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Population genetic difference of pharmacogenomic VIP gene variants in the Lisu population from Yunnan Province

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Abstract

Individual differences in drug clinical response are related to pharmacogenomics. The genetic variation of drug-metabolizing enzymes, drug receptors, and their downstream protein genes is the main factor causing individual differences in drug response. The genetic backgrounds among different ethnic groups are quite different. In this study, we aimed to detect the distribution difference of genotype frequency in very important pharmacogenetic (VIP) gene variants in the Lisu.

Using the chi-squared test, we compared the genotype frequencies of the VIP variants in 105 Lisu people with those in 26 populations from the 1000 Genome project separately. Bonferroni's multiple adjustment was also conducted ($P < .05/(26*49)$). Moreover, Arlequin v3.5 and Structure v2.3.4 software were used to analyze the genetic distance and genetic structure.

There were 9, 9, 11, 12, 11, 11, 9, 17, 13, 13, 16, 5, 3, 5, 3, 4, 17, 14, 16, 17, 16, 10, 13, 12, 10, and 9 single nucleotide polymorphisms that differed in frequency distribution, when Lisu people compared with the 26 populations separately. Only *CYP2E1* rs2070676 was different in the Lisu population compared with the 26 groups from the 1000 Genome project. *PTGS2* rs5275 and *CYP2D6* rs1065852 were different in the Lisu population compared with most of the populations. Additionally, genetic backgrounds of Lisu and Han Chinese in Beijing were closest according to the lowest F-statistics value and resemblance in genetic structures.

Our results complete the information of the Lisu population in pharmacogenomics database.

Abbreviations: ACB = African Caribbean in Barbados, ASW = Americans of African Ancestry in southwest United States, BEB = Bengali from Bangladesh, CDX = Chinese Dai in Xishuangbanna, China, CEU = Utah Residents (CEPH) with Northern and Western European Ancestry, CHB = Han Chinese in Beijing, China, CHS = Southern Han Chinese, CLM = Colombians from Medellin, Colombia, ESN = Esan in Nigeria, FIN = Finnish in Finland, GBR = British in England and Scotland, GIH = Gujarati Indian from Houston, Texas, GWD = Gambian in Western Divisions in the Gambia, IBS = Iberian Population in Spain, ITU = Indian Telugu from the United Kingdom, JPT = Japanese in Tokyo, Japan, KHV = Kinh in Ho Chi Minh City, Vietnam, LWK = Luhya in Webuye, Kenya, MSL = Mende in Sierra Leone, MXL = Mexican Ancestry from Los Angeles, United States, PEL = Peruvians from Lima, Peru, PJL = Punjabi from Lahore, Pakistan, PUR = Puerto Ricans from Puerto Rico, SNP = single nucleotide polymorphism, STU = Sri Lankan Tamil from the United Kingdom, TSI = Toscani in Italia, VIP = very important pharmacogenetic, YRI = Yoruba in Ibadan, Nigeria.

Keywords: Lisu population, population genetic, very important pharmacogenetic

1. Introduction

The Pharmacogenetics and Pharmacogenomics knowledgebase (PharmGKB) is a national research alliance, which examines how variations in genes lead to individual differences in drug response. Pharmacogenomics Knowledge Base (PharmGKB: http://www.pharmgkb.org) has as many as 621 drugs with PGx-related gene polymorphism annotation information, and up to 128 Pathways involving pharmacokinetics and pharmacodynamics. There are 65 very important pharmacogenetics (VIP) involved in this database. VIP summaries provide an overview of a significant gene involved in metabolism or response to one or several drugs. VIPs play roles in the metabolism of many drugs, and contain variants, which potentially contribute to a severe drug response.

Comparing the effects of drugs among different races has become a major direction of pharmacogenomics research. Cytochrome P450 is the main family enzyme system in the drug-metabolism enzyme system in human body. The difference in the ability of metabolizing substrates among individuals in different populations will lead to differences in individual clinical treatment effects and disease susceptibility. Studies have shown that the vast majority of statins metabolism is closely related to Cytochrome P450 Family 3 Subfamily A Member 4 (*CYP3A4*),[1] Cytochrome P450 Family 2 Subfamily C Member 9 (*CYP2C9*),[2] and Cytochrome P450 Family 2 Subfamily D Member 6 (*CYP2D6*)[3] single nucleotide polymorphisms (SNPs). The presence of these SNPs may affect drug efficacy or cause adverse drug reactions.

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Chan Zhang and Xiaochun Jiang have contributed equally to this work.

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As a result, different races and their subpopulations have different effects on the same dose of the same drug. Therefore, the study of differences in pharmacogenomics among different populations can provide valuable theoretical basis for individualized drug treatment based on different populations. At present, there are studies on population genetics of genes related to drug-metabolism enzymes in ethnic minorities in Yunnan Province (Zhuang population[4] and Yi population[5]), Tibet Autonomous Region (Lhoba population,[6] Deng population,[7] Sherpa population,[8] and Tibetan population[9]), Xinjiang Uygur Autonomous Region (Uygur population,[10] Tajik ethnic population,[11] and Kyrgyz population),[12] and Guizhou province (Miao people[13]). The Lisu originates from the ancient Diqiang family and has the origin relationship with the Yi people. The national language belongs to the Sino-Tibetan Tibetan-Burmese language group. The Lisu ethnic group mainly distributes along the Nu River and the Kaijiang River (Irrawaddy River Branch) basin areas, which are the border area of Yunnan, Tibet, and Burma Kachin.[14,15] The rest are scattered in the other regions of Yunnan, the east and the north of India, and the border between Thailand and Burma.

The ethnic composition of various regions in China is complex, and the genetic backgrounds among different ethnic groups are quite different. There are few population genetic studies on the drug-metabolism genes of the Lisu population. The incidence of drug efficacy and adverse reactions in minority populations are not optimistic.[16] So, in this study, we want to detect the allele frequencies of 49 VIP variants in the Lisu population, and further determine the allele frequency differences between the Lisu and 26 populations reported in the 1000 Human Genome Project. We hope that the results of this study will extend our understanding of ethnic diversity, pharmacogenomics, and enable medical professionals to use genomic and molecular data to effectively implement personalized medicine in the future.

2. Materials and methods

2.1. Ethical statement

All procedures involving human participants were in accordance with the ethical standards of the First People's Hospital of Yunnan Province, and the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. Informed consent was obtained from each individual who participated in the study.

2.2. Study participants

A total of 105 healthy, randomly selected individuals from Lisu ethnic group were enrolled in this study, and had exclusively Lisu ancestry for at least the last 3 generations. One hundred five blood samples were collected from the First People's Hospital of Yunnan Province.

2.3. Variant selection and genotyping

From the Pharmacogenetics and Pharmacogenomics knowledgebase, we selected 25 genes, which previously reported to be related to drug metabolism. And, based on the frequency of minor allele >0.05 in the global population from the 1000 Human Genome Project, we selected 49 SNPs. Genomic DNA was extracted from the peripheral blood of the participants using the GoldMag whole blood genomic DNA purification kit (GoldMag Co Ltd, Xi'an, China), as recommended by the manufacturer's instructions. DNA concentration was determined using a NanoDrop 2000C spectrophotometer (Thermo Scientific, Waltham, MA). The Agena MassARRAY Assay Design 3.0 Software (Agena Bioscience, Inc., San Diego, CA) was used to design Multiplexed SNPMassEXTEND assays.[17] SNP genotyping analysis was performed using the standard protocol recommended by the manufacturer with an Agena MassARRAY RS1000 (San Diego, CA). Agena Typer 4.0 Software (San Diego, CA) was used to manage and analyze the SNP genotyping data as described in a previous report.[18]

2.4. 1000 Human Genome Project genotype data

The 1000 Genomes Project ran between 2008 and 2015, creating the largest public catalog of human variation and genotype data. The genotype data of individuals from 26 populations were downloaded from the 1000 Genomes Project website (http://www.internationalgenome.org/data). The following table lists these populations. The 26 populations comprised Han Chinese in Beijing, China (CHB), Japanese in Tokyo, Japan (JPT), Southern Han Chinese (CHS), Chinese Dai in Xishuangbanna, China (CDX), Kinh in Ho Chi Minh City, Vietnam (KHV), Utah Residents (CEPH) with Northern and Western European Ancestry (CEU), Toscani in Italia (TSI), Finnish in Finland (FIN), British in England and Scotland (GBR), Iberian Population in Spain (IBS), Yoruba in Ibadan, Nigeria (YRI), Luhya in Webuye, Kenya (LWK), Gambian in Western Divisions in the Gambia (GWD), Mende in Sierra Leone (MSL), Esan in Nigeria (ESN), Americans of African Ancestry in southwest United States (ASW), African Caribbean in Barbados (ACB), Mexican Ancestry from Los Angeles, United States (MXL), Puerto Ricans from Puerto Rico (PUR), Colombians from Medellin, Colombia (CLM), Peruvians from Lima, Peru (PEL), Gujarati Indian from Houston, Texas (GIH), Punjabi from Lahore, Pakistan (PJL), Bengali from Bangladesh (BEB), Sri Lankan Tamil from the United Kingdom (STU), and Indian Telugu from the United Kingdom (ITU).

2.5. Statistical analyses

We compared the genotype frequencies of the VIP in the Lisu people with those in the 26 populations separately using the chi-squared test. All P values obtained in this study were 2-sided and Bonferroni's multiple adjustment was applied to improve the significance, which was set at $P < .05/(26*49)$. With the chi-squared test, we wanted to find some significant different SNPs.

The values of Fst and P were calculated by Arlequin v3.5 software.[19] The population genetic differentiation factor (Fst) reflects the degree of difference between the average heterozygosity of each subpopulation and the heterozygosity of the total population. The Fst value is 0 to 1, the larger the value, the more obvious the genetic differentiation among the subgroups.

A Bayesian model based on allelic frequency correlation among populations was used to estimate the K of population classification by Structure v2.3.4 software.[20] According to the recommendation of the Structure software manual, the K value was 5 to 8, each K value was repeated 3 times, and the Markov Chain Monte Carlo reaction times was set to 100,000, and the subsequent burn-in reaction times was set to 10,000. When software running to complete and getting results, we drew bar chart through drawing software.

3. Results

The basic information of the 49 selected variants is shown in Table 1, including the position, allele, alternative amino acids, and minor allele frequency of the selected SNPs.

Table 1

Basic characteristic of selected variants and the MAF in Lisu people.

SNP ID	Chr	Position	Gene	Alleles	Functional consequence	Function	MAF
rs1801159	1	97515839	*DPYD*	A/G	Ile506Val	Intron variant, missense	0.18
rs1801158	1	97515865	*DPYD*	A/G	Ser369Asn	Intron variant, missense	0.00
rs1801265	1	97883329	*DPYD*	C/T	Cys29Arg	Intron variant, missense	0.13
rs5275	1	18667392	*PTGS2*	A/C/T	3′UTR	UTR variant 3 prime	0.00
rs1800462	6	18143724	*TPMT*	C/G	Ala80Pro	Missense	0.00
rs34130495	6	16013979	*SLC22A1*	A/G	Gly401Ser	Missense	0.00
rs34059508	6	16015480	*SLC22A1*	A/G	Gly465Arg	Intron variant, missense	0.00
rs776746	7	99672916	*CYP3A5*	A/G	Intronic	Intron variant	0.44
rs12721627	7	99768470	*CYP3A4*	C/G	Thr185Ser	Missense	0.00
rs4986908	7	99769769	*CYP3A4*	A/C/G	Asp174Asn	Missense	0.50
rs4986907	7	99769804	*CYP3A4*	A/G	Arg162Gln	Missense	0.00
rs12190875	7	11758780	*CFTR*	A/G/T	Ser579Asn	Intron variant	0.00
rs4646244	8	18390208	*NAT2*	A/T	5 Flanking	Intron variant	0.20
rs4271002	8	18390758	*NAT2*	C/G	5 Flanking	Intron variant	0.16
rs1801279	8	18400194	*NAT2*	A/G	Arg64Gln	Missense	0.00
rs1801280	8	18400344	*NAT2*	C/T	Ile114Thr	Missense	0.10
rs1799929	8	18400484	*NAT2*	C/T	Leu161=	Synonymous codon	0.11
rs1208	8	18400806	*NAT2*	A/G/T	Arg268Lys	Missense	0.11
rs1799931	8	18400860	*NAT2*	A/G	Gly286Glu	Missense	0.16
rs2115819	10	45405641	*ALOX5*	C/T	Intronic	Intron variant	0.18
rs12248560	10	94761900	*CYP2C19*	A/C/T	5 Flanking	Upstream variant 2 kb	0.03
rs1057910	10	94981296	*CYP2C9*	A/C/G	Ile359Leu	Missense	0.02
rs10509681	10	95038992	*CYP2C8*	C/T	Lys329Arg	Missense	0.00
rs1058930	10	95058362	*CYP2C8*	A/C/G	Ile194Met	Missense	0.00
rs11572080	10	95067273	*CYP2C8*	A/G/T	Arg69Lys	Missense	0.00
rs7909236	10	95069673	*CYP2C8*	G/T	5 Flanking	Upstream variant 2 kb	0.06
rs17110453	10	95069772	*CYP2C8*	A/C	5 Flanking	Upstream variant 2 kb	0.36
rs2031920	10	13352634	*CYP2E1*	C/T	5 Flanking	Upstream variant 2 kb	0.08
rs6413432	10	13353504	*CYP2E1*	A/T	Intronic	Intron variant	0.25
rs2070676	10	13353763	*CYP2E1*	C/G	Intronic	Intron variant	0.30
rs1801028	11	11341276	*DRD2*	C/G	Ser311Cys	Missense	0.01
rs4149015	12	21130388	*SLCO1B1*	A/C/G	5 Flanking	Upstream variant 2 kb	0.03
rs2306283	12	21176804	*SLCO1B1*	A/C/T	Asn130Asp	Missense	0.29
rs731236	12	47844974	*VDR*	C/T	Ile352=	Synonymous codon	0.00
rs4516035	12	47906043	*VDR*	C/T	5 Flanking	Upstream variant 2 kb	0.03
rs12720461	15	74749010	*CYP1A2*	C/T	Intronic	Intron variant	0.00
rs762551	15	74749576	*CYP1A2*	A/C	Intronic	Intron variant	0.34
rs9282861	16	28606193	*SULT1A1*	A/G	Arg213His	Missense	0.09
rs750155	16	28609251	*SULT1A1*	C/T	Intronic	Intron variant	0.29
rs1800566	16	69711242	*NQO1*	C/T	Pro115Ser	Missense	0.32
rs2108622	19	15879621	*CYP4F2*	C/T	Val433Met	Missense	0.19
rs118192172	19	38457545	*RYR1*	C/T	Arg614Cys	Missense	0.00
rs8192726	19	40848591	*CYP2A6*	G/T	Intron variant	Intron variant	0.22
rs5629	20	49513169	*PTGIS*	A/C/T	Arg373=	Synonymous codon	0.17
rs1051298	21	45514912	*SLC19A1*	C/T	3′UTR	Intron variant	0.45
rs1051296	21	45514947	*SLC19A1*	G/T	3′UTR	Intron variant	0.45
rs3892097	22	42128945	*CYP2D6*	A/G	Intronic	Intron variant	0.02
rs1065852	22	42130692	*CYP2D6*	C/T	Pro34Ser	Intron variant, missense	0.31
rs28358569	MT	827	None	A/G	None	None	0.17

Chr = chromosome, MAF = minor allele frequency.

Through the chi-squared test, the differences in genotype frequencies of the 49 variants between the Lisu population and the 26 individuals from the 1000 Genomes Project were compared, and further Bonferroni adjustment was performed. The results showed that there were 9, 9, 11, 12, 11, 11, 9, 17, 13, 13, 16, 5, 3, 5, 3, 4, 17, 14, 16, 17, 16, 10, 13, 12, 10, and 9 SNPs that different in the frequency distribution, when Lisu people compared with the ACB, ASW, ESN, GWD, LWK, MSL, YRI, CLM, MXL, PEL, PUR, CDX, CHB, CHS, JPT, KHV, CEU, FIN, GBR, IBS, TSI, BEB, GIH, ITU, PJL, and STU populations, respectively (Table 2; Fig. 1). Two SNPs (*CYP3A5* rs776746 and *CYP2E1* rs2070676) were different in the Lisu population compared with East Asian population; 11 SNPs (*PTGS2* rs5275, *CYP3A5* rs776746, *NAT2* rs1801280, *NAT2* rs1799929, *NAT2* rs1208, *ALOX5* rs2115819, *CYP2C19* rs12248560, *CYP2E1* rs2070676, *SLCO1B1* rs2306283, *VDR* rs731236, and *VDR* rs4516035) were different in the Lisu population compared with European population; 7 loci (*PTGS2* rs5275, *NAT2* rs1208, *ALOX5* rs2115819, *CYP2C19* rs12248560, *CYP2C8* rs17110453, *CYP2E1* rs2070676, and *VDR* rs731236) were different in the Lisu population compared with African population; 8 SNPs (*PTGS2* rs5275, *CYP3A5* rs776746, *NAT2*

Table 2

Significant variants in Lisu people compared with the 26 populations from 1000 genomes project (phase 3) by chi-squared test.

SNP ID	Gene	ACB	ASW	ESN	GWD	LWK	MSL	YRI	CLM	MXL	PEL	PUR	CDX	CHB
rs1801159	*DPYD*	—	—	—	—	—	—	—	—	—	3.97E−08	—	—	—
rs5275	*PTGS2*	2.64E−34	4.99E−29	8.10E−38	1.18E−31	1.29E−33	3.21E−36	4.78E−39	4.55E−21	1.33E−17	1.44E−22	6.57E−18	8.63E−11	–
rs776746	*CYP3A5*	—	—	3.19E−12	1.17E−05	2.38E−11	2.04E−10	7.89E−09	4.97E−12	1.15E−07	1.52E−15	2.64E−08	9.75E−06	4.97E−06
rs1801279	*NAT2*	—	—	1.07E−06	9.57E−08	—	—	—	—	—	—	—	—	—
rs1801280	*NAT2*	—	1.83E−05	—	2.59E−07	9.28E−09	—	—	5.66E−09	5.78E−08	3.90E−05	2.40E−09	—	—
rs1799929	*NAT2*	—	—	—	2.43E−05	4.45E−07	—	—	3.55E−08	1.57E−07	—	7.69E−08	—	—
rs1208	*NAT2*	2.90E−08	6.27E−07	5.07E−09	6.13E−12	3.09E−12	2.64E−07	8.70E−09	5.30E−09	3.71E−10	—	5.72E−09	—	—
rs1799931	*NAT2*	—	—	—	—	—	—	—	—	—	—	—	—	—
rs2115819	*ALOX5*	2.13E−26	1.44E−17	6.01E−26	8.18E−29	3.87E−22	5.84E−22	3.55E−28	8.68E−10	2.06E−07	—	1.05E−08	—	—
rs12248560	*CYP2C19*	2.82E−11	1.03E−06	1.93E−09	2.27E−09	5.77E−06	6.41E−10	4.79E−09	—	—	—	3.99E−06	—	—
rs10509681	*CYP2C8*	—	—	—	—	—	—	—	2.02E−06	—	—	8.16E−08	—	—
rs11572080	*CYP2C8*	—	—	—	—	—	—	—	2.02E−06	—	—	8.16E−08	—	—
rs7909236	*CYP2C8*	—	—	—	—	—	—	—	2.33E−09	8.60E−09	3.34E−12	—	—	—
rs17110453	*CYP2C8*	3.20E−16	7.77E−12	1.23E−16	4.51E−20	3.96E−18	3.85E−16	1.31E−18	1.92E−08	9.42E−06	4.51E−11	1.27E−07	—	—
rs6413432	*CYP2E1*	1.09E−05	—	—	—	1.96E−07	1.44E−05	—	—	—	—	—	—	—
rs2070676	*CYP2E1*	7.03E−17	2.05E−14	1.90E−20	4.16E−22	9.93E−23	1.79E−17	4.54E−21	1.26E−11	5.42E−06	1.50E−07	1.07E−12	1.30E−12	6.10E−13
rs1801028	*DRD2*	—	—	—	—	—	—	—	—	—	—	—	—	—
rs2306283	*SLCO1B1*	—	—	3.37E−05	—	—	—	—	8.41E−06	9.18E−09	7.33E−06	—	—	—
rs731236	*VDR*	2.19E−17	2.57E−13	5.57E−16	3.69E−14	8.61E−15	1.22E−12	1.49E−17	2.98E−13	1.08E−10	4.64E−06	7.12E−22	—	—
rs4516035	*VDR*	—	—	—	—	—	—	—	9.71E−11	1.27E−08	3.28E−05	8.12E−15	—	—
rs762551	*CYP1A2*	—	—	—	—	—	—	—	—	—	1.99E−06	—	—	—
rs750155	*SULT1A1*	—	—	—	—	—	—	—	—	—	1.01E−18	—	2.63E−05	—
rs1800566	*NQO1*	—	—	—	—	—	8.89E−06	—	—	—	—	—	—	—
rs2108622	*CYP4F2*	—	—	—	—	—	—	—	—	—	—	—	—	—
rs8192726	*CYP2A6*	—	—	—	—	—	—	—	4.05E−06	—	—	4.76E−06	—	—
rs3892097	*CYP2D6*	—	—	—	—	—	—	—	7.08E−06	—	—	3.21E−05	—	—
rs1065852	*CYP2D6*	4.19E−08	4.16E−07	6.44E−12	4.76E−11	—	1.31E−07	9.10E−11	7.67E−08	1.81E−05	6.34E−12	4.13E−09	3.67E−13	1.81E−11
SNP ID	Gene	CHS	JPT	KHV	CEU	FIN	GBR	IBS	TSI	BEB	GIH	ITU	PJL	STU
rs1801159	*DPYD*	—	—	—	—	—	—	—	—	—	—	—	—	—
rs5275	*PTGS2*	3.68E−10	1.17E−13	7.85E−12	3.33E−22	7.85E−12	2.32E−15	1.47E−18	3.64E−16	1.10E−20	1.18E−21	1.03E−20	1.89E−24	7.17E−23
rs776746	*CYP3A5*	6.88E−08	3.83E−09	4.79E−07	9.97E−25	1.06E−22	6.12E−22	4.99E−22	1.80E−24	—	1.14E−07	3.74E−05	—	—
rs1801279	*NAT2*	—	—	—	—	—	—	—	—	—	—	—	—	—
rs1801280	*NAT2*	—	—	—	4.06E−11	1.82E−12	1.65E−12	4.08E−14	1.12E−11	9.02E−08	2.17E−07	1.33E−07	4.53E−11	2.95E−05
rs1799929	*NAT2*	—	—	—	8.30E−11	2.48E−11	2.05E−11	9.68E−14	1.54E−11	4.11E−07	8.00E−06	3.33E−06	1.89E−09	—
rs1208	*NAT2*	—	—	—	7.51E−10	6.73E−11	2.31E−11	7.88E−14	1.10E−11	7.24E−09	3.85E−07	1.08E−07	3.32E−11	5.90E−06
rs1799931	*NAT2*	—	—	—	—	—	—	—	—	—	—	—	—	—
rs2115819	*ALOX5*	—	—	—	5.70E−15	1.55E−11	1.20E−11	7.18E−12	2.80E−12	1.22E−09	9.15E−14	2.36E−12	6.34E−09	4.12E−07
rs12248560	*CYP2C19*	—	—	—	1.00E−08	4.29E−08	4.53E−09	3.55E−08	3.29E−08	—	—	—	—	—
rs10509681	*CYP2C8*	—	—	—	1.07E−06	—	—	3.58E−08	9.13E−07	—	—	—	—	—
rs11572080	*CYP2C8*	—	—	—	1.07E−06	—	—	3.58E−08	9.13E−07	—	—	—	—	—
rs7909236	*CYP2C8*	—	—	—	1.00E−08	1.17E−07	9.10E−06	—	—	—	2.86E−07	2.49E−06	—	—
rs17110453	*CYP2C8*	—	—	—	4.87E−09	—	9.30E−08	—	2.69E−08	—	—	—	—	—
rs6413432	*CYP2E1*	—	—	—	—	—	—	2.62E−05	—	—	—	—	—	—
rs2070676	*CYP2E1*	2.87E−11	2.04E−12	1.10E−13	2.52E−12	3.94E−09	4.98E−09	8.55E−11	4.61E−14	9.61E−11	9.75E−11	1.73E−11	1.10E−10	3.85E−10

(*continued*)

Table 2
(continued).

rs1801028	*DRD2*	—	—	—	—	—	—	—	—	7.35E−07	—	—	—	—	—	—
rs2306283	*SLCO1B1*	—	—	—	6.73E−09	2.19E−07	7.95E−11	6.50E−09	1.01E−10	—	—	—	—	1.01E−06	—	—
rs731236	*VDR*	—	—	—	3.23E−24	4.54E−18	1.18E−16	5.85E−24	5.38E−23	6.97E−16	1.09E−17	1.37E−26	2.04E−15	7.32E−24		
rs4516035	*VDR*	—	—	—	4.43E−15	1.29E−24	2.08E−17	1.38E−15	5.77E−19	5.43E−06	2.62E−06	1.81E−06	2.77E−08	2.57E−07		
rs762551	*CYP1A2*	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
rs750155	*SULT1A1*	8.05E−07	—	—	—	1.59E−05	—	—	—	—	—	—	—	—	—	—
rs1800566	*NQO1*	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
rs2108622	*CYP4F2*	—	—	—	—	—	—	—	—	1.11E−05	9.84E−07	1.77E−05	—	4.89E−06		
rs8192726	*CYP2A6*	—	—	—	2.49E−05	—	9.68E−06	1.54E−05	—	—	—	—	—	—		
rs3892097	*CYP2D6*	—	—	—	1.59E−09	—	3.83E−09	2.93E−05	3.51E−07	—	—	—	—	—		
rs1065852	*CYP2D6*	7.05E−13	—	3.06E−14	—	4.25E−08	6.29E−06	4.48E−07	9.72E−06	1.89E−06	4.61E−07	1.94E−06	1.44E−09	1.44E−08		

ACB = African Caribbean in Barbados, ASW = Americans of African Ancestry in southwest United States, BEB = Bengali from Bangladesh, CDX = Chinese Dai in Xishuangbanna, China, CEU = Utah Residents (CEPH) with Northern and Western European Ancestry, CHB = Han Chinese in Beijing, China, CHS = Southern Han Chinese, CLM = Colombians from Medellin, Colombia, ESN = Esan in Nigeria, FIN = Finnish in Finland, GBR = British in England and Scotland, GIH = Gujarati Indian from Houston, Texas, GWD = Gambian in Western Divisions in the Gambia, IBS = Iberian Population in Spain, ITU = Indian Telugu from the United Kingdom, JPT = Japanese in Tokyo, Japan, KHV = Kinh in Ho Chi Minh City, Vietnam, LWK = Luhya in Webuye, Kenya, MSL = Mende in Sierra Leone, MXL = Mexican Ancestry from Los Angeles, United States, PEL = Peruvians from Lima, Peru, PJL = Punjabi from Lahore, Pakistan, PUR = Puerto Ricans from Puerto Rico, STU = Sri Lankan Tamil from the United Kingdom, TSI = Toscani in Italia, YRI = Yoruba in Ibadan, Nigeria.
$P < .05/(49*26)$ indicates statistical significance.

rs1801280, *CYP2C8* rs17110453, *CYP2E1* rs2070676, *VDR* rs731236, *VDR* rs4516035, and *CYP2D6* rs1065852) were different in the Lisu population compared with Ad Mixed American population; 8 SNPs (*PTGS2* rs5275, *NAT2* rs1801280, *NAT2* rs1208, *ALOX5* rs2115819, *CYP2E1* rs2070676, *VDR* rs731236, *VDR* rs4516035, and *CYP2D6* rs1065852) were different in the Lisu population compared with South Asian population. Only *CYP2E1* rs2070676 was different in the Lisu population compared with 26 individuals from the 1000 Genomes Project. *PTGS2* rs5275 and *CYP2D6* rs1065852 were different in the Lisu population compared with most of the populations.

We used a model-based clustering approach, as implemented in STRUCTURE, to infer population structures among the 27 populations. When the K value runs 5, the genetic structure of Lisu people was much similar to the Chinese Dai in Xishuangbanna (CDX), CHB, and CHS (Fig. 2). Pairwise Fst values were calculated for all population comparisons. The Fst value is 0 to 1, the larger the value, the more obvious the genetic differentiation among the subgroups. From Table 3, we found that the value of Fst (Lisu and CHB) was 0.047, which was the smallest. Therefore, we considered that the genetic backgrounds of Lisu and CHB were similar. We speculated that the genetic backgrounds of Lisu and CHB were resemblance.

4. Discussion

The emergence of pharmacogenomics has promoted correlation analysis of drug-metabolism, target-related genes, and various adverse drug reactions. Meanwhile, a number of genetic polymorphisms have gradually gained attention and laid the foundation for clinical individualized medication. In this study, we detected the allele frequencies of 49 VIP variants in the Lisu population, and further determined the allele frequency differences with populations reported in the 1000 Genome Project. In the end, we found that only *CYP2E1* rs2070676 was different in the Lisu population compared with 26 individuals from the 1000 genome project. *PTGS2* rs5275 and *CYP2D6* rs1065852 were different in the Lisu population compared with most of the populations. And genetic backgrounds of Lisu and CHB were similar through the Structure and the Fst analysis.

Cytochrome P450 2E1 (*CYP2E1*) is an important phase I metabolizing enzyme in the body. It is mainly located in liver microsomes and can metabolize many low-molecular weight potentially carcinogenic chemicals including benzene. This enzyme can be induced by ethanol and other compounds. *CYP2E1* affects the metabolism and efficacy of various drugs, such as cisplatin[21] and antituberculosis drug.[22] There are substantial differences among different populations in their reaction to drugs. Besides, *CYP2E1* also affects the occurrence of diseases. Chang et al researched the association between *CYP2E1* gene polymorphisms and formation of Coronary Artery Lesions in Kawasaki Disease, and the results pointed that the GG genotype of rs2070676 was strongly associated with the risk of coronary artery lesions formation in Kawasaki disease patients.[23] Shahabi et al presented significantly results indicating an association between the rs2070676 polymorphism and Parkinson disease.[24] The allele frequencies of the variants are different in populations. In our research, the frequency of minor allele (G) in rs2070676 is 0.30. From the ALelle FREquency Database, we found that the frequency range of minor allele (G) was 0.030 to 0.426 in Asian population, while in African population, was 0.500 to 0.811.[25] The same rule was found in

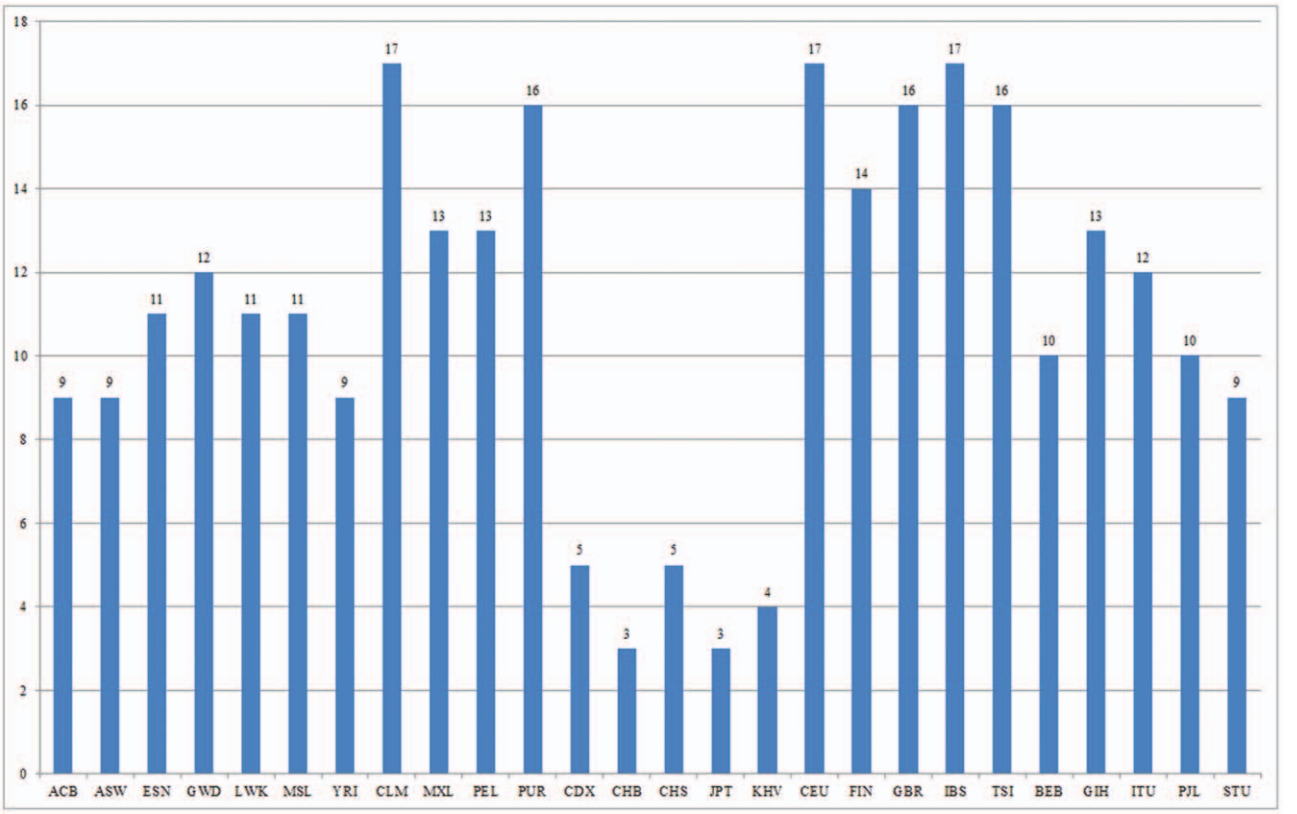

Figure 1. Number of variants significantly differed with 26 populations after multiple adjustment.

the 1000 Genome project. There was very wide variation in population allele frequency of this variant because several African populations were reported to have the G variant as the major allele, whereas European and Asian populations to have the G variant as the minor allele.

Prostaglandin-endoperoxide synthase 2 (*PTGS2*) gene is a paralog of *PTGS1*, which located on 1q31.1. *PTGS2* gene encodes the prostaglandin-endoperoxide synthase, regulates prostaglandin levels at the level of cyclooxygenase, and catalyzes the first 2 steps in the metabolism of arachidonic acid. The *PTGS2* gene is associated with the efficacy of various drugs, such as anti-inflammatory agents, nonsteroids, rofecoxib, ibuprofen, Cobra drugs, aspirin, and other drugs. The *PTGS2* gene also has a close relationship with a variety of cancers and diseases. *PTGS2* rs5275 (8473 T>C) variant is located in the 3′-UTR in which it may stabilize the mRNA. One research found that rs5275 TT genotype was associated with better progression-free survival and overall survival in patients with advanced colorectal cancer treated with XELOX (capecitabine and oxaliplatin) chemotherapy.[26] The TT genotype was also associated with lower risk for severe pain in lung cancer patients.[27] The C variant occurs at a frequency of 0.355 in Caucasians, 0.435 in Native American/Hispanic, and 0.667 in African/African American sample sets in the SNP500Cancer control sample set from the Coriell collection.[28] The C variant is also observed at a frequency of 0.291 in German Caucasians.[29] In our research, the frequency of minor allele (T) of rs5275 is 0.00.

Cytochrome P450 2D6 (CYP2D6) mediates several important metabolic pathways in the body and is a key enzyme in metabolism. As the most polymorphic enzyme system, approximate 55 mutants of *CYP2D6* influence the final enzyme activity and quantity changes, resulting in considerable individualized differences in human response to drugs. The frequencies of *CYP2D6* gene mutations are extremely different among races. In

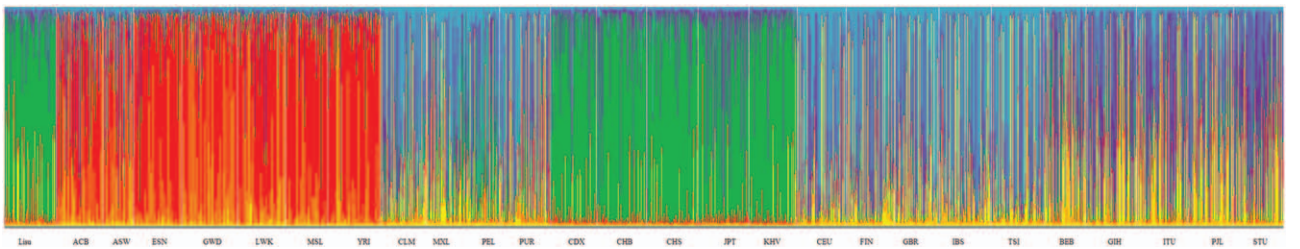

Figure 2. Bayesian clustering of cod sampled from 27 populations. Each vertical bar denotes an individual, whilst colors denote inferred clusters.

Table 3

Fst values between population pairs.

	Lisu	ACB	ASW	ESN	GWD	LWK	MSL	YRI	CLM	MXL	PEL	PUR	CDX	CHB	CHS	JPT	KHV	CEU	FIN	GBR	IBS	TSI	BEB	GIH	ITU	PJL	STU
Lisu	0.000																										
ACB	0.154	0.000																									
ASW	0.119	0.002	0.000																								
ESN	0.172	0.009	0.012	0.000																							
GWD	0.166	0.006	0.008	0.006	0.000																						
LWK	0.174	0.014	0.016	0.007	0.012	0.000																					
MSL	0.165	0.004	0.010	0.004	0.005	0.013	0.000																				
YRI	0.162	0.004	0.008	−0.001	0.004	0.012	0.001	0.000																			
CLM	0.103	0.106	0.071	0.144	0.128	0.138	0.140	0.132	0.000																		
MXL	0.103	0.123	0.082	0.158	0.144	0.149	0.156	0.145	0.002	0.000																	
PEL	0.126	0.174	0.134	0.213	0.205	0.201	0.212	0.198	0.039	0.028	0.000																
PUR	0.093	0.084	0.055	0.119	0.104	0.111	0.116	0.109	0.005	0.010	0.057	0.000															
CDX	0.052	0.165	0.135	0.199	0.192	0.201	0.184	0.183	0.099	0.106	0.104	0.096	0.000														
CHB	0.047	0.175	0.142	0.200	0.196	0.207	0.193	0.187	0.101	0.105	0.114	0.101	0.007	0.000													
CHS	0.051	0.181	0.149	0.211	0.205	0.215	0.200	0.197	0.104	0.113	0.114	0.105	0.003	0.001	0.000												
JPT	0.039	0.159	0.126	0.185	0.179	0.189	0.176	0.170	0.086	0.094	0.103	0.085	0.019	0.010	0.010	0.000											
KHV	0.059	0.180	0.149	0.211	0.205	0.215	0.200	0.198	0.104	0.115	0.117	0.106	0.004	0.005	0.000	0.017	0.000										
CEU	0.139	0.126	0.092	0.166	0.149	0.163	0.164	0.155	0.009	0.022	0.077	0.012	0.142	0.143	0.146	0.124	0.146	0.000									
FIN	0.130	0.142	0.104	0.182	0.162	0.173	0.179	0.170	0.012	0.022	0.070	0.015	0.137	0.137	0.138	0.117	0.141	0.007	0.000								
GBR	0.136	0.139	0.100	0.180	0.160	0.172	0.176	0.168	0.009	0.017	0.072	0.012	0.142	0.143	0.146	0.126	0.147	0.000	0.004	0.000							
IBS	0.132	0.124	0.093	0.166	0.146	0.154	0.162	0.153	0.010	0.023	0.080	0.010	0.137	0.139	0.138	0.116	0.140	0.006	0.006	0.004	0.000						
TSI	0.129	0.127	0.093	0.166	0.150	0.157	0.164	0.154	0.009	0.017	0.073	0.008	0.131	0.132	0.135	0.112	0.136	0.003	0.006	0.001	−0.001	0.000					
BEB	0.077	0.090	0.062	0.120	0.104	0.117	0.115	0.108	0.022	0.030	0.091	0.018	0.087	0.081	0.086	0.064	0.089	0.040	0.040	0.039	0.028	0.032	0.000				
GIH	0.087	0.098	0.071	0.129	0.115	0.126	0.126	0.118	0.021	0.035	0.087	0.023	0.097	0.088	0.090	0.066	0.094	0.037	0.035	0.040	0.027	0.031	0.002	0.000			
ITU	0.083	0.090	0.064	0.117	0.104	0.117	0.116	0.108	0.025	0.037	0.098	0.019	0.101	0.091	0.096	0.070	0.100	0.036	0.037	0.040	0.028	0.031	0.001	0.001	0.000		
PJL	0.097	0.092	0.062	0.121	0.106	0.114	0.117	0.110	0.017	0.024	0.087	0.016	0.118	0.112	0.116	0.086	0.118	0.031	0.029	0.031	0.020	0.024	0.003	0.005	0.004	0.000	
STU	0.080	0.102	0.075	0.129	0.118	0.129	0.125	0.118	0.037	0.050	0.111	0.028	0.099	0.089	0.092	0.062	0.097	0.049	0.049	0.054	0.037	0.041	0.003	0.004	0.000	0.008	0.000

ACB = African Caribbean in Barbados, ASW = Americans of African Ancestry in southwest United States, BEB = Bengali from Bangladesh, CDX = Chinese Dai in Xishuangbanna, China, CEU = Utah Residents (CEPH) with Northern and Western European Ancestry, CHB = Han Chinese in Beijing, China, CHS = Southern Han Chinese, CLM = Colombians from Medellin, Colombia, ESN = Esan in Nigeria, FIN = Finnish in Finland, GBR = British in England and Scotland, GIH = Gujarati Indian from Houston, Texas, GWD = Gambian in Western Divisions in the Gambia, IBS = Iberian Population in Spain, ITU = Indian Telugu from the United Kingdom, JPT = Japanese in Tokyo, Japan, KHV = Kinh in Ho Chi Minh City, Vietnam, LWK = Luhya in Webuye, Kenya, MSL = Mende in Sierra Leone, MXL = Mexican Ancestry from Los Angeles, United States, PEL = Peruvians from Lima, Peru, PJL = Punjabi from Lahore, Pakistan, PUR = Puerto Ricans from Puerto Rico, STU = Sri Lankan Tamil from the United Kingdom, TSI = Toscani in Italia, YRI = Yoruba in Ibadan, Nigeria.

Asian populations, *CYP2D6**10 (rs1065852, Pro34Ser) is a common mutant with a mutation frequency of approximately 17.4% and an allelic mutation frequency of 45.7%.[30] Based on the data from the 1000 Genomes project, the frequency of minor allele (G) in Chinese population CHB, CDX, and CHS is 0.371 to 0.398, respectively, while the minor allele is A base in other races. In our research, the frequency of minor allele (G) was 0.31 in Lisu. The change of amino acid leads to decreased enzyme activity and metabolic conversion rate. A meta-analysis,[31] including 15 research with 1794 Asian breast cancer patients, revealed that the enzyme activity and metabolic conversion rate was low among the patients with *CYP2D6**10/*10 (TT) genotype. A correlation study between *CYP2D6* gene polymorphism and Tam and metabolite plasma concentrations revealed that the mean plasma concentration of Tamoxifen in poor metabolizers and intermediate metabolizers was only 25% and 55%, respectively, of the fast metabolizing type (extensive metabolizers + ultrarapid metabolizers).[32] Accordingly, female breast cancer patients with *10 variants should increase their Tamoxifen dose of >20 mg/d. In a study of the association between the *CYP2D6* gene polymorphism and early onset preeclampsia patients and the effect of labetalol, the frequency of the *CYP2D6* rs1065852 "G" allele in patients with ineffective labetalol is higher than that of the therapeutically effective one. Thus, the "G" allele of the rs1065852 may be associated with the therapeutic effect of labetalol.[33] The optimal drug dose should be based on the genotype of individual Lisu patients.

However, intrinsic limitations still exist in our study. Our sample size of Lisu is relative small, and further investigation in a larger cohort of Lisu is necessary to ascertain the generalizability and extrapolation of our results to these and other conditions in the Lisu population. In the follow-up study, we will conduct an in-depth study of the polymorphic sites with differences, and analyze the effects of mutations on the dose of different diseases.

In conclusion, *CYP2E1* rs2070676 was different in the Lisu population compared with 26 individuals from the 1000 Genome project. *PTGS2* rs5275 and *CYP2D6* rs1065852 were different in the Lisu population compared with most of the populations. Due to the difference in genotype distribution frequency of SNPs in genes affecting drug metabolism, the appropriate drug dose should be chosen to ensure the safety and efficacy of the drug in certain group.

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