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## Copy Number Variants in pharmacogenetic genes

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### Abstract

Variation in drug efficacy and toxicity remains an important clinical concern. Presently, single nucleotide polymorphisms (SNP) only explain a portion of this problem, even in situations where the pharmacological trait is clearly heritable. The Human CNV Project identified copy number variations (CNVs) across approximately 12% of the human genome, and these CNVs were considered causes of diseases. Although the contribution of CNVs to the pathogenesis of many common diseases is questionable, CNVs play a clear role in drug related genes by altering drug metabolizing and drug response. Here we provide a comprehensive review of the clinical relevance of CNVs to drug efficacy, toxicity, disease prevalence in world populations and discuss the implication of using CNVs as diagnosis in clinical intervention.

### Keywords

pharmacogenetics; copy number variants; drug efficacy; drug toxicity

### Pharmacogenetics

Pharmacogenetics aims to study the gene variants associated with drug metabolism enzymes, transporters, drug receptors and relieve the burden of sickness caused by interindividual differences in drug response or vulnerabilities to drug toxicity [1, 2]. Since the completion of the Human Genome Project in 2000, there has been a burst in the number of pharmacogenetic research studies published, with some of the findings now being introduced into clinical practice. To practice pharmacogenetics testing in the clinic, the United States Food and Drug Administration (FDA) approved the first DNA microarray molecular diagnostic test in 2004, the AmpliChip CYP450 (Roche Molecular Diagnostics, Alameda, CA, USA), for the analysis of single nucleotide polymorphisms (SNPs) and copy number variants (CNVs) of *CYP2D6* and *CYP2C19* genes which are closely related with the drug therapy of depression, gastroesophageal reflux and cardiovascular diseases. [3]. The FDA also issued “Draft Guidelines for Industry: Pharmacogenomic Data Submission”, making the submission of pharmacogenomic data mandatory for new drug applications from drug companies [4]. Since 2008, the FDA has issued a list of valid pharmacogenetic

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#### Conflicts of interest

Dr McLeod is a consultant to Medco Health Solutions, Gentris corporation, and Myriad Genetics.

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biomarkers in the context of approved drug labels that are updated every year [5], which accelerates the translation of pharmacogenetics from bench to bedside.

At present, pharmacogenetics can be applied to the treatment of patients with psychiatric illnesses, cancer, cardiovascular disease, pain, HIV infection, and microorganism infection, to enhance drug efficacy and decrease drug toxicities [6–9]. According to a survey of 1200 drug labels for the years 1945–2005, 121 drugs labels contained pharmacogenetic information [10]. Of those, 69 labels referred to human genomic biomarkers (e.g. atomoxetine, fluoxetine and codeine are labeled with *CYP2D6* variants associated drug efficacy and toxicity change) [10]. Over 20 of these markers were either required or recommended by the FDA to be tested in patients before starting drug therapy [11]. For example, tramadol is an orally administered synthetic analogue of codeine and its product label warns that patients with cytochrome P450 2D6 (*CYP2D6*) gene (*CYP2D6*) duplications are at increased risk of respiratory depression [12].

## What is a copy number variant?

Human DNA has one copy of autosomal regions on each chromosome. However, as discovered by the Human Genome Project, many genetic regions display a variation in the number of copies (more or less than two copies totally). Alleles containing 0–13 gene copies have been reported across the human population [13]. These genetic variants are termed copy number variants (CNVs) and are defined as DNA segments ranging in size from one kilobase to several megabases among individuals due to deletion, insertion, inversion, duplication or complex recombination [14] (Figure 1). Many groups have addressed the presence of CNV in the human genome and their associations with common human diseases including neuropsychiatric, autoimmune, infectious and cardiovascular diseases [15, 16], while others found evidence that CNVs are not associated with common diseases [17]. While the contribution of CNVs to the pathogenesis of common diseases is questionable, CNVs in some pharmacogenetic genes play a clear role in drug efficacy and toxicity [18]. Inspired by the increasing interest in CNVs, here we review our current knowledge of CNVs in relation to drug efficacy and toxicity, their prevalence in ethnically diverse populations, and the potential utilization of CNV knowledge in the clinical setting.

## CNVs associated with drug efficacy in the clinic

### CYP2D6

*CYP2D6* is predominantly expressed in human liver and it metabolizes over 25% of drugs currently used in the clinic [19]. *CYP2D6* is highly polymorphic and its variants account for the majority of the variation in enzyme activity that is observed within and between populations. To date, more than 75 *CYP2D6* alleles have been documented in the Human Cytochrome P450 (CYP) Allele Nomenclature database (<http://www.cypalleles.ki.se/cyp2d6.htm>). Alleles are comprised of a combination of polymorphisms, including SNPs, insertion/deletions and/or gene conversions, which influence *CYP2D6* enzyme activity. These alleles include normal activity alleles (\*1, \*2, \*33, \*35), reduced activity alleles (\*9, \*10, \*17, \*29, \*41, \*59), nonfunctional alleles, which encode enzymes with no activity or produce no enzyme at all (\*3, \*4, \*5–\*8, \*11–\*16, \*18–\*21, \*36, \*38, \*40, \*42, \*44, \*56, \*62), and as yet functionally undetermined alleles (\*22–\*28, \*30–\*32, \*34, \*37, \*39, \*43, \*45–\*55, \*70, \*71, \*73, \*74) [20]. The functional polymorphisms of the nonfunctional and reduced activity alleles introduce splice defects, non-synonymous amino acid changes or frameshifts, which alter enzyme activity. In addition, CNVs of the normal, nonfunctional and reduced activity *CYP2D6* alleles are observed and these can also alter *in vivo* *CYP2D6* enzyme activity. For example, more than two copies of *CYP2D6* normal alleles (e.g. \*1×N, \*2×N, \*33×N, \*35×N, 13>N>2, Figure 2)

have been shown to elevate CYP2D6 enzyme activity [21]. Individuals who inherit these alleles have more rapid metabolism of CYP2D6 substrates and are classified as ultrarapid-metabolizers (UMs) [21]. CNVs are also found for nonfunctional ( $*4 \times N$ ,  $*36 \times N$ ) and reduced activity alleles ( $*10 \times N$ ,  $*17 \times N$ ), but individuals who inherit these alleles do not have increased metabolism of CYP2D6 substrates [22]. It is noteworthy that *CYP2D6\*36* is absent in white populations but in Asian and African American populations it exists on each chromosome in three forms: 1. *CYP2D6\*36* $\times 2$  (2 copies of nonfunctional allele); 2. *CYP2D6\*36*+*\*10* (1 copy of nonfunctional allele in a tandem arrangement with *\*10*); and 3. *CYP2D6\*36* alone (Figure 2) [23]. Only the first situation is considered a CNV. In addition to gaining extra copies of the *CYP2D6* gene, *CYP2D6\*5* is a loss of gene copy that produces no CYP2D6 protein [24]. Individuals with two nonfunctional *CYP2D6* alleles, including *CYP2D6\*5*, have no CYP2D6 activity, in turn have impaired metabolism of CYP2D6 substrates and are classified as poor-metabolizers (PMs). If two normal activity alleles are inherited, subjects have normal metabolism of CYP2D6 substrates and are classified as extensive metabolisers (EMs) [21]. *CYP2D6* CNV is a significant cause of CYP2D6 phenotype diversity in the clinic.

In Caucasian and Asian populations, the frequency of duplicated normal activity alleles, *CYP2D6\*1* $\times 2$  and  $*2 \times 2$ , is approximately 0.5–1.5% [25], whereas in African populations, the frequency is approximately 1.6–3.3% [26, 27]. The frequency of *CYP2D6\*5* is similar in Caucasian, Asian and African populations (2–7%) [28]. Although the frequency of duplication of *CYP2D6* normal alleles is quite low, the clinical impact of the corresponding phenotype cannot be ignored.

Nortriptyline, a tricyclic antidepressant, is prescribed for the treatment of depression and acts by inhibiting the reuptake of norepinephrine [29]. Taken orally, nortriptyline is well absorbed from the gastrointestinal tract and metabolized in the liver by CYP2D6 [30]. Dalen and colleagues found that the plasma exposure and clearance of nortriptyline was clearly related to the number of copies of the *CYP2D6* gene of subjects (ranging from 0 to 13) [31]. Subjects with 13 copies of functional *CYP2D6* gene displayed a 17-fold higher oral clearance and 36-fold lower plasma concentrations of nortriptyline than those carrying no copies of the functional gene [31]. Interestingly, a recent genome-wide study showed that CNVs in two intergenic regions (chromosome 1 and 10) were associated with nortriptyline response in unipolar depression patients of European ancestry [32]. However, *CYP2D6* CNVs were not shown to be associated with the nortriptyline response. Further studies are required to validate the associations between CNVs and nortriptyline efficacy for *CYP2D6* and new CNV candidates in the clinic.

Tamoxifen is a tissue selective estrogen receptor (ER) modulator, which serves as an antagonist of ERs in breast tissue; however, it is an agonist in endometrial tissue [33]. It is prescribed to treat and prevent hormone-dependent breast cancer in premenopausal and postmenopausal women [34]. Since the 1990s, tamoxifen has been approved by the FDA as the standard anti-estrogen treatment for metastatic breast cancer and adjuvant treatment of primary breast cancer for both female and male patients [35]. Tamoxifen is a prodrug and undergoes complex metabolism *in vivo* and *in vitro*, primarily catalysed by CYPs [36]. CYP2D6 is considered the key enzyme that converts N-desmethyltamoxifen (NDM), a primary tamoxifen metabolite, to endoxifen. Endoxifen has 10-fold higher affinity for ERs than tamoxifen, and is therefore considered the active form of tamoxifen *in vivo* [37]. Indeed, endoxifen/NDM plasma concentration ratios are significantly higher in UMs who carried multiple copies of *CYP2D6* functional alleles than in EMs and PMs who carried two or less copies of *CYP2D6* alleles [38]. It is possible that UM patients may derive more therapeutic benefit from tamoxifen than other patients; however, this has not yet been tested. The recent Italian Tamoxifen Prevention Trial genotyped 47 breast cancer patients using the

AmpliChip CYP450 chip [39]. Subjects who carried more than two functional *CYP2D6* alleles ( $*1/*1 \times N$ ,  $*1/*2A \times N$  and  $*1 \times N/*2A$ ) were classified as UMs and those with one or two nonfunctional alleles ( $*3/*5$ ,  $*41/*5$  and  $*4A/*5$ ) were classified as PMs. Although the small sample size limited the statistical power of the study, PMs had a higher risk of developing breast cancer than other subjects [39]. In addition, a larger German study genotyped 492 ER<sup>+</sup> breast cancer patients who had received tamoxifen treatment. In this study patients with more than two *CYP2D6* functional genes ( $*1/*1 \times N$ ,  $*2/*1 \times N$  and  $*1/*2 \times N$ ) were also classified as UMs. A Kaplan-Meier survival analyses displayed highest benefit from tamoxifen treatment in UM patients. Meanwhile, PM patients who carried no copies of *CYP2D6* functional alleles had poorer outcomes than EMs (hazard ratio, 2.77; 95% confidence interval 1.31–5.89) [40]. Due to the low frequency of *CYP2D6* CNV in Caucasians, small sample size is a common limitation in these studies, which contributes to low statistical power in data analysis. Clinical trials with larger sample sizes are required to further validate these findings.

Metoprolol is a selective beta1-adrenergic antagonist, which is prescribed to decrease heart rate in patients with acute myocardial infarction (AMI), hypertension or dilated cardiomyopathy [41]. It is extensively metabolized in human liver and predominantly by *CYP2D6* [42]. In a pharmacokinetics study, subjects with three copies of *CYP2D6* functional alleles, classified as UMs, showed significantly lower metoprolol plasma concentrations and higher drug clearances after a single dose of 100 mg metoprolol compared with EMs and PMs ( $P < 0.0001$ ) [43]. For multi-doses and long-term (42 days) drug administration, metoprolol plasma concentrations (normalized for daily dose) and metoprolol/a-hydroxymetoprolol ratios at steady state were markedly lower in UMs than EMs and PMs ( $P < 0.0001$ ), which suggested that UMs are at potential risk of treatment failure [42]. In another study, 187 AMI patients received metoprolol and heart rate (HR) was recorded at administration and discharge. PM patients achieved pronounced HR reduction, while UM patients ( $*1/*1 \times N$ ) failed the treatment [44]. Pre-screening for *CYP2D6* CNVs may help physicians evaluate the potential benefit that patients can receive from metoprolol treatment.

In summary, associations between *CYP2D6* CNVs and therapeutic response to drugs were found in wide range of drug classes including antidepressants, endocrine therapy agents, and beta-blocking agents. Pro-drugs that depend on an activation process via *CYP2D6* may demonstrate a better response in individuals with multiple copies of functional *CYP2D6* genes (UMs). Individuals with two copies of nonfunctional *CYP2D6* alleles (PMs) may benefit from low metabolizing efficiency of drugs exempt from activation process; however, they could be at increased risk of toxicities.

## CNV associations with drug toxicity in the clinic

### CYP2D6

Codeine is a widely used analgesic and antitussive agent in the clinic. It is activated by *CYP2D6* to a strong central nervous system (CNS) sedative, morphine. Kirchheiner and colleagues found that after a single 30 mg dose of codeine, the plasma concentrations of morphine are significantly higher in *CYP2D6* UMs than EMs [45]. High plasma concentrations of morphine can lead to several fatal effects including hypotension and respiratory depression, especially in pediatric patients. A healthy 2-year-old boy recently died after elective adenotonsillectomy because of the commonly prescribed pain-killer, codeine [46]. Postmortem genotyping revealed that the boy had a duplication of a functional *CYP2D6* gene, which most likely made him a UM of codeine and the subsequent elevated morphine exposure was considered the likely cause of respiratory depression and death in this tragic case [46]. There was a similar case where an infant died with high morphine

concentrations in his blood. The morphine was transferred via breastmilk from a mother with a UM genotype who was taking codeine for postpartum pain [47]. Breastfed infants are at increased risk of potentially life-threatening CNS suppression in such cases [48]. Codeine and its analogues have been labeled with *CYP2D6* CNVs markers by FDA.

### Glutathione S-transferases (GSTs)

Glutathione S-transferases (GSTs) are a superfamily of enzymes that catalyze the detoxification of carcinogens, therapeutic chemicals and environmental toxins. GSTs catalyze the reaction of glutathione (GSH) with an acceptor molecule to form a sulfur-substituted glutathione. GST enzymes are encoded by eight distinct loci including alpha, kappa, mu, omega, pi, sigma, theta, and zeta [49]. The GST Mu-1 and Theta-1 enzymes are encoded by the *GSTM1* and *GSTT1* genes, respectively. Complete gene deletions (loss of functional gene copy) of *GSTM1* and *GSTT1* are relatively common. About 50% and 30% of Caucasians are homozygous for *GSTM1* and *GSTT1* gene deletions respectively, compared with >22% and >14% in Asians, and >27% and >37% in Africans [50, 51]. In addition to gene deletions, a multiplex real-time PCR method was developed to detect *GSTM1* and *GSTT1* gene duplications [52]. However, *GSTT1* duplications have not been observed, and a *GSTM1* duplication has only been detected in one of 1320 Caucasian samples. No further functional studies on *GSTM1/GSTT1* gene duplications have been reported.

Consistent with the important roles that *GSTM1* and *GSTT1* play in the detoxification of exogenous compounds, carriers of gene deletions have elevated risks of various cancers (e.g. colorectal cancer, acute myeloid leukemia), especially in Caucasians [53, 54], and drug-related toxicities. Cho and colleagues investigated the associations between *GSTM1* and *GSTT1* gene deletions and toxicity from the standard chemotherapy regimen for lymphoma, R-CHOP (rituximab with cyclophosphamide, doxorubicin, vincristine and prednisone), in 94 diffuse large B-cell lymphoma (DLBCL) patients [55]. R-CHOP caused more grade III-IV toxicities, including leukocytopenia (odds ratio (OR)=3.12), fever (OR=5.27), and mucositis (OR=4.61), in patients homozygous for *GSTT1* gene deletion, than other patients. Patients with a *GSTM1/GSTT1* double-deletion genotype had an even higher risk of grade III-IV thrombocytopenia than other patients (OR=7.75). Loss of either or both *GSTM1* and *GSTT1* genes might be expected to increase toxicity along with improved efficacy of chemotherapy, however no difference was observed in this study. *GSTM1* and *GSTT1* gene deletions are likely to predict R-CHOP induced toxicity in DLBCL patients.

Tacrine was the first drug for the treatment of mild to moderate dementia of Alzheimer's disease [56], which is the most common form of dementia characterized for a progressive and fatal neurodegenerative disorder [57]. Tacrine-induced hepatotoxicity is primarily caused by one of its metabolites, which is de-activated by *GSTM1* and *GSTT1* [58]. A study in 141 Caucasian Alzheimer's disease patients found that patients with the *GSTM1* and *GSTT1* double-deletion had alanine aminotransferase (ALT) levels that were three times the upper limit of normal (relative hazard: 2.84, 95% confidence interval: 1.51–5.35, P=0.001), which is a marker of tacrine-induced hepatotoxicity [59]. In addition, a patient's susceptibility to tacrine-induced hepatotoxicity could not be predicted by either *GSTM1* or *GSTT1* deletion alone suggesting redundancy in the enzymes [59]. Due to the high prevalence of *GSTM1* and *GSTT1* gene deletions in Caucasians, over 10% of patients have a *GSTM1* and *GSTT1* double deletion and are therefore at increased risk of tacrine-induced hepatotoxicity. A nitric oxide donor-tacrine hybrid compound may be an alternative therapy for these patients to achieve both drug efficacy and low risk of hepatotoxicity [60].

In summary, many phase I and II drug metabolism enzymes are involved in the detoxification of drugs used in clinic. CNVs can lead to the over-activation or loss of



detoxification of a drug which may cause increased drug toxicities in patients. Precautions should be adopted based on individual genotyping information.

## Prevalence of CNVs in drug related gene in human populations

Understanding the prevalence of major genetic variations related to drug efficacy and toxicity is critical for both health providers and patients. Both the Ministry of Health in each nation and physicians in clinics can use this knowledge to maximize benefit and minimize harm for patients prior to and during drug therapy [61], which is widely accepted as the base for personalized genomic medicine [62].

The frequency of CNVs is shown in Table 1. These data can help each population to select the most appropriate drugs and proper doses for specific treatments. For example, in the United States, over 50% and 30% of Caucasian populations carry either *GSTM1* or *GSTT1* gene deletions [50], which leads to increased susceptibility to colorectal cancer, acute myeloid leukemia and chemotherapy-induced toxicities [53]. Using the CNVs frequency data, physicians will be aware of the probability of achieving benefit or developing a drug-induced toxicity in his/her patients and thus precautions can be taken. Meanwhile, multi-copies of *CYP2D6* functional genes are more prevalent in Europe, Saudi Arab, and Ethiopia than in Asia and the U.S. (Caucasian). In former regions, more people may respond to tamoxifen treatment, and less codeine induced CNS suppression will be observed in the latter. The data of CNVs global prevalence can help to avoid translating a regional drug problem for a specific population into a common problem for all other populations [63].

## Which drugs should be labeled with CNV markers?

Since 2008, the FDA has released a list of valid genomic biomarkers in the context of approved drug labels [5]. The term “valid biomarkers” has been described in the previously released “Guidance for Industry: Pharmacogenomic Data Submissions” [4] as a “biomarker that is measured in an analytical test system with well established performance characteristics and for which there is an established scientific framework or body of evidence that elucidates the physiologic, toxicological, pharmacologic, or clinical significance of the test results.” Drugs whose therapeutic response or toxicity is affected by these markers were recorded in this list including one CNV marker: *CYP2D6*\*2×2. The package insert for codeine sulfate and other drugs containing codeine as an ingredient, such as Fiorinal® with codeine (butalbital, aspirin, caffeine, and codeine phosphate) and Fioricet® with codeine (butalbital, acetaminophen, caffeine, and codeine phosphate) warns of elevated risk of codeine-induced toxicity in individuals with more copies of *CYP2D6* functional genes. However, none of the other drugs (tamoxifen, tacrine, etc) or other CNVs (*CYP2D6*\*5, *GSTM1*, *GSTT1* deletions) discussed in this review are included in this list yet. Meanwhile, an FDA-approved analytical test system for *GSTM1* and *GSTT1* deletions is not available at this time. The only approved genotyping platform for CNV markers diagnosis in clinic is the AmpliChip CYP450 chip [3], which detects *CYP2D6* CNVs. There is still a gap between the knowledge of drug-related CNVs and implementation of drug label changes. The association of pharmacogenetic CNVs with drugs and clinical indications are summarized in Table 2.

## Concluding remarks

The translation of CNV knowledge into clinic intervention remains a big challenge, with a growing number of pharmacogenetics studies providing robust evidence for the impact of CNV on the efficacy and toxicity of drug therapy [18]. However, few comparative effectiveness studies have been conducted in this field, which has limited the expansion of genetic testing of CNVs into primary health care system. In addition, investigations on the

cost versus benefits of genetic testing have been questioned. Additionally, health providers and insurance companies quite understandably are reluctant to reimburse genetic tests for low frequency variants to identify only a few patients who may benefit. That includes *CYP2D6* CNVs and over 10 other clinically important pharmacogenetic variants (*CYP2C19*\*3, *CYP2C9*\*2,\*3, *CYP2D6*\*3,\*6, *DPYD*\*2A, *TPMT*\*2,\*3,\*4, etc) that are considered rare. These variants exist in every population and may cause severe drug-induced toxicities or reduce the therapeutic benefit that a patient derives from a drug. As a result of this economic reality, diffusion of genetic tests for drug treatment has been slow. Unless we take a coordinated approach to CNV translational research, by adding policy and economics members to the investigation team, we will never see the promise of genome-guided therapy.

### Box 1

Currently available methodologies for CNV detection  
Many PCR based methods have been developed for the detection of CNVs. However, the low-throughput and low reproductively rate limit the use of these methods in the clinic. Recently, new methods have been developed based on microarray hybridization technology.

#### Conventional methods

##### Long distance and multiplex PCR

This is the first method developed for the detection of *CYP2D6* CNV. Long PCR is used to amplify a > 4000 bp fragment of the *CYP2D6* gene. Specific primers for *CYP2D6*\*5 have been designed. Two separate PCR reactions for either wild-type or mutated allele primers are run for each sample using the long PCR product of *CYP2D6* gene. Following up with gel electrophoresis, the *CYP2D6*\*5 can be identified [64].

##### TaqMan Real-Time PCR

This is a PCR quantification method. TaqMan probes emit a specific report fluorophore (usually a short-wavelength colored dye, such as green) during the elongation process of PCR reactions. The fluorophore can be detected by a corresponding detection system and compared with the signal of reference genes, such as albumin, to calculate the amount of specific genes in each sample. This is a low-through put detection method for CNVs [65].

#### Microarrays hybridization-based methods

##### Array comparative genomic hybridization (aCGH)

This is a genome-wide screening technique for CNVs [66] with higher resolution than chromosome-based comparative genomic hybridization [67], which allows detection of copy number changes of 5–10 kb of DNA sequence.

##### Roche AmpliChip CYP450 system

This is an oligonucleotide microarray hybridization method for genotyping 27 *CYP2D6* variants (including CNVs) and two *CYP2C19* variants. It has been developed by Roche based on Affymetrix microarray technology [68]. A logarithmic scale is used to predict *CYP2D6* and *CYP2C19* phenotypes [69]. This is the first FDA approved pharmacogenetics test for clinic use.

##### Genome-wide association SNP microarrays

These arrays cover 300K to more than 1 million genetic markers including thousands of probes for the detection of CNVs [70].

### Affymetrix DMET plus microarray

This is a novel genotyping tool customized for pharmacogenetics research. It covers 225 essential pharmacogenetics genes and 1936 common or rare variants including five CNVs belongs to *CYP2D6*, *CYP2A6*, *UGT2B17*, *GSTM1* and *GSTT1* [71].

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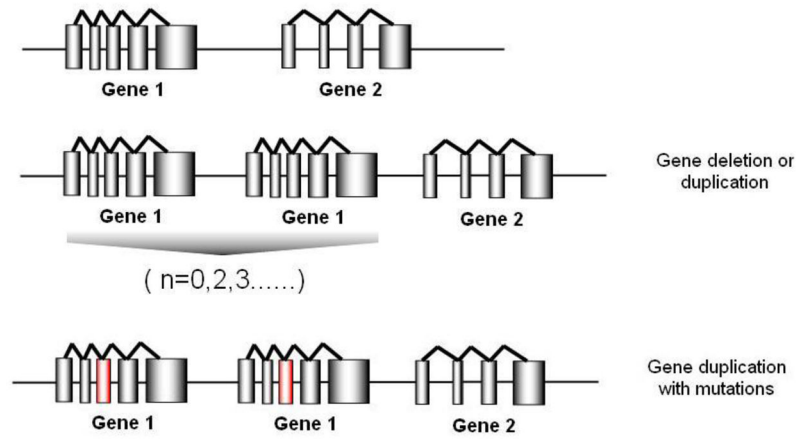


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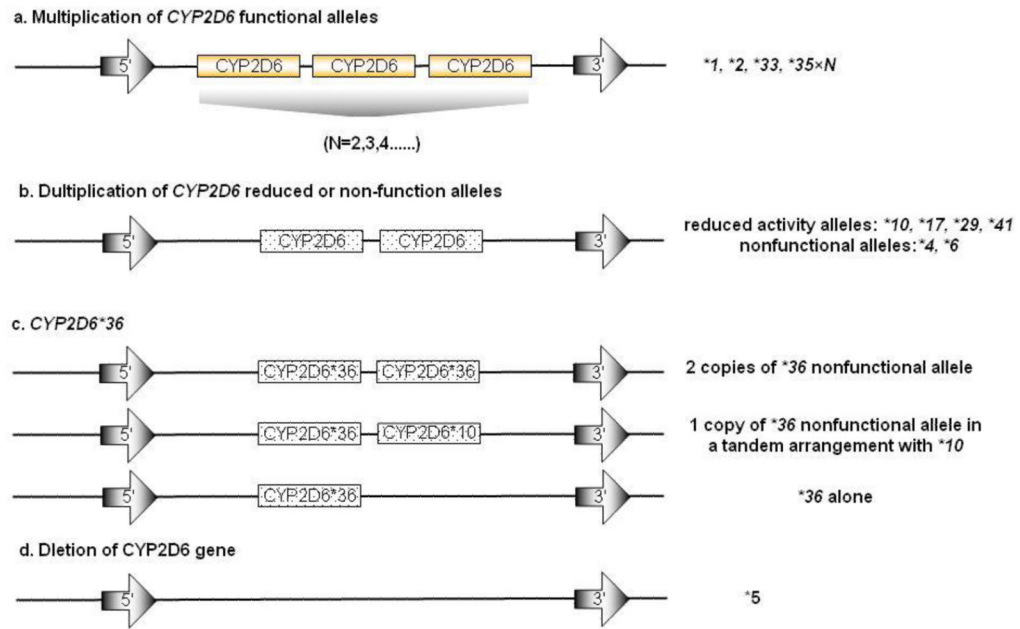
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**Figure 1.**

A diagram for copy number variants in human genome. If the gene recombination event occurs between two genes, a gene duplication or multiplication ( $n=2,3..$ ) or a gene deletion ( $n=0$ ) could happen. A duplication of gene could also carry mutations from the original copy (show in red column).





**Figure 2.**

A summary of *CYP2D6* CNV structure[18]. (a) Functional *CYP2D6* CNVs are shown in gold. (b) Duplication of *CYP2D6* reduced activity or nonfunctional alleles are shown in dotted background. (c) Three forms of *CYP2D6*\*36 allele. The first arrangement is considered as a CNV for this allele. (d) Deletion of *CYP2D6* gene.

Table 1

A summary of CNV frequencies in pharmacogenetic genes in different ethnic populations.

Population	CNV frequencies (%)				
	<i>CYP2D6</i> *1×N	<i>CYP2D6</i> *2×N	<i>CYP2D6</i> *5	<i>GSTMI</i> deletion	<i>GSTT1</i> deletion
<b>North America</b>					
U.S.A. (Caucasian)	0.2 [72]	0.7 [72]	6.2 [72]	54.3 [50]	27.6 [50]
U.S.A. (African-American)	1.2 [72]	1.6 [72]	4.0 [72]	23.7 [73]	17.5 [73]
Mexico (Mestizo)			1.3–2.7 [74, 75]	33.5 [76]	12.1 [76]
<b>Africa</b>					
Cameroon	3.3 [26]	3.3 [26]		27.8 [51]	46.8 [51]
Ethiopia		10–16 [21, 77]	3.3 [77]	43.8 [51]	37.3 [51]
Ghana		1.6 [27]	6 [27]	19.3 [78]	73.7 [78]
<b>East Asia</b>					
China ( Han)	2.2 [25]	0–2 [21]	7.2 [79]	52 [80]	38.7 [80]
Japan	0.5 [81]	0.5 [81]	6.2 [82]	50.8 [83]	45.8 [83]
Korea	0.13 [84]	0.5 [84]	6.1 [84]	51.4 [85]	51.6 [85]
<b>Europe</b>					
France	2.0 [25]		4.0 [25]	49 [86]	26 [86]
Germany	0.51 [87]	1.34 [87]	1.95 [87]	51.6 [50]	19.5 [50]
<b>Middle East</b>					
Saudi Arabia		10.4 [88]	1.0 [88]	8.3 [89]	4.2 [89]

Table 2

Major clinical information for CNVs in pharmacogenetic genes.

	CNV type	Drug affected	Phenotype	Clinical indication	PharmGKB gene category	Reference
<i>CYP2A6</i> *4	Deletion	Nicotine Coumarin Tegafur	No enzyme activity	Unknown	VIP	[90–92]
	Duplication		Increased enzyme activity	Unknown	VIP	
<i>CYP2A6</i> *1x2	Multiplication	Anticholinesterases Antidepressants Antipsychotics Beta Blocking Agents Opioids	Increased enzyme activity	Alzheimer Disease Depression Hypertension Breast cancer Codeine dependence	VIP	[19]
			Increased enzyme activity		VIP	
<i>CYP2D6</i> *5	Deletion		No enzyme activity		VIP	
<i>GSTM1</i> *0	Deletion	Antineoplastic agent Xenobiotics	No enzyme activity	Breast cancer Non-small cell lung cancer Leukemia Drug toxicity		[93, 94]
	Deletion		No enzyme activity			
<i>SULT1A1</i>	Multiplication	Unclear clinic impact on drugs Unclear clinic impact on drugs	Increased enzyme activity	Unknown	VIP	
	Duplication		Unknown	Unknown		
<i>UGT2B17</i>	Deletion	Estradiol Testosterone	Decreased enzyme activity	Androgen Excretion		[95]

VIP = very important pharmacogene.