of *NAT2* or *CYP2E1* is significantly associated with isoniazid-driven hepatotoxicity.[72] Thus, a combination of polymorphisms could impact the pharmacokinetics of isoniazid. For most antituberculous drugs, both efficacy and toxicity are most likely determined by multiple factors, with both genetic and non-genetic factors playing a role. Large clinical trials will be needed to evaluate prospectively the merits of using genotyping and adjustment of tuberculosis drug doses during treatment. More studies are also required to understand the influence of *SLCO1B1* gene polymorphism on rifampicin pharmacokinetics, as well as polymorphisms affecting the metabolism of other first-line and second-line antituberculous drugs. These studies must link pharmacogenetic data with tuberculosis treatment outcomes. Ultimately, we also have to bear in mind the cost, cost-effectiveness, and feasibility of using pharmacogenomics in routine tuberculosis control programs. Unless the benefits of genetic testing and individualizing treatment are worth the cost of such an exercise, the approach will not find many takers, because tuberculosis is mainly a disease of poverty.

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Disclosure

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