Farmacia
HOSPITALARIA
Órgano oficial de expresión científica de la Sociedad Española de Farmacia Hospitalaria

ORIGINALS
Bilingual edition English/Spanish

Results of the implementation of a pharmacogenomics platform based on NGS technologies. Combining clinical and research approaches

Resultados de implementación de una plataforma farmacogenómica basada en tecnologías NGS. Combinación de abordajes asistencial y de investigación

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Received 14 June 2021;
Accepted 30 July 2021.
DOI: 10.7399/fh.11762

How to cite this paper
Ramudo-Cela L, Busto-Fernández F, Outeda-Macías M, Antolín S, Calvo-Martínez L, Martín-Herranz I. Results of the implementation of a pharmacogenomics platform based on NGS technologies. Combining clinical and research approaches. Farm Hosp. 2021;45(Suppl 1):S11-37

Abstract

Objective: As more genes are incorporated into pharmacogenomic care processes and more importance is given to rare variants, the use of targeted capture sequencing panels has been proposed as a very efficient alternative due to their affordability, high throughput, and deep coverage, all of them characteristics of high-quality next-generation sequencing data. The purpose of this study is to describe the prevalence of clinically actionable pharmacogenetic variants previously described in the scientific literature, as well as that of new variants identified by next-generation sequencing technologies, and to evaluate the drugs potentially affected by such variants.

Method: A panel of 18 clinically actionable pharmacogenomics-related genes was evaluated in 41 subjects diagnosed with breast cancer undergoing neoadjuvant treatment. The prevalence of previously descri-

KEYWORDS

Pharmacogenetics; Pharmacogenomics; Personalized medicines; High throughput nucleotide sequencing; Germline mutation; Health plan implementation; Clinical guidelines; Genome structural variants.

Resumen

Objetivo: A medida que se incorporan más genes a los procesos farmacogenómicos asistenciales y se otorga más importancia a las variantes raras, el uso de paneles de secuenciación dirigida por captura se ha propuesto como una alternativa muy eficiente atendiendo a sus costes, su rendimiento y la cobertura profunda, característica de los datos de secuenciación de nueva generación de alta calidad. El objeto de este trabajo es describir la prevalencia de variantes farmacogenéticas clínicamente procesables descritas previamente en la literatura científica, así como de nuevas variantes identificadas mediante tecnologías de secuenciación de nueva generación y evaluar los fármacos potencialmente afectados por estas variantes.

Método: Se evaluó un panel de 18 genes relacionados con la farmacogenómica clínicamente procesables en 41 individuos con diagnóstico de cáncer de mama que van a recibir tratamiento adyuvante y neoadyuvante. Se estudió

PALABRAS CLAVE

Farmacogenética; Farmacogenómica; Medicina personalizada; Secuenciación de nucleótidos de alto rendimiento; Mutación de la línea germinal; Aplicación del plan de salud; Guías de práctica clínica; Variante genómica estructural.

bed clinically actionable variants as well as of phenotypes classified according to current interpretation standards was studied. The pharmacological treatments potentially affected by the identified variants were also evaluated. An estimation was made of the prevalence of not previously described, possibly deleterious, variants selected using bioinformatics criteria.

Results: All subjects carried clinically actionable variants, with a mean of 4.02 genes affected by each variant per individual. *VKORC1, CYP4F2, CYP2C19, CYP2D6* and *CYP2B6* were the most polymorphic genes and were present with actionable phenotypes in more than 50% of patients; 15-50% had actionable phonotypes in *UGT1A1, SLCO1B1, CYP2C9* and *TPMT* and 2-15% in *HLA-B, CYP3A5, HLA-A* and *DPYD*. No actionable variants were identified in *RYR1, CACNA1S, G6PD, F5 and NUDT15*. These variants had the potential to affect response to 84% of the drugs described in the leading pharmacogenetic guidelines. Possibly deleterious variants not previously described accounted for 11.4% of all clinically actionable variants and were present in 12.2% of patients.

Conclusions: The results obtained show a high prevalence of clinically actionable variants, both common, i.e., previously described in the literature, and rare, i.e., not previously studied with conventional technological approaches. The latter are candidates for a more exhaustive molecular and/or clinical characterization.

la prevalencia de variantes clínicamente procesables previamente descritas en la literatura científica, así como de los fenotipos farmacogenéticos clasificados según los estándares de interpretación actuales. Asimismo, se evaluaron los tratamientos farmacológicos potencialmente afectados por las variantes identificadas. Se estimó la prevalencia de variantes posiblemente deletéreas no descritas previamente seleccionadas con criterios bioinformáticos.

Resultados: Todos los individuos fueron portadores de variantes clínicamente procesables, con una media de 4,02 genes afectados por alguna variante por individuo. Los genes *VKORC1, CYP4F2, CYP2C19, CYP2D6* y *CYP2B6* fueron los más polimórficos, con más de un 50% de pacientes con fenotipos procesables; un 15-50% en *UGT1A1, SLCO1B1, CYP2C9* y *TPMT* y un 2-15% *HLA-B, CYP3A5, HLA-A* y *DPYD*. No se identificaron variantes procesables en *RYR1, CACNA1S, G6PD, F5* y *NUDT15*. Estas variantes afectarían a la respuesta de un 84% de los fármacos descritos en las principales guías de farmacogenética. Las variantes posiblemente deletéreas no descritas previamente supusieron un 11,4% del total de variantes clínicamente procesables y están presentes en un 12,2% de los pacientes.

Conclusiones: Los resultados obtenidos constatan una alta prevalencia de variantes clínicamente procesables tanto comunes, previamente descritas en la literatura, como raras, no estudiadas con abordajes tecnológicos convencionales y candidatas a una caracterización molecular y/o clínica más exhaustiva.

Introduction

Pharmacogenetic research has, since its initial stages, identified numerous genes related to the metabolism and transport of, and the response to, drugs showing that many of the genomic variables in these genes are associated with inter-individual pharmacological response variations. Multiple clinical guidelines and other sources of information have been published in the last few years that have helped identify a number of key genes that contain clinically actionable variants, with patients carrying such variants requiring dose adjustments or specific therapeutic strategies[1]. These gene-drug pairs include metabolizing enzymes (CYP2C19 and clopidogrel[2]) and transporting (SLCO1B1 and simvastatin[3]) and other proteins involved in the pharmacological response (RYR1 and halogenated anesthetics)[4].

Although pharmacogenetic studies are becoming increasingly popular in clinical centers, most of the genomic variations analyzed are common (i.e. with an allele frequency > 1%)[1,5]. In fact, most of the currently available high-throughput pharmacogenomic platforms are focused mainly on common variations[6]. However, several studies based on next generation sequencing (NGS) have confirmed the existence of rare deleterious variants (i.e., with an allele frequency < 1%), which are very frequently found in drug metabolizing enzymes and in the genes coding for pharmacological target proteins. It has been estimated that up to 17% of individuals harbor this kind of variant[6,7]. Moreover, rare variants have been directly associated with more severe drug response variations than common variants[6], as well as with unusual adverse reactions[8]. For that reason, it is a priority to endow clinical processes with technologies able to identify and manage information not only on the widely studied common variants but particularly on the less known rare variants.

NGS techniques are becoming increasingly popular for the performance of routine genetic studies. Indeed, their cost has been going down in the last few years, the equipment needed is available in a growing number of centers, and there is a rising awareness that rare variants play an important role in the development of disease and in the patients' response to their medication[6]. Most pharmacogenomic studies based on NGS techniques correspond to whole exome and whole genome sequencing projects led by large research consortia[9,10]. Whole exome and whole genome sequencing is still associated with high costs and with problems related to the processing and storage of the large amounts of data generated[1,6,11,12]. The use of targeted high-throughput sequencing panels, capable of capturing and sequencing a small set of genomic targets to high depth has been proposed as an ideal alternative as it represents a middle ground that maximizes throughput while maintaining the deep coverage characteristic of high-quality NGS data[1,6,11,12].

The A Coruña University Hospital Complex has developed a previously described and validated NGS-based pharmacogenomic platform[13] intended to support clinical practice and research studies. The platform was designed with a view to studying high evidence, clinically actionable genes and pharmacogenetic regions in addition to genomic regions related to clinical research projects currently underway in the hospital. The idea is to improve the effectiveness of the work carried out in the molecular biology laboratory.

The purpose of this study is to use the NGS platform to identify the prevalence of clinically actionable pharmacogenetic variants in a previously studied population and use NGS to study the new variants identified in the genes that contain clinically actionable variants. In addition, an analysis will be made of the drugs included in pharmacogenetic clinical guidelines that may potentially be affected by such variants.

Methods

Design

This was a descriptive cross-sectional pharmacogenetic variant prevalence study of a population of 41 patients. The sample was selected based on the availability of genomic sequencing data obtained using the NGS platform developed by the A Coruña University Hospital Complex. The studied population corresponded to the total number of patients recruited by the Hospital within the framework of a project geared towards validating pharmacokinetic and pharmacogenetic biomarkers related with the risk of developing neuropathy following administration of taxanes in the context of the neoadjuvant breast cancer therapy.

Genetic study

The genomic regions of clinical interest were captured using a personalized capture probe library (SureSelect Target Enrichment Kit for the Illumina paired-end multiplex sequencing method; Agilent Technologies, Santa Clara, California, USA) and sequenced on the HiSeq 1500 platform (Illumina, San Diego, California) following Illumina protocols[14,15]. The read depth (number of times a base was sequenced by independent reads) of every nucleotide of genes from the defined genomic regions of interest was >30× (mean: 250×-400×). Analytical validation of this platform has been previously described[13]. The capture probe library allows sequencing of a total of 433,000 bases. The genes and regions of interest evaluated in this study correspond to a subset of all the genomic regions included in the capture probe library.

Selection of candidate genomic regions of interest

A group of genomic regions was selected from 18 pharmacogenomics-related genes that were considered clinically actionable (*CACNA1S, CYP2B6, CYP2C19, CYP2C9, CYP2D6, CYP3A5, CYP4F2, DPYD, F5, G6PD, HLA-A, HLA-B, NUDT15, RYR1, SLCO1B1, TPMT, UGT1A1* and *VKORC1*). These genes have been described in several clinical guidelines, including CPIC (Clinical Pharmacogenetics Implementation Consortium)[16], DPWG (Dutch Pharmacogenomics Working Group)[17] and CPNDS (Canadian Pharmacogenomics Network for Drug Safety)[18]. A mixed research strategy was developed, which consisted of: a) the development of a specific allele-variant database that allowed an automatic evaluation of the genetic variants and the pharmacogenetic alleles described in the literature; this database comprised 1,027 variants and was developed based on the PharmVar[19] and PharmGKB[20] databases, and on the GeT-RM pharmacogenomic projects[21-23]; b) an analysis of the candidate functional variants in the coding regions of genes *CACNA1S, CYP2B6, CYP2C19, CYP2C9, CYP2D6, DPYD, NUDT15, RYR1, SLCO1B1, TPMT, UGT1A1* and *VKORC1*.

Bioinformatic analysis

The sequence analysis was carried out using a purpose-developed bioinformatic algorithm that included the demultiplexing of the samples as well as all the steps needed to obtain a validated report of the annotated variants, together with their coverage and quality parameters[13]. Haplotypes were assigned following a purpose-designed algorithm that used variant-allele translation tables developed together with the variant files (vcf format) and the coverage data (cov format) obtained from each sample[13].

The analysis of the copy number variants (CNVs) and the structural variants of *CYP2D6* was carried out using a previously-described and validated comparative coverage depth strategy[13,24].

Genotype interpretation

It was done using the genotype-to-phenotype prediction classification system described in pharmacogenomic prescription guidelines and recommendations. These standards are summarized below. Phenotypes were determined by genotyping sets (haplotypes) of genetic variants known as star alleles "*". Every patient has two star alleles that are collectively referred to as a diplotype or genotype (e.g., *1/*2). Each star allele was then assigned a function (i.e., no, decreased, normal or increased function) and a corresponding numerical activity level based on the evidence available on databases and in leading publications such as PharmVar. The activity levels of the two alleles in each individual were combined and translated into a phenotype (poor, intermediate, normal, rapid, ultrarapid) that was then linked to a selection of specific drugs and a dosing recommendation[22,25].

Clinical actionability

It was determined based on the prescription recommendations described in the CPIC, DPWG and CPNDS clinical guidelines[16-18]. Three different categories were established: "non actionable", "conditional" and "actionable". Table 2 in the Annex includes a detailed description of this classification.

Data analysis

In the first place, a clinically actionable allele prevalence study was conducted; alleles were grouped by patient and by gene. Secondly, an analysis was carried out of the prevalence of the different pharmacogenetic phenotypes obtained from the genotype interpretation process. Thirdly, the clinical actionability of the pharmacogenetic phenotypes identified foe each of the drugs described in pharmacogenetic clinical guidelines was established. Lastly, a bioinformatic algorithm was used to select the potentially deleterious candidate variables using the following filtering criteria: they had to be rare variants (whose gnomAD population frequency was below 1%) located in coding regions (gene coding exons), which could bring about changes in the protein sequence (nonsense, missense) and with a phred score above 20 for the CADD bioinformatic predictor (the phred score is used to select the most deleterious 1% of all possible variants of the gene).

Ethical-legal aspects

The present study was approved by the Drug Research Ethics Committee of Galicia (CEIm-G ID 2017/437). All the patients included gave their informed consent to participate in the study.

Results

The patient sample was made up of a total of 41 individuals of whom 40 were female (97.6%). Mean age was 57.05 ± 11.23 years (range 36-77 years).

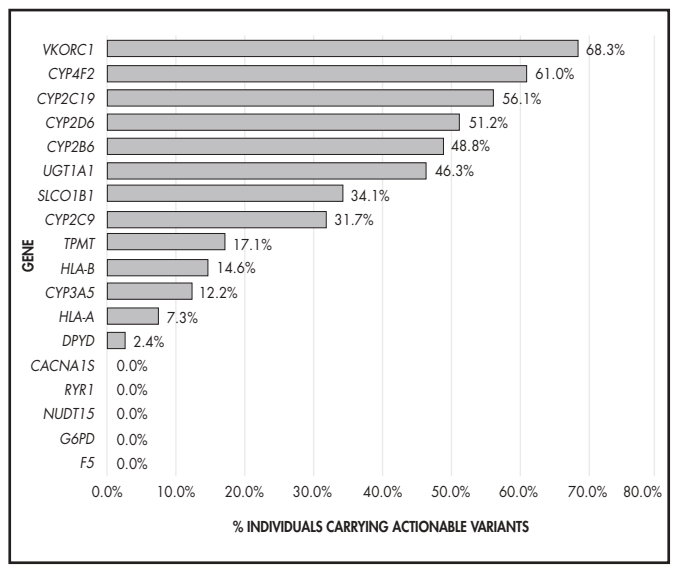
The sequencing and bioinformatic analysis process of the 41 analyzed patients resulted in the identification of 6,802 variants in the genes for whose coding regions there was sequencing data available. A total of 2,216 of these variants were found in the coding genes that had been coded in full. Removing duplications, a total of 175 unique variants were identified. Table 3 in the Annex includes a list of the genetic variants identified in this population.

Distribution of clinically actionable alleles in the studied population

An analysis of clinically actionable alleles, grouped by gene and by individual, showed that all the subjects carried alleles of clinical interest in at least one of the 18 genes studied, with a mean of 4.02 ± 1.68 genes and a maximum number of seven genes; 4.8% of patients were carriers of clinically actionable alleles in one gene, 14.6% in two genes, 22% in three genes, 22% in four genes, 14.6% in five genes, 12.2% in six genes and 9.8% in seven genes.

The analysis of clinically actionable alleles grouped by gene (Figure 1) showed that over 50% of subjects carried alleles of clinical interest in the *VKORC1, CYP4F2, CYP2C19, CYP2D6* and *CYP2B6* genes; between 15 and 50% of subjects carried such alleles in the *UGT1A1, SLCO1B1, CYP2C9* and *TPMT* genes, and between 2 and 15% carried them in the *HLA-B, CYP3A5, HLA-A* and *DPYD* genes. None of the patients carried the *RYR1, CACNA1S, G6PD, F5* or *NUDT15* genes.

Figure 1. *Percentage of individuals carrying clinically actionable alleles in the different genes.*



Distribution of pharmacogenetic phenotypes in the population and their potential influence (clinical actionability) on treatment

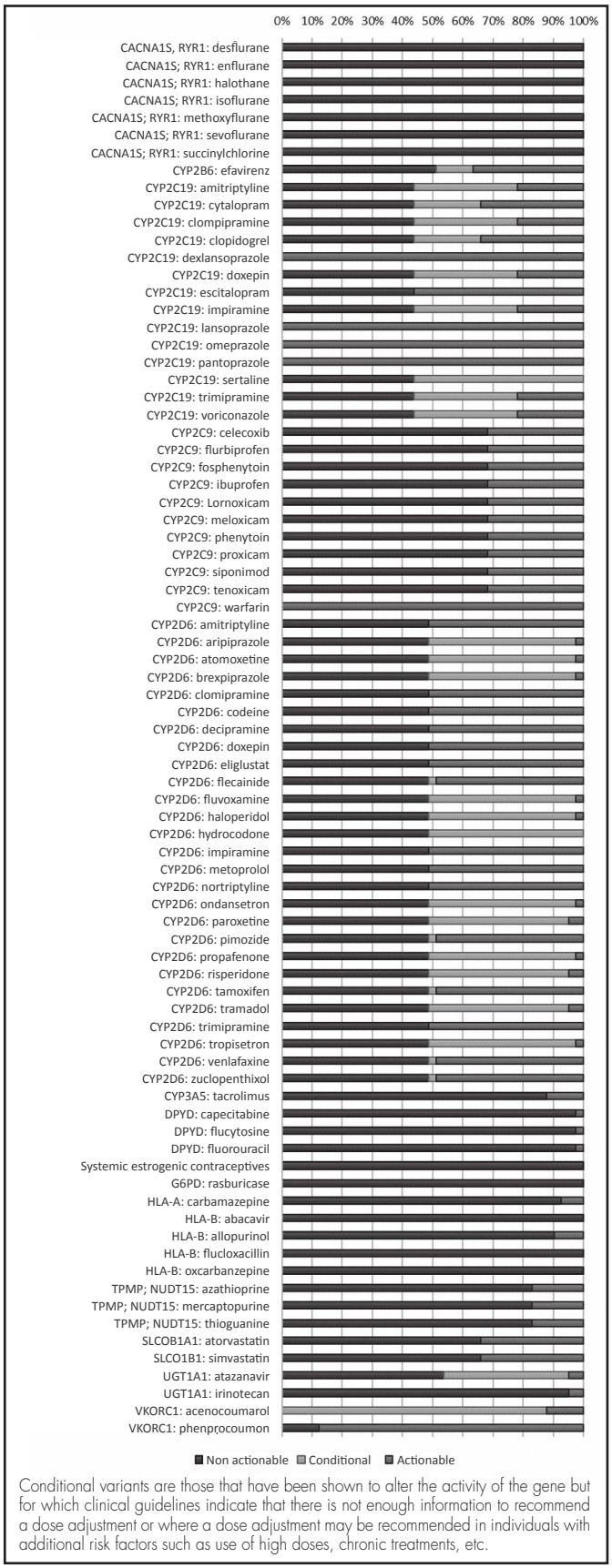
Table 1 shows the pharmacogenetic categories or phenotypes identified in the population. The identified genotypes, together with their frequency in the studied population, are shown in Table 2 in the Annex. Clinical guidelines establish prescription recommendations or strategies for specific medications within each one of these pharmacogenetic categories or phenotypes. A total of 75 different drugs were found to be discussed in the CPIC, DPWG, CPNDS guidelines; 63 of them (84%) appear to be potentially affected by one of the genetic variants identified in the sample.

Table 1. Distribution of pharmacogenetic phenotypes in the analyzed genes

Gene	Category	Nr (%)	Smith *et al.*[30]	McInnes *et al.*[7] (Eur)
CACNA1S	Negative (MH susceptibility)	41 (100)	667 (100)	
CYP2B6	Intermediate metabolizer	15 (36.6)	247 (37)	157,574 (35.3)
	Normal metabolizer	21 (51.2)	355 (53)	235,044 (52.6)
	Rapid metabolizer	5 (12.2)	65 (10)	10,474 (2.3)
CYP2C19	Intermediate metabolizer	14 (34.1)	186 (29)	116,100 (26)
	Normal metabolizer	18 (43.9)	269 (40)	177,971 (39.8)
	Rapid metabolizer	6 (14.6)	160 (24)	121,160 (27.1)
	Ultrarapid metabolizer	3 (7.3)	27 (4)	20,788 (4.7)
CYP2C9	Intermediate metabolizer	13 (31.7)	218 (33)	144,156 (32.3)
	Normal metabolizer	28 (68.3)	434 (65)	284,032 (63,6)
CYP2D6	Intermediate metabolizer	19 (46.3)	248 (37)	113,670 (25.4)*
	Normal metabolizer	20 (48.8)	351 (53)	167,876 (37.6)*
	Poor metabolizer	1 (2.4)	34 (5)	23,220 (5.2)*
	Ultrarapid metabolizer	1 (2.4)	19 (3)	*
CYP3A5	Intermediate metabolizer	5 (12.2)	125 (19)	5,683 (1.3)
	Poor metabolizer	36 (87.8)	496 (74)	436,556 (97.6)
CYP4F2	Intermediate metabolizer	22 (53.7)		95,254 (21.3)**
	Normal metabolizer	16 (39)		217,127 (48.6)**
	Poor metabolizer	3 (7.3)		
DPYD	Intermediate metabolizer	1 (2.4)	8 (1)	30,181 (6.8)
	Normal metabolizer	40 (97.6)	659 (99)	416,050 (93.2)
F5	Negative (FVL)	41 (100)		
G6PD	Normal activity	41 (100)		
HLA-A	Negative	38 (92.7)		
	Positive (HLA-A*31:01 het.)	3 (7.3)		
HLA-B	Negative	37 (90,2)		
	Positive (HLA-B*58:01 het.)	4 (9.8)		
NUDT15	Normal metabolizer	41 (100)		444,955 (99.4)
RYR1	Negative (HM susceptibility)	41 (100)	662 (99)	
SLCO1B1	Increased function	1 (2.4)	158 (24)	120,720 (27)
	Normal function	16 (39)	495 (74)	171,380 (38.3)
	Normal function; increased function	10 (24.4)		
	Poor function	1 (2.4)	14 (2)	10,304 (2.3)
	Decreased function	13 (31.7)		83,552 (18.7)
TPMT	Intermediate metabolizer	2 (4.9)	59 (9)	
	Intermediate metabolizer; poor metabolizer	5 (12.2)		
	Normal metabolizer	34 (82.9)	607 (91)	
UGT1A1	Intermediate metabolizer	17 (41.5)		204 (0)***
	Normal metabolizer	21 (51.2)		142,438 (31.8)***
	Poor metabolizer	1 (2.4)		***
	Rapid metabolizer	1 (2.4)		***
VKORC1	NP c.-1639G>A	13 (31.7)	274 (41)	175,737 (39.3)
	hom. c.-1639G>A	5 (12.2)	88 (13)	62,474 (14)
	het. c.-1639G>A	23 (56.1)	305 (46)	209,357 (46.8)

FVL: factor V Leiden; het.: heterozygous carrier; hom.: homozygous carrier; MH: malignant hyperthermia; NC: non-carrier.
*McInnes et al did not analyze CNVs in CYP2D6. A total of 17.1% of subjects were classified as intermediate metabolizers. **McInnes et al classified intermediate and poor metabolizers as part of the same group. ***McInnes et al reported that 68.1% were "unavailable" among the population.

Figure 2. *Clinical actionability of the identified pharmacogenetic alleles for different drugs.*



Conditional variants are those that have been shown to alter the activity of the gene but for which clinical guidelines indicate that there is not enough information to recommend a dose adjustment or where a dose adjustment may be recommended in individuals with additional risk factors such as use of high doses, chronic treatments, etc.

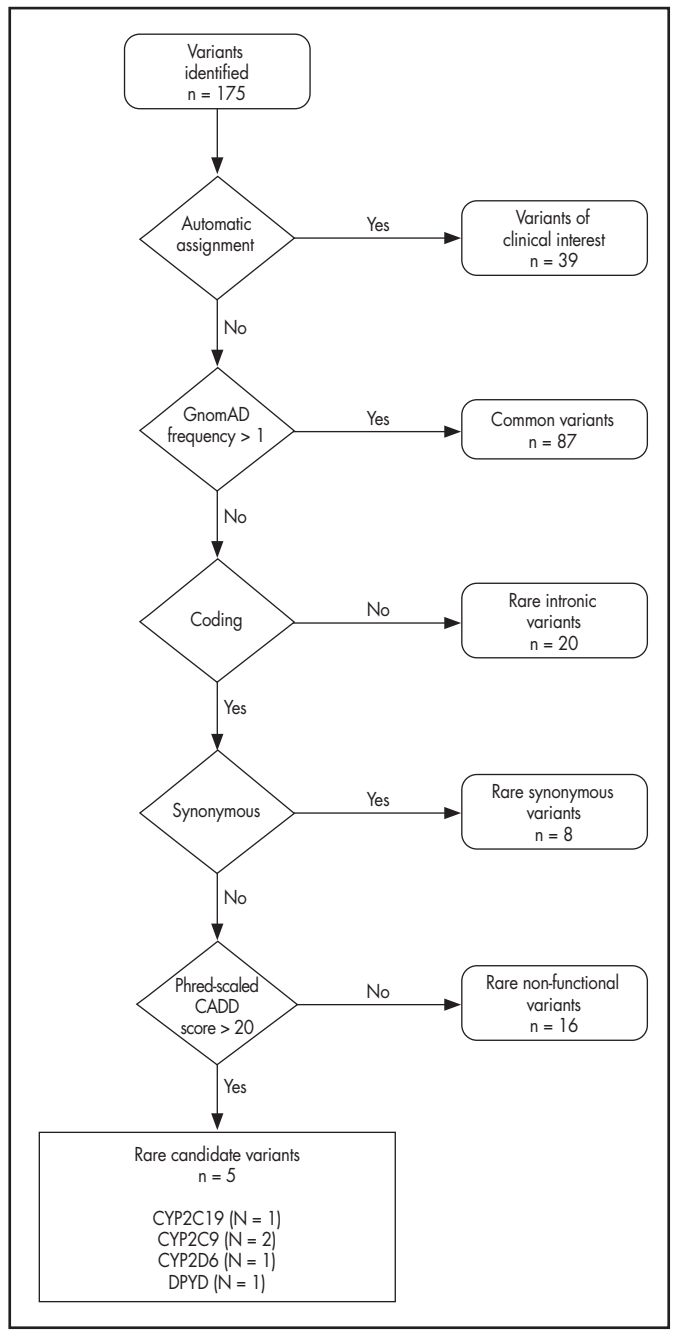
Figure 2 shows the proportion of patients with actionable variants in the different treatment categories included in the clinical guidelines.

Apart from the SNV and INDEL variants usually considered in conventional techniques, 14 samples (34.15%) were found to contain CNVs in the *CYP2D6* gene. Five samples carried heterozygous deletions (*5), three samples had heterozygous duplications (x2) and 7 samples possessed hybrid *CYP2D6/2D7* tandem alleles. One of the samples presented with a deletion and a *36 hybrid; the remaining hybrids identified were of the *68 class.

Candidate variants identified

By recourse to the analysis described in Figure 3, a total of five variants were identified that had not been included in the previously described sets

Figure 3. *Bioinformatic process followed to filter candidate variants that may potentially exert a functional effect on the protein.*



of clinically actionable variants (11.4% of all clinically actionable variants, present in 12.2% of subjects). These variants were *CYP2C19* p.Arg125His / c.374G>A, *CYP2C9* p.Pro33Ser / c.97C>T and p.Val153Ala / c.458T>C, *CYP2D6* p.Tyr355Cys / c.1064A>G and *DPYD* p.Lys259Glu / c.775A>G. Table 3 in the Annex includes a more detailed description of these variants including their location in the gene and their population frequency according to the gnomAD database.

Discussion

The present article describes the prevalence of clinically actionable pharmacogenetic variants and alleles in the genes most commonly covered by clinical guidelines as well as the prevalence of new candidate variants in those same genes. The study was carried out using a purpose-developed pharmacogenetic platform based on NGS technologies aimed at providing support to studies seeking to advance both clinical practice and scientific inquiry[13]. A cohort of 41 patients was analyzed, which corresponded to the accessible patients from whom genomic sequencing data had been obtained using the same platform. Patients with breast cancer in their first cycle of neoadjuvant chemotherapy were deemed to be an appropriate population (proof of concept) to test the implementation of this kind of screening in clinical practice as genetic studies could be added to other diagnostic tests in these patients, the results provided by genomic biomarkers possibly changing future therapeutic management.

This study has shown that pharmacogenetic variants of clinical interest in key genes are highly prevalent, and that the majority of individuals in the studied population exhibited multiple clinically actionable variants. This high prevalence was already reported by other authors such as Van Driest *et al.* who identified one such variant in the majority of individuals studied (98%)[26]. Bush *et al.*, who used the eMERGE cohort with an NGS capture sequencing panel (PGRNseq), identified one or more level A actionable variants (CPIC) in 96.19% of all samples, with a median of two actionable variants per individual[27]. Likewise, McInnes *et al.*, who analyzed a cohort of patients from the UK Biobank using a whole genome approach, identified one variant in 99.5% of individuals with a mean of 3.7 genes per individual containing clinically actionable variants[7].

Table 1 shows that, for some genes such as *SLCO1B1* or *TPMT*, variant combinations have been identified for which multiple classifications are possible. These ambiguous results, reported previously by other authors and present in other pharmacogenetic analysis platforms, are due to the fact that certain combinations of functional variants may be identified in the same or in different alleles therefore affecting one or both alleles of the gene[21-23].

NGS massive sequencing technologies allow identification of rare variants that have not been described previously and that are not included in conventional genotyping platforms. Although these variants are extremely rare in isolation, when taken as a whole they are apt to affect a large number of individuals[6,8]. The role of these variants has been scarcely studied in the literature, with most of the information available being based on proofs of concept[9,10]. This study identified a total of five candidate variants in 41 subjects (i.e., in 12.2% of the sample) using an algorithm that takes into consideration the variant's allelic frequency, the location of the gene, any changes in the protein sequence, and the *in silico* bioinformatic prediction (CADD). The accuracy of the *in silico* bioinformatic predictor used (CADD) has been estimated at 84%[28]. In addition to displaying a phred-scaled CADD score above 20, these variants are considered deleterious by at least three additional bioinformatic predictors: SIFT, Poliphen-2 and DANN. The p.Lys259Glu/c.775A>G DPYD variant is included in the pharmVar database as due for classification and, with the exception of the variants above, the bioinformatic Poliphen-2 predictor classifies it as benign. We believe that these *in silico* results do not preclude the performance of confirmation studies. As regards the potential clinical application of the technology, these variants could result in a decrease in the genes' activity and patients carrying them may benefit from a closer follow-up when prescribed a drug that may be affected. Furthermore, 17% of the population studied by McInnes *et al.* carried at least one deleterious variant of one of the 14 genes analyzed that was not included in the existing allelic definitions[7]. In the limited sample included in this analysis, the frequency of potentially deleterious rare variants vs. the already established ones was low (5 vs. 39), which contrasts with previous reports, which found rare variants to account for half of total variants[27]. Lastly, rare genetic variants could be the key for applying the information about the better biologically preserved genes, e.g., those coding for pharmacological targets, as the only variants identified for many of these genes are rare[29].

This is one of the first studies to analyze the results of implementing a pharmacogenomics-specific NGS sequencing platform to support clinical and research activities. The PGRN (Pharmacogenomics Research Network), in collaboration with several US centers involved in the eMERGE-PGx pharmacogenomic sequencing implementation project, has developed a similar platform to the one presented here[11,27]. For these projects to be successful, multi-center studies are needed that generate a broader knowledge base. It is to be hoped that more centers can join this initiative and benefit from the use of this platform for clinical and research purposes.

When selecting the technology to be used for pharmacogenetic implementation, it is essential to take into consideration numerous factors, most of them related to the assets that must be available to the molecular biology laboratory of the participating center. At the same time, it must be remembered that the different technologies are complementary and the decision to prioritize one over another should be made based on the specific clinical condition of the patient. A detailed description of pharmacogenetic technologies may be found in van der Lee M *et al.*[6]. The Annex included here provide a detailed explanation of the reasons behind the choice of the technological approach used.

The main limitations of this study are its small sample size and the failure to validate the genomic findings identified by means of *in vitro*, *ex vivo* or *in vivo*, molecular functional studies and subsequently validate the genotype-phenotype correlation in the studied patients. Another limitation is the failure to obtain information about the pharmacological treatment that patients were on at the time of —or before— the study, which could have been influenced by the pharmacogenetic alleles identified. Nor was there any intervention made regarding prescription of the medication, or were the patients' health outcomes analyzed to evaluate the therapeutic interventions or their clinical relevance. Although that was not one of the goals of the study and the clinical impact of the pharmacogenetic alleles studied has admittedly been well-described in the literature, such evaluations could be useful to clinically validate the platform. It is estimated that around 24% of the general population receive a medication affected by their genotype[7]. What is more, this prevalence could be even higher in patients with the characteristics of the subject included in this study.

Funding

This article was funded by an AII SEFF grant for hospital pharmacy research.

Acknowledgements

The authors wish to thank the staff at the Hospital Pharmacy Department of the A Coruña Hospital who were in change of looking after the subjects of this study. A special thanks goes to Teresa Calleja Chuclá, María Mateos Salvador and Elena Fernández Gabriel.

We would also like to thank the Health in Code staff members, who contributed to the processing and analysis of the samples. A special thanks goes to the members of the pharmacogenetics team (Sara Santana, Maite García, Iria Gómez and Rosalía Peteiro), tue quality team (Dolores Salvado), the laboratory team (Lisi Santiago, Isaac Rosa, Raquel Calvo, Marta Neira, María del Pilar González and Luis Santomé) and the bioinformatics

team (Andrea Grana, Roberto Noya, Pablo García, Pablo Cabaleiro and David de Uña).

Conflict of interest

Luis Ramudo Cela is a member of the scientific committee at Health in Code. The other authors declare to have no conflict of interest.

Contribution to the scientific literature

The present study demonstrates the usefulness of implementing next generation sequencing-based pharmacogenomic processes in clinical practice with a view to identifying both common and rare clinically actionable variants not studied previously by conventional approaches.

ANNEX 1

Selection of the best technological approach

When selecting the technology for implementing a pharmacogenetic approach, it is crucial to consider a series of factors, most of them dependent on the assets and resources available to the molecular biology laboratory where the testing will be carried out. It is also important to bear in mind that the different technologies are complementary, and the decision to prioritize one over another should be made on the basis of the patients' clinical situation.

Our hospital was already using NGS technology for diagnostic studies, and had obtained a large volume of samples prior to the setting up of the pharmacogenetics panel used in the study. Moreover, the team of molecular biologists, IT specialists and physicians who participated in the study had already gained significant experience in the management of purpose-developed "tailor-made" panels, databases and bioinformatic algorithms. This made it easier to carry out the required NCS processing and to acquire enough affordable reagents to deal with the high volume of work involved. Also, the possibility of combining pharmacogenetic and diagnostic samples in one single sequencing pool makes it possible to work with smaller batches of pharmacogenetic samples without the risk of excessive sample accumulations (which is an important limitation to the use of dedicated processes and is particularly important for array rtPCR technologies, where sample volumes are typically low). Although the theoretical per-sample cost for array rtPCR procedures is lower than the sample cost for NGS procedures, when such factors are considered as the depreciation of new equipment, the development of new workflows, the extra cost of personalizing the arrays and the need to use a larger number of sample batches, the implementation of this technology usually turns out to be more disadvantageous than adapting an already-implemented NGS procedure. New process automation systems such as the Magnis NGS Prep System, or Agilent's Bravo Automated Liquid Handling Platform allow a reduction of NGS preparation times in the wet lab of up to 48 hours. Mean NGS response times in our center are of 3-5 weeks from the arrival of the sample (including bioinformatic data processing and preparation of the clinical report). Emergency clinical scenarios (e.g., DPYD studies prior to treatment with fluoropyrimidines) may be addressed with more targeted complementary technologies such as Sanger sequencing or low-scale rtPCR.

While NGS panels that include a set of genes of interest do not need to have their design updated following the publication of new variants of interest for those genes, targeted technologies require either updating their design with any non-included variant or applying a complementary technique to ensure the success of the study.

The strengths of the platform include the fact that the laboratory is UNE-EN ISO 15189 and CLIA (Clinical Laboratory Improvement Amendments) certified, both accreditations covering NGS pharmacogenetic procedures. In addition, the platform has been validated by comparative studies performed by institutions from different geographical areas (the College of American Pathologists [CAP]), the European Molecular Genetics Quality Network [EMQN]) and the Spanish Society of Pharmacogenetics and Pharmacogenomics (SEFFT). The platform could be used by centers wishing to outsource the process or may alternatively be implemented in centers that possess the required equipment.

Table 1. Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Abacavir	HLA-B	Positive *57:01	Yes	Yes	
		Negative 15:01	No	No	
Acenocoumarol	VKORC1	het. c.-1639G>A		Yes	
		hom. c.-1639G>A		Conditional	
		NC c.-1639G>A		Conditional	
Alopurinol	HLA-B	*58:01 negative	No		
		*58:01 positive	Si		
Amitriptyline	CYP2C19	IM	Conditional		
		NM	No		
		PM	Si		
		RM	Si		
		UM	Si		
	CYP2D6	IM	Si	Si	
		NM	No	No	
		PM	Yes	Yes	
		UM	Yes	Yes	
Aripiprazole	CYP2D6	IM		Conditional	
		NM		No	
		PM		Yes	
		UM		Conditional	

Table 1 *(cont.).* Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Atazanavir	UGT1A1	IM	Conditional		
		NM	No		
		PM	Yes		
Atomoxetine	CYP2D6	IM	Conditional	Conditional	
		NM	Conditional	No	
		PM	Conditional	Yes	
		UM	Conditional	Conditional	
Atorvastatin	SLCO1B1	DF		Yes	
		NF		No	
		PF		Yes	
Azathioprine	NUDT15	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
	TPMT	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
Brexpiprazole	CYP2D6	IM		Conditional	
		NM		No	
		PM		Yes	
		UM		Conditional	
Capecitabine	DPYD	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
Carbamazepine	HLA-A	31:01 negative	No		No
		31:01 positive	Yes		Yes
	HLA-B	*15:02 negative	No		No
		*15:02 negative	Yes		Yes
Celecoxib	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
Citalopram	CYP2C19	IM	Conditional	Yes	
		NM	No	No	
		PM	Yes	Yes	
		RM	Yes	Conditional	
		UM	Yes	Conditional	
Clomipramine	CYP2C19	IM	Conditional	Conditional	
		NM	No	No	
		PM	Yes	Conditional	
		RM	Yes	Yes	
		UM	Yes	Yes	
	CYP2D6	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
		UM	Yes	Yes	
Clopidogrel	CYP2C19	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
		RM	Conditional	Conditional	
		UM	Conditional	Conditional	

Table 1 *(cont.).* Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Codeine	CYP2D6	IM	Yes	Conditional	No
		NM	No	No	No
		PM	Yes	Yes	Yes
		UM	Yes	Yes	Yes
Desflurane	CACNA1S	MH negative	No		
		MH positive	Yes		
	RYR1	MH negative	No		
		MH positive	Yes		
Desipramine	CYP2D6	IM	Yes		
		NM	No		
		PM	Yes		
		UM	Yes		
Dexlansoprazole	CYP2C19	IM	Yes		
		NM	Yes		
		PM	Yes		
		RM	Yes		
		UM	Yes		
Doxepin	CYP2C19	IM	Conditional		
		NM	No		
		PM	Yes		
		RM	Yes		
		UM	Yes		
	CYP2D6	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
		UM	Yes	Yes	
Efavirenz	CYP2B6	IM	Yes	Conditional	
		NM	No	No	
		PM	Yes	Yes	
		RM	Conditional	Conditional	
		UM	Conditional	Conditional	
Eliglustat	CYP2D6	IM		Yes	
		NM		No	
		PM		Yes	
		UM		Yes	
Enflurane	CACNA1S	MH negative	No		
		MH positive	Yes		
	RYR1	MH negative	No		
		MH positive	Yes		
Escitalopram	CYP2C19	IM	Conditional	Yes	
		NM	No	No	
		PM	Yes	Yes	
		RM	Yes	Yes	
		UM	Yes	Yes	
Flecainide	CYP2D6	IM		Yes	
		NM		No	
		PM		Yes	
		UM		Conditional	

Table 1 *(cont.).* Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Flucytosine	DPYD	IM		Yes	
		NM		No	
		PM		Yes	
Flucloxacillin	HLA-B	*57:01 negative		No	
		*57:01 positive		Yes	
Fluorouracil	DPYD	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
Flurbiprofen	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
Fluvoxamine	CYP2D6	IM	Conditional		
		NM	No		
		PM	Yes		
		UM	Conditional		
Fosphenytoin	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
	HLA-B	*15:02 negative	No		
		*15:02 positive	Yes		
Haloperidol	CYP2D6	IM		Conditional	
		NM		No	
		PM		Yes	
		UM		Conditional	
Halothane	CACNA1S	MH negative	No		
		MH positive	Yes		
	RYR1	MH negative	No		
		MH positive	Yes		
Hydrocodone	CYP2D6	IM	Conditional		
		NM	No		
		PM	Conditional		
		UM	Conditional		
Ibuprofen	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
Imipramine	CYP2C19	IM	Conditional	Conditional	
		NM	No	No	
		PM	Yes	Yes	
		RM	Yes	Conditional	
		UM	Yes	Conditional	
	CYP2D6	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
		UM	Yes	Yes	
Irinotecan	UGT1A1	IM		No	
		NM		No	
		PM		Yes	

Table 1 *(cont.).* Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Isoflurane	CACNA1S	MH negative	No		
		MH positive	Yes		
	RYR1	MH negative	No		
		MH positive	Yes		
Lansoprazole	CYP2C19	IM	Yes	Conditional	
		NM	Yes	No	
		PM	Yes	Conditional	
		RM	Yes	Yes	
		UM	Yes	Yes	
Lornoxicam	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
Meloxicam	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
Mercaptopurine	NUDT15	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
	TPMT	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
Methoxiflurane	CACNA1S	MH negative	No		
		MH positive	Yes		
	RYR1	MH negative	No		
		MH positive	Yes		
Metoprolol	CYP2D6	IM		Yes	
		NM		No	
		PM		Yes	
		UM		Yes	
Nortriptyline	CYP2D6	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
		UM	Yes	Yes	
Omeprazole	CYP2C19	IM	Yes	Conditional	
		NM	Yes	No	
		PM	Yes	Conditional	
		RM	Yes	Yes	
		UM	Yes	Yes	
Ondansetron	CYP2D6	IM	Conditional		
		NM	No		
		PM	Conditional		
		UM	Yes		
Oxcarbazepine	HLA-B	*15:02 negative	No		
		*15:02 positive	Yes		
Pantoprazole	CYP2C19	IM	Yes	Conditional	
		NM	Yes	No	
		PM	Yes	Conditional	
		RM	Yes	Yes	
		UM	Yes	Yes	

Table 1 *(cont.).* Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Paroxetine	CYP2D6	IM	Conditional	Conditional	
		NM	No	No	
		PM	Yes	Conditional	
		UM	Yes	Yes	
Phenprocoumon	VKORC1	het. c.-1639G>A		No	
		hom. c.-1639G>A		Yes	
		NC c.-1639G>A		Yes	
Phenytoin	CYP2C9	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
	HLA-B	*15:02 negative	No		
		*15:02 positive	Yes		
Pimozide	CYP2D6	IM		Yes	
		NM		No	
		PM		Yes	
		UM		Conditional	
Piroxicam	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
Propafenone	CYP2D6	IM		Conditional	
		NM		No	
		PM		Yes	
		UM		Conditional	
Rasburicase	G6PD	DA	Yes		
		NA	No		
		VA	Conditional		
Risperidone	CYP2D6	IM		Conditional	
		NM		No	
		PM		Yes	
		UM		Yes	
Sertraline	CYP2C19	IM	Conditional	Conditional	
		NM	No	No	
		PM	Yes	Yes	
		RM	Conditional	Conditional	
		UM	Conditional	Conditional	
Sevoflurane	CACNA1S	MH negative	No		
		MH positive	Yes		
	RYR1	MH negative	No		
		MH positive	Yes		
Simvastatin	SLCO1B1	DF	Yes	Yes	
		NF	No	No	
		PF	Yes	Yes	
Siponimod	CYP2C9	IM		Yes	
		NM		No	
		PM		Yes	

Table 1 *(cont.).* Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Succinylcholine	CACNA1S	MH negative	No		
		MH positive	Yes		
	RYR1	MH negative	No		
		MH positive	Yes		
Systemic estrogenic contraceptives	F5	FVL negative		No	
		FVL positive		Yes	
Tacrolimus	CYP3A5	IM	Yes	Yes	
		NM	Yes	Yes	
		PM	No	No	
Tamoxifen	CYP2D6	IM	Yes	Yes	Yes
		NM	No	No	No
		PM	Yes	Yes	Yes
		UM	Conditional	Conditional	Conditional
Tenoxicam	CYP2C9	IM	Yes		
		NM	No		
		PM	Yes		
Thioguanine	NUDT15	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
	TPMT	IM	Yes	Yes	
		NM	No	No	
		PM	Yes	Yes	
Tramadol	CYP2D6	IM	Conditional	Conditional	
		NM	No	No	
		PM	Yes	Conditional	
		UM	Yes	Yes	
Trimipramine	CYP2C19	IM	Conditional		
		NM	No		
		PM	Yes		
		RM	Yes		
		UM	Yes		
	CYP2D6	IM	Yes		
		NM	No		
		PM	Yes		
		UM	Yes		
Tropisetron	CYP2D6	IM	Conditional		
		NM	No		
		PM	Conditional		
		UM	Yes		
Venlafaxine	CYP2D6	IM		Yes	
		NM		No	
		PM		Yes	
		UM		Conditional	
Voriconazole	CYP2C19	IM	Conditional	Conditional	
		NM	No	No	
		PM	Yes	Yes	
		RM	Yes	Yes	
		UM	Yes	Yes	

Table 1 *(cont.).* Clinical actionability of the recommendations of pharmacogenetic clinical guidelines grouped by drug, gene, and phenotype (or genotype, if appropriate)

Drug	Gene	Phenotype / genotype	CPIC	DPWG	CPNDS
Warfarin	CYP2C9	IM	Yes	Yes	Yes
		NM	Yes	No	Yes
		PM	Yes	Yes	Yes
	CYP4F2	IM	Yes		
		NM	No		
		PM	Yes		
	VKORC1	het. c.-1639G>A	Yes	No	Yes
		hom. c.-1639G>A	Yes	Yes	Yes
		NC c.-1639G>A	Yes	Yes	Yes
Zuclopenthixol	CYP2D6	IM		Yes	
		NM		No	
		PM		Yes	
		UM		Conditional	

CPIC: Clinical Pharmacogenetics Implementation Consortium; CPNDS: Canadian Pharmacogenomics Network for Drug Safety; DA: decreased activity; DF: decreased function; DPWG: Royal Dutch Association for the Advancement of Pharmacy-Pharmacogenetics Working Group; FVL: factor V Leiden; het.: heterozygosis; hom.: homozygosis; IF: increased function; IM: intermediate metabolizer; MH: malignant hyperthermia; NA: normal activity; NC: non-carrier; NF: normal function; NM: normal metabolizer; PF: poor function; PM: poor metabolizer; RM: rapid metabolizer; UM: ultrarapid metabolizer; VA: variable activity.
Clinical actionability classification. Non-actionable: no pharmacogenic alleles are present; conditional: there is a pharmacogenic allele that alters the activity of the coded enzyme or protein, but changes to the prescription are either contingent on the concomitant presence of other risk factors such as disease, use of high doses, chronic use of the treatment, etc. or not recommended due to lack of sufficient information. In the latter case, the guidelines recommend close monitorization. Actionable: the guidelines recommend dose adjustments or selection of an alternative treatment in carriers with no other risk factors.

Table 2. Genotypes identified and their frequency in the studied population, grouped by gene and phenotype

Gene	Phenotype	Genotype	Nr (%)
CACNA1S	**Negative**	No high-risk MH variant present (rs1800559, rs772226819)	41 (100) 41 (100)
CYP2B6	**Intermediate metabolizer**	*14/*6[a]	1 (2.4)
		*1A/*6A; *4A/*9A	7 (17.1)
		*1A/*7A; *5A/*6A	1 (2.4)
		*4A/*9A; *1A/*6A	4 (9,8)
		*5A/*6A; *1A/*7A	1 (2.4)
		*6A/*6[a]	1 (2.4)
	Normal metabolizer	*1A/*1[a]	14 (34.1)
		*1A/*2[a]	3 (7.3)
		*1A/*5[a]	3 (7.3)
		*5A/*5[a]	1 (2.4)
	Rapid metabolizer	*1A/*4[a]	2 (4.9)
		*22A/*5[a]	1 (2.4)
		*2A/*4[a]	1 (2.4)
		*4A/*5[a]	1 (2.4)
CYP2C19	**Intermediate metabolizer**	*17/*2[a]	1 (2.4)
		*1A/*2[a]	9 (22)
		*1A/*2B	4 (9,8)
	Normal metabolizer	*1A/*1[a]	18 (43.9)
	Rapid metabolizer	*1A/*17	6 (14.6)
	Ultrarapid metabolizer	*17/*17	3 (7.3)
CYP2C9	**Intermediate metabolizer**	*1/*2	7 (17.1)
		*1/*3	6 (14.6)
	Normal metabolizer	*1/*1	28 (68.3)

Table 2 *(cont.).* Genotypes identified and their frequency in the studied population, grouped by gene and phenotype

Gene	Phenotype	Genotype	Nr (%)
CYP2D6	Intermediate metabolizer	*10A/*5	1 (2.4)
		*1A/*3A	1 (2.4)
		*1A/*4A	6 (14.6)
		*1A/*5	2 (4.9)
		*2A/*4A	7 (17.1)
		*2A/*5	1 (2.4)
		*4A/*41	1 (2.4)
	Normal metabolizer	*1A/*1A	5 (12.2)
		*1A/*2A	9 (22)
		*1A/*41	2 (4.9)
		*2A/*41	1 (2.4)
		*2A/*9	1 (2.4)
		*2Ax2/*4A	2 (4.9)
	Poor metabolizer	*6A/*5	1 (2.4)
	Ultrarapid metabolizer	*1Ax2/*1A	1 (2.4)
CYP3A5	Intermediate metabolizer	*1A/*3C	5 (12.2)
	Poor metabolizer	*3C/*3C	35 (85.4)
		*3C/*6	1 (2.4)
CYP4F2	Intermediate metabolizer	*1/*3	10 (24.4)
		*2/*3	11 (26.8)
		*3/*3	1 (2.4)
	Normal metabolizer	*1/*1	16 (39)
	Poor metabolizer	*2+3/*2+3	2 (4.9)
		*2+3/*3	1 (2.4)
DPYD	Intermediate metabolizer	*1/c.1905+1G>A (*2A)	1 (2.4)
	Normal metabolizer	*1/*1	40 (97.6)
F5	Negative	Non-carrier FVL	41 (100)
G6PD	Normal activity	B (homozygosis)	1 (2.4)
		B (homozygosis)	40 (97.6)
HLA-A	Negative	c.*66A= (rs1061235-A)/c.*66A= (rs1061235-A)	38 (92.7)
	Positive (HLA-A*31:01 het.)	c.*66A= (rs1061235-A)/c.*66A>T (rs1061235-T) (*31:01)	3 (7.3)
HLA-B	Negative	B*07:02:01/B*35:08:01	1 (2.4)
		B*07:02:01/B*37:01:01	1 (2.4)
		B*07:02:01/B*38:01:01	1 (2.4)
		B*08:01:01/B*14:02:01	1 (2.4)
		B*08:01:01/B*15:01:01:01	1 (2.4)
		B*08:01:01/B*18:01:01:01	1 (2.4)
		B*08:01:01/B*35:08:01	1 (2.4)
		B*08:01:01/B*44:02:01:01	1 (2.4)
		B*13:02:01/B*14:02:01	1 (2.4)
		B*15:01:01:01/B*49:01:01	1 (2.4)
		B*15:16:01/B*44:03:01	1 (2.4)
		B*18:01:01:01/B*53:01:01	1 (2.4)
		B*35:01:01:01/B*14:02:01	1 (2.4)
		B*35:01:01:01/B*18:01:01:01	1 (2.4)
		B*40:02:01/B*14:02:01	1 (2.4)
		B*40:02:01/B*55:01:01	2 (4.9)
		B*40:04/B*14:02:01	1 (2.4)

Table 2 *(cont.).* Genotypes identified and their frequency in the studied population, grouped by gene and phenotype

Gene	Phenotype	Genotype	Nr (%)
HLA-B	Negative	B*41:01/B*44:03:01	1 (2.4)
		B*44:02:01:01/B*15:16:01	1 (2.4)
		B*44:02:01:01/B*18:01:01:01	1 (2.4)
		B*44:02:01:01/B*27:02:01	1 (2.4)
		B*44:02:01:01/B*51:01:07	1 (2.4)
		B*44:03:01/B*44:02:01:01	1 (2.4)
		B*44:03:01/B*49:01:01	1 (2.4)
		B*44:03:01/B*51:01:01	1 (2.4)
		B*49:01:01/B*49:01:01	1 (2.4)
		B*49:01:01/B*51:01:01	1 (2.4)
		B*49:01:01/B*55:01:01	2 (4.9)
		B*50:01:01/B*51:01:01	1 (2.4)
		B*51:01:01/B*35:01:01:01	1 (2.4)
		B*51:01:01/B*40:02:01	1 (2.4)
		B*51:01:01/B*44:02:01:01	1 (2.4)
		B*51:01:01/B*50:01:01	1 (2.4)
		B*53:01:01/B*38:01:01	1 (2.4)
		B*55:01:01/B*15:16:01	1 (2.4)
	Positive (HLA-B*58:01 het.)	B*13:02:01/B*58:01:01	1 (2.4)
		B*37:01:01/B*58:01:01	1 (2.4)
		B*44:02:01:01/B*58:01:01	1 (2.4)
		B*58:01:01/B*27:05:02	1 (2.4)
NUDT15	Normal metabolizer	*1A/*1A	41 (100)
RYR1	Negative	No high-risk MH variant present	41 (100)
SLCO1B1	Increased function	*14/*1B	1 (2.4)
	Normal function	*1A/*1A	2 (4.9)
		*1A/*1B	11 (26.8)
		*1A/*21	3 (7.3)
		*1B/*21	1 (2.4)
	Normal function; increased function	*1A/*14; *1B/*4	1 (2.4)
		*1B/*4; *1A/*14	8 (19,5)
	Poor function	*15/*5	1 (2.4)
	Decreased function	*14/*15	2 (4.9)
		*14/*17	1 (2.4)
		*14/*5; *15/*4	1 (2.4)
		*15/*1B	1 (2.4)
		*15/*4; *14/*5	1 (2.4)
		*1A/*15; *1B/*5	1 (2.4)
		*1A/*17; *21/*5	1 (2.4)
		*1A/*5	3 (7.3)
		*1B/*5; *1A/*15	2 (4.9)
TPMT	Intermediate metabolizer	*1/*2	2 (4.9)
	Intermediate metabolizer; Poor metabolizer	*1/*3A; *3B/*3C	3 (7.3)
		*3B/*3C; *1/*3A	34 (82.9)
	Normal metabolizer	*1/*1	2 (4.9)

Table 2 *(cont.).* Genotypes identified and their frequency in the studied population, grouped by gene and phenotype

Gene	Phenotype	Genotype	Nr (%)
UGT1A1	Intermediate metabolizer	*1/*28+60; *28/*60	2 (4.9)
		*1/*28+60+93; *28+60/*93	4 (9,8)
		*28/*60; *1/*28+60	1 (2.4)
		*28+60/*93; *1/*28+60+93	5 (12.2)
		*28+60+93/*60	5 (12.2)
	Normal metabolizer	*1/*1	13 (31.7)
		*1/*60	8 (19,5)
	Poor metabolizer	*28+60+93/*28+60+93	1 (2.4)
	Rapid metabolizer	*1/*36+60; *36/*60	1 (2.4)
VKORC1	Normal sensitivity to coumarins	Non-carrier c.-1639G>A (rs9923231)	13 (31.7)
	Highly increased sensitivity to coumarins	Homozygous carrier c.-1639G>A (rs9923231)	5 (12.2)
	Increased sensitivity to coumarins	Heterozygous carrier c.-1639G>A (rs9923231)	23 (56.1)

FVL: factor V Leiden; het.: heterozygosis; hom.: homozygosis; MH: malignant hyperthermia.
The B allele in the *G6PD* gene corresponded to the wild-type reference allele.

Table 3. Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
CYP2B6	NP_000758.1:p.(Leu238=)	NM_000767.4:c.714G>A	NC_000019.9:g.41515192G>A	Coding exon	Synonymous	NO	NO	0,031	0,0891	0,3094	0,0891	2	4,87804878		0	1,219512195
CYP2B6	NP_000758.1:p.(Pro72=)	NM_000767.4:c.216G>C	NC_000019.9:g.41509950G>C	Coding exon	Synonymous	NO	NO	3,718	5,043	4,9602	5,0489	4	9,756097561		0	2,43902439
CYP2B6	NP_000758.1:p.Arg140Gln	NM_000767.4:c.419G>A	NC_000019.9:g.41510286G>A	Coding exon	Nonsynonymous	NO	YES	20,9	0,3455	0,3428	0,3357	2	4,87804878		0	1,219512195
CYP2B6	NP_000758.1:p.Arg22Cys	NM_000767.4:c.64C>T	NC_000019.9:g.41497274C>T	Coding exon	Nonsynonymous	NO	YES	17,44	4,896	4,8301	4,8903	1	2,43902439		0	0,609756098
CYP2B6	NP_000758.1:p.Arg487Cys	NM_000767.4:c.1459C>T	NC_000019.9:g.41522715C>T	Coding exon	Nonsynonymous	NO	YES	0,31	9,0906	8,7843	8,9391	1	2,43902439		0	0,609756098
CYP2B6	NP_000758.1:p.Gln172His	NM_000767.4:c.516G>T	NC_000019.9:g.41512841G>T	Coding exon	Nonsynonymous	NO	YES	0,001	27,319	27,0857	27,4879	14	34,14634146	1	2,43902439	9,756097561
CYP2B6	NP_000758.1:p.Gln21Leu	NM_000767.4:c.62A>T	NC_000019.9:g.41497272A>T	Coding exon	Nonsynonymous	NO	NO	0,117	0,3857	0,4042	0,3846	1	2,43902439		0	0,609756098
CYP2B6	NP_000758.1:p.Lys262Arg	NM_000767.4:c.785A>G	NC_000019.9:g.41515263A>G	Coding exon	Nonsynonymous	NO	YES	0,001	5,6317	14,7183	5,6317	2	4,87804878		0	1,219512195
CYP2B6	NP_000758.1:p.Lys61Thr	NM_000767.4:c.182A>C	NC_000019.9:g.41509916A>C	Coding exon	Nonsynonymous	YES	NO	15,67	0,0017	0,0032	0,0017	20	48,7804878	5	12,19512195	18,29268293
CYP2B6		NM_000767.4:c.1153-9C>T	NC_000019.9:g.41518570C>T	Intron		YES	NO	1,345	0,019	0,0163	0,019	20	48,7804878	5	12,19512195	18,29268293
CYP2B6		NM_000767.4:c.1294+53C>T	NC_000019.9:g.41518773C>T	Intron		NO	NO	1,133		26,6997	31,6893	2	4,87804878		0	1,219512195
CYP2B6		NM_000767.4:c.334+34T>G	NC_000019.9:g.41510102T>G	Intron		NO	NO	11,39	0,0009	0,3152		21	51,2195122	16	39,02439024	32,31707317
CYP2B6		NM_000767.4:c.335-14C>G	NC_000019.9:g.41510188C>G	Intron		NO	NO	6,01	0,2709	0,2791	0,2641	1	2,43902439		0	0,609756098
CYP2B6		NM_000767.4:c.485-1007C>G	NC_000019.9:g.41511803C>G	Intron		NO	NO	9,023		28,318	29,1334	1	2,43902439		0	0,609756098
CYP2B6		NM_000767.4:c.485-18C>T	NC_000019.9:g.41512792C>T	Intron		NO	NO	4,528	33,5284	33,1657	33,3375	21	51,2195122	16	39,02439024	32,31707317
CYP2B6		NM_000767.4:c.646-17C>T	NC_000019.9:g.41515107C>T	Intron		NO	NO	4,245	1,8462	1,7689	2,0419	6	14,63414634		0	3,658536585
CYP2B6		NM_000767.4:c.822+183G>A	NC_000019.9:g.41515483G>A	Intron		NO	NO	2,561		68,8008	76,1581	14	34,14634146		0	8,536585366
CYP2B6		NM_000767.4:c.822+40A>T	NC_000019.9:g.41515340A>T	Intron		NO	NO	2,446				1	2,43902439		0	0,609756098
CYP2B6		NM_000767.4:c.822+50G>A	NC_000019.9:g.41515350G>A	Intron		NO	NO	5,156	1,3805	0,2056	1,3805	1	2,43902439		0	0,609756098
CYP2B6		NM_000767.4:c.823-197T>C	NC_000019.9:g.41515702T>C	Intron		NO	NO	1,759		66,5122	73,4824	6	14,63414634		0	3,658536585
CYP2B6		NM_000767.4:c.-82T>C	NC_000019.9:g.41497129T>C	Intron		NO	YES			1,6546	1,6374	14	34,14634146		0	8,536585366
CYP2C19	NP_000760.1:p.(Pro227=)	NM_000769.2:c.681G>A	NC_000010.10:g.96541616G>A	Coding exon	Synonymous	NO	YES	5,686	18,5627	17,4893	18,7069	14	34,14634146		0	8,536585366
CYP2C19	NP_000760.1:p.(Pro33=)	NM_000769.2:c.99T>C	NC_000010.10:g.96522561T>C	Coding exon	Synonymous	NO	NO	0,096	7,8891	7,6424	7,9405	6	14,63414634		0	3,658536585
CYP2C19	NP_000760.1:p.(Val330=)	NM_000769.2:c.990C>T	NC_000010.10:g.96602622C>T	Coding exon	Synonymous	NO	NO	7,62	18,3515	17,7101	18,501	1	2,43902439		0	0,609756098
CYP2C19	NP_000760.1:p.Arg125His	NM_000769.2:c.374G>A	NC_000010.10:g.96535189G>A	Coding exon	Nonsynonymous	NO	NO	23	0,0297	0,0343	0,0297	1	2,43902439		0	0,609756098
CYP2C19	NP_000760.1:p.Glu92Asp	NM_000769.2:c.276G>C	NC_000010.10:g.96534922G>C	Coding exon	Nonsynonymous	NO	YES	0,026	2,3597	2,2587	2,3019	1	2,43902439		0	0,609756098
CYP2C19	NP_000760.1:p.Ile222Val	NM_000769.2:c.664A>G	NC_000010.10:g.96541599A>G	Coding exon	Nonsynonymous	NO	NO	0,02				1	2,43902439		0	0,609756098
CYP2C19	NP_000760.1:p.Val331Ile	NM_000769.2:c.991G>A	NC_000010.10:g.96602623G>A	Coding exon	Nonsynonymous	NO	NO	0,001	6,2417	5,9734	6,1866	7	17,07317073		0	4,268292683
CYP2C19		NM_000769.2:c.332-23A>G	NC_000010.10:g.96535124A>G	Intron		NO	NO	5,31	18,6267	17,9538	18,7942	7	17,07317073	3	7,317073171	7,926829268
CYP2C19		NM_000769.2:c.-806C>T	NC_000010.10:g.96521657C>T	Intron		NO	YES			20,5184	15,3155	13	31,70731707		0	7,926829268
CYP2C19		NM_000769.2:c.820-51C>G	NC_000010.10:g.96580202C>G	Intron		NO	NO	1,805	18,6111	17,9388	18,7809	3	7,317073171	1	2,43902439	3,048780488
CYP2C9	NP_000762.2:p.(Gly475=)	NM_000771.3:c.1425A>T	NC_000010.10:g.96748737A>T	Coding exon	Synonymous	NO	NO	0,01	6,3769	6,1589	6,3181	29	70,73170732		0	17,68292683
CYP2C9	NP_000762.2:p.(Phe267=)	NM_000771.3:c.801C>T	NC_000010.10:g.96709023C>T	Coding exon	Synonymous	NO	NO	15,43	0,0812	0,0831	0,0811	2	4,87804878		0	1,219512195
CYP2C9	NP_000762.2:p.Arg144Cys	NM_000771.3:c.430C>T	NC_000010.10:g.96702047C>T	Coding exon	Nonsynonymous	NO	YES	29,1	9,1435	9,0956	8,971	13	31,70731707		0	7,926829268

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
CYP2C9	NP_000762.2:p.Ile359Leu	NM_000771.3:c.1075A>C	NC_000010.10:g.96741053A>C	Coding exon	Nonsynonymous	NO	YES	20,4	6,3706	6,1539	6,3104		0	1	2,43902439	1,219512195
CYP2C9	NP_000762.2:p.Pro33Ser	NM_000771.3:c.97C>T	NC_000010.10:g.96698536C>T	Coding exon	Nonsynonymous	NO	NO	24,5				28	68,29268293		0	17,07317073
CYP2C9	NP_000762.2:p.Val153Ala	NM_000771.3:c.458T>C	NC_000010.10:g.96702075T>C	Coding exon	Nonsynonymous	NO	NO	23,8	0,0049	0,0056	0,0049	1	2,43902439		0	0,609756098
CYP2C9	NP_000762.2:p.Val5Ala	NM_000771.3:c.14T>C	NC_000010.10:g.96698453T>C	Coding exon	Nonsynonymous	NO	NO	13,63	0,0305	0,0273	0,0301	1	2,43902439		0	0,609756098
CYP2C9		NM_000771.3:c.169-14G>C	NC_000010.10:g.96701601G>C	Intron		NO	NO	0,859	9,4384	9,4439	9,2589	1	2,43902439		0	0,609756098
CYP2C9		NM_000771.3:c.482-2313A>T	NC_000010.10:g.96705223A>T	Intron		NO	NO	0,482		19,4066	14,4768	16	39,02439024		0	9,756097561
CYP2C9		NM_000771.3:c.820-6326A>C	NC_000010.10:g.96725535A>C	Intron		NO	NO	0,336		18,5248	16,3938		0	1	2,43902439	1,219512195
CYP2C9		NM_000771.3:c.962-32T>C	NC_000010.10:g.96740908T>C	Intron		NO	NO	8,36	4,7688	4,5508	4,6612	18	43,90243902	10	24,3902439	23,17073171
CYP2D6	NP_000097.3:p.(Gly176=)	NM_000106.5:c.528T>C	NC_000022.10:g.42524924A>G	Coding exon	Synonymous	NO	NO	0,044	28,1514	14,5047	28,1514	1	2,43902439		0	0,609756098
CYP2D6	NP_000097.3:p.(His232=)	NM_000106.5:c.696T>C	NC_000022.10:g.42524323A>G	Coding exon	Synonymous	NO	NO	0,006	0,7238	0,6731		19	46,34146341		0	11,58536585
CYP2D6	NP_000097.3:p.(His361=)	NM_000106.5:c.1083T>C	NC_000022.10:g.42523539A>G	Coding exon	Synonymous	NO	NO	2,934	1,0587	0,6735		19	46,34146341		0	11,58536585
CYP2D6	NP_000097.3:p.(Phe112=)	NM_000106.5:c.336C>T	NC_000022.10:g.42525756G>A	Coding exon	Synonymous	NO	NO	4,543	7,7657	7,7645			0	1	2,43902439	1,219512195
CYP2D6	NP_000097.3:p.(Phe219=)	NM_000106.5:c.657T>C	NC_000022.10:g.42524795A>G	Coding exon	Synonymous	YES	NO	0,002	34,3437	28,8157		1	2,43902439		0	0,609756098
CYP2D6	NP_000097.3:p.(Pro267=)	NM_000106.5:c.801C>A	NC_000022.10:g.42524218G>T	Coding exon	Synonymous	NO	NO	5,175	1,0593	0,9592		1	2,43902439		0	0,609756098
CYP2D6	NP_000097.3:p.(Pro325=)	NM_000106.5:c.975G>A	NC_000022.10:g.42523854C>T	Coding exon	Synonymous	YES	NO	12,86	0,3337	0,2929		1	2,43902439		0	0,609756098
CYP2D6	NP_000097.3:p.(Ser401=)	NM_000106.5:c.1203G>A	NC_000022.10:g.42522965C>T	Coding exon	Synonymous	NO	NO	10,41	0,3912	0,4404		10	24,3902439		0	6,097560976
CYP2D6	NP_000097.3:p.(Thr98=)	NM_000106.5:c.294C>G	NC_000022.10:g.42525798G>C	Coding exon	Synonymous	NO	NO	1,268	12,3687	11,4005	87,8645	16	39,02439024		0	9,756097561
CYP2D6	NP_000097.3:p.(Val119=)	NM_000106.5:c.357G>A	NC_000022.10:g.42525183C>T	Coding exon	Synonymous	YES	NO	7,5	0,0009	0,0033	0,0009	16	39,02439024		0	9,756097561
CYP2D6	NP_000097.3:p.(Val136=)	NM_000106.5:c.408C>G	NC_000022.10:g.42525132G>C	Coding exon	Synonymous	NO	NO	0,421	45,1474	44,843	55,0625	18	43,90243902	10	24,3902439	23,17073171
CYP2D6	NP_000097.3:p.Ala122Ser	NM_000106.5:c.364G>T	NC_000022.10:g.42525176C>A	Coding exon	Nonsynonymous	NO	NO	6,612	0,0681	0,0724		11	26,82926829		0	6,707317073
CYP2D6	NP_000097.3:p.Arg259Glyfs*2	NM_000106.5:c.775delA	NC_000022.10:g.42524244delT	Coding exon	Frame Shift	NO	YES	24,2	1,3082	1,247		2	4,87804878		0	1,219512195
CYP2D6	NP_000097.3:p.Arg329Leu	NM_000106.5:c.986G>T	NC_000022.10:g.42523636C>A	Coding exon	Nonsynonymous	YES	NO	23,4	7,5641	2,8174		10	24,3902439		0	6,097560976
CYP2D6	NP_000097.3:p.Arg365His	NM_000106.5:c.1094G>A	NC_000022.10:g.42523528C>T	Coding exon	Nonsynonymous	NO	NO	35	12,1059	9,3852		1	2,43902439		0	0,609756098
CYP2D6	NP_000097.3:p.Asn141Ser	NM_000106.5:c.422A>G	NC_000022.10:g.42525118T>C	Coding exon	Nonsynonymous	NO	NO	15,79		0,0008		2	4,87804878		0	1,219512195
CYP2D6	NP_000097.3:p.Cys296Arg	NM_000106.5:c.886T>C	NC_000022.10:g.42523943A>G	Coding exon	Nonsynonymous	NO	YES	0,042	65,6656	65,5519	65,6025	17	41,46341463		0	10,36585366
CYP2D6	NP_000097.3:p.Gln151Glu	NM_000106.5:c.451C>G	NC_000022.10:g.42525089G>C	Coding exon	Nonsynonymous	NO	NO	0,002	0,2392	0,2333	99,7663	1	2,43902439		0	0,609756098
CYP2D6	NP_000097.3:p.Glu216Ala	NM_000106.5:c.647A>C	NC_000022.10:g.42524805T>G	Coding exon	Nonsynonymous	NO	NO	12,96	0,0039	0,0032	0,0039	22	53,65853659	17	41,46341463	34,14634146
CYP2D6	NP_000097.3:p.Gly212Glu	NM_000106.5:c.635G>A	NC_000022.10:g.42524817C>T	Coding exon	Nonsynonymous	NO	NO	0,001	0,7325	0,7045		10	24,3902439		0	6,097560976
CYP2D6	NP_000097.3:p.Gly373Ser	NM_000106.5:c.1117G>A	NC_000022.10:g.42523505C>T	Coding exon	Nonsynonymous	NO	NO	15,94	1,7085	0,3559	80,7692	28	68,29268293		0	17,07317073
CYP2D6	NP_000097.3:p.His94Arg	NM_000106.5:c.281A>G	NC_000022.10:g.42525811T>C	Coding exon	Nonsynonymous	NO	NO	0,001	11,562	9,9999		22	53,65853659	17	41,46341463	34,14634146
CYP2D6	NP_000097.3:p.Leu91Met	NM_000106.5:c.271C>A	NC_000022.10:g.42525821G>T	Coding exon	Nonsynonymous	NO	NO	23,1	11,3402	9,5161		1	2,43902439		0	0,609756098
CYP2D6	NP_000097.3:p.Lys281del	NM_000106.5:c.841_843delAAG	NC_000022.10:g.42524178_42524180delTCT	Coding exon	Deletion	YES	YES	18,02	1,8972	1,5486		20	48,7804878	20	48,7804878	36,58536585
CYP2D6	NP_000097.3:p.Pro34Ser	NM_000106.5:c.100C>T	NC_000022.10:g.42526694G>A	Coding exon	Nonsynonymous	NO	YES	24,9	24,6687	20,6826		22	53,65853659	17	41,46341463	34,14634146
CYP2D6	NP_000097.3:p.Thr486Ser	NM_000106.5:c.1457C>G	NC_000022.10:g.42522613G>C	Coding exon	Nonsynonymous	NO	NO	0,001	45,556	44,7491	54,6687	22	53,65853659	17	41,46341463	34,14634146
CYP2D6	NP_000097.3:p.Trp152Glyfs*2	NM_000106.5:c.454delT	NC_000022.10:g.42525086delA	Coding exon	Frame Shift	NO	YES	23,5	0,7929	0,8049		22	53,65853659	17	41,46341463	34,14634146
CYP2D6	NP_000097.3:p.Tyr355Cys	NM_000106.5:c.1064A>G	NC_000022.10:g.42523558T>C	Coding exon	Nonsynonymous	NO	NO	22,1	0,7697	0,2337	0,7697	22	53,65853659	17	41,46341463	34,14634146
CYP2D6	NP_000097.3:p.Val11Met	NM_000106.5:c.31G>A	NC_000022.10:g.42526763C>T	Coding exon	Nonsynonymous	NO	NO	5,192	5,3012	3,9348		22	53,65853659	17	41,46341463	34,14634146
CYP2D6	NP_000097.3:p.Val370Ile	NM_000106.5:c.1108G>A	NC_000022.10:g.42523514C>T	Coding exon	Nonsynonymous	NO	NO	2,428	1,3867	0,1439	1,3867	16	39,02439024	12	29,26829268	24,3902439
CYP2D6	NP_000097.3:p.Val7Met	NM_000106.5:c.19G>A	NC_000022.10:g.42526775C>T	Coding exon	Nonsynonymous	NO	NO	2,415	0,328	0,2351	0,3067	16	39,02439024	1	2,43902439	10,97560976
CYP2D6		NM_000106.5:c.*112C>T	NC_000022.10:g.42522464G>A	Intron		NO	NO			0,5889		1	2,43902439		0	0,609756098
CYP2D6		NM_000106.5:c.*184C>T	NC_000022.10:g.42522392G>A	Intron		NO	NO			18,8933		1	2,43902439		0	0,609756098
CYP2D6		NM_000106.5:c.*227A>G	NC_000022.10:g.42522349T>C	Intron		NO	NO			0,1053	0,3395	4	9,756097561		0	2,43902439
CYP2D6		NM_000106.5:c.*264A>G	NC_000022.10:g.42522312T>C	Intron		NO	NO			71,749	76,0982	3	7,317073171		0	1,829268293
CYP2D6		NM_000106.5:c.*26C>T	NC_000022.10:g.42522550G>A	UTR		NO	NO	2,417	2,2075	1,5757	2,2075	28	68,29268293		0	17,07317073
CYP2D6		NM_000106.5:c.1173+40C>A	NC_000022.10:g.42523409G>T	Intron		NO	NO	0,591	34,0283	33,3998	34,0282	1	2,43902439		0	0,609756098
CYP2D6		NM_000106.5:c.1174-9T>C	NC_000022.10:g.42523003A>G	Intron		YES	NO	2,238	5,9767	37,7906		2	4,87804878		0	1,219512195
CYP2D6		NM_000106.5:c.1316-20C>T	NC_000022.10:g.42522774G>A	Intron		NO	NO	0,842	0,277	0,1163	0,277	6	14,63414634		0	3,658536585
CYP2D6		NM_000106.5:c.-1589G>C	NC_000022.10:g.42528382C>G	Intron		NO	NO			79,6524	16,254	16	39,02439024	3	7,317073171	13,41463415
CYP2D6		NM_000106.5:c.-1775A>G	NC_000022.10:g.42528568T>C	Intron		NO	NO			72,4672		12	29,26829268	2	4,87804878	9,756097561
CYP2D6		NM_000106.5:c.180+34C>G	NC_000022.10:g.42526580G>C	Intron		NO	NO	0,38	67,1945	66,5612	32,6762	3	7,317073171		0	1,829268293
CYP2D6		NM_000106.5:c.180+41A>C	NC_000022.10:g.42526573T>G	Intron		NO	NO	1,108	65,6614	66,3199		12	29,26829268	2	4,87804878	9,756097561
CYP2D6		NM_000106.5:c.180+43G>C	NC_000022.10:g.42526571C>G	Intron		NO	NO	1,754	65,3955	66,3607		21	51,2195122	4	9,756097561	17,68292683

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
CYP2D6		NM_000106.5:c.180+47C>T	NC_000022.10:g.42526567G>A	Intron		NO	NO	0,227	64,926	66,3613		4	9,756097561		0	2,43902439
CYP2D6		NM_000106.5:c.181-41G>T	NC_000022.10:g.42525952C>A	Intron		NO	NO	1,647	47,3976	44,0387		12	29,26829268	26	63,41463415	39,02439024
CYP2D6		NM_000106.5:c.-2183G>A	NC_000022.10:g.42528976C>T	Intron		NO	NO			21,9583	26,9369	14	34,14634146	1	2,43902439	9,756097561
CYP2D6		NM_000106.5:c.-44_-43insG	NC_000022.10:g.42526841_42526842insC	UTR		NO	NO	4,969	1,5689	1,4032		1	2,43902439		0	0,609756098
CYP2D6		NM_000106.5:c.505+32A>G	NC_000022.10:g.42525003T>C	Intron		NO	NO	5,149	0,2129	0,1292		7	17,07317073	2	4,87804878	6,707317073
CYP2D6		NM_000106.5:c.506-1G>A	NC_000022.10:g.42524947C>T	Intron		YES	YES	23	17,0761	13,8417		1	2,43902439		0	0,609756098
CYP2D6		NM_000106.5:c.506-29G>A	NC_000022.10:g.42524975C>T	Intron		NO	NO	6,845	3,6561	1,3377	3,6561	2	4,87804878		0	1,219512195
CYP2D6		NM_000106.5:c.506-36G>A	NC_000022.10:g.42524982C>T	Intron		NO	NO	6,256	3,6651	0,812	3,6651	2	4,87804878		0	1,219512195
CYP2D6		NM_000106.5:c.666+43C>T	NC_000022.10:g.42524743G>A	Intron		NO	NO	2,822	33,1398	29,4826	33,1398	7	17,07317073	2	4,87804878	6,707317073
CYP2D6		NM_000106.5:c.985+39G>A	NC_000022.10:g.42523805C>T	Intron		NO	YES	6,015	8,0816	7,8544		14	34,14634146	1	2,43902439	9,756097561
CYP3A5	NP_000768.1:p.(Lys208=)	NM_000777.4:c.624G>A	NC_000007.13:g.99262835C>T	Coding exon	Synonymous	NO	NO	9,935	1,1982	1,2956	1,3273	20	48,7804878	15	36,58536585	30,48780488
CYP3A5	NP_000768.1:p.Thr398Asn	NM_000777.4:c.1193C>A	NC_000007.13:g.99250236G>T	Coding exon	Nonsynonymous	NO	NO	0,084	0,3535	0,3243		11	26,82926829		0	6,707317073
CYP3A5		NM_000777.4:c.219-237G>A	NC_000007.13:g.99270539C>T	Intron		NO	NO	3,375		26,3653			0	41	100	50
CYP4F2	NP_001073.3:p.(His343=)	NM_001082.4:c.1029C>T	NC_000019.9:g.15996820G>A	Coding exon	Synonymous	NO	NO	3,495	28,6405	28,2101	28,429	10	24,3902439		0	6,097560976
CYP4F2	NP_001073.3:p.(Pro55=)	NM_001082.4:c.165A>G	NC_000019.9:g.16008257T>C	Coding exon	Synonymous	NO	NO	0,007	16,8004	16,5553		3	7,317073171		0	1,829268293
CYP4F2	NP_001073.3:p.Gly185Val	NM_001082.4:c.554G>T	NC_000019.9:g.16001215C>A	Coding exon	Nonsynonymous	NO	NO	23,2	4,7139	4,757		5	12,19512195		0	3,048780488
CYP4F2	NP_001073.3:p.Trp12Gly	NM_001082.4:c.34T>G	NC_000019.9:g.16008388A>C	Coding exon	Nonsynonymous	NO	NO	0,001	16,0631	15,7477		39	95,12195122	2	4,87804878	26,2195122
CYP4F2	NP_001073.3:p.Val433Met	NM_001082.4:c.1297G>A	NC_000019.9:g.15990431C>T	Coding exon	Nonsynonymous	NO	NO	26,1	27,2576	26,6086	27,1159	19	46,34146341	15	36,58536585	29,87804878
DPYD	NP_000101.2:p.(Phe632=)	NM_000110.3:c.1896T>C	NC_000001.10:g.97915624A>G	Coding exon	Synonymous	YES	NO	0,005	4,6849	5,043	4,7058	3	7,317073171	38	92,68292683	48,17073171
DPYD	NP_000101.2:p.Arg29Cys	NM_000110.3:c.85C>T	NC_000001.10:g.98348885G>A	Coding exon	Nonsynonymous	NO	NO	23,7	76,5172	76,602		12	29,26829268		0	7,317073171
DPYD	NP_000101.2:p.Ile543Val	NM_000110.3:c.1627A>G	NC_000001.10:g.97981395T>C	Coding exon	Nonsynonymous	NO	NO	9,639	19,2959	19,5184		9	21,95121951	2	4,87804878	7,926829268
DPYD	NP_000101.2:p.Lys259Glu	NM_000110.3:c.775A>G	NC_000001.10:g.98144726T>C	Coding exon	Nonsynonymous	NO	NO	23,4	1,0218	0,608	0,9753	7	17,07317073		0	4,268292683
DPYD	NP_000101.2:p.Met166Val	NM_000110.3:c.496A>G	NC_000001.10:g.98165091T>C	Coding exon	Nonsynonymous	NO	NO	24,5	8,6366	8,585	8,5182	14	34,14634146	1	2,43902439	9,756097561
DPYD	NP_000101.2:p.Met406Ile	NM_000110.3:c.1218G>A	NC_000001.10:g.98039437C>T	Coding exon	Nonsynonymous	NO	NO	17,28	0,6046	0,6736	0,6646	15	36,58536585		0	9,146341463
DPYD	NP_000101.2:p.Ser534Asn	NM_000110.3:c.1601G>A	NC_000001.10:g.97981421C>T	Coding exon	Nonsynonymous	NO	NO	23,4	1,4159	1,4336		34	82,92682927		0	20,73170732
DPYD	NP_000101.2:p.Val732Ile	NM_000110.3:c.2194G>A	NC_000001.10:g.97770920C>T	Coding exon	Nonsynonymous	NO	NO	25,9	4,6473	4,5309		1	2,43902439		0	0,609756098
DPYD		NM_000110.3:c.1129-15T>C	NC_000001.10:g.98039541A>G	Intron		NO	NO	5,766	10,4577	9,6922	10,2832	28	68,29268293	11	26,82926829	30,48780488
DPYD		NM_000110.3:c.1740+39C>T	NC_000001.10:g.97981243G>A	Intron		NO	NO	3,247	18,9927	18,8407	18,9222	13	31,70731707		0	7,926829268
DPYD		NM_000110.3:c.1740+40A>G	NC_000001.10:g.97981242T>C	Intron		NO	NO	1,556	66,0886	63,284		5	12,19512195		0	3,048780488
DPYD		NM_000110.3:c.1905+1G>A	NC_000001.10:g.97915614C>T	Intron		YES	YES	23,7	0,5229	0,5689		18	43,90243902		0	10,97560976
DPYD		NM_000110.3:c.2300-39G>A	NC_000001.10:g.97700589C>T	Intron		NO	NO	6,088	11,761	11,6491	11,6432	7	17,07317073		0	4,268292683
G6PD	NP_001035810.1:p.(Tyr437=)	NM_001042351.2:c.1311C>T	NC_000023.10:g.153760654G>A	Coding exon	Synonymous	NO	NO	6,988	16,7207	16,356		1	2,43902439		0	0,609756098
HLA-A		NM_001242758.1:c.*66A>T	NC_000006.11:g.29913298A>T	UTR		NO	YES	10,48		4,7044	8,5064	7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.(Ala15=)	NM_005514.7:c.45G>A	NC_000006.11:g.31324891C>T	Coding exon	Synonymous	NO	YES	15,43	11,6352	9,1342		3	7,317073171	38	92,68292683	48,17073171
HLA-B	NP_005505.2:p.(Ala159=)	NM_005514.7:c.477C>G	NC_000006.11:g.31324086G>C	Coding exon	Synonymous	NO	YES	6,543	44,9056	48,8756	53,5536	6	14,63414634		0	3,658536585
HLA-B	NP_005505.2:p.(Ala16=)	NM_005514.7:c.48C>A	NC_000006.11:g.31324888G>T	Coding exon	Synonymous	NO	YES	2,301	25,0682	30,1801		2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.(Ala206=)	NM_005514.7:c.618T>G	NC_000006.11:g.31323945A>C	Coding exon	Synonymous	YES	YES	1,19	80,3351	84,5789		26	63,41463415	7	17,07317073	24,3902439
HLA-B	NP_005505.2:p.(Ala24=)	NM_005514.7:c.72C>T	NC_000006.11:g.31324864G>A	Coding exon	Synonymous	YES	YES	10,39	2,3516	3,8903		40	97,56097561		0	24,3902439
HLA-B	NP_005505.2:p.(Ala5=)	NM_005514.7:c.15G>A	NC_000006.11:g.31324921C>T	Coding exon	Synonymous	NO	YES	15,82	15,204	13,4031		9	21,95121951		0	5,487804878
HLA-B	NP_005505.2:p.(Ala95=)	NM_005514.7:c.285A>G	NC_000006.11:g.31324523T>C	Coding exon	Synonymous	NO	YES	6,396	4,0786	2,8874		1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.(Arg103=)	NM_005514.7:c.309G>C	NC_000006.11:g.31324499C>G	Coding exon	Synonymous	NO	YES	9,174	2,5521	3,0411	97,2402	15	36,58536585	4	9,756097561	14,02439024
HLA-B	NP_005505.2:p.(Arg180=)	NM_005514.7:c.540G>C	NC_000006.11:g.31324023C>G	Coding exon	Synonymous	NO	YES	8,168	0,0027	0,0009	0,6043	18	43,90243902	3	7,317073171	14,63414634
HLA-B	NP_005505.2:p.(Arg258=)	NM_005514.7:c.774A>G	NC_000006.11:g.31323215T>C	Coding exon	Synonymous	NO	YES	0,052	25,0375	6,727		9	21,95121951		0	5,487804878
HLA-B	NP_005505.2:p.(Arg59=)	NM_005514.7:c.175A>C	NC_000006.11:g.31324633T>G	Coding exon	Synonymous	NO	YES	9,514	0,1033	0,2354		21	51,21951122	1	2,43902439	14,02439024
HLA-B	NP_005505.2:p.(Arg68=)	NM_005514.7:c.204A>G	NC_000006.11:g.31324604T>C	Coding exon	Synonymous	NO	YES	6,274	13,0273	15,518	13,0273	23	56,09756098		0	14,02439024
HLA-B	NP_005505.2:p.(Asn151=)	NM_005514.7:c.453C>T	NC_000006.11:g.31324110G>A	Coding exon	Synonymous	NO	YES	10,05	4,835	2,5737		20	48,7804878	16	39,02439024	31,70731707
HLA-B	NP_005505.2:p.(Asn198=)	NM_005514.7:c.594C>T	NC_000006.11:g.31323969G>A	Coding exon	Synonymous	NO	YES	7,319	1,8941	0,9147		22	53,65853659	6	14,63414634	20,73170732
HLA-B	NP_005505.2:p.(Asp153=)	NM_005514.7:c.459C>T	NC_000006.11:g.31324104G>A	Coding exon	Synonymous	NO	YES	15,07	22,1871	14,7826		18	43,90243902	19	46,34146341	34,14634146
HLA-B	NP_005505.2:p.(Asp262=)	NM_005514.7:c.786T>C	NC_000006.11:g.31323203A>G	Coding exon	Synonymous	NO	YES	0,172	7,3409	8,1132		2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.(Gln78=)	NM_005514.7:c.234G>A	NC_000006.11:g.31324574C>T	Coding exon	Synonymous	NO	YES	10,62	3,6213	2,4897	3,5743	5	12,19512195		0	3,048780488
HLA-B	NP_005505.2:p.(Glu288=)	NM_005514.7:c.864G>A	NC_000006.11:g.31323125C>T	Coding exon	Synonymous	NO	YES	9,834	0,0049	0,0044		8	19,51219512	32	78,04878049	43,90243902
HLA-B	NP_005505.2:p.(Glu82=)	NM_005514.7:c.246G>A	NC_000006.11:g.31324562C>T	Coding exon	Synonymous	NO	YES	12,07	10,5664	10,2713		20	48,7804878	6	14,63414634	19,51219512

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
HLA-B	NP_005505.2:p.(Gly231=)	NM_005514.7:c.693T>C	NC_000006.11:g.31323296A>G	Coding exon	Synonymous	NO	YES	7,193	84,891	85,0889		15	36,58536585	4	9,756097561	14,02439024
HLA-B	NP_005505.2:p.(Gly245=)	NM_005514.7:c.735C>G	NC_000006.11:g.31323254G>C	Coding exon	Synonymous	NO	YES	6,808	7,6505	0,9069	92,4079	5	12,19512195		0	3,048780488
HLA-B	NP_005505.2:p.(His137=)	NM_005514.7:c.411T>C	NC_000006.11:g.31324152A>G	Coding exon	Synonymous	NO	YES	0,062	3,7801	3,3778		5	12,19512195		0	3,048780488
HLA-B	NP_005505.2:p.(His212=)	NM_005514.7:c.636C>T	NC_000006.11:g.31323353G>A	Coding exon	Synonymous	NO	YES	1,129	39,6858	39,8976		39	95,12195122	2	4,87804878	26,2195122
HLA-B	NP_005505.2:p.(His287=)	NM_005514.7:c.861T>C	NC_000006.11:g.31323128A>G	Coding exon	Synonymous	NO	YES	0,162	17,3756	4,5488		33	80,48780488	7	17,07317073	28,65853659
HLA-B	NP_005505.2:p.(Ile47=)	NM_005514.7:c.141C>T	NC_000006.11:g.31324667G>A	Coding exon	Synonymous	NO	YES	9,471	7,4514	5,7783		32	78,04878049		0	19,51219512
HLA-B	NP_005505.2:p.(Leu102=)	NM_005514.7:c.306G>T	NC_000006.11:g.31324502C>A	Coding exon	Synonymous	NO	YES	12,65	0	0		18	43,90243902	15	36,58536585	29,26829268
HLA-B	NP_005505.2:p.(Leu119=)	NM_005514.7:c.357C>G	NC_000006.11:g.31324206G>C	Coding exon	Synonymous	NO	YES	0,613			92,1658	23	56,09756098	13	31,70731707	29,87804878
HLA-B	NP_005505.2:p.(Leu119=)	NM_005514.7:c.357C>T	NC_000006.11:g.31324206G>A	Coding exon	Synonymous	NO	YES	2,647	7,1791	8,6844		34	82,92682927		0	20,73170732
HLA-B	NP_005505.2:p.(Lys145=)	NM_005514.7:c.435G>A	NC_000006.11:g.31324128C>T	Coding exon	Synonymous	NO	YES	10,74	8,2054	6,8021		9	21,95121951		0	5,487804878
HLA-B	NP_005505.2:p.(Pro129=)	NM_005514.7:c.387G>C	NC_000006.11:g.31324176C>G	Coding exon	Synonymous	NO	YES	10,76	7,772	8,9058	92,228	19	46,34146341	19	46,34146341	34,75609756
HLA-B	NP_005505.2:p.(Pro291=)	NM_005514.7:c.873G>A	NC_000006.11:g.31323116C>T	Coding exon	Synonymous	NO	YES	12,46	4,7356	3,3684		2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.(Pro300=)	NM_005514.7:c.900G>A	NC_000006.11:g.31322996C>T	Coding exon	Synonymous	YES	YES	12,23	57,593	57,7534		12	29,26829268	1	2,43902439	8,536585366
HLA-B	NP_005505.2:p.(Pro71=)	NM_005514.7:c.213G>C	NC_000006.11:g.31324595C>G	Coding exon	Synonymous	NO	YES	11,63	16,2895	16,1718	81,6462	19	46,34146341	12	29,26829268	26,2195122
HLA-B	NP_005505.2:p.(Pro74=)	NM_005514.7:c.222G>A	NC_000006.11:g.31324586C>T	Coding exon	Synonymous	NO	YES	14,2	29,0403	34,9386		20	48,7804878	5	12,19512195	18,29268293
HLA-B	NP_005505.2:p.(Ser121=)	NM_005514.7:c.363C>T	NC_000006.11:g.31324200G>A	Coding exon	Synonymous	NO	YES	4,694	3,2343	3,3339		11	26,82926829	28	68,29268293	40,85365854
HLA-B	NP_005505.2:p.(Ser26=)	NM_005514.7:c.78C>T	NC_000006.11:g.31324730G>A	Coding exon	Synonymous	YES	YES	11,68	1,3664	0,386		6	14,63414634	33	80,48780488	43,90243902
HLA-B	NP_005505.2:p.(Ser336=)	NM_005514.7:c.1008T>C	NC_000006.11:g.31322888A>G	Coding exon	Synonymous	YES	YES	4,089	77,1272	76,5535		1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.(Ser48=)	NM_005514.7:c.144A>C	NC_000006.11:g.31324664T>G	Coding exon	Synonymous	NO	YES	8,045	12,6143	15,7518	13,0793	1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.(Thr118=)	NM_005514.7:c.354C>T	NC_000006.11:g.31324209G>A	Coding exon	Synonymous	YES	YES	7,178	0			20	48,7804878	3	7,317073171	15,85365854
HLA-B	NP_005505.2:p.(Thr158=)	NM_005514.7:c.474C>T	NC_000006.11:g.31324089G>A	Coding exon	Synonymous	NO	YES	11,16	3,6372	2,2775		17	41,46341463	13	31,70731707	26,2195122
HLA-B	NP_005505.2:p.(Thr162=)	NM_005514.7:c.486G>A	NC_000006.11:g.31324077C>T	Coding exon	Synonymous	NO	YES	13,48	1,5883	1,6212		40	97,56097561	1	2,43902439	25,6097561
HLA-B	NP_005505.2:p.(Thr162=)	NM_005514.7:c.486G>C	NC_000006.11:g.31324077C>G	Coding exon	Synonymous	NO	YES	10,69	38,6657	41,2848	59,7864	2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.(Thr252=)	NM_005514.7:c.756T>C	NC_000006.11:g.31323233A>G	Coding exon	Synonymous	NO	YES	0,035	44,5239	43,3704		13	31,70731707		0	7,926829268
HLA-B	NP_005505.2:p.(Thr282=)	NM_005514.7:c.846A>G	NC_000006.11:g.31323143T>C	Coding exon	Synonymous	NO	YES	0,138	18,071	4,7174		15	36,58536585		0	9,146341463
HLA-B	NP_005505.2:p.(Thr55=)	NM_005514.7:c.165C>G	NC_000006.11:g.31324643G>C	Coding exon	Synonymous	NO	YES	8,619	37,883	42,6687	62,1316	16	39,02439024		0	9,756097561
HLA-B	NP_005505.2:p.(Tyr123=)	NM_005514.7:c.369C>T	NC_000006.11:g.31324194G>A	Coding exon	Synonymous	NO	YES	0,212	23,9087	33,508		15	36,58536585		0	9,146341463
HLA-B	NP_005505.2:p.(Val285=)	NM_005514.7:c.855A>G	NC_000006.11:g.31323134T>C	Coding exon	Synonymous	NO	YES	13,32	17,7441	3,9315		34	82,92682927	3	7,317073171	24,3902439
HLA-B	NP_005505.2:p.(Val285=)	NM_005514.7:c.855A>T	NC_000006.11:g.31323134T>A	Coding exon	Synonymous	NO	YES	2,456	0,0442	0,0437	82,2117	18	43,90243902		0	10,97560976
HLA-B	NP_005505.2:p.Ala15Gly	NM_005514.7:c.44C>G	NC_000006.11:g.31324892G>C	Coding exon	Nonsynonymous	NO	YES	7,314	39,9684	46,0736	57,8691	3	7,317073171		0	1,829268293
HLA-B	NP_005505.2:p.Ala182Thr	NM_005514.7:c.544G>A	NC_000006.11:g.31324019C>T	Coding exon	Nonsynonymous	NO	YES	11,22	0,2065	0,6119		5	12,19512195		0	3,048780488
HLA-B	NP_005505.2:p.Ala223Val	NM_005514.7:c.668C>T	NC_000006.11:g.31323321G>A	Coding exon	Nonsynonymous	NO	YES	7,797	9,7979	9,6858		32	78,04878049		0	19,51219512
HLA-B	NP_005505.2:p.Ala329Thr	NM_005514.7:c.985G>A	NC_000006.11:g.31322911C>T	Coding exon	Nonsynonymous	NO	YES	22,3	44,2739	43,5388		4	9,756097561		0	2,43902439
HLA-B	NP_005505.2:p.Ala65Thr	NM_005514.7:c.193G>A	NC_000006.11:g.31324615C>T	Coding exon	Nonsynonymous	NO	YES	13,89	9,9304	13,0787		20	48,7804878	3	7,317073171	15,85365854
HLA-B	NP_005505.2:p.Ala93Thr	NM_005514.7:c.277G>A	NC_000006.11:g.31324531C>T	Coding exon	Nonsynonymous	NO	YES	6,739	70,371	76,0648		20	48,7804878	4	9,756097561	17,07317073
HLA-B	NP_005505.2:p.Ala95Thr	NM_005514.7:c.283G>A	NC_000006.11:g.31324525C>T	Coding exon	Nonsynonymous	NO	YES	11,07	0,0393	0,0016		7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Arg103_Asn104insGlu	NM_005514.7:c.308_309insAGA	NC_000006.11:g.31324499_31324500insTCT	Coding exon	Insertion	NO	YES					16	39,02439024	1	2,43902439	10,97560976
HLA-B	NP_005505.2:p.Arg103Thrfs*49	NM_005514.7:c.306_307insAC	NC_000006.11:g.31324501_31324502insGT	Coding exon	Frame Shift	NO	YES					7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Arg106Alafs*45	NM_005514.7:c.315delG	NC_000006.11:g.31324493delC	Coding exon	Frame Shift	NO	YES	22,9	12,9825	9,353	12,9825	14	34,14634146	1	2,43902439	9,756097561
HLA-B	NP_005505.2:p.Arg106Leu	NM_005514.7:c.317G>T	NC_000006.11:g.31324491C>A	Coding exon	Nonsynonymous	NO	YES	5,414	7,4281	6,2526		17	41,46341463	13	31,70731707	26,2195122
HLA-B	NP_005505.2:p.Arg155Ser	NM_005514.7:c.463C>A	NC_000006.11:g.31324100G>T	Coding exon	Nonsynonymous	NO	YES	8,954	41,3205	44,2724		17	41,46341463	13	31,70731707	26,2195122
HLA-B	NP_005505.2:p.Arg169Leu	NM_005514.7:c.506G>T	NC_000006.11:g.31324057C>A	Coding exon	Nonsynonymous	NO	YES	25	1,9414	1,8143		28	68,29268293	6	14,63414634	24,3902439
HLA-B	NP_005505.2:p.Arg180Aspfs*35	NM_005514.7:c.537_538insGA	NC_000006.11:g.31324025_31324026insTC	Coding exon	Frame Shift	NO	YES	16,81	14,1501	14,679	17,614	2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Arg180Gln	NM_005514.7:c.539G>A	NC_000006.11:g.31324024C>T	Coding exon	Nonsynonymous	NO	YES	11,86	0,1175	0,1105		3	7,317073171		0	1,829268293
HLA-B	NP_005505.2:p.Arg180Glnfs*27	NM_005514.7:c.539_540delGG	NC_000006.11:g.31324023_31324024delCC	Coding exon	Frame Shift	NO	YES	22,6	15,8133	14,2816	15,8133	11	26,82926829	30	73,17073171	43,29268293
HLA-B	NP_005505.2:p.Arg180Gly	NM_005514.7:c.538C>G	NC_000006.11:g.31324025G>C	Coding exon	Nonsynonymous	NO	YES	4,17			0,4825	2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Arg180Leu	NM_005514.7:c.539G>T	NC_000006.11:g.31324024C>A	Coding exon	Nonsynonymous	NO	YES	7,935	50,2561	47,0553		4	9,756097561		0	2,43902439
HLA-B	NP_005505.2:p.Arg180Trp	NM_005514.7:c.538C>T	NC_000006.11:g.31324025G>A	Coding exon	Nonsynonymous	NO	YES	0,206	19,6659	17,8451	22,5899	16	39,02439024	24	58,53658537	39,02439024
HLA-B	NP_005505.2:p.Arg181Glufs*33	NM_005514.7:c.540delG	NC_000006.11:g.31324024delC	Coding exon	Frame Shift	NO	YES					22	53,65853659	14	34,14634146	30,48780488
HLA-B	NP_005505.2:p.Arg243Trp	NM_005514.7:c.727C>T	NC_000006.11:g.31323262G>A	Coding exon	Nonsynonymous	NO	YES	29,5	7,5581	0,5349	7,5581	18	43,90243902	3	7,317073171	14,63414634

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
HLA-B	NP_005505.2:p.Arg263Gly	NM_005514.7:c.787A>G	NC_000006.11:g.31323202T>C	Coding exon	Nonsynonymous	NO	YES	0,04	24,272	5,5489		18	43,90243902	6	14,63414634	18,29268293
HLA-B	NP_005505.2:p.Arg86Gly	NM_005514.7:c.256C>G	NC_000006.11:g.31324552G>C	Coding exon	Nonsynonymous	NO	YES	0,669	3,2927	2,0834	3,5043	7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Asn104_Leu105delinsMet	NM_005514.7:c.311_313delACC	NC_000006.11:g.31324495_31324497delGGT	Coding exon	Insertion/Deletion	NO	YES	8,006	14,6059	10,4071	14,6072	2	4,87804878	1	2,43902439	2,43902439
HLA-B	NP_005505.2:p.Asn104Ile	NM_005514.7:c.311A>T	NC_000006.11:g.31324497T>A	Coding exon	Nonsynonymous	NO	YES	6,927	0,0056	0,0006	76,0583	7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Asn104Serfs*46	NM_005514.7:c.311_314delACCT	NC_000006.11:g.31324494_31324497delAGGT	Coding exon	Frame Shift	NO	YES	21,5	0,0013		0,0013	11	26,82926829	28	68,29268293	40,85365854
HLA-B	NP_005505.2:p.Asn104Thr	NM_005514.7:c.311A>C	NC_000006.11:g.31324497T>G	Coding exon	Nonsynonymous	NO	YES	5,825	6,929	4,7476		8	19,51219512	33	80,48780488	45,12195122
HLA-B	NP_005505.2:p.Asn104Thrfs*34	NM_005514.7:c.311_312delAC	NC_000006.11:g.31324496_31324497delGT	Coding exon	Frame Shift	NO	YES					11	26,82926829	28	68,29268293	40,85365854
HLA-B	NP_005505.2:p.Asn104Thrfs*47	NM_005514.7:c.311delA	NC_000006.11:g.31324498delT	Coding exon	Frame Shift	NO	YES	9,509	3,2488	3,3109	3,4633	7	17,07317073	33	80,48780488	44,51219512
HLA-B	NP_005505.2:p.Asn87Asp	NM_005514.7:c.259A>G	NC_000006.11:g.31324549T>C	Coding exon	Nonsynonymous	NO	YES	1,09	33,9071	35,5134		2	4,87804878	1	2,43902439	2,43902439
HLA-B	NP_005505.2:p.Asn87Lys	NM_005514.7:c.261C>G	NC_000006.11:g.31324547G>C	Coding exon	Nonsynonymous	NO	YES	12,57	34,0051	35,61	65,432	7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Asp138Asn	NM_005514.7:c.412G>A	NC_000006.11:g.31324151C>T	Coding exon	Nonsynonymous	NO	YES	0,007	45,4023	45,9411		2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Asp138His	NM_005514.7:c.412G>C	NC_000006.11:g.31324151C>G	Coding exon	Nonsynonymous	NO	YES	0,357	3,5677	3,6749	51,0258	1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Asp201Asn	NM_005514.7:c.601G>A	NC_000006.11:g.31323962C>T	Coding exon	Nonsynonymous	NO	YES	21,1	0,0014		0,0231	32	78,04878049	1	2,43902439	20,73170732
HLA-B	NP_005505.2:p.Asp201Glu	NM_005514.7:c.603C>G	NC_000006.11:g.31323960G>C	Coding exon	Nonsynonymous	NO	YES	0,014	57,6933	68,279	40,9375	35	85,36585366	2	4,87804878	23,7804878
HLA-B	NP_005505.2:p.Asp201Lysfs*14	NM_005514.7:c.600_601insAA	NC_000006.11:g.31323962_31323963insTT	Coding exon	Frame Shift	NO	YES	24,9	0,0726	0,0183	0,0726	33	80,48780488	1	2,43902439	21,34146341
HLA-B	NP_005505.2:p.Asp54Gly	NM_005514.7:c.161A>G	NC_000006.11:g.31324647T>C	Coding exon	Nonsynonymous	NO	YES	23,6	4,7592	3,6628	4,6675	35	85,36585366	2	4,87804878	23,7804878
HLA-B	NP_005505.2:p.Asp98Tyr	NM_005514.7:c.292G>T	NC_000006.11:g.31324516C>A	Coding exon	Nonsynonymous	NO	YES	0,533	59,3112	61,8419		33	80,48780488	1	2,43902439	21,34146341
HLA-B	NP_005505.2:p.Cys349Ser	NM_005514.7:c.1046G>C	NC_000006.11:g.31322303C>G	Coding exon	Nonsynonymous	YES	YES	0,001	52,4683	52,5197	47,2634	4	9,756097561		0	2,43902439
HLA-B	NP_005505.2:p.Gln120Glyfs*32	NM_005514.7:c.357_358insGG	NC_000006.11:g.31324205_31324206insCC	Coding exon	Frame Shift	NO	YES	25,7	7,0835	8,4468	7,0835	10	24,3902439	31	75,6097561	43,90243902
HLA-B	NP_005505.2:p.Gln56Leu	NM_005514.7:c.167A>T	NC_000006.11:g.31324641T>A	Coding exon	Nonsynonymous	NO	YES	25,1	10,1749	14,6787	89,3553	9	21,95121951		0	5,487804878
HLA-B	NP_005505.2:p.Gln89Arg	NM_005514.7:c.266A>G	NC_000006.11:g.31324542T>C	Coding exon	Nonsynonymous	NO	YES	2,645	3,8034	2,6479		17	41,46341463	5	12,19512195	16,46341463
HLA-B	NP_005505.2:p.Gln94*	NM_005514.7:c.280C>T	NC_000006.11:g.31324528G>A	Coding exon	Nonsense	NO	YES	0,7	0,001			22	53,65853659	6	14,63414634	20,73170732
HLA-B	NP_005505.2:p.Gln94Argfs*4	NM_005514.7:c.281_282delAG	NC_000006.11:g.31324526_31324527delCT	Coding exon	Frame Shift	NO	YES	18,5	23,9284	3,2512	23,9405	17	41,46341463	5	12,19512195	16,46341463
HLA-B	NP_005505.2:p.Gln94Asnfs*58	NM_005514.7:c.279_280insAA	NC_000006.11:g.31324528_31324529insTT	Coding exon	Frame Shift	NO	YES	14,16	70,5354	76,1003	73,59	7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Gln94His	NM_005514.7:c.282G>C	NC_000006.11:g.31324526C>G	Coding exon	Nonsynonymous	NO	YES	9,392	0,1118	0,0125	71,3835	28	68,29268293	11	26,82926829	30,48780488
HLA-B	NP_005505.2:p.Gln94Hisfs*4	NM_005514.7:c.282_283delGG	NC_000006.11:g.31324525_31324526delCC	Coding exon	Frame Shift	NO	YES	22,8	69,6503	72,857	69,6503	7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Gln94Lys	NM_005514.7:c.280C>A	NC_000006.11:g.31324528G>T	Coding exon	Nonsynonymous	NO	YES	0,346	4,1499	3,7558		7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Gln94Pro	NM_005514.7:c.281A>C	NC_000006.11:g.31324527T>G	Coding exon	Nonsynonymous	NO	YES	9,561				20	48,7804878	3	7,317073171	15,85365854
HLA-B	NP_005505.2:p.Gln94Serfs*58	NM_005514.7:c.279_280insTC	NC_000006.11:g.31324529_31324530insAG	Coding exon	Frame Shift	NO	YES	14,16	4,0074	3,2488	4,1809	14	34,14634146	1	2,43902439	9,756097561
HLA-B	NP_005505.2:p.Glu100Val	NM_005514.7:c.299A>T	NC_000006.11:g.31324509T>A	Coding exon	Nonsynonymous	NO	YES	11,74	1,2622	1,2118	98,5299	17	41,46341463	13	31,70731707	26,2195122
HLA-B	NP_005505.2:p.Glu176Ala	NM_005514.7:c.527A>C	NC_000006.11:g.31324036T>G	Coding exon	Nonsynonymous	NO	YES	0,327	0,2216	0,1532		14	34,14634146	1	2,43902439	9,756097561
HLA-B	NP_005505.2:p.Glu176Val	NM_005514.7:c.527A>T	NC_000006.11:g.31324036T>A	Coding exon	Nonsynonymous	NO	YES	0,001	41,6419	43,5173	58,1054	1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Glu187Ala	NM_005514.7:c.560A>C	NC_000006.11:g.31324003T>G	Coding exon	Nonsynonymous	NO	YES	0,001	31,8706	32,8644	0,0339	32	78,04878049	4	9,756097561	24,3902439
HLA-B	NP_005505.2:p.Glu187Gln	NM_005514.7:c.559G>C	NC_000006.11:g.31324004C>G	Coding exon	Nonsynonymous	NO	YES	0,16	37,7301	39,6141	30,077	17	41,46341463	2	4,87804878	12,80487805
HLA-B	NP_005505.2:p.Glu187Lys	NM_005514.7:c.559G>A	NC_000006.11:g.31324004C>T	Coding exon	Nonsynonymous	NO	YES	2,834	31,973	32,8101		7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Glu187Val	NM_005514.7:c.560A>T	NC_000006.11:g.31324003T>A	Coding exon	Nonsynonymous	NO	YES	0,023	37,6498	39,5155	30,2167	2	4,87804878	1	2,43902439	2,43902439
HLA-B	NP_005505.2:p.Glu197Lys	NM_005514.7:c.589G>A	NC_000006.11:g.31323974C>T	Coding exon	Nonsynonymous	NO	YES	25,3	1,8855	1,1719		14	34,14634146	1	2,43902439	9,756097561
HLA-B	NP_005505.2:p.Glu204Gln	NM_005514.7:c.610G>C	NC_000006.11:g.31323953C>G	Coding exon	Nonsynonymous	YES	YES	0,021	65,4	72,4059	33,7911	3	7,317073171		0	1,829268293
HLA-B	NP_005505.2:p.Glu299Lys	NM_005514.7:c.895G>A	NC_000006.11:g.31323094C>T	Coding exon	Nonsynonymous	YES	YES	24,1	0,0182	0,0615		15	36,58536585	14	34,14634146	26,2195122
HLA-B	NP_005505.2:p.Glu69Ala	NM_005514.7:c.206A>C	NC_000006.11:g.31324602T>G	Coding exon	Nonsynonymous	NO	YES	5,248	0,0065	0,0015		4	9,756097561		0	2,43902439
HLA-B	NP_005505.2:p.Glu69Argfs*8	NM_005514.7:c.204delA	NC_000006.11:g.31324604delT	Coding exon	Frame Shift	NO	YES	14,13	17,5781	19,0359	19,445	15	36,58536585	14	34,14634146	26,2195122
HLA-B	NP_005505.2:p.Glu69Aspfs*30	NM_005514.7:c.206_207insC	NC_000006.11:g.31324601_31324602insG	Coding exon	Frame Shift	NO	YES	22,4	12,1956	11,5165	11,4642	15	36,58536585	4	9,756097561	14,02439024
HLA-B	NP_005505.2:p.Glu69Aspfs*30	NM_005514.7:c.206_207insT	NC_000006.11:g.31324601_31324602insA	Coding exon	Frame Shift	NO	YES	22,4	2,6037	3,1117	2,4883	27	65,85365854	3	7,317073171	20,12195122
HLA-B	NP_005505.2:p.Glu69Lys	NM_005514.7:c.205G>A	NC_000006.11:g.31324603C>T	Coding exon	Nonsynonymous	NO	YES	7,692	9,5995	15,1153	48,4824	13	31,70731707	6	14,63414634	15,24390244
HLA-B	NP_005505.2:p.Glu69Val	NM_005514.7:c.206A>T	NC_000006.11:g.31324602T>A	Coding exon	Nonsynonymous	NO	YES	6,536	0,0033	0,0007		18	43,90243902	3	7,317073171	14,63414634

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
HLA-B	NP_005505.2:p.Glu70Ala	NM_005514.7:c.209A>C	NC_000006.11:g.31324599T>G	Coding exon	Nonsynonymous	NO	YES	24,3	2,5646	2,9318		13	31,70731707		0	7,926829268
HLA-B	NP_005505.2:p.Gly107Alafs*33	NM_005514.7:c.319_320insCTCC	NC_000006.11:g.31324488_31324489insGGAG	Coding exon	Frame Shift	NO	YES	24,5	13,3328	10,7929		19	46,34146341	15	36,58536585	29,87804878
HLA-B	NP_005505.2:p.Gly107Alafs*45	NM_005514.7:c.319_320insCC	NC_000006.11:g.31324488_31324489insGG	Coding exon	Frame Shift	NO	YES	23,1	2,9093	3,419		11	26,82926829	28	68,29268293	40,85365854
HLA-B	NP_005505.2:p.Gly107Arg	NM_005514.7:c.319G>C	NC_000006.11:g.31324489C>G	Coding exon	Nonsynonymous	NO	YES	5,228	7,2419	5,8869	87,6388	2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Gly107Cys	NM_005514.7:c.319G>T	NC_000006.11:g.31324489C>A	Coding exon	Nonsynonymous	NO	YES	23,3	4,2489	4,4768		5	12,19512195	36	87,80487805	46,95121951
HLA-B	NP_005505.2:p.Gly107Ilefs*46	NM_005514.7:c.317_318insGATCG	NC_000006.11:g.31324492_31324493insATCCG	Coding exon	Frame Shift	NO	YES					40	97,56097561		0	24,3902439
HLA-B	NP_005505.2:p.His137Tyr	NM_005514.7:c.409C>T	NC_000006.11:g.31324154G>A	Coding exon	Nonsynonymous	NO	YES	0,003	27,3148	26,549		11	26,82926829	28	68,29268293	40,85365854
HLA-B	NP_005505.2:p.Ile218Val	NM_005514.7:c.652A>G	NC_000006.11:g.31323337T>C	Coding exon	Nonsynonymous	NO	YES	0,001	23,29	23,3893		1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Ile90_Tyr91delinsAsn	NM_005514.7:c.269_271delTCT	NC_000006.11:g.31324537_31324539delAGA	Coding exon	Insertion/Deletion	NO	YES	11,3	3,8699	3,2439	15,7045	16	39,02439024	15	36,58536585	28,04878049
HLA-B	NP_005505.2:p.Ile90Asn	NM_005514.7:c.269T>A	NC_000006.11:g.31324539A>T	Coding exon	Nonsynonymous	NO	YES	0,003	0,0986	0,207	99,353	1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Leu105Argfs*46	NM_005514.7:c.314delT	NC_000006.11:g.31324494delA	Coding exon	Frame Shift	NO	YES	19,97	3,244	3,2546	6,5895	3	7,317073171		0	1,829268293
HLA-B	NP_005505.2:p.Leu105del	NM_005514.7:c.314_316delTGC	NC_000006.11:g.31324493_31324495delCAG	Coding exon	Deletion	NO	YES					13	31,70731707	28	68,29268293	42,07317073
HLA-B	NP_005505.2:p.Leu105Pro	NM_005514.7:c.314T>C	NC_000006.11:g.31324494A>G	Coding exon	Nonsynonymous	NO	YES	23,2	0,0032	0,0006	28,8938	2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Leu105Profs*33	NM_005514.7:c.314_315delTG	NC_000006.11:g.31324493_31324494delCA	Coding exon	Frame Shift	NO	YES					5	12,19512195		0	3,048780488
HLA-B	NP_005505.2:p.Leu105Val	NM_005514.7:c.313C>G	NC_000006.11:g.31324495G>C	Coding exon	Nonsynonymous	NO	YES	7,54	0,0014	0,0006	1,9518	12	29,26829268	2	4,87804878	9,756097561
HLA-B	NP_005505.2:p.Leu119Arg	NM_005514.7:c.356T>G	NC_000006.11:g.31324207A>C	Coding exon	Nonsynonymous	NO	YES	23,6				20	48,7804878	16	39,02439024	31,70731707
HLA-B	NP_005505.2:p.Leu119Ile	NM_005514.7:c.355C>A	NC_000006.11:g.31324208G>T	Coding exon	Nonsynonymous	NO	YES	0,001	24,1648	25,4092		3	7,317073171		0	1,829268293
HLA-B	NP_005505.2:p.Leu119Phe	NM_005514.7:c.355C>T	NC_000006.11:g.31324208G>A	Coding exon	Nonsynonymous	NO	YES	0,382	0			17	41,46341463	22	53,65853659	37,19512195
HLA-B	NP_005505.2:p.Leu119Profs*19	NM_005514.7:c.354_355delCC	NC_000006.11:g.31324209_31324210delGG	Coding exon	Frame Shift	YES	YES	23,4	7,3031	8,6855		26	63,41463415	3	7,317073171	19,51219512
HLA-B	NP_005505.2:p.Leu119Serfs*32	NM_005514.7:c.355delC	NC_000006.11:g.31324210delG	Coding exon	Frame Shift	NO	YES					20	48,7804878	6	14,63414634	19,51219512
HLA-B	NP_005505.2:p.Leu17Val	NM_005514.7:c.49C>G	NC_000006.11:g.31324887G>C	Coding exon	Nonsynonymous	NO	YES	22,6	25,3964	30,3761	72,5647	26	63,41463415	2	4,87804878	18,29268293
HLA-B	NP_005505.2:p.Leu2Arg	NM_005514.7:c.5T>G	NC_000006.11:g.31324931A>C	Coding exon	Nonsynonymous	NO	YES	6,401	59,0995	61,9014	60,3469	13	31,70731707		0	7,926829268
HLA-B	NP_005505.2:p.Lys202Alafs*5	NM_005514.7:c.604_605delAA	NC_000006.11:g.31323958_31323959delTT	Coding exon	Frame Shift	NO	YES	23,2	0,2112	0,0185	0,2112	13	31,70731707		0	7,926829268
HLA-B	NP_005505.2:p.Lys202Thr	NM_005514.7:c.605A>C	NC_000006.11:g.31323958T>G	Coding exon	Nonsynonymous	NO	YES	0,005	74,1991	81,5323		1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Lys292Glu	NM_005514.7:c.874A>G	NC_000006.11:g.31323115T>C	Coding exon	Nonsynonymous	NO	YES	0,002	18,7265	5,9152		2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Met4Thr	NM_005514.7:c.11T>C	NC_000006.11:g.31324925A>G	Coding exon	Nonsynonymous	NO	YES	6,638	58,9501	61,9114		6	14,63414634		0	3,658536585
HLA-B	NP_005505.2:p.Ser101_Leu102insArg	NM_005514.7:c.303_304insAGA	NC_000006.11:g.31324504_31324505insTCT	Coding exon	Insertion	NO	YES					14	34,14634146		0	8,536585366
HLA-B	NP_005505.2:p.Ser101Asn	NM_005514.7:c.302G>A	NC_000006.11:g.31324506C>T	Coding exon	Nonsynonymous	NO	YES	4,204	22,1306	20,6561		24	58,53658537	1	2,43902439	15,85365854
HLA-B	NP_005505.2:p.Ser101Asnfs*51	NM_005514.7:c.301_302insAC	NC_000006.11:g.31324506_31324507insGT	Coding exon	Frame Shift	NO	YES					19	46,34146341	1	2,43902439	12,80487805
HLA-B	NP_005505.2:p.Ser101Gly	NM_005514.7:c.301A>G	NC_000006.11:g.31324507T>C	Coding exon	Nonsynonymous	NO	YES	0,918	4,5549	3,7244		18	43,90243902		0	10,97560976
HLA-B	NP_005505.2:p.Ser121Arg	NM_005514.7:c.363C>G	NC_000006.11:g.31324200G>C	Coding exon	Nonsynonymous	NO	YES	0,085	65,3352	70,9502	30,6883	5	12,19512195		0	3,048780488
HLA-B	NP_005505.2:p.Ser121Asn	NM_005514.7:c.362G>A	NC_000006.11:g.31324201C>T	Coding exon	Nonsynonymous	NO	YES	0,004	3,1026	3,2685	2,1554	21	51,2195122		0	12,80487805
HLA-B	NP_005505.2:p.Ser121Cys	NM_005514.7:c.361A>T	NC_000006.11:g.31324202T>A	Coding exon	Nonsynonymous	NO	YES	0,002	1,0155	1,4367	96,8779	2	4,87804878	1	2,43902439	2,43902439
HLA-B	NP_005505.2:p.Ser121Thr	NM_005514.7:c.362G>C	NC_000006.11:g.31324201C>G	Coding exon	Nonsynonymous	NO	YES	0,002	6,9778	7,2488	87,43	7	17,07317073		0	4,268292683
HLA-B	NP_005505.2:p.Ser14Trp	NM_005514.7:c.41C>G	NC_000006.11:g.31324895G>C	Coding exon	Nonsynonymous	NO	YES	23,7	27,5617	33,5403	70,4301	2	4,87804878	1	2,43902439	2,43902439
HLA-B	NP_005505.2:p.Ser28Phe	NM_005514.7:c.83C>T	NC_000006.11:g.31324725G>A	Coding exon	Nonsynonymous	YES	YES		0,0883	0,128		9	21,95121951		0	5,487804878
HLA-B	NP_005505.2:p.Ser35Ala	NM_005514.7:c.103T>G	NC_000006.11:g.31324705A>C	Coding exon	Nonsynonymous	NO	YES	1,503	56,4428	63,6734		18	43,90243902	4	9,756097561	15,85365854
HLA-B	NP_005505.2:p.Ser48Ala	NM_005514.7:c.142T>G	NC_000006.11:g.31324666A>C	Coding exon	Nonsynonymous	NO	YES	0,21	46,7136	42,8675		24	58,53658537	7	17,07317073	23,17073171
HLA-B	NP_005505.2:p.Ser48Thr	NM_005514.7:c.142T>A	NC_000006.11:g.31324666A>T	Coding exon	Nonsynonymous	NO	YES	0,34	12,4078	16,3262	40,4096	20	48,7804878	2	4,87804878	14,63414634
HLA-B	NP_005505.2:p.Thr118Ile	NM_005514.7:c.353C>T	NC_000006.11:g.31324210G>A	Coding exon	Nonsynonymous	YES	YES	17,54	21,3299	23,0229		1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Thr162Lys	NM_005514.7:c.485C>A	NC_000006.11:g.31324078G>T	Coding exon	Nonsynonymous	NO	YES	13,4	6,451	4,1867		19	46,34146341	12	29,26829268	26,2195122
HLA-B	NP_005505.2:p.Trp191Ser	NM_005514.7:c.572G>C	NC_000006.11:g.31323991C>G	Coding exon	Nonsynonymous	NO	YES	8,923	9,7552	8,0324	90,2448	18	43,90243902	19	46,34146341	34,14634146
HLA-B	NP_005505.2:p.Tyr123Phe	NM_005514.7:c.368A>T	NC_000006.11:g.31324195T>A	Coding exon	Nonsynonymous	NO	YES	0,886	0,3753	0,4723	99,5325	23	56,09756098	4	9,756097561	18,90243902
HLA-B	NP_005505.2:p.Tyr123Ser	NM_005514.7:c.368A>C	NC_000006.11:g.31324195T>G	Coding exon	Nonsynonymous	NO	YES	14,75	0,091	0,4551	0,0011	2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Tyr140*	NM_005514.7:c.420C>A	NC_000006.11:g.31324143G>T	Coding exon	Nonsense	NO	YES	29,5	6,9791	5,5843		1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Tyr140Asp	NM_005514.7:c.418T>G	NC_000006.11:g.31324145A>C	Coding exon	Nonsynonymous	NO	YES	0,001	14,0091	12,3637	14,0091	22	53,65853659	6	14,63414634	20,73170732

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
HLA-B	NP_005505.2:p.Tyr140Phe	NM_005514.7:c.419A>T	NC_000006.11:g.31324144T>A	Coding exon	Nonsynonymous	NO	YES	0,001	20,1943	19,0754	20,3351	13	31,70731707	2	4,87804878	10,36585366
HLA-B	NP_005505.2:p.Tyr140Ser	NM_005514.7:c.419A>C	NC_000006.11:g.31324144T>G	Coding exon	Nonsynonymous	NO	YES	0,001	19,2367	22,2731	19,5806	1	2,43902439	1	2,43902439	1,829268293
HLA-B	NP_005505.2:p.Tyr195His	NM_005514.7:c.583T>C	NC_000006.11:g.31323980A>G	Coding exon	Nonsynonymous	NO	YES	4,606	7,0066	7,5071		1	2,43902439	1	2,43902439	1,829268293
HLA-B	NP_005505.2:p.Tyr33Asp	NM_005514.7:c.97T>G	NC_000006.11:g.31324711A>C	Coding exon	Nonsynonymous	NO	YES	0,01	4,63	5,8449		8	19,51219512		0	4,87804878
HLA-B	NP_005505.2:p.Tyr33His	NM_005514.7:c.97T>C	NC_000006.11:g.31324711A>G	Coding exon	Nonsynonymous	NO	YES	0,027	16,8116	17,1135		20	48,7804878	2	4,87804878	14,63414634
HLA-B	NP_005505.2:p.Tyr91*	NM_005514.7:c.273C>G	NC_000006.11:g.31324535G>C	Coding exon	Nonsense	NO	YES	35	0,001	0,0004	99,602	3	7,317073171		0	1,829268293
HLA-B	NP_005505.2:p.Tyr91_Lys92insMet	NM_005514.7:c.274_275insTGA	NC_000006.11:g.31324534_31324535insCAT	Coding exon	Insertion	NO	YES	7,12	3,8577	2,7891		15	36,58536585	3	7,317073171	12,80487805
HLA-B	NP_005505.2:p.Tyr91Asn	NM_005514.7:c.271T>A	NC_000006.11:g.31324537A>T	Coding exon	Nonsynonymous	NO	YES	2,907	0,0011	0,0038	99,5942	2	4,87804878		0	1,219512195
HLA-B	NP_005505.2:p.Tyr91Cys	NM_005514.7:c.272A>G	NC_000006.11:g.31324536T>C	Coding exon	Nonsynonymous	NO	YES	6,514	14,551	15,5791	36,6813	13	31,70731707	2	4,87804878	10,36585366
HLA-B	NP_005505.2:p.Tyr91Phe	NM_005514.7:c.272A>T	NC_000006.11:g.31324536T>A	Coding exon	Nonsynonymous	NO	YES	8,641	29,4641	29,7045	18,8898	1	2,43902439		0	0,609756098
HLA-B	NP_005505.2:p.Tyr91Ser	NM_005514.7:c.272A>C	NC_000006.11:g.31324536T>G	Coding exon	Nonsynonymous	NO	YES	8,067	31,3427	34,5332	9,8243	13	31,70731707	2	4,87804878	10,36585366
HLA-B	NP_005505.2:p.Val127Leu	NM_005514.7:c.379G>C	NC_000006.11:g.31324184C>G	Coding exon	Nonsynonymous	NO	YES	0,011	4,683	10,0588	94,1052	9	21,95121951	6	14,63414634	12,80487805
HLA-B	NP_005505.2:p.Val272Met	NM_005514.7:c.814G>A	NC_000006.11:g.31323175C>T	Coding exon	Nonsynonymous	NO	YES	25,7	0,0058	0,0011		9	21,95121951	6	14,63414634	12,80487805
HLA-B	NP_005505.2:p.Val306Ile	NM_005514.7:c.916G>A	NC_000006.11:g.31322980C>T	Coding exon	Nonsynonymous	NO	YES	0,039	44,7249	44,0241		6	14,63414634		0	3,658536585
HLA-B	NP_005505.2:p.Val36Met	NM_005514.7:c.106G>A	NC_000006.11:g.31324702C>T	Coding exon	Nonsynonymous	NO	YES	17,12	49,5751	57,0086		17	41,46341463	5	12,19512195	16,46341463
HLA-B	NP_005505.2:p.Val9Leu	NM_005514.7:c.25G>C	NC_000006.11:g.31324911C>G	Coding exon	Nonsynonymous	NO	YES	9,781	7,8903	9,3882	91,0713	3	7,317073171	38	92,68292683	48,17073171
HLA-B		NM_005514.7:c.*4+27_*4+34delTGGGGTGG	NC_000006.11:g.31322224_31322231delACCCCACC	Intron		NO	YES	3,004	4,2482	1,3615	4,4417	1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.*4+27delT	NC_000006.11:g.31322229delA	Intron		NO	YES	0,02	0,0011	0,0886	0,0011	5	12,19512195		0	3,048780488
HLA-B		NM_005514.7:c.*4+27T>C	NC_000006.11:g.31322229A>G	Intron		NO	YES	0,159	19,4564	20,1396		12	29,26829268	1	2,43902439	8,536585366
HLA-B		NM_005514.7:c.*4+32_*4+35delTGGC	NC_000006.11:g.31322221_31322224delGCCA	Intron		NO	YES	3,238	14,2031	16,9559	20,8299	6	14,63414634		0	3,658536585
HLA-B		NM_005514.7:c.*4+32_*4+42delTGGCGGGTCTG	NC_000006.11:g.31322215_31322225delAGACCCGCCAC	Intron		NO	YES	3,12				17	41,46341463	5	12,19512195	16,46341463
HLA-B		NM_005514.7:c.*4+32_*4+43delTGGCGGGTCTGG	NC_000006.11:g.31322215_31322226delAGACCCGCCACC	Intron		NO	YES					6	14,63414634	1	2,43902439	4,87804878
HLA-B		NM_005514.7:c.*4+32delT	NC_000006.11:g.31322224delA	Intron		NO	YES	4,482	0,0945	0,5779	0,0945	7	17,07317073		0	4,268292683
HLA-B		NM_005514.7:c.*4+32T>G	NC_000006.11:g.31322224A>C	Intron		NO	YES	3,848	8,0337	14,5852	8,3692	14	34,14634146	25	60,97560976	39,02439024
HLA-B		NM_005514.7:c.*4+35delC	NC_000006.11:g.31322221delG	Intron		NO	YES	3,299	0,1394	2,2648		2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.*4+36G>A	NC_000006.11:g.31322220C>T	Intron		NO	YES	4,246	29,639	17,6763		2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.*4+39_*4+41delTCT	NC_000006.11:g.31322215_31322217delAGA	Intron		NO	YES	4,979	10,378	9,0655		10	24,3902439	2	4,87804878	8,536585366
HLA-B		NM_005514.7:c.*4+39T>G	NC_000006.11:g.31322217A>C	Intron		NO	YES	0,744	11,7196			11	26,82926829	1	2,43902439	7,926829268
HLA-B		NM_005514.7:c.*4+40_*4+42delCTG	NC_000006.11:g.31322214_31322216delCAG	Intron		NO	YES	4,552	0,0062	0,2049		2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.*4+41_*4+42delTG	NC_000006.11:g.31322214_31322215delCA	Intron		NO	YES	4,577	10,0843	3,0674		9	21,95121951		0	5,487804878
HLA-B		NM_005514.7:c.*4+41T>G	NC_000006.11:g.31322215A>C	Intron		NO	YES	1,314	0,0168	0,2244		1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.*4+45G>A	NC_000006.11:g.31322211C>T	Intron		NO	YES	3,632	0,0081	0,0014		9	21,95121951	32	78,04878049	44,51219512
HLA-B		NM_005514.7:c.1012+29G>A	NC_000006.11:g.31322855C>T	Intron		NO	YES	0,921	7,5948	7,4886		19	46,34146341	3	7,317073171	15,24390244
HLA-B		NM_005514.7:c.1013-17A>G	NC_000006.11:g.31322459T>C	Intron		NO	YES	15,15	26,5818	26,5702		10	24,3902439		0	6,097560976
HLA-B		NM_005514.7:c.1013-28G>C	NC_000006.11:g.31322470C>G	Intron		NO	YES	0,535	84,6565	84,7442	15,2418	4	9,756097561		0	2,43902439
HLA-B		NM_005514.7:c.1013-32C>T	NC_000006.11:g.31322474G>A	Intron		NO	YES	2,029	0,1195	0,1253	0,1171	1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.1013-45C>T	NC_000006.11:g.31322487G>A	Intron		NO	YES	1,101	10,2244	10,1143		2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.1045+15T>C	NC_000006.11:g.31322395A>G	Intron		NO	YES	7,58	9,9476	9,7782	9,8715	1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.1045+43A>C	NC_000006.11:g.31322367T>G	Intron		NO	YES	4,843	7,526	7,5566		12	29,26829268	1	2,43902439	8,536585366
HLA-B		NM_005514.7:c.1045+8G>A	NC_000006.11:g.31322402C>T	Intron		YES	YES	10,81	26,6203	26,5963		5	12,19512195		0	3,048780488
HLA-B		NM_005514.7:c.1046-37C>A	NC_000006.11:g.31322340G>T	Intron		NO	YES	0,447	3,3586	3,3758		3	7,317073171	38	92,68292683	48,17073171
HLA-B		NM_005514.7:c.1046-37C>T	NC_000006.11:g.31322340G>A	Intron		NO	YES	1,072	7,7087	8,443		2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.-18G>A	NC_000006.11:g.31324953C>T	UTR		NO	YES	2,871	59,8493	60,2889	60,9438	3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.-20G>A	NC_000006.11:g.31324955C>T	UTR		NO	YES	9,762	7,4508	5,791	7,673	7	17,07317073		0	4,268292683
HLA-B		NM_005514.7:c.343+17C>T	NC_000006.11:g.31324448G>A	Intron		NO	YES	8,14	9,2551	8,4386		3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.343+50T>G	NC_000006.11:g.31324415A>C	Intron		NO	YES	5,211	15,2245	15,1806		2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.344-10C>G	NC_000006.11:g.31324229G>C	Intron		YES	YES	4,958	8,7411	10,4829	91,2589	21	51,2195122	14	34,14634146	29,87804878

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
HLA-B		NM_005514.7:c.344-16G>A	NC_000006.11:g.31324235C>T	Intron		NO	YES	9,304	1,9563	1,8634	1,9563	20	48,7804878	2	4,87804878	14,63414634
HLA-B		NM_005514.7:c.344-24G>T	NC_000006.11:g.31324243C>A	Intron		NO	YES	9,475	4,0708	6,0333		11	26,82926829	28	68,29268293	40,85365854
HLA-B		NM_005514.7:c.344-26delT	NC_000006.11:g.31324245delA	Intron		NO	YES	7,813	0,0316	0,0059	0,0316	11	26,82926829	28	68,29268293	40,85365854
HLA-B		NM_005514.7:c.344-26T>G	NC_000006.11:g.31324245A>C	Intron		NO	YES	2,187	70,8559	79,3142	72,3828	12	29,26829268	1	2,43902439	8,536585366
HLA-B		NM_005514.7:c.344-29_344-28insG	NC_000006.11:g.31324252_31324253insC	Intron		NO	YES	5,185	29,8479	30,6508	29,8507	4	9,756097561		0	2,43902439
HLA-B		NM_005514.7:c.344-36_344-35insGGGGC	NC_000006.11:g.31324270_31324271insCCCCG	Intron		NO	YES	2,12	0,0453	1,3438	0,0453	8	19,51219512		0	4,87804878
HLA-B		NM_005514.7:c.344-42_344-41insGGGGG	NC_000006.11:g.31324264_31324265insCCCCC	Intron		NO	YES					3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.344-46_344-45insTGGGC	NC_000006.11:g.31324268_31324269insAGCCC	Intron		NO	YES					5	12,19512195		0	3,048780488
HLA-B		NM_005514.7:c.344-47_344-46insGGGGG	NC_000006.11:g.31324269_31324270insCCCCC	Intron		NO	YES					5	12,19512195		0	3,048780488
HLA-B		NM_005514.7:c.344-48_344-47insTCGGG	NC_000006.11:g.31324273_31324274insCGACC	Intron		NO	YES					3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.344-8G>T	NC_000006.11:g.31324227C>A	Intron		YES	YES	9,536	16,2335	12,8601	16,2029	2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.-3G>A	NC_000006.11:g.31324938C>T	UTR		NO	YES	3,754	4,574	3,95	4,4083	2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.620-40A>G	NC_000006.11:g.31323409T>C	Intron		NO	YES	2,821	84,6702	84,7984		5	12,19512195		0	3,048780488
HLA-B		NM_005514.7:c.620-43T>G	NC_000006.11:g.31323412A>C	Intron		NO	YES	9,442				4	9,756097561		0	2,43902439
HLA-B		NM_005514.7:c.620-45C>T	NC_000006.11:g.31323414G>A	Intron		NO	YES	3,456	2,5757	2,4822		1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.620-47C>G	NC_000006.11:g.31323416G>C	Intron		NO	YES	0,817	9,1626	9,3164	90,9516	7	17,07317073	34	82,92682927	45,73170732
HLA-B		NM_005514.7:c.-6G>A	NC_000006.11:g.31324941C>T	UTR		NO	YES	7,363	1,8921	1,7678	1,9372	2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.73+11_73+12insA	NC_000006.11:g.31324851_31324852insT	Intron		NO	YES	8,95	1,4629	1,6387		3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.73+11_73+12insG	NC_000006.11:g.31324854_31324855insC	Intron		NO	YES	8,95	37,1184	42,2209		2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.73+16G>C	NC_000006.11:g.31324847C>G	Intron		NO	YES	9,718	25,384	27,2645	26,0563	2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.73+33C>T	NC_000006.11:g.31324830G>A	Intron		NO	YES	7,984	61,7311	65,8285	63,1271	4	9,756097561		0	2,43902439
HLA-B		NM_005514.7:c.73+34C>G	NC_000006.11:g.31324829G>C	Intron		NO	YES	7,021	61,0417	64,3156	62,1829	2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.73+43C>A	NC_000006.11:g.31324820G>T	Intron		NO	YES	3,93	3,9893	4,092	4,3354	21	51,2195122		0	12,80487805
HLA-B		NM_005514.7:c.74-10_74-9insTG	NC_000006.11:g.31324743_31324744insCA	Intron		YES	YES	8,688	3,7915	2,9408	3,8248	40	97,56097561		0	24,3902439
HLA-B		NM_005514.7:c.74-15C>A	NC_000006.11:g.31324749G>T	Intron		NO	YES	8,48	6,4129	1,2942	6,4129	38	92,68292683		0	23,17073171
HLA-B		NM_005514.7:c.74-16C>T	NC_000006.11:g.31324750G>A	Intron		NO	YES	11,98	1,31	2,9103		9	21,95121951	2	4,87804878	7,926829268
HLA-B		NM_005514.7:c.74-22C>T	NC_000006.11:g.31324756G>A	Intron		NO	YES	13,91	4,2382	3,4719		3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.74-30G>T	NC_000006.11:g.31324764C>A	Intron		NO	YES	14,05	3,5534	3,3596		1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.74-3delC	NC_000006.11:g.31324741delG	Intron		YES	YES	9,822			2,5959	26	63,41463415		0	15,85365854
HLA-B		NM_005514.7:c.74-42G>T	NC_000006.11:g.31324776C>A	Intron		NO	YES	9,736	0,7512	0,1388	0,7512	2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.74-7C>G	NC_000006.11:g.31324741G>C	Intron		YES	YES	15,47				1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.74-7C>T	NC_000006.11:g.31324741G>A	Intron		YES	YES	8,956	0,9247	0,1403	0,9247	5	12,19512195		0	3,048780488
HLA-B		NM_005514.7:c.74-8_74-6delACC	NC_000006.11:g.31324741_31324743delGTG	Intron		YES	YES	7,955	3,6015	2,8825	3,6015	1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.74-8_74-7delAC	NC_000006.11:g.31324742_31324743delTG	Intron		YES	YES					8	19,51219512		0	4,87804878
HLA-B		NM_005514.7:c.74-8A>G	NC_000006.11:g.31324742T>C	Intron		YES	YES	6,55	81,8712	84,3621	82,3537	1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.74-8A>T	NC_000006.11:g.31324742T>A	Intron		YES	YES	11,09	0,0177	0,0092	0,0166	16	39,02439024	2	4,87804878	12,19512195
HLA-B		NM_005514.7:c.74-8delA	NC_000006.11:g.31324742delT	Intron		YES	YES	6,414				20	48,7804878	2	4,87804878	14,63414634
HLA-B		NM_005514.7:c.74-9_74-7delCACinsTG	NC_000006.11:g.31324741_31324743delGTGinsCA	Intron		YES	YES					3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.74-9C>G	NC_000006.11:g.31324743G>C	Intron		YES	YES	7,618	0,8695	0,8713	98,789	2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.74-9C>T	NC_000006.11:g.31324743G>A	Intron		YES	YES	9,199	0,0014			1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.74-9delC	NC_000006.11:g.31324743delG	Intron		YES	YES	8,447				33	80,48780488		0	20,12195122
HLA-B		NM_005514.7:c.895+22C>G	NC_000006.11:g.31323072G>C	Intron		NO	YES	2,2	0,0151	0,093	0,0151	6	14,63414634		0	3,658536585
HLA-B		NM_005514.7:c.895+25A>G	NC_000006.11:g.31323069T>C	Intron		NO	YES	5,464	0,6167	0,4186	0,6167	19	46,34146341	13	31,70731707	27,43902439
HLA-B		NM_005514.7:c.895+27C>G	NC_000006.11:g.31323067G>C	Intron		NO	YES	5,848	0,6385	0,4195	0,6385	2	4,87804878		0	1,219512195

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
HLA-B		NM_005514.7:c.895+29C>G	NC_000006.11:g.31323065G>C	Intron		NO	YES	6,588	14,5771	13,9235	85,2636		0	1	2,43902439	1,219512195
HLA-B		NM_005514.7:c.895+44_895+45insTGAGCCCTTCT	NC_000006.11:g.31323049_31323050insAGAAGGGCTCA	Intron		NO	YES					1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.895+44_895+45insTGATCCCTTCT	NC_000006.11:g.31323049_31323050insAGAAGGGATCA	Intron		NO	YES					1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.895+45_895+46insAAGTCCTG	NC_000006.11:g.31323049_31323050insAGGACTTC	Intron		NO	YES	14,66	0,0643	0,0911	0,0643	13	31,70731707	1	2,43902439	9,146341463
HLA-B		NM_005514.7:c.895+45_895+46insCAGCCCTTCTG	NC_000006.11:g.31323049_31323050insAGAAGGGCTGC	Intron		NO	YES					6	14,63414634		0	3,658536585
HLA-B		NM_005514.7:c.895+45_895+46insTAGCCCTGCTG	NC_000006.11:g.31323049_31323050insAGCAGGGCTAC	Intron		NO	YES					1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.895+45_895+46insTAGCCCTTCTG	NC_000006.11:g.31323049_31323050insAGAAGGGCTAC	Intron		NO	YES					24	58,53658537	12	29,26829268	29,26829268
HLA-B		NM_005514.7:c.895+46_895+47insCGCCCTTCTGG	NC_000006.11:g.31323049_31323050insAGAAGGGCGCC	Intron		NO	YES					19	46,34146341	9	21,95121951	22,56097561
HLA-B		NM_005514.7:c.895+46G>A	NC_000006.11:g.31323048C>T	Intron		NO	YES	15,28			0,85	16	39,02439024		0	9,756097561
HLA-B		NM_005514.7:c.895+47_896-46insTCCCTTCTGGA	NC_000006.11:g.31323049_31323050insAGAAGGGATCC	Intron		NO	YES			0,0004		3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.896-12C>T	NC_000006.11:g.31323012G>A	Intron		NO	YES	7,537	17,7809	17,903		1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.896-20A>G	NC_000006.11:g.31323020T>C	Intron		NO	YES	11,09	25,899	26,3527		1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.896-26_896-25insTGAGGCTTGGAGGTCAGGGC	NC_000006.11:g.31323034_31323035insTCCAAGCCTCAGCCCTGACC	Intron		NO	YES					2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.896-27G>A	NC_000006.11:g.31323027C>T	Intron		NO	YES	12,6	1,3617	1,2155		1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.896-35G>A	NC_000006.11:g.31323035C>T	Intron		NO	YES	15,39					0	2	4,87804878	2,43902439
HLA-B		NM_005514.7:c.896-36A>C	NC_000006.11:g.31323036T>G	Intron		NO	YES	15,31		0,0019		1	2,43902439	1	2,43902439	1,829268293
HLA-B		NM_005514.7:c.896-36A>T	NC_000006.11:g.31323036T>A	Intron		NO	YES	16,39				2	4,87804878		0	1,219512195
HLA-B		NM_005514.7:c.896-40_896-39insTGGAGCCCTTC	NC_000006.11:g.31323049_31323050insAGAAGGGCTCC	Intron		NO	YES	12,94	48,4352	48,1478	48,9402	1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.896-43_896-42insGTCTGGAGCCC	NC_000006.11:g.31323049_31323050insAGACGGGCTCC	Intron		NO	YES					1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.896-44_896-43insATTCTGGAGCC	NC_000006.11:g.31323049_31323050insAGAATGGCTCC	Intron		NO	YES					1	2,43902439		0	0,609756098
HLA-B		NM_005514.7:c.896-45_896-44insACTTCTGGAGC	NC_000006.11:g.31323049_31323050insAGAAGTGCTCC	Intron		NO	YES					3	7,317073171		0	1,829268293
HLA-B		NM_005514.7:c.896-46_896-45insACCTTCTGGAG	NC_000006.11:g.31323049_31323050insAGAAGGTCTCC	Intron		NO	YES					16	39,02439024	1	2,43902439	10,97560976
NUDT15		NM_018283.3:c.*7G>A	NC_000013.10:g.48619942G>A	UTR		NO	NO	0,33	6,624	6,7361	6,4937	29	70,73170732		0	17,68292683
NUDT15		NM_018283.3:c.158+52_158+53insGGGGCGTGCGCAGAGGGACGATCTC	NC_000013.10:g.48612092_48612093insGGGGCGTGCGCAGAGGGACGATCTC	Intron		NO	NO	1,513	4,101	4,2305	4,1031	12	29,26829268		0	7,317073171
SLCO1B1	NP_006437.3:p.(Leu191=)	NM_006446.4:c.571T>C	NC_000012.11:g.21331599T>C	Coding exon	Synonymous	NO	NO	0,006	52,6046	52,195	51,9758	1	2,43902439		0	0,609756098
SLCO1B1	NP_006437.3:p.(Phe199=)	NM_006446.4:c.597C>T	NC_000012.11:g.21331625C>T	Coding exon	Synonymous	NO	NO	12,16	38,5138	38,9939	38,6343	20	48,7804878	6	14,63414634	19,51219512
SLCO1B1	NP_006437.3:p.(Ser137=)	NM_006446.4:c.411G>A	NC_000012.11:g.21329761G>A	Coding exon	Synonymous	NO	NO	6,028	11,2778	11,0057	11,0351	10	24,3902439	26	63,41463415	37,80487805
SLCO1B1	NP_006437.3:p.Asn130Asp	NM_006446.4:c.388A>G	NC_000012.11:g.21329738A>G	Coding exon	Nonsynonymous	NO	YES	0,002	47,9486	47,9938		8	19,51219512		0	4,87804878
SLCO1B1	NP_006437.3:p.Leu643Phe	NM_006446.4:c.1929A>C	NC_000012.11:g.21391976A>C	Coding exon	Nonsynonymous	NO	NO	3,415	4,6322	4,5844	4,6241	20	48,7804878	12	29,26829268	26,82926829
SLCO1B1	NP_006437.3:p.Pro155Thr	NM_006446.4:c.463C>A	NC_000012.11:g.21329813C>A	Coding exon	Nonsynonymous	NO	YES	2,73	11,6632	11,3856	11,4573	12	29,26829268	21	51,2195122	32,92682927
SLCO1B1	NP_006437.3:p.Val174Ala	NM_006446.4:c.521T>C	NC_000012.11:g.21331549T>C	Coding exon	Nonsynonymous	NO	YES	22,9	12,9434	13,3191	12,7777	3	7,317073171		0	1,829268293
SLCO1B1		NM_006446.4:c.1682+7A>C	NC_000012.11:g.21370244A>C	Intron		YES	NO	13,09				5	12,19512195		0	3,048780488
SLCO1B1		NM_006446.4:c.1747+26_1747+38delAAAAAAAAATATA	NC_000012.11:g.21375324_21375336delAAAAAAAAATATA	Intron		NO	NO					1	2,43902439		0	0,609756098
SLCO1B1		NM_006446.4:c.1747+33A>T	NC_000012.11:g.21375331A>T	Intron		NO	NO	3,677				12	29,26829268	21	51,2195122	32,92682927
SLCO1B1		NM_006446.4:c.1747+34_1747+42delATATATATA	NC_000012.11:g.21375332_21375340delATATATATA	Intron		NO	NO					29	70,73170732	6	14,63414634	25
SLCO1B1		NM_006446.4:c.1747+35_1747+37delTAT	NC_000012.11:g.21375333_21375335delTAT	Intron		NO	NO	9,537		0,0977		29	70,73170732		0	17,68292683
SLCO1B1		NM_006446.4:c.1747+35_1747+39delTATAT	NC_000012.11:g.21375333_21375337delTATAT	Intron		NO	NO	9,339		0,014		1	2,43902439		0	0,609756098
SLCO1B1		NM_006446.4:c.1747+35T>A	NC_000012.11:g.21375333T>A	Intron		NO	NO	8,75	14,5833	12,3148	14,8936	2	4,87804878		0	1,219512195
SLCO1B1		NM_006446.4:c.1747+39T>A	NC_000012.11:g.21375337T>A	Intron		NO	NO	7,347	0,2865	2,3016	0,2865	17	41,46341463	21	51,2195122	35,97560976

Table 3 *(cont.).* Genetic variants identified, frequency in the study population and other associated data

GENE	PROTEIN_NAME	CDNA_NAME	CHROMOSOMIC_NAME	GENE_ZONE	PROTEIN_TYPE	SPLICING_REGION	IN_DATASET	CADD_PHRED	EXAC_FREQ	GNOMAD_FREQ	DBSNP_FREQ	NUMBER_HETEROZYGOTES	HETEROZYGOTES_FREQ	NUMBER_HOMOZYGOTES	HOMOZYGOTES_FREQ	ALLELE_FREQ
SLCO1B1		NM_006446.4:c.1747+41T>A	NC_000012.11:g.21375339T>A	Intron		NO	NO	3,377		0,5593		17	41,46341463	23	56,09756098	38,41463415
SLCO1B1		NM_006446.4:c.1747+43T>A	NC_000012.11:g.21375341T>A	Intron		NO	NO	4,076		0,2046		16	39,02439024	21	51,2195122	35,36585366
SLCO1B1		NM_006446.4:c.1747+9A>G	NC_000012.11:g.21375307A>G	Intron		YES	NO	11,87	5,2566	10,1819	4,5991	20	48,7804878	7	17,07317073	20,73170732
SLCO1B1		NM_006446.4:c.1865+4846T>C	NC_000012.11:g.21382619T>C	Intron		NO	NO	1,565		21,0258	21,9249	17	41,46341463	2	4,87804878	12,80487805
SLCO1B1		NM_006446.4:c.359+23_359+24insA	NC_000012.11:g.21327666_21327667insA	Intron		NO	NO	9,211	40,0171	42,1978	5,758	17	41,46341463	2	4,87804878	12,80487805
SLCO1B1		NM_006446.4:c.359+23_359+24insAA	NC_000012.11:g.21327666_21327667insAA	Intron		NO	NO	9,108	9,0865	9,2002	44,9076	1	2,43902439		0	0,609756098
SLCO1B1		NM_006446.4:c.481+1G>T	NC_000012.11:g.21329832G>T	Intron		YES	NO	22,7	0,2889	0,2997	0,314	3	7,317073171		0	1,829268293
SLCO1B1		NM_006446.4:c.727+33C>T	NC_000012.11:g.21331987C>T	Intron		NO	NO	2,492	40,6972	40,4735	40,2466	2	4,87804878		0	1,219512195
SLCO1B1		NM_006446.4:c.-910G>A	NC_000012.11:g.21283322G>A	Intron		NO	YES			6,3214	5,4713	14	34,14634146	1	2,43902439	9,756097561
TPMT	NP_000358.1:p.(Ile158=)	NM_000367.3:c.474C>T	NC_000006.11:g.18139214G>A	Coding exon	Synonymous	NO	NO	14,07	76,3337	76,3961	76,2927	22	53,65853659	14	34,14634146	30,48780488
TPMT	NP_000358.1:p.Ala154Thr	NM_000367.3:c.460G>A	NC_000006.11:g.18139228C>T	Coding exon	Nonsynonymous	NO	YES	28,4	2,7492	2,7671						
TPMT	NP_000358.1:p.Ala80Pro	NM_000367.3:c.238G>C	NC_000006.11:g.18143955C>G	Coding exon	Nonsynonymous	YES	YES	29,5	0,1381	0,1685	99,8586					
TPMT	NP_000358.1:p.Tyr240Cys	NM_000367.3:c.719A>G	NC_000006.11:g.18130918T>C	Coding exon	Nonsynonymous	NO	YES	28,3	3,6689	3,7185						
TPMT		NM_000367.3:c.141-10delT	NC_000006.11:g.18148166delA	Intron		YES	NO	0,451	40,6003	19,7605	0,138					
TPMT		NM_000367.3:c.233+35C>T	NC_000006.11:g.18148019G>A	Intron		NO	NO	4,774	52,0288	52,5022	52,0839					
TPMT		NM_000367.3:c.367-17delT	NC_000006.11:g.18139973delA	Intron		NO	NO	3,202	65,2649	58,4255	0,0057					
TPMT		NM_000367.3:c.367-25T>A	NC_000006.11:g.18139973A>T	Intron		NO	NO	0,019	1,3037	1,1009						
TPMT		NM_000367.3:c.367-27_367-26delAA	NC_000006.11:g.18139984_18139985delTT	Intron		NO	NO	1,777	5,3655	5,6909	5,4753					
TPMT		NM_000367.3:c.580+14delG	NC_000006.11:g.18134023delC	Intron		NO	NO	0,167	1,266	0,1178	1,266					
TPMT		NM_000367.3:c.580+14G>T	NC_000006.11:g.18134021C>A	Intron		NO	NO	1,345	61,1539	66,4879	61,0312					
TPMT		NM_000367.3:c.580+26_580+27insT	NC_000006.11:g.18134020_18134021insA	Intron		NO	NO	0,788	51,2875	55,8348	41,7252					
TPMT		NM_000367.3:c.580+26_580+27insTT	NC_000006.11:g.18134020_18134021insAA	Intron		NO	NO	0,726	6,3537	6,3233	51,3512					
UGT1A1	NP_000454.1:p.His203_Lys211delinsGln	NM_000463.2:c.609_632del	NC_000002.11:g.234669542_234669565del	Coding exon	Insertion/Deletion	NO	NO									
UGT1A1	NP_000454.1:p.Thr168Ala	NM_000463.2:c.502A>G	NC_000002.11:g.234669435A>G	Coding exon	Nonsynonymous	NO	NO	12,11	0,0008	0,0014	0,0008					
UGT1A1		NM_000463.2:c.*211T>C	NC_000002.11:g.234681416T>C	UTR		NO	NO	0,737		74,7718	75,2396					
UGT1A1		NM_000463.2:c.*339G>C	NC_000002.11:g.234681544G>C	UTR		NO	NO	0,051		81,2089	82,1086					
UGT1A1		NM_000463.2:c.*440G>C	NC_000002.11:g.234681645G>C	UTR		NO	NO	1,174		73,3231	74,5008					
UGT1A1		NM_000463.2:c.-1352A>C	NC_000002.11:g.234667582A>C	Intron		NO	NO	2,587		51,6751						
UGT1A1		NM_000463.2:c.-2951A>G	NC_000002.11:g.234665983A>G	Intron		NO	NO	5,241		35,4615						
UGT1A1		NM_000463.2:c.-3152G>A	NC_000002.11:g.234665782G>A	Intron		NO	YES			29,971	30,2117					
UGT1A1		NM_000463.2:c.-3275T>G	NC_000002.11:g.234665659T>G	Intron		NO	YES			54,8473						
UGT1A1		NM_000463.2:c.-364C>T	NC_000002.11:g.234668570C>T	Intron		NO	NO	4,544		36,3619						
UGT1A1		NM_000463.2:c.-40_-39insTA	NC_000002.11:g.234668894_234668895insTA	Intron		NO	YES	6,723		34,6576	32,528					
UGT1A1		NM_000463.2:c.-41_-40delTA	NC_000002.11:g.234668893_234668894delTA	Intron		NO	YES	7,661		2,2006						
UGT1A1		NM_000463.2:c.996+18C>T	NC_000002.11:g.234675829C>T	Intron		NO	NO	5,081	1,1561	1,2209	1,2791					
UGT1A1		NM_000463.2:c.997-37T>C	NC_000002.11:g.234676458T>C	Intron		NO	NO	5,189	3,4911	3,7776	3,4873					
VKORC1	NP_076869.1:p.(Arg12=)	NM_024006.5:c.36G>A	NC_000016.9:g.31106015C>T	Coding exon	Synonymous	NO	NO	15,25	1,7664	1,5078	1,7148					
VKORC1	NP_076869.1:p.(Leu120=)	NM_024006.5:c.358C>T	NC_000016.9:g.31102589G>A	Coding exon	Synonymous	NO	YES	11,89	1,9094	1,9988	2,0719					
VKORC1		NM_001311311.1:c.284-6_284-5insT	NC_000016.9:g.31104201_31104202insA	Intron		YES	NO	6,408	14,9733	16,1791	31,3233					
VKORC1		NM_024006.5:c.-1639G>A	NC_000016.9:g.31107689C>T	Intron		NO	YES			32,5975	35,5631					
VKORC1		NM_024006.5:c.173+324T>G	NC_000016.9:g.31105554A>C	Intron		NO	YES	0,371	19,1899	18,9878	18,7661					
VKORC1		NM_024006.5:c.173+525C>T	NC_000016.9:g.31105353G>A	Intron		NO	YES	7,872		16,7808	9,365					
VKORC1		NM_024006.5:c.174-136C>T	NC_000016.9:g.31104878G>A	Intron		NO	YES	10,42		32,6143	35,5831					
VKORC1		NM_024006.5:c.-1877A>G	NC_000016.9:g.31107927T>C	Intron		NO	YES			10,5002						
VKORC1		NM_024006.5:c.283+124G>C	NC_000016.9:g.31104509C>G	Intron		NO	YES	5,156		37,492	41,6334					
VKORC1		NM_024006.5:c.283+837T>C	NC_000016.9:g.31103796A>G	Intron		NO	YES	8,857		64,309	60,9625					
VKORC1		NM_024006.5:c.-4931C>T	NC_000016.9:g.31110981G>A	Intron		NO	NO			57,5756	52,5559					

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