Cytochrome P450 2D6 Phenoconversion Is Common in Patients Being Treated for Depression: Implications for Personalized Medicine

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ABSTRACT

Objective: Determine the point prevalence of phenoconversion to cytochrome P450 2D6 (CYP2D6) poor metabolizer status in clinical practice.

Method: This multicenter, open-label, single-visit naturalistic study was conducted from October 2008 to July 2009 in adult patients (≥ 18 years) who had been receiving venlafaxine extended-release (ER) (37.5–225 mg/d) treatment for up to 8 weeks. A 15-mL blood sample was drawn 4 to 12 hours after patients' last venlafaxine ER dose. Plasma *O*-desmethylvenlafaxine and venlafaxine concentrations were determined for each patient. CYP2D6 poor metabolizer phenotype was defined as *O*-desmethylvenlafaxine to venlafaxine ratio < 1 based on published data. *CYP2D6* genotype was determined for each patient; patients were classified as poor metabolizer, intermediate metabolizer, extensive metabolizer, and ultrarapid metabolizer. Agreement between poor metabolizer phenotype and genotype classifications was assessed using the McNemar test.

Results: Phenoconversion to CYP2D6 poor metabolizer status occurred in 209 of 865 individuals (24%) with a CYP2D6 non–poor metabolizer genotype. The incidence of CYP2D6 poor metabolizer status based on phenotype was almost 7 times higher than that expected based on genotype: only 4% (35/900) of patients were genotypic CYP2D6 poor metabolizers, but 27% (243/900) were phenotypic CYP2D6 poor metabolizers (McNemar test, $P < .0001$).

Conclusions: CYP2D6 phenotype conversion is common in patients being treated for depression. These results are important because differences in CYP2D6 drug metabolic capacity, whether genetically determined or due to phenoconversion, can affect clinical outcomes in patients treated with drugs substantially metabolized by CYP2D6. These results demonstrate that personalized medicine based solely on genetics can be misleading and support the need to consider drug-induced variability as well.

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The concept of "personalized medicine" has gained widespread interest, as genes that may underlie individual differences in clinical response to medications have been identified.[1–4] According to this concept, clinicians could select among treatment options the medication and dosage with the greatest efficacy and fewest side effects for an individual patient based on his or her genetic profile. However, the patient's current functional status (ie, phenotype) may be more clinically relevant than his/her genotype (eg, cytochrome P450 [CYP] 2D6–mediated oxidative drug metabolism). Thus, application of personalized medicine requires understanding and consideration of the implications of relevant nongenetic factors (including environmental and personal variables) in addition to genetic factors.[5]

The hepatic CYP system is the principal phase 1 metabolic pathway for most clinically used drugs[6,7]; CYP2D6 is responsible for approximately 15% of oxidative metabolism of drugs in humans.[8] The CYP2D6 isozymes are among the most extensively studied gene polymorphisms associated with individual differences in medication efficacy or tolerability.[9–16] Over 100 different human *CYP2D6* alleles have been identified,[17] including nonfunctional alleles and alleles with decreased, normal, or increased function.[9] Accordingly, individuals can be classified as CYP2D6 poor metabolizers, intermediate metabolizers, extensive metabolizers, or ultrarapid metabolizers based on the combination of *CYP2D6* alleles they carry.[6,9] A CYP2D6 poor metabolizer clears CYP2D6 substrate drugs slowly and, therefore, has greater than expected exposure to the parent drug and lower than expected exposure to active metabolites. Both of these phenomena can negatively affect tolerability or efficacy, depending on the nature of the CYP2D6 drug the patient is taking.

In formal pharmacokinetic studies, administration of drugs that are substantial CYP2D6 inhibitors (eg, paroxetine, fluoxetine) has been shown to convert up to 80% of individuals with a non–poor metabolizer genotype to poor metabolizer phenotype in small trials (N = 12–31) of healthy volunteers[18,19] and patients with depression.[20] Such findings are clinically important because multiple medication use is common in clinical practice[21] and increases the risk of CYP2D6 phenoconversion. Thus, genotyping is likely to underestimate the true prevalence of the CYP2D6 poor metabolizer phenotype, particularly in patient populations in which multiple medication use is high, such as those being treated with antidepressants.[21]

To date, no large-scale antidepressant studies have assessed the incidence of phenoconversion to CYP2D6 poor metabolizer status in patients in clinical practice. The objective of this study was to provide those data by using a large clinical sample of depressed outpatients receiving treatment with venlafaxine extended release (ER). Venlafaxine ER (a mild inhibitor of CYP2D6) was chosen because it is principally metabolized by CYP2D6, which catalyzes *O*-demethylation

Clinical Points

- Genotyping in clinical practice may significantly underestimate the incidence of CYP2D6 poor metabolic capacity in depressed patients for whom multiple drugs are prescribed.
- Differences in CYP2D6 drug metabolic capacity, whether genetically determined or due to phenoconversion, can affect clinical outcomes in patients treated with drugs substantially metabolized by CYP2D6.
- When prescribing treatment for depressed patients, particularly those with chronic medical disorders taking concomitant medications, it is important for clinicians to consider their current functional capacity (ie, phenotype) to metabolize and clear a drug, which is more clinically relevant than their genetic potential capacity to do so.

to the major active metabolite, *O*-desmethylvenlafaxine and, hence, the ratio of *O*-desmethylvenlafaxine to venlafaxine can be used to determine extensive metabolizer and poor metabolizer phenotype status.[22,23]

METHOD

The study protocol received institutional review board approval before the study began. The study was designed and performed in compliance with Good Clinical Practice and applicable regulatory requirements and was conducted according to the ethical principles in the Declaration of Helsinki.[24] The study was registered on ClinicalTrials.gov (identifier: NCT00788944). Patients' identities were kept confidential; written informed consent was obtained from all participants before enrollment.

Patients

Eligible patients were depressed men and women aged ≥ 18 years receiving outpatient treatment with venlafaxine ER (Effexor XR only; generic venlafaxine ER, which was not widely available at the time of this study, was not permitted) at a dosage within the range approved by the US Food and Drug Administration for depression (37.5–225 mg/d) for ≤ 8 weeks at time of enrollment (because the study recruited patients who were already being treated with venlafaxine ER, no test article was provided by the sponsor to study participants). Patients were enrolled if they were able to have blood drawn within 4 to 12 hours of the most recent dose of venlafaxine ER. Patients were excluded if they had been previously treated with venlafaxine ER within 6 months prior to the study visit (other than the current regimen, to ensure a total treatment duration of ≤ 8 weeks) or had used desvenlafaxine or any unknown investigational study drug within 30 days prior to the study visit. All other concomitant medications were permitted.

Study Design

This open-label, single-visit, naturalistic study was conducted from October 2008 to July 2009 at 50 US sites. At the study visit, patients' demographic information, medical history, and use of tobacco, alcohol, caffeine, and grapefruit were recorded. All concomitant medications used within 7 days prior to the study visit (≤ 30 days for fluoxetine, paroxetine, terbinafine, or bupropion), start and stop dates, dosages, frequency of administration, and medical indications were recorded. A 15-mL whole blood sample for determination of plasma *O*-desmethylvenlafaxine and venlafaxine concentrations and *CYP2D6* genotyping was drawn 4 to12 hours after the patient's most recent venlafaxine ER dose. Protocol-related adverse events were collected from the time informed consent was signed until 24 hours after the blood draw.

Bioanalytical Methodology

Venlafaxine and *O*-desmethylvenlafaxine concentrations were assayed in 5-mL plasma samples by Cetero Research (Houston, Texas) (formerly known as BA Research International) using a validated liquid chromatography with tandem mass spectrometry method.[25] The interday precision (percentage coefficient of variation) for the venlafaxine standards was 4.3% or better and the accuracy (%Bias) ranged from –1.5% to 1.6%. For the *O*-desmethylvenlafaxine standards, the percentage coefficient of variation was 1.3% or better and %Bias ranged from –1.9% to 1.0%. The lower and upper limits of quantitation were 2.0 ng/mL and 500.0 ng/mL, respectively, for both compounds.

CYP2D6 Phenotyping and Genotyping

Phenotype (poor metabolizer vs non–poor metabolizer) was assigned based on the ratio of *O*-desmethylvenlafaxine to venlafaxine concentrations in plasma. On the basis of published data,[26] the *O*-desmethylvenlafaxine to venlafaxine ratio is clearly distinguished in subjects genotyped as extensive metabolizers (1 or 2 fully active *CYP2D6* gene alleles) versus poor metabolizers (no active *CYP2D6* gene alleles). *O*-desmethylvenlafaxine to venlafaxine ratios (based on plasma concentrations) were consistently ≥ 1 in genotypic extensive metabolizers and < 1 in genotypic poor metabolizers for blood samples obtained at least 4 hours after administration of venlafaxine.[26] This ratio of *O*-desmethylvenlafaxine to venlafaxine was found to be an effective means of phenotyping individuals according to their CYP2D6 metabolizer status. In the current study, the patient was classified as a poor metabolizer if the *O*-desmethylvenlafaxine to venlafaxine ratio at 4–12 hours postdose was < 1 and a non–poor metabolizer if the ratio was ≥ 1. Because phenotype determination was based on the ratio of *O*-desmethylvenlafaxine to venlafaxine levels rather than absolute levels, the dose strength would not be expected to influence the results, and the full range of approved doses (37.5–225 mg/d) was permitted. Achievement of a steady state venlafaxine level was not a requirement prior to sampling because the method had been shown to be effective in both single- and multiple-dose settings.[26]

Ethylenediaminetetraacetic acid whole blood samples (5 mL) were shipped frozen to QPS (Newark, Delaware) for *CYP2D6* genotyping and stored at –80°C until processed. To isolate genomic DNA, lysis buffer (Buffer AL; Qiagen, Valencia, California) and proteinase K were incubated with

200 µL of whole blood at 56°C for 10 minutes, 200 µL of 100% ethanol was added to each sample, and samples were spun at 6,000 × g for 1 minute in a QIAamp (Qiagen) spin column. All samples were processed in parallel and washed using 500 µL of Buffer AW1 and AW2 (Qiagen), spinning at 8,000 rpm and 13,200 rpm, respectively. Spin columns were then centrifuged for 1 minute at 13,200 rpm to dry the membranes, and samples were eluted by using 200 µL of elution Buffer AE (Qiagen) by placing each spin column into a fresh 1.5-mL microfuge tube and centrifuging at 8,000 rpm for 1 minute.

Genomic DNA concentration and purity (A_{260}/A_{280} ratio) were determined using the NanoDrop ND-1000 spectrophotometer (Thermo Scientific, Waltham, Massachusetts), and samples were screened for the following polymorphisms: **1* Wild Type, **3* (2549delA), **4* (1846G > A), **5* (*CYP2D6* deletion), **6* (1707delT), **7* (2935A > C), **8* (1758G > T), **10* (100C > T), **17* (1023C > T), **29* (3183G > A), **40* (1863_1864ins[TTT CGC CCC]2), **41* (2988G > A), and **2xN* (*CYP2D6* duplication). Positive control genomic DNA samples were purchased from the Coriell Institute for Medical Research (Camden, New Jersey) or ParagonDx (Morrisville, North Carolina).

Screening for the **3*, **6*, **7*, **8*, and **10* polymorphisms was performed by a validated assay using pyrosequencing technology (Qiagen). Genomic DNA samples were standardized at 2 ng/µL, and the polymerase chain reaction amplification was performed by using HotStart DNA Polymerase Reagents (Qiagen) and protocols. Primers used for polymerase chain reaction amplification and downstream sequencing were obtained from Integrated DNA Technologies, Inc (Coralville, Iowa). The amplified DNA region of interest was denatured into a single-stranded fragment, and a sequencing primer was annealed to the fragment in a 96-well plate format. Sequencing reactions were performed by using the PSQ HS96 (Qiagen).

TaqMan assays (Applied Biosystems Inc, Foster City, California) were used to screen for the **4* (ID# C_27102431_B0), **17* (ID# C_2222771_40), **29* (ID# C_34816113_20), **40* (ID# C_32407240_20), **41* (ID# C_34816116_20), and **5/*2xN* (Hs00010001_cn) polymorphisms. Genomic DNA samples were standardized at 2 ng/µL, and samples were amplified and analyzed on the ABI 7900HT (Applied Biosystems Inc).

CYP2D6 metabolizer genotype designation was determined (blinded) at the Wyeth Biomarker Laboratory, Collegeville, Pennsylvania. The **1* allele was defined as the absence of alleles that completely abolish CYP2D6 activity (**3*, **4*, **5*, **6*, **7*, and **8*), alleles that decrease CYP2D6 activity (**9*, **10*, **17*, **29*, **40*, or **41*), and gene duplication. Allele combinations having at least one **1* allele, except with gene duplication in the presence of a **1/*1* genotype, resulted in a genotypic extensive metabolizer designation. Patients with gene duplication in the presence of a **1/*1* genotype were designated genotypic ultrarapid metabolizers. Patients with 2 alleles that completely abolish CYP2D6 activity were designated genotypic poor metabolizers. Those with an allele that decreases CYP2D6 activity paired with a like allele, or one that abolishes activity, were designated genotypic intermediate metabolizers.

Table 1. Demographic Characteristics of Study Completers[a]

Characteristic	Genotypic Non–Poor Metabolizer (n = 865)	Genotypic Poor Metabolizer (n = 35)	Total (N = 900)
Age, y			
Mean (SD)	44.6 (14.5)	44.9 (15.1)	44.7 (14.5)
Range	18–92	18–77	18–92
Sex, n (%)			
Female	531 (61)	21 (60)	552 (61)
Male	334 (39)	14 (40)	348 (39)
Race, n (%)			
White	733 (85)	34 (97)	767 (85)
Black or African American	107 (12)	1 (3)	108 (12)
American Indian or Alaska Native	7 (< 1)	0	7 (< 1)
Asian	7 (< 1)	0	7 (< 1)
Other	11 (1)	0	11 (1)
Ethnicity, n (%)			
Hispanic/Latino	68 (8)	3 (9)	71 (8)
Non–Hispanic/Latino	733 (92)	32 (91)	829 (92)

[a]Completers (N = 900) comprised all patients who had both *O*-desmethylvenlafaxine to venlafaxine ratio and genotype results available for analysis. Missing data were due to errors in blood sample collection, processing, or analysis.

Statistical Analysis

Prevalence rates were calculated for designated extensive metabolizer, poor metabolizer, intermediate metabolizer, and ultrarapid metabolizer genotypes and for poor metabolizer and non–poor metabolizer phenotypes. Agreement between phenotype and genotype (poor metabolizer vs non–poor metabolizer) classifications was assessed using the McNemar test. The poor metabolizer versus non–poor metabolizer genotype classification used for this analysis was consistent with the phenotype classification; determination of phenotype based on *O*-desmethylvenlafaxine to venlafaxine ratio has not been tested for its potential to distinguish among extensive metabolizer, ultrarapid metabolizer, and intermediate metabolizer groups.[26] The proportion of patients taking concomitant medications was determined with a focus on the proportion taking concomitant inhibitors and substrates of CYP2D6 and CYP3A4, a minor metabolic pathway for venlafaxine,[23] to allow for examination of the potential relationship to poor metabolizer phenotype prevalence.

RESULTS

A total of 970 patients were enrolled in the study; 900 patients had both *O*-desmethylvenlafaxine to venlafaxine ratio and genotype results available for analysis (completers). Table 1 shows demographic characteristics of study completers.

Prevalence of Genotypic and Phenotypic *CYP2D6* Poor Metabolizer Status

Overall, 35/900 patients (3.9%; 95% CI, 2.63%–5.15%) were classified as genotypic CYP2D6 poor metabolizers, whereas 243/900 (27.0%; 95% CI, 24.1%–29.9%) were

classified as phenotypic poor metabolizers based on the ratio of *O*-desmethylvenlafaxine to venlafaxine < 1. There was a statistically significant lack of agreement between the genotypic and phenotypic CYP2D6 poor metabolizer status classification (McNemar test, $P < .0001$; Figure 1A). Prevalence rates for the poor metabolizer genotype in black/African-American (108 completers) and white patients (767 completers) were 0.9% (95% CI, 0%–2.73%) and 4.4% (95% CI, 2.98%–5.89%), respectively; poor metabolizer phenotype rates were 22.2% (95% CI, 14.38%–30.06%) and 27.2% (95% CI, 24.1%–30.4%), respectively.

Phenoconversion

Rate of conversion to phenotypic poor metabolizer status in patients with CYP2D6 extensive metabolizer, intermediate metabolizer, or ultrarapid metabolizer genotypes was 24.2%. Although 34/35 (97%) genotypic CYP2D6 poor metabolizers had a concordant poor metabolizer phenotype (for 1 genotypic poor metabolizer, *O*-desmethylvenlafaxine to venlafaxine ratio = 1.58), 160/784 (20%) genotypic extensive metabolizer or ultrarapid metabolizer patients were classified as phenotypic poor metabolizers. A greater proportion of genotypic extensive metabolizers (159/748; 21%) converted to phenotypic CYP2D6 poor metabolizer status; only 1/36 (3%) ultrarapid metabolizers (*O*-desmethylvenlafaxine to venlafaxine ratio = 0.180) was classified as a phenotypic poor metabolizer (Figure 1B).

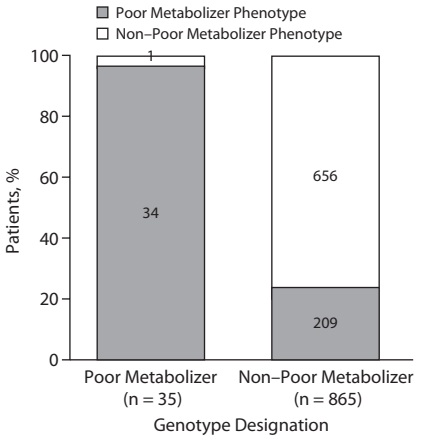
Median *O*-desmethylvenlafaxine to venlafaxine ratios for patients who took no concomitant CYP2D6 inhibitors (Table 2) were consistent with published phenotyping decision rules.[26] Median *O*-desmethylvenlafaxine to venlafaxine ratio was greater than 1 for all extensive metabolizer genotype allele combinations, and less than 1 for all poor metabolizer genotype allele combinations. Of allele combinations with an intermediate metabolizer genotype designation, however, 2 had median *O*-desmethylvenlafaxine to venlafaxine ratios greater than 1; 3 had median *O*-desmethylvenlafaxine to venlafaxine ratios less than 1 (Table 2). This result was not unexpected, given that the decision rule was developed based on pharmacokinetic data from extensive metabolizers and poor metabolizers only.[26] Since the expected phenotype based on *O*-desmethylvenlafaxine to venlafaxine ratio is unknown, assessment of phenoconversion in the intermediate metabolizer individuals (n = 81) was problematic. Some individuals who were genetically classified as intermediate metabolizers (particularly those with **4*41*) had *O*-desmethylvenlafaxine to venlafaxine ratios as low as genotypic poor metabolizers, and, thus, this genotype may need further study to determine whether they should be classified as poor metabolizers or intermediate metabolizers. For these reasons, the effect of concomitant medications on phenoconversion rates was examined in patients with CYP2D6 extensive metabolizer and ultrarapid metabolizer genotypes.

Concomitant Medications

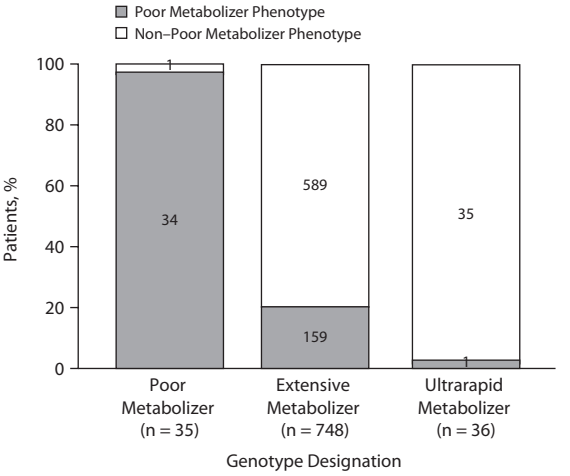
A total of 705/900 completers (78%) were taking medications other than venlafaxine ER (Table 3). In all, 247/900 patients (27%) reported using known concomitant CYP2D6 substrates or inhibitors. *O*-desmethylvenlafaxine to venlafaxine ratios for patients with non–poor metabolizer genotypes who used concomitant CYP2D6 substrates or inhibitors were, in general, smaller (ie, closer to 1 or less) compared with those who did not use CYP2D6 substrates or inhibitors (Table 2). The frequency distribution of *O*-desmethylvenlafaxine to venlafaxine ratio values was shifted toward lower values for patients taking concomitant CYP2D6 substrates or inhibitors compared with patients who were not (data not shown). There was no apparent additional effect of concomitant CYP3A4 substrates and inhibitors on the *O*-desmethylvenlafaxine to venlafaxine

Figure 1. Phenotypic Poor Metabolizer Rates by Genotype Designation in Study Completers[a,b]

A. Poor Metabolizer Phenotype Rates for Poor Metabolizer Versus Non–Poor Metabolizer Genotype Designations in Study Completers



B. Poor Metabolizer Phenotype Rates by Poor Metabolizer, Extensive Metabolizer, or Ultrarapid Metabolizer Genotype Designations in Study Completers[c]



[a]Numbers within bars indicate the number of patients of each phenotype.
[b]There was a statistically significant lack of agreement between the genotypic and phenotypic CYP2D6 poor metabolizer classification among genotypic poor metabolizers, extensive metabolizers, or ultrarapid metabolizers (McNemar test, $P < .0001$).
[c]Total N differs from panel A because intermediate metabolizers are not included in panel B.

Table 2. *O*-Desmethylvenlafaxine to Venlafaxine Ratios[a] According to *CYP2D6* Alleles[b] for Patients Taking Versus Not Taking CYP2D6 Substrates or Known Inhibitors

		No CYP2D6 Known Inhibitors		Taking CYP2D6 Known Inhibitors		No CYP2D6 Substrates and/or Known Inhibitors		Taking CYP2D6 Substrates and/or Known Inhibitors	
CYP2D6 Alleles	Genotype Designation	n	*O*-desmethylvenlafaxine to Venlafaxine (median)	n	*O*-desmethylvenlafaxine to Venlafaxine (median)	n	*O*-desmethylvenlafaxine to Venlafaxine (median)	n	*O*-desmethylvenlafaxine to Venlafaxine (median)
**1*1 (xN)*	UM	31	5.637	5	1.986	26	6.175	10	3.184
**1*1*	EM	276	3.767	80	1.249	258	3.810	98	1.577
**1*3*	EM	16	1.792	6	1.193	16	1.792	6	1.193
**1*4*	EM	131	1.542	32	0.889	118	1.534	45	0.985
**1*4 (xN)*	EM	7	1.518	…	…	7	1.518	…	…
**1*5*	EM	25	1.969	5	0.405	23	2.039	7	0.824
**1*6*	EM	7	1.638	…	…	7	1.638	…	…
**1*6 (xN)*	EM	3	1.002	…	…	3	1.002	…	…
**1*10*	EM	14	2.695	…	…	10	1.794	5	2.927
**1*17*	EM	18	1.939	4	1.521	18	1.939	4	1.521
**1*17 (xN)*	EM	3	2.007	…	…	3	2.007	…	…
**1*29*	EM	9	2.365	…	…	8	2.170	…	…
**1*29 (xN)*	EM	4	2.196	…	…	3	2.829	…	…
**1*41*	EM	81	2.135	20	0.589	73	2.153	28	1.034
**4*10*	IM	9	1.199	3	1.213	8	1.148	4	1.461
**4*41*	IM	21	0.424	8	0.236	20	0.452	9	0.259
**41*41*	IM	9	1.328	…	…	9	1.328	…	…
**5*10*	IM	3	0.887	…	…	3	0.887	…	…
**5*41*	IM	5	0.716	…	…	5	0.716	…	…
**3*4*	PM	3	0.648	…	…	3	0.648	…	…
**4*4*	PM	9	0.060	7	0.096	8	0.055	8	0.108
**4*4 (xN)*	PM	3	0.300	…	…	…	…	…	…
**4*5*	PM	4	0.022	…	…	4	0.022	…	…

[a]Poor metabolizer phenotype status was defined as *O*-desmethylvenlafaxine to venlafaxine ratio < 1, based on published results.[25]
[b]Only *CYP2D6* allele groupings with data for 3 or more participants are presented; total N is therefore less than 900.
Abbreviations: CYP2D6 = cytochrome P450 2D6, EM = extensive metabolizer, IM = intermediate metabolizer, PM = poor metabolizer, UM = ultrarapid metabolizer.
Symbol: … = Allele groups for which n < 3.

Table 3. Concomitant Medications (ATC classification or preferred term) Taken by at Least 5% of Study Completers (N = 900)

ATC Classification Preferred Term	n (%)
Any nonstudy medication	705 (78.3)
Antidepressants (other than venlafaxine extended release)	227 (25.2)
Desyrel/trazodone/trazodone hydrochloride	54 (6.0)
Cholesterol and triglyceride reducers	145 (16.1)
Analgesics and antipyretics	142 (15.8)
acetylsalicylic acid/ASA/aspirin, enteric coated/Ecotrin	61 (6.8)
Antiinflammatory/antirheumatic products, nonsteroids	130 (14.4)
Advil/ibuprofen/Motrin	75 (8.3)
Anxiolytics	113 (12.6)
alprazolam/Xanax	65 (7.2)
Drugs for treatment of peptic ulcer	113 (12.6)
Antiepileptics	101 (11.2)
β-blocking agents	88 (9.8)
Renin-angiotensin system, agents acting on	82 (9.1)
lisinopril/Prinivil/Zestril	56 (6.2)
Thyroid preparations	78 (8.7)
Levothyroid/levothyroxine/Synthroid/thyroxine	76 (8.4)
Antipsychotics	79 (8.8)
Multivitamins, combinations	74 (8.2)
Adrenergics, inhalants	71 (7.9)
Albuterol/Proventil/Salbuterol/Ventolin	52 (5.8)
Hypnotics and sedatives	68 (7.6)
zolpidem/zolpidem tartrate	60 (6.7)
Oral blood glucose lowering drugs	59 (6.6)
Hormonal contraceptives for systemic use	55 (6.1)
Antihistamines for systemic use	46 (5.1)
Opioids	46 (5.1)

Abbreviation: ATC = anatomic therapeutic chemical.

frequency distribution compared with the distribution for patients taking CYP2D6 substrates or inhibitors only.

Conversion to the poor metabolizer phenotype was significantly more common among genotypic extensive metabolizers and ultrarapid metabolizers who reported taking known concomitant CYP2D6 substrates or inhibitors (82/210 [40%]) versus those who did not (77/574 [13%]; $P < .0001$, Fisher exact test). In patients who did not use known concomitant CYP2D6 substrates or inhibitors, 77/548 genotypic extensive metabolizers (14%) and 0 genotypic ultrarapid metabolizers had a poor metabolizer phenotype (*O*-desmethylvenlafaxine to venlafaxine ratio < 1). In contrast, 82/200 genotypic extensive metabolizers (41%) and 1/10 genotypic ultrarapid metabolizers (10%) who took known concomitant CYP2D6 substrates or inhibitors had a poor metabolizer phenotype. Phenoconverters used a greater mean number of concomitant nonstudy medications compared with nonconverters (Table 4).

DISCUSSION

To our knowledge, this is the first large-scale assessment of CYP2D6 phenoconversion in patients treated for depression in real-world clinical practice. We demonstrate that CYP2D6 phenoconversion is common: 27% of patients had a poor metabolizer phenotype based on *O*-desmethylvenlafaxine to venlafaxine ratio, compared with 4% of individuals with the poor metabolizer genotype. These results indicate that genotyping in clinical practice would significantly underestimate

Table 4. Number of Concomitant Medications, in Addition to Venlafaxine, Taken by Patients by Genotype, Phenotype, and Phenoconversion

Comparison Group	n	Mean (SD)[a]	Median[a]	Minimum-Maximum[a]
Total	900	3.02 (3.35)	2	0–22
Genotype				
Poor metabolizer	35	4.17 (3.49)	3	0–13
Non–poor metabolizer	865	2.97 (3.34)	2	0–22
Phenotype				
Poor metabolizer	243	4.07 (3.72)	3	0–22
Non–poor metabolizer	657	2.63 (3.11)	2	0–22
Phenoconversion				
Converters[b]	210[c]	4.04 (3.75)	3	0–20
Nonconverters	690	2.71 (3.15)	2	0–22

[a]Values do not include venlafaxine extended release.
[b]Patients with discordance between genotype and phenotype.
[c]This value represents 209 individuals who had a non–poor metabolizer genotype and a poor metabolizer phenotype + 1 individual who had a poor metabolizer genotype and a non–poor metabolizer phenotype.

the incidence of CYP2D6 poor metabolic capacity in patients for whom multiple drugs are prescribed. They underscore an important limitation of genotyping: genotyping establishes the genetic potential of an individual but not necessarily his or her functional capacity at any given moment in time. In this study, the focus was CYP2D6 functional capacity, but this general principle may apply to other physiological parameters as well. Practicing clinicians should understand this limitation, given the otherwise deserved enthusiasm for the application of genotyping to the practice of medicine. When treating patients with medications, clinicians should place more importance on the patients' functional capacity to metabolize and clear the drug at any given moment than on their genetic potential capacity to do so.

In a prior cross-sectional study of psychiatric inpatients and outpatients receiving risperidone treatment (N = 325),[16] *CYP2D6* genotype was determined to assess the relationship between poor metabolizer status and poor tolerability. The plasma concentration ratio of risperidone and its metabolite 9-hydroxyrisperidone was calculated as an index of CYP2D6 activity (with a ratio of > 1 indicating functional poor metabolizer status). Consistent with the current study, the incidence of functional poor metabolizer phenotype was higher than the genotypic rate. Of those taking risperidone, 8% (27/325) were found to be genotypic poor metabolizers, whereas 19% (53/281) of those for whom risperidone/9-hydroxyrisperidone ratio was calculated were found to be functional poor metabolizers. Among those genotyped as non–poor metabolizers, 34 of 260 (13%) were found to have phenoconverted to poor metabolizer status based on risperidone to 9-hydroxyrisperidone ratio. Several smaller studies (N = 7–72)[20,27–30] have also evaluated the incidence of functional CYP2D6 poor metabolizer status in genotypic CYP2D6 extensive metabolizers in clinical populations. In patients treated with the known CYP2D6 inhibitor paroxetine (mean ± SD dose: 20.8 ± 5.6 mg/d),[31] 24 of 30 genotyped extensive metabolizers (80%) had converted to the poor metabolizer phenotype.[20] Phenoconversion was reported in 7 of 14 psychiatric patients[28] and 4 of 20 psychiatric patients[27] who were genotypic CYP2D6 extensive metabolizers chronically treated with psychotropic drugs (including paroxetine, moclobemide, chlorpromazine, levomepromazine) in 2 smaller studies. The incidence of the CYP2D6 poor metabolizer phenotype (36%) was also higher than expected compared with the incidence of the poor metabolizer genotype (6%) in 36 patients treated with neuroleptic agents (including thioridazine and haloperidol).[30] Concordance between *CYP2D6* genotype and phenotype was also assessed in 2 small studies[32,33] of human immunodeficiency virus–positive patients (N = 61 and N = 17, respectively), in which reduced CYP2D6 activity was observed in several patients. In those studies, however, the investigators assumed the cause was the disease itself.

CYP2D6 metabolizer status can contribute to clinically significant differences in drug efficacy in patients prescribed CYP2D6 substrate medications.[10,12,15,34] CYP2D6 poor metabolizers (genotypically or due to phenoconversion) may not respond to treatment with opioids such as codeine, oxycodone, and hydrocodone, which are prodrugs that require metabolism to active forms for efficacy. Likewise, tamoxifen, the standard treatment for estrogen receptor–positive breast cancer,[35] is metabolized by CYP2D6 to the active metabolite endoxifen.[36,37] Patients with the poor metabolizer genotype are exposed to lower levels of the active drug,[38] and the poor metabolizer genotype is associated with increased risk of mortality in breast cancer patients taking tamoxifen, as is the concomitant use of the substantial CYP2D6 inhibitor paroxetine.[15,34] Genotypic extensive metabolizers taking paroxetine have plasma endoxifen levels similar to those of genotypic poor metabolizers.[38] A clinician who assesses only genotype when considering tamoxifen therapy for breast cancer risks undertreating patients with a drug-induced poor metabolizer phenotype, with potentially life-threatening consequences.

Previous work by our group and others[12,13] has shown that the antidepressant efficacy of venlafaxine (ER or immediate release) is greater in phenotypic extensive metabolizers compared with poor metabolizers. In an analysis of 830 patients enrolled in 4 clinical trials for major depressive disorder, statistically significant differences in venlafaxine efficacy were observed for phenotypic extensive metabolizers versus poor metabolizers after 6 to 12 weeks of treatment (75–375 mg/d).[12] Both venlafaxine-treated extensive metabolizer and poor metabolizer groups improved compared with placebo, but extensive metabolizers scored significantly better than poor metabolizers on 4 of 5 depression scales. Response and remission rates were significantly higher for venlafaxine-treated extensive metabolizers compared with venlafaxine-treated poor metabolizers.[12] The reasons for the lower antidepressant efficacy in CYP2D6 poor metabolizers versus extensive metabolizers has not been established, but venlafaxine, like tamoxifen, is metabolized via CYP2D6 to an active metabolite, *O*-desmethylvenlafaxine. Phenotypic poor metabolizers have lower exposure to this active metabolite after venlafaxine ER administration.[25] In the current study, we limited inclusion to those taking venlafaxine for

less than 8 weeks to limit potential bias based on differential response in poor metabolizers versus extensive metabolizers. Assuming patients taking the drug for longer durations would represent those who have achieved a response to the drug, we believed that including patients treated for longer than 8 weeks might have biased against enrolling a sufficient number of poor metabolizers in the study.

The use of CYP2D6 (but not CYP3A4) inhibitors and substrates in this study was associated with an approximate 50% reduction in median *O*-desmethylvenlafaxine to venlafaxine ratio, and the distribution of *O*-desmethylvenlafaxine to venlafaxine ratio values was shifted toward lower values in patients with non–poor metabolizer genotypes. Conversion to the poor metabolizer phenotype was observed in 40% of genotypic extensive metabolizers and ultrarapid metabolizers who were taking concomitant medications known to be CYP2D6 inhibitors, substrates, or both compared with 13% of genotypic extensive metabolizers and ultrarapid metabolizers who were not. Further, phenoconverters were taking, on average, 4.0 drugs in addition to venlafaxine ER compared with 2.7 concomitant drugs used by nonconverters. These results indicate that, in patients using multiple medications, genotyping would yield an inaccurate representation of CYP2D6 metabolic capacity in a sizable percentage of individuals. Patients were not excluded from this study for multiple medication use; therefore, the study population reflects a real-world clinical population.

In this analysis, genotypic intermediate metabolizers were likely to be classified as the poor metabolizer phenotype based on the ratio of *O*-desmethylvenlafaxine to venlafaxine, whether or not they had taken concomitant CYP2D6 substrates or inhibitors. Overall, 60% of intermediate metabolizers had an *O*-desmethylvenlafaxine to venlafaxine ratio < 1. This finding suggests that the *O*-desmethylvenlafaxine to venlafaxine rule may identify a subset of genotypic intermediate metabolizers as phenotypic poor metabolizers. In the original study examining the use of *O*-desmethylvenlafaxine to venlafaxine ratio for phenotyping, Nichols and colleagues[26] enrolled only extensive metabolizers and poor metabolizers based on either genotype or dextromethorphan metabolism. This is a limitation for the interpretation of intermediate metabolizer results in the current study. The observation that most intermediate metabolizer patients in this population had *O*-desmethylvenlafaxine to venlafaxine ratios similar to that of poor metabolizers, however, underscores the importance of considering phenotype, rather than genotype, in clinical practice.

CONCLUSIONS

In this study, almost 1 of 4 patients (24%) with a CYP2D6 non–poor metabolizer genotype was converted to phenotypic poor metabolism status as a result of the other medications they were taking. Phenotype conversion was associated with use of concomitant medications and, specifically, the use of CYP2D6 substrates or inhibitors. The results of this study have clinical implications for patient populations with chronic medical disorders, particularly those who are likely to use concomitant medications that are CYP2D6 substrates or inhibitors.

Drug names: albuterol (Proventil, Ventolin, and others), alprazolam (Xanax, Niravam, and others), bupropion (Wellbutrin, Aplenzin, and others), desvenlafaxine (Pristiq), fluoxetine (Prozac and others), oxycodone (OxyContin, Oxecta, and others), paroxetine (Paxil, Pexeva, and others), risperidone (Risperdal and others), tamoxifen (Soltamox and others), terbinafine (Lamisil and others), trazodone (Oleptro and others), venlafaxine (Effexor and others), zolpidem (Ambien, Edluar, and others).

Author affiliations: Department of Psychiatry, Kansas University School of Medicine, Wichita (Dr Preskorn), and Pfizer Inc, formerly Wyeth Research, Collegeville, Pennsylvania (Drs Kane, Lobello, Nichols, Fayyad, and Guico-Pabia and Mss Buckley and Focht). Dr Kane is now with the Reading Hospital and Medical Center, Reading, Pennsylvania. Dr Guico-Pabia is now retired.

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Additional information: The FDA notified pharmaceutical companies of objectionable conditions at Cetero's Houston, Texas bioanalytical facility following several inspections. Plasma concentration samples for determination of venlafaxine and desvenlafaxine were assayed at Cetero during a time period when the FDA is requesting an independent third party audit to confirm the validity of the data. The independent third party audit will be conducted as requested by the FDA. Pfizer has conducted a thorough examination and verification of the bioanalytical data generated by Cetero's Houston, Texas bioanalytical facility for this study and has determined that concentration data obtained for this study are accurate.

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