Applying Pharmacogenomics in Drug Therapy of Cardiovascular Disease

Ye Zhu[a], Paul Y Takahashi[b,c], Naveen L Pereira[c,d], Eric T Matey[e], and Bijan J Borah[a,c], [a]Robert D. and Patricia E. Kern Center for the Science of Health Care Delivery, Mayo Clinic, Rochester, MN, United States; [b]Division of Community and Internal Medicine, Department of Internal Medicine, Mayo Clinic, Rochester, MN, United States; [c]Mayo Clinic College of Medicine and Science, Rochester, MN, United States; [d]Division of Circulatory Failure, Department of Cardiovascular Medicine, Mayo Clinic, Rochester, MN, United States; [e]Ambulatory Care Pharmacist-Pharmacogenomics, Department of Pharmacy, Mayo Clinic, Rochester, MN, United States

Glossary

Cost-effectiveness A measure in economic evaluation to report the relative values of social resource needed in one intervention compared to the return of the investment in health outcomes. It is usually used to compare the economic efficiency between new interventions and the standard intervention when the effectiveness of the new intervention is comparable or higher.

Genetic frequency The relative frequency of a specific gene variant or allele in a population compared to total copies of the specific gene or allele in that population.

Genotype Collection of genes inherited from individual's biological parents. It can also be referred to the specific gene sequences on two alleles. Expressed genotype is referred to the encoded gene(s) being used to generate RNAs and amino acids and consequently to make up proteins. Different gene sequences determine different genotypes.

Phenotype The individual's observed characteristics, such as sex, skin color, blood type, etc. In pharmacogenomics, phenotype can be observed speed the drug was distributed or metabolized in the body. It can be affected by the enzyme activity. Phenotype can be the results of the genotype, gene regulation and other biological factors, and it can change over time.
Polymorphism The variants of DNA sequence for a particular gene. For majority of the times, DNA sequences should be the same to lead to the same function. Mutations occur to some genes which lead to the change of one or more nucleotides (A, T, C, or G). The change could (but not necessary) lead to the different expression level of genes and consequently result in abnormal function of the body. In pharmacogenomics, polymorphism can lead to change in drug responses.
Quality-adjusted life year A measure of one's life span with both of quality of life and health status taken into consideration. It aggregates one's quality of life and the length of life into a single number. Living in perfect health for 1 year is usually recorded as the value of 1, while death is usually recorded as the value of 0. It can be used to report one's health outcomes with disease burden incorporated.

Abbreviations

ACMG	American College of Medical Genetics and Genomics
CAD	Coronary artery disease
CPIC	Clinical Pharmacogenetics Implementation Consortium
CPNDS	Canadian Pharmacogenomics Networks for Drug Safety
CVD	Cardiovascular disease
DPWG	Dutch Pharmacogenetics Working Group
FDA	US Food and Drug Administration
NHLBI	The National Heart, Lung, and Blood Institute
PCI	Percutaneous coronary revascularization
PCSK 9	Proprotein convertase subtilisin/kexin type 9
PGx	Pharmacogenomics
QALY	Quality-adjusted life year

1 Current status of pharmacogenomic application in cardiovascular disease

1.1 Overview of pharmacogenomics in cardiovascular disease

Pharmacogenomics (PGx) is a broad study area that investigates the association between individual's genetic polymorphisms and drug distribution, metabolism, drug-drug interactions and variations in the body's responses to the drug (Weinshilboum, 2003; Weinshilboum and Wang, 2004).

Pharmacogenomics is sometimes used interchangeably with pharmacogenetics, but these two terms have slight differences. The term of pharmacogenetics was initially used to describe the individual's DNA sequences of genes and their regulations which are found to be related to drug responses. Patients with mutated genes cannot metabolize the corresponding drug into a deactivated or activated form with a rate that was expected in clinical practice. This differential drug metabolization rate results in an either higher or lower drug level in patient's body after the patient is given a clinically standard dosage, and consequently lead to undesired adverse events. Incorporating patient genotype information into clinical decision-making can help predicting patient's responses to the drug, adjusting dosages, and potentially preventing drug-related adverse events, thus helping to facilitate an individualized disease management plan. However, genotype of a single gene was found to only address a part of the impact. The polymorphisms of multiple genes were found contributing to variations in drug response (CPIC, 2019). In fact, majority of individuals carry multiple genes that are associated with varying dose responses, and only a few carry single gene variants. It was reported that approximately 58% of individuals carried three or more of identified genes, 31% had two genetic variants, and only 10% had one gene variant (Ji et al., 2016). Using information from multiple genes provide a more comprehensive understanding of patient's treatment responses.

With the recent developments in genomic science, most significantly the achievement of Human Genome Project, and the expansion of our knowledge on gene sequencing, pharmacogenetics has transformed into PGx (Evans and McLeod, 2003). Drug response can now be investigated at the genome-wide sequencing level instead of analyzing microarray of genes at a time. This transformation parallels the development of studies on gene regulation and expression (e.g., transcriptomics, proteomics, etc.), which allow phenotypes of complex drug response to be explored and is a considerable future direction of PGx therapy.

1.1.1 Pharmacokinetics in cardiovascular disease management: Drug metabolism

As mentioned in previous chapters, pharmacokinetics is the field of study that investigates how a drug is metabolized in the body. It provides knowledge on drug absorption, distribution, metabolism and elimination (Evans and McLeod, 2003; Weinshilboum, 2003). Majority of the PGx-guided CVD management are under this category, where genetic polymorphisms are identified and the relationship between these polymorphisms and variations in the pharmacokinetics processes are established. For example, warfarin metabolism is involved in multiple enzymes. *CYP2C9* gene encodes a liver enzyme that metabolizes warfarin into inactive metabolites. *CYP2C9* genotypes are found to affect the plasma level in the body (Rettie et al., 1994; Scordo et al., 2002; Van Schie et al., 2012). The wild type is the allele CYP2C9*1, which is carried by majority of people and is considered as "normal" genotype. Genotypes of CYP2C9*2 and CYPE2C9*3 are recognized as two most common variants among European ancestry, and CYP2C9*5, *6, *8, and *11 are the most common variants in African ancestry (CPIC, 2019). These variants are associated with decreased activity of CYP2C9 enzyme, slowing down the clearance rate of warfarin, and lead to higher warfarin levels when standard clinical dose of warfarin is prescribed. If the variant information is available before a patient initiates warfarin, clinician could adjust the dosage or select an alternative medication (e.g., novel anti-coagulants) for the patient and could prevent bleeding events due to higher warfarin levels. Thus, a patient's genetic information can be used to understand his or her potential response to the drug and help control the drug's pharmacokinetic process.

1.1.2 Pharmacodynamics in cardiovascular diseases management: Drug targets

Pharmacodynamics studies the mechanism of a drug targeting the body, pathogen, or cancer cells. It outlines the molecular, biochemical, and physiologic effect of the drug. Recently, a few PGx-guided CVD therapies under this category have emerged. For example, it was reported that medications targeting the renin-angiotensin-aldosterone system (RAAS), including angiotensin converting enzyme (ACEIs), antagonists of angiotensin receptors (ARBs), renin inhibitors and aldosterone antagonists, were more effective in individuals with certain genotypes and gender (Franconi and Campesi, 2014). However, the evidence is inconclusive and PGx-related information for these medications has not been approved or ready to be used in practice.

1.1.3 Pharmacogenomics-guided therapy for cardiovascular diseases

PGx-guided drug therapy holds considerable potential for individualizing cardiovascular disease management and improving quality of care by reducing the patient's risks of experiencing drug-related adverse events. First, cardiovascular diseases are one of the most prevalent diseases in the United States and worldwide (Benjamin et al., 2018). Any advancement of disease treatment and patient management will benefit a wide range of population. PGx-guided therapies provide opportunities for tailored treatments to reach patient populations with variety of genetic background and maximize the benefit of treatment on a large scale. Second, cardiovascular diseases, such as hypertension and coagulation disorders, are carried in a chronic form, which demand large efforts in disease management. Therapies that help reduce the treatment burden on patients and provide fastest drug responses could help with delivering care and improve quality. PGx holds the potential to simplify the disease diagnosis and monitoring process, shorten the clinical decision-making time, and at the same time ensure the quality of care. Third, cardiovascular disease, such as coagulation disorders and stroke, have high risk of developing emergency cases and fatal outcomes, which require short response time and fast yet accurate clinical decisions. Any lack of information or misinformation will lead to potentially harmful care and undesired treatment outcomes. For example, patients who receive anticoagulation treatments (e.g., warfarin) require close monitoring of drug responses, in order to maintain a safe and effective therapeutic coagulation state. Unintentional low dosage (without presence of other anticoagulants) may lead to thromboembolic events, while high dosage may lead to bleeding events. Both are undesired outcomes and potentially fatal to patients. PGx-guided therapies can help clinicians predict patient's response to drug, select appropriate therapies, and/or take proactive actions to prevent adverse events. In this way, PGx can play a critical role in improving patient safety especially for the treatment with narrow therapeutic windows. Fourth, cardiovascular diseases impose a significant economic burden both to the patients and to the US health care system, with the systemwide costs of $555 billion annually and was projected to cost $1.1 trillion in 2035 (Dunbar et al., 2018; American Heart Assocation, 2017).

Advancement in targeted therapies and tailored disease management in a cost-effective manner will potentially help reduce the costs and improve patients' quality of life. With the advancing of genomic technologies, the cost of PGx testing, which relies largely on new genetic sequencings technologies, have decreased approximately by 1000 folds during the past decades (Wetterstrand, 2020). Recent studies report that PGx-guided therapies have become more attractive to the stake holders including patients and payers and can be beneficial from societal perspective (Zhu et al., 2020b; Dong et al., 2020; Ademi et al., 2014). Thus, gene-drug response research in cardiovascular disease is among one of the frontiers in the pharmacogenomic research. The most well-established gene-drug pairs supported by evidence in clinical implementation in CVD management are clopidogrel-CYP2C9, warfarin-CYP2C9/VKROC1/CYP4F2, simvastatin-SLCO1B1; other emerging PGx-guided therapies that have been studied but evidence for implementation are yet well established are ACEI, ivabradine, NOAC and diuretics (Zhu et al., 2020a). There is emerging evidence that genetic testing for multiple genes included in a panel while managing CVD patients can be useful and cost-effective (Zhu et al., 2020b; Dong et al., 2020).

In this chapter, we will discuss current status and the new developments in pharmacogenomic implementation in cardiovascular disease management. Major barriers and future directions will be discussed subsequently.

1.2 Clinical evidence

While tremendous efforts are being made in broad and specific PGx research areas, there is a significant demand to review, incorporate, integrate all the efforts into one harmonized environment. In this sense, further efforts can be made to transform these research findings into implementable approaches and introduce them to fill the chasm between research and practice.

The Genomics and Targeted Therapy Group at US Food and Drug Administration (FDA) has been collecting and reviewing current evidence on pharmacogenomics biomarkers in drug discovery and clinical studies for years. This regulatory body unites the works from government, academia and industry, and provides advice on the applications of the genomic biomarkers into clinical practice. The FDA Table of Pharmacogenomic Biomarkers provides detailed information on the pharmacokinetics and pharmacodynamics of genes which can lead to heterogenous drug responses among individuals with various genotypes. While updated continuously, as of March 2021, a total of 431 drug-gene pairs have been listed as therapeutic products that may contain their pharmacogenomic information in their labeling (https://www.fda.gov/drugs/science-and-research-drugs/table-pharmacogenomic-biomarkers-drug-labeling). Among them, 19 drug-gene pairs are used in CVD therapies as shown in Table 1. For hydralazine, there are no specific genes identified to be associated with the drug responses. Instead, the absorption of hydralazine is associated with acetylation and the metabolism is associated with acetylation, ring oxidation and conjugation. Individuals who are fast acetylators can process hydralazine faster and will have lower exposure to the drug. Procainamide is listed as having association with *NAT*; however, the drug label information was not available in the FDA table. For nebivolol, prasugrel and ticagrelor, labeling should suggest that their absorption and metabolism are NOT dependent on the corresponding genotypes.

Table 1 lists the selected drug-gene pairs from FDA Table of Pharmacogenomic Biomarkers in CVD management. The labeling text includes multiple sections and specific details on pharmacokinetics. For some drugs, the labeling text include some specific actions. Some others provide information that can be used in specific actions in clinical practice. We abstract the potential actions implied in the drug labeling and list them in the right-side column of the table.

The FDA Table of Pharmacogenomic Biomarkers elaborates the evidence that are at the pharmacokinetics and pharmacodynamics level, while contents that are more closely relevant to care outcomes are needed. In response to this demand, FDA further provides advice on the established impact of the pharmacogenetic association to guide the genetic testing. All this information is listed in the FDA Table of Pharmacogenetic Associations which is updated on a continuous basis (https://www.fda.gov/medical-devices/precision-medicine/table-pharmacogenetic-associations). As of March 2021, there are three types of impacts identified for a total of 102 medications and 107 drug-gene pairs: (1) support therapeutic management recommendations, (2) indicate a potential impact on safety or response, or (3) demonstrate a potential impact on pharmacokinetic properties only. Table 2 lists the 9 medications and 11 drug-gene pairs that are relevant to CVD management.

1.3 Clinical implementation

1.3.1 Clinical studies

With the growing body of evidence on pharmacogenomic associations based on pharmacokinetics and pharmacodynamics, efforts have been made to transform these findings into patient management. Clinical studies examine the impact of gene-drug associations in clinical environments and the effectiveness of genetic testing as a key factor (observational studies) or interventions (clinical trials). There is mixed evidence for which whether PGx-guided therapies can benefit patients in terms of disease control, adverse event prevention and decreasing mortality rate. The differences may be partially due to distinct study designs, perspectives and the observed timeframe for the study, but unidentified environmental factors and social factors could play important roles as well.

A case in point is the management of coronary artery disease (CAD). Clopidogrel-CYP2C19 associations have been proven in pharmacokinetic studies to impact the CAD management. Clopidogrel is a type of P2Y12 inhibitors which are used to prevent platelet aggregation for patients who are prone to thrombotic events. It is typically used in patients who experienced coronary artery stenosis and after percutaneous coronary revascularization (i.e., PCI). The standard procedure for the new onset coronary artery stenosis is to go under PCI and reopen the clotted artery as soon as possible (ideally less than 90 min). After the PCI procedure, patients are prescribed clopidogrel to prevent coronary artery restenosis. In the body, clopidogrel needs to be activated by liver enzyme CYP2C19 in order to be functional. It is found that the loss-of-function of *CYP2C19* leads to reduced CYP2C19 enzyme activity so that the bioactivation of clopidogrel is impaired in such patients (Table 2). This group of patients are recommended to use other types of P2Y12 inhibitors, which are not associated with CYP2C19 genotypes, such as prasugrel or ticagrelor (Table 1).

In a meta-analysis conducted by Holmes et al., 32 studies on CYP2C19 and patient outcomes were synthesized and 42,016 patients who underwent PCI, clopidogrel therapy and genetic testing were followed-up with a mean time of 24 months (Holmes et al., 2011). This study did a very comprehensive review of how CYP2C19 genotypes were associated with clopidogrel metabolism and CVD adverse events. The results suggested that even though the platelet function was reduced with the CYP2C19 mutation, there were no significantly higher CVD adverse events, including all-cause mortality, myocardial infarction, stent thrombosis and bleeding. The study also compared the outcomes of the most outstanding minor alleles that reduce the CYP2C19 function (*2 and *8) with the wildtype alleles (*1 and *17), and the results didn't suggest significant differences. However, the stent thrombosis potentially had a strongest association with CYP2C19 mutation. This study provided us information that even with *CYP2C19* gene mutated, this group of patients still had outcomes similar to patients with normal genotypes. This indifference could accredit the standardization of the medical care and the quality control at clinical practice. However, in this case, one can raise the question that

Table 1 FDA Table of Pharmacogenomic Biomarkers in drug labeling and implied actions (cardiovascular therapeutics).

Drug	*Biomarker*	*Implied actions in drug labeling*[a]	*Genotype info*
Carvedilol	CYP2D6	• Potent inhibitors of CYP2D6 isoenzyme (such as quinidine, fluoxetine, paroxetine, and propafenone) would be expected to increase blood levels of the R(+) enantiomer of carvedilol • CYP2D6 poor metabolizers had a higher rate of dizziness during up-titration	NA
Clopidogrel	CYP2C19	• CYP2C19 poor metabolizers consider use of another platelet P2Y12 inhibitor Approximately 2% of White and 4% of Black patients are poor metabolizers; the prevalence of poor metabolism is higher in Asian patients (e.g., 14% of Chinese) • Homozygous for nonfunctional alleles = CYP2C19 poor metabolizers	Yes
Hydralazine	Nonspecific (NAT)	• Absolute bioavailability dependent on the acetylator phenotype	NA
Isosorbide dinitrate	CYB5R	• Overdose leads to methemoglobinemia. The treatment of choice is methylene blue, 1–2 mg/kg intravenously • In patients with poor or no CYPB5 reductase activity, about 1 mg/kg of isosorbide dinitrate should be required before any of these patients manifests clinically significant (≥10%) methemoglobinemia In patients with normal CYPB5 reductase activity, larger doses of isosorbide required (up to 73.1–4.4 mg/h in clinical studies)	NA
Isosorbide mononitrate	CYB5R	• Overdose leads to methemoglobinemia. The treatment of choice is methylene blue, 1–2 mg/kg intravenously • In patients with poor or no CYPB5 reductase activity, about 2 mg/kg of isosorbide dinitrate should be required before any of these patients manifests clinically significant (≥10%) methemoglobinemia In patients with normal CYPB5 reductase activity, larger doses of isosorbide required (up to 7.8–11.1 mg/h in clinical studies)	NA
Metoprolol	CYP2D6	• Potent inhibitors of CYP2D6 isoenzyme (such as quinidine, fluoxetine, paroxetine, and propafenone) are likely to increase metoprolol concentration. These increases in plasma concentration would decrease the cardioselectivity of metoprolol • CYP2D6 enzyme is absent in about 8% of Caucasians (poor metabolizers) and about 2% of most other populations • Poor metabolizers will have increased (sevenfold) metoprolol level and decreased cardioselectivity	NA
Nebivolol	CYP2D6	• No dose adjustments are necessary for patients who are CYP2D6 poor metabolizers. The clinical effect and safety profile observed in poor metabolizers were similar to those of extensive metabolizers (19 h of half-life in poor metabolizers vs 12 h in extensive metabolizers)	NA
Prasugrel	CYP2B6/CYP2C9/ CYP2C19/CYP3A5	• In healthy individual, patients with stable atherosclerosis, or patients with ACS, no effect on pharmacokinetics is relevant to genetic variants of CYP2B6, CYP2C9, CYP2C19, or CYP3A5	NA
Procainamide	Nonspecific (NAT)	Labeling not electronically available on Drugs@FDA	NA
Propafenone	CYP2D6	• Drugs that inhibit these CYP pathways (such as desipramine, paroxetine, ritonavir, sertraline for CYP2D6; ketoconazole, erythromycin, saquinavir, and grapefruit juice for CYP3A4; and amiodarone and tobacco smoke for CYP1A2) can be expected to cause increased plasma levels of propafenone • Propafenone pharmacokinetics is linear relationship in slow metabolizers, while there is a greater-than-linear in extensive metabolizers • Approximately 6% of Caucasians in the U.S. population are naturally deficient in CYP2D6 activity and to a somewhat lesser extent in other demographic groups	NA
Propranolol	CYP2D6	• In healthy subjects, no difference was observed between CYP2D6 extensive metabolizers and poor metabolizers with respect to oral clearance or elimination half-life • In poor metabolizers, partial clearance to 4-hydroxy propranolol was significantly lower while higher to naphthyloxy lactic acid	NA
Quinidine	CYP2D6	• Inhibit CYP2D6 enzyme, effectively converting CYP2D6 extensive metabolizer into poor metabolizers. Take caution and reduce the dosage of CYP2D6 dependent therapy when use quinidine • Less than 1% of Asians, in about 2% of American blacks, and in about 8% of American whites	NA
Rivaroxaban	F5 (Factor V Leiden)	• Factor V Leiden gene mutation (4%) in population composed of 56% male, 70% Caucasian, 14% Asian and 3% Black, with mean age was approximately 59 years High EVT and/or PE risks	NA
Tafamidis	TTR	• Stabilized both the wild type TTR tetramer and the tetramers of 14 TTR variants tested clinically after once-daily dosing • Tafamidis also stabilized the TTR tetramer for 25 variants tested ex vivo	NA
Ticagrelor	CYP2C19	• Ticagrelor adverse events did not depend on CYP2C19 loss of function status	NA
Warfarin	CYP2C9/VKORC1	• If the patient's CYP2C9 and/or VKORC1 genotype are known, FDA provided dose range for initial dose • Patients with CYP2C9 *1/*3, *2/*2, *2/*3, and *3/*3 may require more prolonged time (>2–4 weeks) to achieve maximum INR effect for a given dosage regimen than patients without these CYP variants • Patients with one or more variant CYP2C9 alleles (CYP2C9*2 and CYP2C9*3) have decreased S-warfarin clearance • CYP2C9 alleles associated with reduced enzymatic activity frequencies: 11% for CYP2C9*2 and 7% for CYP2C9*3 in Caucasians. *5, *6, and *11 alleles in populations of African ancestry and *5, *9, and *11 alleles in Caucasians	Yes

(*Continued*)

Table 1 (Continued)

Drug	*Biomarker*	*Implied actions in drug labeling*	*Genotype info*
	PROS1/PROC	• FDA provides initial dose rang for CYP2C9 and VKORC1 genotype information • Low or deficiency of protein C (coded by gene PROC) and protein S (coded by gene PROS1), have been associated with tissue necrosis following warfarin administration. Concomitant anticoagulation therapy with heparin for 5–7 days during initiation of therapy with COUMADIN in these patients	NA

[a]Implied actions were abstracted from FDA labeling text, accessed at https://www.fda.gov/drugs/science-and-research-drugs/table-pharmacogenomic-biomarkers-drug-labeling, in the downloadable detailed version.

Table 2 FDA Table of Pharmacogenetic Associations (cardiovascular therapies).

Drug	*Gene*	*Affected subgroups*	*Description of gene-drug interaction*	*Pharmacogenetic associations for which the data*		
				(A) Support therapeutic management recommendations	*(B) Indicate a potential impact on safety or response*	*(C) Demonstrate a potential impact on pharmacokinetic properties only*
Carvedilol	CYP2D6	Poor	metabolizers	Results in higher systemic concentrations and higher adverse reaction risk (dizziness)	–	Yes
Clopidogrel	CYP2C19	Intermediate or poor	metabolizers	Results in lower systemic active metabolite concentrations, lower antiplatelet response, and may result in higher cardiovascular risk. Consider use of another platelet P2Y12 inhibitor	Yes	–
Hydralazine	Nonspecific (NAT)	Poor	metabolizers	Results in higher systemic concentrations	–	–
Metoprolol	CYP2D6	Poor	metabolizers	Results in higher systemic concentrations	–	–
Nebivolol	CYP2D6	Poor	metabolizers	May result in higher systemic concentrations	–	–
Procainamide	Nonspecific (NAT)	Poor	metabolizers	Alters systemic parent drug and metabolite concentrations. May result in higher adverse reaction risk	–	Yes
Propafenone	CYP2D6	Poor	metabolizers	Results in higher systemic concentrations and higher adverse reaction risk (arrhythmia). Avoid use in poor metabolizers taking a CYP3A4 inhibitor	Yes	–
Propranolol	CYP2D6	Poor	metabolizers	May affect systemic concentrations	Yes	–
Warfarin	CYP2C9	Intermediate or poor	metabolizers	Alters systemic concentrations and dosage requirements. Select initial dosage, taking into account clinical and genetic factors. Monitor and adjust dosages based on INR	Yes	–
Warfarin	CYP4F2	V433M variant carriers	May affect dosage requirements. Monitor and adjust doses based on INR	Yes	–	–
Warfarin	VKORC1	-1639G > A variant carriers	Alters dosage requirements. Select initial dosage, taking into account clinical and genetic factors. Monitor and adjust dosages based on INR	Yes	–	–

Note: Information listed in the left four columns were abstracted from the FDA table texts.
FDA Table of Pharmacogenetic Associations (https://www.fda.gov/medical-devices/precision-medicine/table-pharmacogenetic-associations).

what if patients with CYP2C19 loss-of-function were treated differently with other genotype insensitive P2Y12 inhibitors, would they receive better outcomes if medications were changed? If so, we could improve the quality of care for this group of patients.

In a recently completed clinical trials led by Pereira et al., the effect of PGx-guided clopidogrel on patients who received percutaneous coronary intervention was examined (the TAILOR-PCI Randomized Clinical Trial) (Pereira et al., 2020). This study included 5302 patients with acute coronary syndromes (ACS) or stable coronary artery disease (CAD). In the TAILOR-PCI trial, patients in the treatment arm received CYP2C19 genetic testing before they were prescribed P2Y12 inhibitors. If patients were identified to carry CYP2C19 loss-of-function, they were given ticagrelor. Otherwise, they were prescribed clopidogrel and under usual care. For the patients under control arm, none was given the PGx test and everyone received clopidogrel treatment. The results suggested that after 12 months of follow-up, patients with genetic testing did not receive significantly better outcomes than non-genetic testing group. This clinical trial suggested that between genotype and patient outcomes, there might be "broken links" that stopped the effect of patient's drug-responses passing on to disease progression and adverse events. These broken links might be strengthened in the PGx implementation process other than pharmacokinetics and pharmacodynamics. For example, patients who are taking warfarin were under close monitoring of blood coagulation function may show stabilized coagulation status. During this time, patients with higher risk of bleeding might need more efforts in dosage adjustment and longer monitoring time compared to patients with lower risk of bleeding. In this case, higher risk patients don't receive worse outcome, but they were under more treatment burden which may not have been reflected in clinical trials. In addition, management of these higher risk patients requires more provider efforts and resources from health care system, which are not often reflected in clinical trials as well.

In order to identify the challenges in the PGx implementation, there are some programs pioneered in this area. The Right Drug, Right Dose, Right Time-Using Genomic Data to Individualize Treatment (RIGHT 10K Study) is a study of clinical implementation of PGx-guided therapy. This study enrolled 10,074 participants with genetic information incorporated into the electronic health records (Bielinski et al., 2014). This study is making continuous efforts to synthesize multiple expertise together to support clinical decision making. Patients were surveyed for demographic information and other social factors. Blood samples were collected, genes were sequenced, and results were captured in patient's electronic health records regardless of patient's disease diagnosis and the need for medication therapies. When any of these patients is prescribed a PGx-relevant medication, the electronic health system alerts the prescribing clinician about the patient's genetic information. Patients' outcomes will be evaluated consequently. The RIGHT study provides important information for clinical implementation of genetic information and be influential in evolving best clinical practice and improve quality of care.

In an effort to explore and develop personalized medicine, Vanderbilt University launched a PGx implementation project, the Pharmacogenomic Resource for Enhanced Decisions in Care and Treatment (PREDICT) (Pulley et al., 2012). This program explored preemptively PGx testing and incorporating the genotypes into patients' electronic health record system in a large-scale patient population. Initially 184 gene variants were included in the analytical plan for genotyping. This program not only focused on PGx implementations on the practice side, but also explored patient education and preparedness (MyHealthAtVanderbilt.com). The PREDICT program provides very comprehensive information from multiple perspectives, including institutional patient, provider, institutional and clinical laboratory perspectives.

Another stream of efforts has been made in order to link the genetic testing with patient outcome. The Phenome-Wide Association Studies (PheWAS) utilize electronic health record-based information including ICD diagnoses, patient symptoms and epidemiology-based data, to generate patients' phenotype (Bush et al., 2016). The phenotype is different from the genotype. Genotype reflects patient's drug-responses in relation with patient's genetic sequences (e.g., CYP2C19 loss-of-function). Phenotype reflects the actual drug's responses in patient's body (clopidogrel stopped platelet from aggregating). Theoretically, phenotype has better association with patient outcome, since it directly reflects patient's drug responses. However, the measurement of phenotype takes tremendous amount of efforts. For example, in order to measure clopidogrel's effect, researchers must monitor patient's blood sample or patient's healthcare utilizations frequently in order to understand patient's status. It is very hard to predict patient's adverse events since the results only reflect the moment when blood is collected. On the other hand, patient's genotype is consistent throughout patient's life. Once tested, results will be valid during lifetime in most of the cases. Therefore, if phenotype could be predicted by genotype with other factors, it would greatly reduce the measurement burden and improve our ability in predicting adverse events. The PheWAS study is promising in linking PGx genotype with phenotypes and is expected to create substantial impact in clinical practice.

1.3.2 PGx testing in real-world clinical practice

There are multiple ways to conduct PGx testing in the real-world practices. When making clinical decision to conduct a PGx test, one should make multiple considerations: When to test? What to test? Who to test? We will introduce the test strategies that have been studied in CVD management in this section. Each of the test strategies has its advantages and disadvantages, and its level of uptake in the real-world clinical practice will depend on the cost-effectiveness analysis, which will be discussed in Section 1.6.1.

1.3.2.1 Testing timing : Reactive and proactive (preemptive) pharmacogenomic testing

1.3.2.1.1 Reactive testing

Implementation of reactive PGx testing in cardiology involves ordering PGx testing when the situation or need arises. In clinical medicine, this commonly occurs when a medication like clopidogrel or warfarin is needed. These reactive tests are used to avoid any adverse drug reaction like in the anticipated use of mercaptopurine or azathioprine (Weinshilboum and Wang, 2017). The

advantage of reactive ordering of CYP 2C19 for clopidogrel or CYP 2C9/VKORC1 for warfarin involves a higher likelihood of insurance coverage and a more specific test for a specific medication. Reactive testing is the natural method of getting a clinical test for a specific clinical indication or illness. If a patient requires either clopidogrel for prevention of stent re-thrombosis or uses warfarin for anticoagulation, the appropriate PGx test could be ordered.

However, the reactive approach has the inherent challenge of the delay in testing which may limit the potential impact of the testing on medication dosing. Both clopidogrel and warfarin may require immediate medication administration. Providers utilize clopidogrel for acute coronary syndromes and to prevent cardiac stent thrombosis (Levine et al., 2016b). These coronary conditions are often urgent and require immediate initiation of therapy. In warfarin, the potential advantage of reactive PGx testing involves modification of the initiation of warfarin as well as modification of starting doses of warfarin (Gage and Lesko, 2008). Warfarin is often used for acute diseases like deep venous thrombosis (Kearon et al., 2016), pulmonary embolism, stroke prevention in atrial fibrillation (January et al., 2019) and for thrombosis protection in patients with mechanical heart valves (Singh et al., 2019). For some of these conditions like atrial fibrillation, the use of direct oral anticoagulants is recommended and is the standard of care; however, the use of warfarin is still important for many. Providers are often not willing to wait for testing. As another potential challenge, many providers who would initiate either clopidogrel or warfarin may not be knowledgeable with pharmacogenomics (Johansen Taber and Dickinson, 2014).

1.3.2.1.2 Proactive (preemptive) testing

The use of proactive PGx testing has a much broader potential application for pharmacogenomics in the future. As the name suggest, proactive PGx testing requires performance of the PGx test prior to clinical indication and prior to potential medical treatment like clopidogrel (Pulley et al., 2012). In many cases, this PGx testing could be done as a PGx panel or specific test for a previous specific indication. As costs decrease, the use of whole exome or genome sequencing could be done for patients which would include pharmacogenomics testing.

The most important potential advantage of this method involves the application of drug or dosing changes at the time of prescription. Once the proactive testing is completed and in the electronic medical record, there could potentially be a long-standing advantage for clinical prescribing (Mukerjee et al., 2018). Clinical decision support further helps the provider understand the role of the phenotype and helps with dosing of each medication. This type of support will be critical as new knowledge and information are added about medications in the future (Caraballo et al., 2017).

The inherent limitations of proactive testing involve its implementation and cost. With any major clinical implementation, the translation of science to clinical practice often remains challenging. Many clinicians may be interested in PGx and believe it will be useful for patients (Peterson et al., 2016). However, there remain challenges with practical aspects of using proactive testing including management of patients currently tolerating a medication with a newly discovered extreme phenotype (Peterson et al., 2016).

1.3.2.2 Screening strategies: Cascade and general pharmacogenomics testing

1.3.2.2.1 Cascade screening

Cascade genetic testing is the common method for testing for patients with potential adverse phenotypes. In cascade testing, the patient with an adverse phenotype would trigger family members to be tested. In the case of clopidogrel or warfarin, this would involve testing family members if a genetic variant caused clopidogrel to be less effective. The widespread use of cascade testing for extreme pharmacogenomic phenotype is not currently standard practice (Roosan et al., 2021). The use of cascade testing would involve family members discussing their pharmacogenomic phenotype status with other family members. In clinical practice, family history often involves illness and not efficacy or side effects of medications.

Cascade testing has potential advantages as this allows a more focused use of genetic testing. The common usage of cascade testing involves the diagnosis of heritable cancers like hereditary breast and ovarian cancer and Lynch syndrome. The application of cascade testing for pharmacogenomics is less well defined. Thus, in cardiovascular disease, cascade testing may apply to heritable illnesses like cardiac dysrhythmia or familial hyperlipidemia. In cardiovascular disease, cascade testing for familial hyperlipidemia may be cost-effective with a cost of 3564 Australian dollars/QALY (Ademi et al., 2014). For pharmacogenomics, cascade testing may help serious adverse side effects like Stevens Johnson syndrome. The application of cascade testing for efficacy of cardiovascular medications (clopidogrel, warfarin) is not being used to our knowledge.

The potential challenges of cascade testing involve many barriers. Many people may not be willing to undergo the testing even if faced with potential life changing illnesses (Bednar et al., 2020). There are practical matters of contacting family members and who and how that should be accomplished (Sturm, 2016). Medical providers may lack knowledge in genetic testing and may miss key chances to provide cascade testing if they are not familiar with genetics (Bellcross et al., 2011).

1.3.2.2.2 General screening

General screening or proactive screening for cardiovascular pharmacogenomics would involve using pharmacogenomics testing for patients irrespective of family history of reactions to medications or previous known phenotype. The use of proactive screening for cancer conditions is becoming a bit more commonplace (Evans et al., 2020). This is the common method of pharmacogenomics application as many patients who could potentially use pharmacogenomics do not readily know how family members react to medications or if they have an abnormal phenotype. General screening for cardiovascular pharmacogenomics has become more

popular as the cost of testing has decreased. Increasingly, pharmacogenomics will be proactive as patients have either single gene or panels of pharmacogenomics performed. This has the greatest potential for help as information will be available at the point of care about medication use. Reactive general testing is still a potential option as patients move forward.

1.3.2.3 Testing assay strategies: Single-gene assay and general panel testing

1.3.2.3.1 Single-gene assay

In the early days, genetic testing was conducted one gene at a time. It provides only the relevant gene information and is cost-saving. However, if multiple genes need to be tested using numerous single assays, the cost will be higher than multiple genes in one test. Single-gene assay has an advantage as it deals with a specific gene for a specific medication or clinical indication, and therefore the results can be obtained faster than testing multiple genes at the same time (e.g., panel testing).

1.3.2.3.2 Panel testing

With the advancement of gene sequencing technologies, multiple genes assays can be done at one time, which is often described as panel testing. Panel testing can have many advantages. First, the differences in drug response is often associated with a combination of gene variants (e.g., warfarin-CYP2C9/VKORC1/PROS1/PROC). Second, patients often experience multiple diseases comorbidities or diseases which require multiple medications. For example, if patients develop hypercholesteremia and need statin therapy, they are prone to coronary artery disease and likely to need clopidogrel therapy. In this case, testing CYP2C19 could facilitate future clinical decisions. Third, patients often have multiple genetic variants that may be associated with drug responses (Ji et al., 2016). Given all the reasons above, genetic panel testing may be more efficient than single-gene assay if adopted in clinical practice. Furthermore, genetic panels can be used to predict patients' risk for adverse events and death. Pharmacogenomic polygenic response score can be used to predict ischemic events and death (Lewis et al., 2020).

1.3.2.4 PGx test adoption

While the PGx testing provides information from the highest-end technologies using Genomic science, the adoption of these tests should not differ much from the traditional clinical lab tests. However, with PGx testing, the major advantage is the potential benefit for future medication use. Thus, PGx testing, specifically prospective PGx testing, may not be specific to treating a single clinical condition. Table 3 listed the comparison between genetic testing and current clinical lab tests in CVD management.

1.4 Guidelines for practice

Implementing pharmacogenomics into clinical practice is a challenging process that takes significant amount of efforts from all stakeholders. It requires not only the scientific evidence from pharmacokinetics and pharmacodynamics mechanisms, but also considerations from care providers' perspectives regarding how to adopt this process in their routine practice. It requires knowledge on how to use results and how to modify medication choices. Beside this, widespread cooperation from pharmacists and physicians are needed to translate PGx evidence into clinically actionable prescribing information. As we discussed in Section 1.2, FDA provides suggestions on drug dosing and clinically actionable information based on pharmacokinetics and pharmacodynamic, which is considered as established evidence of PGx's efficacy. However, the efficacy is not necessarily translated into patient outcomes. As we have discussed in Section 1.3, there are emerging studies exploring the associations between genetic testing and drug dosing or drug choices and patient outcomes. However, these studies reached divergent conclusions. In light of this circumstances, strategic plans are required to examine the study results, systematically organize evidence, develop recommendations and effectively communicate the recommendation to practicing clinicians, patients and payers for PGx adoption. It will be critical to continuously review the evidence and develop guidelines for the clinical practice. In this section, we discuss some of the prominent efforts being made by several professional societies in the space of guideline development for PGx adoption and organizations piloting PGx implementation with distinct focuses and strategies.

1.4.1 Clinical Pharmacogenetics Implementation Consortium (CPIC)

Clinical Pharmacogenetics Implementation Consortium (CPIC) (https://cpicpgx.org/) is an international society comprising experts in pharmacogenomics and clinical care, which is dedicated to facilitate PGx implementation in patient care. CPIC provides

Table 3 Comparison between pharmacogenomic testing and traditional clinical lab tests in CVD management.

	PGx testing	*Clinical lab test*
Test ordered	Commercial Clinician prescribed Specific or future disease state	Clinician prescribed Specific disease state
Test purpose	Screening	Diagnostic Screening
Test results	Association with disease is usually low	Association with disease can be strong

freely accessible and evidence-based guidelines updated on a semi-annual basis, which include peer-reviewed recommendations that focuses on how to translate the clinical lab results into patient care with the consideration of patient safety (Caudle et al., 2016). These guidelines are written following the Developing Trustworthy Clinical Practice Guidelines from the Institute of Medicine, which includes standardized literature review and grading algorithm, expertise from clinical care and research, and comprehensive peer-review approval process (Caudle et al., 2014). These guidelines are developed with the expectation that PGx will be used in large-scale populations in routine clinical care, PGx testing is available commonly in clinical lab and testing results will be available when the drug therapy needs to be started in a timely manner. These guidelines aim to prepare medical professionals for handling the PGx knowledge when it's translated successfully from bench to bedside and shorten the timeline of PGx research findings to clinical use. The CPIC guidelines do not provide recommendations on the testing strategies (e.g., when to test, who to test, or how to test).

As of March 2021, there are 25 guidelines for 25 medications or drug categories with 23 different genes and 85 drugs in all disease areas. Among them, there are only three guidelines covering three drugs and six genes in CVD management listed in Table 4: CYP2C19-Clopidgrel, CYP2C9/VKORC1/CYP4F2-Warfarin, and SLCO1B1-simvastatin. This is only a small portion of drugs with established pharmacogenomic associations when compared to FDA's guidance: there are 16 drugs and 17 drug-gene pairs in FDA Table of Pharmacogenomic Biomarkers in Drug Labeling (Table 1), and 9 drugs and 11 drug-gene pairs in FDA's Table of Pharmacogenetic Associations (Table 2). The focus of CPIC is to encompass patient outcomes that derive from clinical trials beyond the results from clinical pharmacokinetics. Practically, incorporating PGx with the strongest clinical evidence into clinical practice makes sense as a starting point.

CPIC guidelines provide methods that assign phenotype to each of the genotypes. Clinically actionable recommendations are based on phenotypes, and clinical evidence levels are listed as classification of recommendations (e.g., strong, or moderate). CPIC guideline is highlighted by providing guidance to multiple genetic polymorphisms. For example, the guideline for warfarin is listed by multiple combinations of genetic variants, including CYP2C9*2, *3 carriers, CYP2C9*2,*3 carriers with VKORC1 -1639 G>A, CYP2C9 *2,*3, *5, *6, *8, *11 carriers, CYP2C9*2,*3 carriers with rs2108622 T (CPIC, 2019). All genetic variants are by African or non-African population. The guideline for pediatric patients is examined by European or non-European population.

1.4.2 Recommendations from US professional societies in cardiology

Although the US cardiology societies haven't provided guidelines regarding PGx-guided therapy, they have been seeking ways to prepare the medical professionals for PGx implementation. In the United States, the current clinical guideline from American Heart Association and American College of Cardiology do not recommend testing CYP2C19 genetic polymorphism for the post-PCI patients who need clopidogrel because there is lack of evidence to support this decision (Levine et al., 2016a).

The National Heart, Lung, and Blood Institute (NHLBI) formed a working group meeting in 2011 to identify the priorities of cardiovascular pharmacogenomics. The working group reviewed and summarized the discussions from the preceding conference of *New Frontiers in Personalized Medicine: Cardiovascular Research and Clinical Care*, which was sponsored by NHLBI, Personalized Medicine Coalition, American College of Cardiology, American Medical Association, and Cheney Cardiovascular Institute at George Washington University (Musunuru et al., 2012). The working group reported four major priorities for the PGx implementation in CVD management (Musunuru et al., 2012):

(1) to establish standards of quality for the research enterprise,
(2) to establish robust systems for more rapid evidence generation,
(3) to harmonize regulatory and reimbursement standards, and
(4) to develop innovative partnerships to accelerate the development and implementation of personalized medicine applications.

The working group recommended that further investigation should be focused on three drugs with PGx implications: warfarin, clopidogrel and statins. In the recommendation, Warfarin-*CYP2C9/VKORC1* was suggested as the most promising gene-drug pair for PGx. The working group acknowledged that dabigatran could potentially be an anticoagulant alternative to warfarin. The working group also expressed concern about the uncertainties regarding the associations between *CYP2C19* genotype and patient outcomes in clopidogrel therapy; however, the group expressed special interest in the point-of-care testing for outpatient care. The genes identified to be potentially associated with statins were KIF6 and SLCO1B1, for which the working group called for clinical evidence regarding patient outcomes.

The working group acknowledged the FDA's advice for these three medications based on the pharmacokinetics evidence; however, it noted that evidence should come from the randomized clinical trials, which is considered as the gold standard for pharmacogenomic applications. Therefore, the groups recommended that large scale clinical trials, which is specifically designed for PGx studies to be conducted and funded, and the group called for collaborations among study teams in this areas regarding data sharing and validation.

The working group further recommended to include expertise of cardiovascular pharmacogenomics in guideline committees, such as "(a) American College of Cardiology/American Heart Association Task Force on Practice Guidelines (eg, clopidogrel pharmacogenomics). (b) National Cholesterol Education Program (eg, statin pharmacogenomics). (c) Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (eg, beta-blocker pharmacogenomics)".(Musunuru et al., 2012).

Table 4 CPIC guideline in cardiovascular disease management.

Drug	*Patient population*	*Gene or biomarker*	*Diplotype or variant allele*	*Phenotype*	*Therapeutic recommendation*	*Classification of recommendations*
Clopidogrel https://cpicpgx.org/guidelines/guideline-for-clopidogrel-and-cyp2c19/	Acute Coronary Syndromes undergoing percutaneous coronary intervention	CYP2C19	*1/*17, *17/*17	Ultrarapid metabolizer	Clopidogrel: label-recommended dosage and administration	Strong
			*1/*1	Extensive metabolizer		
			*1/*2, *1/*3, *2/*17	Intermediate metabolizer	Alternative antiplatelet therapy (if no contraindication), e.g., prasugrel, ticagrelor	Moderate
			*2/*2, *2/*3, *3/*3	Poor metabolizer	Alternative antiplatelet therapy (if no contraindication), e.g., prasugrel, ticagrelor	Strong
Warfarin https://cpicpgx.org/guidelines/guideline-for-warfarin-and-cyp2c9-and-vkorc1/	Non-African ancestry	CYP2C9	*2, *3	Abnormal metabolizer[a]	pharmacogenetic algorithm [b]	Strong
		VKORC1	-1639G>A			
		CYP2C9	*2, *3	Abnormal metabolizer[a]	Pharmacogenetic algorithm [b]	Optional
		CYP2C9	*2,*3, *5, *6, *8, *11	Abnormal metabolizer[a]	Decrease the calculated dose by 15–30%	Optional
		CYP2C9	*2, *3	Abnormal metabolizer[a]	Decrease the calculated dose by 5–10%	Optional
		CYP4F2	rs2108622 T			
		Others			Clinical dosage	
	African ancestry	CYP2C9	*2, *3	Abnormal metabolizer[a]	pharmacogenetic algorithm[b]	Moderate
		VKORC1	-1639G>A			
		CYP2C9	*2, *3, *5, *6, *8, *11	Abnormal metabolizer[a]	Decrease the calculated dose by 15–30%	Moderate
		CYP2C9	*2, *3	Abnormal metabolizer[a]	Decrease the calculated dose by 10–25%	Moderate
		VKORC1	-1639G>A rs12777823 A			
		CYP2C9	*2, *3, *5, *6, *8, *11 rs12777823 A	Abnormal metabolizer[a]	Decrease the calculated dose by 10–25%	Moderate
		Others			Clinical dosage	
	Pediatric European ancestry	CYP2C9	*2, *3	Abnormal metabolizer[a]	Pharmacogenetic algorithm[b]	Moderate
		VKORC1	-1639G>A			
		Others			Clinical dosage	
Simvastatin https://cpicpgx.org/guidelines/guideline-for-simvastatin-and-slco1b1/		SLCO1B1	*1a/*1a, *1a/*1b, *1b/*1b	Normal function; homozygous wild type or normal	Prescribe desired starting dose and adjust doses of simvastatin based on disease-specific guidelines[c]	Strong
			rs4149056 TT			
			*1a/*5, *1a/*15, *1a/*17, *1b/*5, *1b/*15, *1b/*17 rs4149056 TC	Intermediate function; heterozygous	Prescribe a lower dose or consider an alternative statin (e.g., pravastatin or rosuvastatin); consider routine CK surveillance	Strong
			*5/*5, *5/*15, *5/*17, *15/*15, *15/*17, *17/*17 rs4149056 CC	Low function; homozygous variant or mutant	Prescribe a lower dose or consider an alternative statin (e.g., pravastatin or rosuvastatin); consider routine CK surveillance	Strong

CPIC (2019) *Clinical Pharmacogenetics Implementation Consortium (CPIC®). Guidelines* . Available from: https://cpicpgx.org/guidelines/ (Accessed 25 April 2019).

[a]Not assigned any phenotype in the CPIC guideline. It is referred as abnormal metabolizer by authors.

[b]Gage BF, et al. (2008) Use of pharmacogenetic and clinical factors to predict the therapeutic dose of warfarin. *Clinical Pharmacology and Therapeutics* 84: 326–331; Klein TE, et al. (2009) Estimation of the warfarin dose with clinical and pharmacogenetic data. *New England Journal of Medicine* 360: 753–764.

[c]The US Food and Drug Administration recommends against 80 mg (unless already tolerated for 12 months).

1.4.3 Dutch Pharmacogenetics Working Group (DPWG)

The Dutch Pharmacogenetics Working Group was founded by Royal Dutch Pharmacist's Association and is a multi-disciplinary group of experts in medicine, pharmacy, clinical pharmacology, clinical chemistry, epidemiology, and toxicology (Swen et al., 2011). They develop recommendations and guidelines for gene-drug interactions in the context of level of evidence and clinical relevance (Swen et al., 2008). This group also provides methods in incorporating PGx information into electronic health care system: the therapeutic dose recommendations with calculation algorithms and the information has been incorporated in Dutch medication system. Alerts would be activated in all the steps during which a drug is prescribed and processed by pharmacists.

As of March 2021, there are 53 drugs and 11 genes included in the DPWG guideline. Among them, 14 drug-gene pairs are relevant to CVD management, which are listed in Table 5. Compared to CPIC, DPWG guideline includes drugs with no association with the identified genes and provides recommendations for "no dosage adjustment" for these drugs. For example, DPWG indicated that amiodarone, which is an antiarrhythmic medication, is associated with *CYP2D6* gene but indicated no dose adjustment is needed. This is very critical information in PGx implementation, that alternative medications need to be the ones that have no association with genetic polymorphisms. For example, it indicated that fluvastatin has no genetic interactions. This suggested that fluvastatin could potentially be used as alternative for simvastatin for patient with SLCO1B1 mutation.

In the DPWG guideline, patient population for clopidogrel therapy has been expanded to stroke and transient ischemic attacks, other than including only post-PCI patients as in CPIC guideline and the NHLBI working group recommendations. Thus, DPWG appears to translate clinical evidence into guideline in a timelier manner as several studies emerged to suggest that CYP2C19 genotype has associations with ischemic stroke and transient ischemic attacks (Patel et al., 2021; Wang et al., 2019; Alhazzani et al., 2017; Jia et al., 2013). The DPWG guideline also elaborates very detailed steps in patient care from the perspective of physicians and pharmacists. For example, for patients requiring lipid lowering therapy, the guideline suggested to choose alternative medication instead of simvastatin. It also provided guidance when alternative is not available, patients should avoid being given the dosage exceeding 40 mg/day. This guideline takes the real-world clinical situations into consideration—one of the barriers to PGx implementation is the un-availability of alternative medications. If alternative therapy is not available and the medication under consideration is the only treatment available, PGx testing is not meaningful. This topic will be further elaborated in Section 1.6.

The DPWG guideline provided guidance for each genetic genotype independently, but not for situations where patients might have multiple gene mutations. For example, the guideline's recommendations for warfarin therapies are listed by CYP2C9 genotypes or VKORC1 genotypes separately. It recommended to use 20% of the standard dosage for CYP2C9*3/*3 carriers, and 60% of the dosage for VKORC1 -1639 AA carriers (Swen et al., 2011). There was, however, no guidance for the patients with both CYP2C9*3/*3 and VKORC1 -1639 AA carriers.

1.4.4 Canadian Pharmacogenomics Network for Drug Safety

Canadian Pharmacogenomics Networks for Drug Safety (CPNDS) is a group that provides clinical guideline for PGx testing. It is a research community and an international network that serve the goal of reducing serious drug adverse reactions for both pediatric and adult population (https://cpnds.ubc.ca/). It developed a national surveillance network to monitor drug-related adverse reactions. DNA samples are collected, and clinical information was captured from all patients, regardless of the drug reaction status. Studies were conducted including both patient groups with and without drug adverse reactions. CPNDS maintains a large database with phenotypic data from a longitudinal cohort. Between 2005 and 2017, the database received 93,974 medication uses, among which 10,475 were adverse drug reactions. The network focus on pediatric population, with 72.6% adverse drug reactions were from pediatric patients.

As of March 2021, CPNDS provided clinical practice recommendations for CYP2C9 and VKORC1 genetic testing in warfarin therapy (Shaw et al., 2015). The recommendation suggested that within first 2 weeks of warfarin therapy, genetic testing of VKORC1 (-1639G>A), CYP2C9*2, and CYP2C9*3 should be conducted for both pediatric and adult patients. The recommendation didn't support testing for CYP2C9*5, *6, *8, or *11 and CYP4F2 (V433M), which is consistent with DPWG group and controversial with CPIC guidelines. One important reason for this discrepancy with CPIC guideline is that the majority races in CPNDS database are European (38.2%), Canadian (9.6%), and East Asian (4.9%), African ancestry was reported relatively low in its database. The focus of pediatric population contributed to this discrepancy as well. The recommendation is based on reviewing scientific evidence and patient outcomes from CPNDS database, which is a unique strategy compared to other professional societies. The starting point of guideline is to develop clinical practice guidance for drug adverse reactions that are most prevalent, while the starting point for other guidelines are the established evidence in pharmacokinetics.

The CPNDS recommendations suggested that potential dosage adjustment and alternative drug therapy need further investigation since the currently identified therapeutic effects can be explained and affected by both clinical and genetic factors that were beyond our current understanding; therefore, the recommendation indicated that it is still immature to provide any clinical therapeutic guidance.

1.4.5 PharmGKB clinical guideline annotations

The PharmGKB (https://www.pharmgkb.org) is pharmacogenomic knowledge resource that plays a prominent role in facilitating PGx knowledge exchange, dissemination and collaboration. In 2009, PharmGKB formed the CPIC in a joint effort with Pharmacogenomics Research Network (https://www.pgrn.org), by including external expertise in pharmacogenomics and laboratory medicine to develop PGx guidelines for clinical patient care. PharmGKB provides annotations of pharmacogenomic guidelines across multiple resources including CPIC, DPWG, CPNDS and other PGx societies. Table 6 lists the comparison of gene-drug pairs in CVD management included in the guidelines across these international societies.

Table 5 DPWG guidelines of PGx-guided CVD drug therapy recommendation.

Drug	*Patient population*	*Gene or biomarker*	*Diplotypes or variant allele*	*Pheno-type*	*Therapeutic recommendation*
Amiodarone	Nonspecific	CYP2D6			Not a gene-drug interaction
Atenolol	Nonspecific	CYP2D6			Not a gene-drug interaction
Atorvastatin		SLCO1B1	521 TC, 521 CC		– Choose an alternative rosuvastatin and pravastatin are influenced to a similar extent by SLCO1B1 polymorphisms but are not influenced by CYP3A4 inhibitors such as amiodarone, verapamil and diltiazem. Fluvastatin is not influenced by SLCO1B1 polymorphisms or CYP3A4 inhibitors – If an alternative is not an option: advise the patient to contact their doctor in the event of muscle symptoms
Bisoprolol	Nonspecific	CYP2D6			Not a gene-drug interaction
Carvedilol	Nonspecific	CYP2D6			Not a gene-drug interaction. Plasma concentration can be elevated, however, does not result in an increase in side effects
Clopidogrel	PCI, stroke, or TIA	CYP2C19		IM	Choose an alternative or double the dose to 150 mg/day (600 mg loading dose) Prasugrel, ticagrelor and acetylsalicylic acid/dipyridamole are not metabolized by CYP2C19 (or to a lesser extent)
				PM	Avoid clopidogrel Prasugrel, ticagrelor and acetylsalicylic acid/dipyridamole are not metabolized by CYP2C19 (or to a lesser extent)
				UM	No action required for this gene-drug interaction
Clonidine	Nonspecific	CYP2D6			Not a gene-drug interaction
Fluvastatin	Nonspecific	SLCO1B1			Not a gene-drug interaction
Metoprolol	Patient requiring gradual reduction in heart rate	CYP2D6		IM	Increase the dose in smaller steps and/or prescribe no more than 50% of the standard dose
				PM	Increase the dose in smaller steps and/or prescribe no more than 25% of the standard dose
				UM	Use the maximum dose for the relevant indication as a target dose If the effectiveness is still insufficient: increase the dose based on effectiveness and side effects to 2.5 times the standard dose or select an alternative Heart failure: bisoprolol or carvedilol. Bisoprolol: advantage: not metabolized by CYP2D6; disadvantage: elimination depends on the kidney function. Carvedilol: advantage: elimination does not depend on the kidney function; disadvantage is metabolized (to a lesser extent than metoprolol) by CYP2D6 Other indications: atenolol or bisoprolol. Neither is metabolized by CYP2D6
Prasugrel	Nonspecific	CYP2C19			Not a gene-drug interaction. Plasma concentration can be elevated, however, does not result in an increase in side effects
Simvastatin		SLCO1B1	521 TC		Choose an alternative. Consider any additional risk factors for statin-induced myopathy. Rosuvastatin and pravastatin are influenced to a lesser extent by SLCO1B1 polymorphisms. They are also not influenced by CYP3A4 inhibitors such as amiodarone, verapamil and diltiazem. Fluvastatin is not influenced by SLCO1B1 polymorphisms or CYP3A4 inhibitors If an alternative is not an option: (1) Avoid simvastatin doses exceeding 40 mg/day. (2) Advise the patient to contact their doctor in the event of muscle symptoms
			521 CC		Consider any additional risk factors for statin-induced myopathy. Rosuvastatin and pravastatin are influenced to a lesser extent by SLCO1B1 polymorphisms. They are also not influenced by CYP3A4 inhibitors such as amiodarone, verapamil and diltiazem. Fluvastatin is not influenced by SLCO1B1 polymorphisms or CYP3A4 inhibitors
Sotalol	Nonspecific	CYP2D6			Not a gene-drug interaction. Plasma concentration can be elevated, however, does not result in an increase in side effects
Ticagrelor	Nonspecific	CYP2C19			Not a gene-drug interaction. Plasma concentration can be elevated, however, does not result in an increase in side effects

(*Continued*)

Table 5 (Continued)

Drug	*Patient population*	*Gene or biomarker*	*Diplotypes or variant allele*	*Phenotype*	*Therapeutic recommendation*
Warfarin	Nonspecific	CYP2C9	*2/*2, *2/*3, *3/*3	IM	Use 65% of the standard initial dose: The genotype-specific initial dose and maintenance dose can be calculated using an algorithm. Algorithms for Caucasian patients usually contain only the *2 and *3 allele. If the activity of the reduced-activity alleles is comparable to the activity of *2 or *3, then the algorithm can be completed as if *1/*2 or *1/*3 is present. See https://www.knmp.nl/producten-en-diensten/gebruiksrecht-gstandaard/medicatiebewaking-g-standaard/background-information-pharmacogenetics for Excel files containing calculation modules for oral and equivalent intravenous doses. From day 6 on the standard algorithm without genotype information can be used to calculate the dose Modified dose algorithms have been developed for patients of African or (East) Asian heritage
			*2/*2, *2/*3, *3/*3	PM	Use 20% of the standard initial dose: The genotype-specific initial dose and maintenance dose can be calculated using an algorithm. Algorithms for Caucasian patients usually contain only the *2 and *3 allele. If the activity of the reduced-activity alleles is comparable to the activity of *2 or *3, then the algorithm can be completed as if *2 or *3 is present. See https://www.knmp.nl/producten-endiensten/gebruiksrecht-g-standaard/medicatiebewaking-g-standaard/background-information-pharmacogenetics for Excel files containing calculation modules for oral and equivalent intravenous doses. From day 6 on the standard algorithm without genotype information can be used to calculate the dose. Modified dose algorithms have been developed for patients of African or (East) Asian heritage
			*1/*3		Use 65% of the standard initial dose: The genotype-specific initial dose and maintenance dose can be calculated using an algorithm, as used in EU-PACT: see https://www.knmp.nl/producten-en-diensten/gebruiksrecht-g-standaard/medicatiebewaking-g-standaard/background-informationpharmacogenetics. From day 6 on the standard algorithm without genotype information can be used to calculate the dose
			*2/*2		Use 65% of the standard initial dose: The genotype-specific initial dose and maintenance dose can be calculated using an algorithm, as used in EU-PACT: see https://www.knmp.nl/producten-en-diensten/gebruiksrecht-g-standaard/medicatiebewaking-g-standaard/background-informationpharmacogenetics. From day 6 on the standard algorithm without genotype information can be used to calculate the dose
			*2/*3		Use 45% of the standard initial dose: The genotype-specific initial dose and maintenance dose can be calculated using an algorithm, as used in EU-PACT: see https://www.knmp.nl/producten-en-diensten/gebruiksrecht-g-standaard/medicatiebewaking-g-standaard/background-informationpharmacogenetics. From day 6 on the standard algorithm without genotype information can be used to calculate the dose
			*3/*3		Use 20% of the standard initial dose: The genotype-specific initial dose and maintenance dose can be calculated using an algorithm, as used in EU-PACT: see https://www.knmp.nl/producten-en-diensten/gebruiksrecht-g-standaard/medicatiebewaking-g-standaard/background-informationpharmacogenetics. From day 6 on the standard algorithm without genotype information can be used to calculate the dose
			*1/*2		No action required for this gene-drug interaction
		VKORC1	-1639 AA		Use 60% of the standard initial dose: The genotype-specific initial dose and maintenance dose can be calculated using an algorithm, as used in EU-PACT: see https://www.knmp.nl/patientenzorg/medicatiebewaking/farmacogenetica. From day 6 on the standard algorithm without genotype information can be used to calculate the dose
			-1639 GA		No action required for this gene-drug interaction

Table contents were extracted from Dutch guidelines November 2018 update, https://www.pharmgkb.org/guidelineAnnotation/PA166182815.

Table 6 Comparison of drug-gene pairs in CPIC, DPWG and CPND in CVD management.

Drug	*CPIC*	*DPWG*	*CPNDS*
Amiodarone	–	CYP2CD6[a]	–
Aspirin	CYP2C9[a]	–	–
Atenolol	–	CYP2D6[a]	–
Atorvastatin	–	SLCO1B1	–
Bisoprolol	–	CYP2D6[a]	–
Carvedilol	–	CYP2D6[a]	–
Clopidogrel	CYP2C19	CYP2C19	–
Clonidine	–	CYP2D6[a]	–
Fluvastatin	–	SLCO1B1[a]	–
Metoprolol	–	CYP2D6[a]	–
Prasugrel	–	CYP2C19[a]	–
Simvastatin	SLCO1B1	SLCO1B1	–
Sotalol	–	CYP2D6[a]	–
Ticagrelor	–	CYP2C19[a]	–
Warfarin	CYP2C9, CYP4F2, VKORC1	CYP2C9, VKORC1	CYP2C9

[a]No recommendation for this gene-drug pair.

1.5 Policy perspective

PGx implementation in large real-world populations is different from implementing PGx in clinical practice, as clinical guidelines play critical role in preparing clinicians how to handle the PGx results according to a patient's individualized situation. Implementing PGx in large-scale populations involves a more comprehensive decision-making process, which needs to be examined from various perspectives, including those of patients, health care providers, payers, ethics, and legal perspectives. These perspectives may not always be aligned which may limit the PGx adoption into large-scale populations. In this section, we will discuss the role of each perspective in policy formulation. The public policy perspective is not distinct for CVD management in terms of decision-making process but can be different regarding disease management, time-frame and priorities.

1.5.1 Economic evaluation

Economic evaluation is used in healthcare policy decision-making process while deciding to allocate scarce resources between alternative treatment options. For the past two decades, healthcare costs in the United States has been the highest among developed countries, but the quality of care has not been commensurate with the high cost (Tikkanen and Abrams, 2020). For example, healthcare costs in the United States in 2018 was 16.9% of GDP, while adjusted for cost of living, and it was 4.7% higher than Switzerland, which was the second highest among developed countries. On the other hand, life expectancy in the United States (78.6 years) is the lowest among all the developed countries, where people are living 2.5–5 years longer than the US residents. This suggested that the US healthcare system has been spending more money while receiving the lowest effects. The prevalence rate of complex patients, which is defined as patients with two or more chronic diseases, are the highest among all the developed countries. US population sufferers from the highest obesity rates compared to these countries as well. Cardiovascular diseases were not immune from this unmatched situation. The annual costs of cardiovascular disease for the US health care system is $214 billion, which is also associated with $138 billion productivity loss on job (Benjamin et al., 2018). Cardiovascular diseases are the cause for one third of all deaths in the United States, which kill 868,000 people every year (Benjamin et al., 2018). Thus, there is substantial need for CVD care, and unfortunately unmet demands from patients, especially from the poor and minority groups. With high costs and high demand for CVD disease management in the United States, healthcare policy decisions often precede economic evaluation, which evaluate the potential outcomes from new policy adoptions and the resources required for implementing such polices. The assumption underlying such economic evaluation is that there are limited societal resources (e.g., financial resource, labor time, spaces, etc.) and a group of patients must give up one/some resources in order to gain others. It is also assumed that decision making is at the population level instead of the individual level. In this way, economic evaluation helps with the allocation of scarce resources into different health sectors that can maximize the benefit in terms of patient outcomes at the population level.

PGx-guided CVD therapy presents new opportunities in tailoring therapies using patient's genetic information, which can potentially prevent undesired adverse events and maximize treatment benefit. With the advancing of the technology, costs of PGx tests have decreased to more affordable range. Alternative medications, usually are newly developed therapies, tend to have higher costs compared to compared to existing therapies but they may offer higher effectiveness. This necessitates evaluating trade-offs between costs and effectiveness, which is what economic evaluation offers.

1.5.1.1 Effectiveness

The outcome benefit in health care is usually defined by the effectiveness of an intervention strategy. For example, what we most care about from an intervention is the amount of health benefit to the patients, including the cumulative life years, quality of life,

and prevention of adverse events (e.g., cardiovascular deaths, hospitalizations, disease cases, drug adverse events, etc.). In studies with short time frame (e.g., infectious diseases), disease prevented is usually adopted as the measure of effectiveness. In severe disease with fast progression (e.g., hypertrophic cardiomyopathy, late stage cancer, etc.), death prevented is usually used as effectiveness measure. In chronic diseases such as CVD and cancer, patients tend to live for longer time but with decreased quality of life from disease progression, health-related quality of life is often used as measure of effectiveness (Karimi and Brazier, 2016). Quality-adjusted life year (QALY) is the one of most widely used measures for health-related quality of life. It aggregates a patient's quality of life and the length of life into a single number and is used to report patient's health outcomes with disease burden incorporated (Chang et al., 2020).

When PGx implementation is examined, the effectiveness of this intervention is compared with the current CVD management strategies, which is without PGx testing and prescribe medications according to clinical dosage. In terms of what new interventions or strategies will be considered, a fundamental assumption of economic evaluation in health care is not different from other medical interventions, which is "do no harm" in Hippocratic Oath, that is to say that only strategies that produce no less benefit for patient will be considered in treatment. In economic evaluation, it refers to being more effective, have less side effects, or could be cheaper given same effectiveness. Otherwise, the consequent steps of cost analysis are invalid.

PGx-guided CVD therapies, including clopidogrel and warfarin have been identified as effective than the current standard of care, which is without genetic testing (Zhu et al., 2020b). Other medications including statins, other coumarin derivatives, ACEI, ivabradine, novel oral anticoagulant, diuretics were reported to be effective in patient care in some studies, however, evidence is still scarce to establish an conclusion.

In economic evaluation, the effectiveness is usually examined at the population level. Like other clinical lab tests, effectiveness of PGx tests can be affected by the prevalence of the tested genotype variations among the population. The higher the genotype variations, the more likely tests will capture a positive result, the more likely tests will be used in clinical decision making and lead to a tailored therapy which is different from usual care. In this way, genetic testing can bring larger effect to the patients at the population level in terms of tailoring treatments, thereby increasing effectiveness of the intervention and proactively preventing treatment side effects. Furthermore, in situations where the prevalence of genotype variants are known to vary by race (CPIC, 2019), PGx testing can only necessarily increase the probability of getting benefit from the test results when the patient's race is known. In the FDA Table of Pharmacogenomic Biomarkers in Drug Labeling, patients with CYP2C9 *1/*3, *2/*2, *2/*3, and *3/*3 were considered as CYP variants, and patients with these variants have decreased warfarin clearance. The frequencies of these variants are not same among different races (USFDA, 2019). As listed in Table 7, Central/south Asian, European and near Eastern patients have the highest frequency of approximately 0.40, while Sub-Saharan African, African American/Afro-Caribbean, and East Asian patients have very low frequency of less than 0.1. In this case, pharmacogenomic testing will be more likely to have "positive" results, which means finding the genetic variances in communities that have higher rates of Central/south Asian, European and near Eastern populations, while the test positivity rate will be much lower in communities with higher rates of Sub-Saharan African, African American/Afro-Caribbean, and East Asian population.

Once positive PGx results are found, patients will be given alternative therapies in order to prevent bleeding events. For these individuals, genetic testing will likely improve treatment effectiveness; however, for the rest of the patients the genetic testing may not be useful, and their treatment along with its effectiveness stay the same. When examined at the population level, higher frequencies of positive variants lead to higher rates of using the alternative therapy instead of the usual care therapy, therefore it will increase the effectiveness of the genetic testing being conducted in general patient populations.

The effectiveness of PGx testing can be affected by test sensitivity and specificity as well. Test sensitivity is the ability that a test can find positive cases among all the true positive cases (some positive cases could be missed and reported as negative, which is called false negative). Specificity is the test's ability to exclude a negative case when it is truly negative (if some are falsely alarmed, then they are negative cases reported as positive, which is called false positive). Unlike effectiveness, which is always measured from patients' perspectives, various perspectives can be adopted while examining costs in economic evaluations. It reflects the resource (e.g., time, labor, space, material, etc.) required from one party to conduct an intervention and the specific efforts one party should make. When examined from another perspective, cost-effectiveness results could be different.

According to recommendations from second panel on cost-effectiveness in health and medicine, healthcare cost analysis can be categorized into two major domains: health care sector and societal perspectives (Sanders et al., 2016). Fig. 1 shows the cost categories under each of the domains. As suggested by the second panel, the components under the health care sectors are included in societal perspective. For example, health care sector costs include direct medical costs reimbursed by third-party payer or patient's out-of-pocket payments, which are also considered to be part of the societal perspectives. The societal perspectives include both medical costs and indirect costs, which are time costs of seeking for care, time costs for caregivers, transportation costs, loss-of-productivity costs, etc. In some of the studies, societal perspective was examined using all-cause costs (Zhu et al., 2020b). Therefore, theoretically societal perspective should be larger than health care sector perspective. The second panel recommended that both perspectives should be reported at the same time to ensure a comprehensive analysis of all the potential impacts of an intervention on the patients. Usually, the impacts within the health sectors are examined and compared, but the impacts outside of the health sector may have significant effect on the patient outcome as well (Sanders et al., 2016).

In CVD management with PGx therapy, cost analysis will be no different from other diseases. Costs for health sector have been increasing over years. The chronic disease trajectory not only results in financial burden to patients and their family, but also lead to productivity loss to both patients and their caregivers. However, the majority of the current PGx study in CVD management still focus on the perspective of health sectors while societal perspective is under reported. A recent study reviewed the current evidence

Table 7 Warfarin CYP2C9 diplotype and phenotype frequency.

	African American/ Afro-Caribbean	*American*	*Central/South Asian*	*East Asian*	*European*	*Latino*	*Near Eastern*	*Oceanian*	*Sub-Saharan African*
Diplotype									
*1/*1	0.7590	0.8310	0.6000	0.8380	0.6290	0.7460	0.6110	0.9120	0.5260
Known variants	0.1298	0.1307	0.3985	0.0785	0.3680	0.2414	0.3805	0.0878	0.3010
*2, *3	0.0706	0.1229	0.3969	0.0779	0.3647	0.2196	0.3797	0.0878	0.0479
Unknown variants	0.1112	0.0383	0.0015	0.0835	0.0030	0.0126	0.0085	0.0002	0.1730
Phenotype									
CYP2C9 normal metabolizer	0.7587	0.8315	0.5996	0.8379	0.6292	0.7458	0.6105	0.9122	0.7311
CYP2C9 intermediate metabolizer	0.2361	0.1641	0.3629	0.1516	0.3452	0.2446	0.3596	0.0866	0.2635
CYP2C9 poor metabolizer	0.0052	0.0044	0.0375	0.0060	0.0256	0.0095	0.0298	0.0012	0.0054
Indeterminate	0	0	0	0.0046	0	0	0	0	0

CPIC (2019) *Clinical Pharmacogenetics Implementation Consortium (CPIC®). Guidelines* [Online]. Available from: https://cpicpgx.org/guidelines/ (Accessed 25 April 2019).

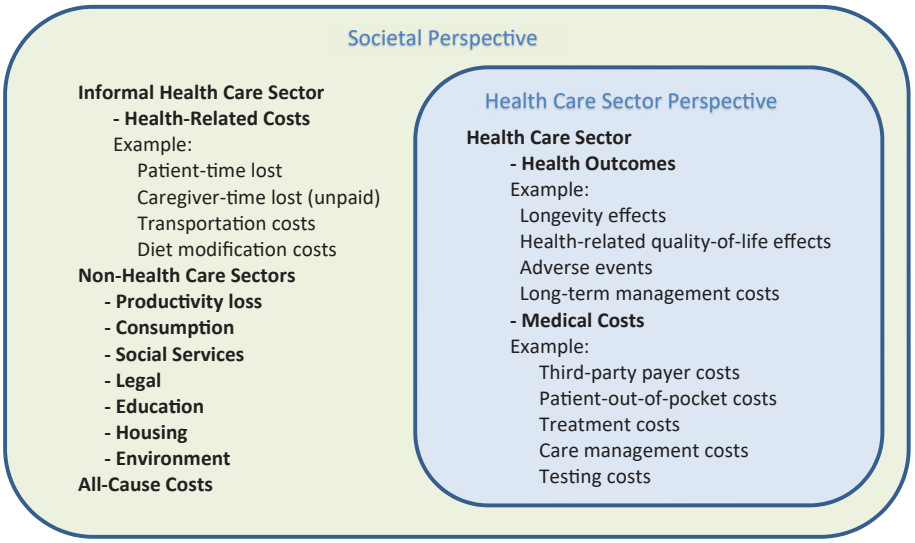

Fig. 1 Cost categories by perspectives. From Sanders GD, Neumann PJ, Basu A, Brock DW, Feeny D, Krahn M, Kuntz KM, Meltzer DO, Owens DK, and Prosser LA (2016) Recommendations for conduct, methodological practices, and reporting of cost-effectiveness analyses: Second panel on cost-effectiveness in health and medicine. *JAMA* 316(10): 1093–1103; Zhu Y, Swanson KM, Rojas RL, Wang Z, St Sauver JL, Visscher SL, Prokop LJ, Bielinski SJ, Wang L, Weinshilboum R, and Borah BJ (2020) Systematic review of the evidence on the cost-effectiveness of pharmacogenomics-guided treatment for cardiovascular diseases. *Genetics in Medicine* 22(3): 475–486.

of PGx testing in CVD management and found that for clopidogrel and warfarin, which are the most frequently studied medications, societal perspective was examined by a few studies (see Fig. 2) (Zhu et al., 2020b). Although societal perspective was included, they are still relatively small in number (1 study out of 16 clopidogrel studies and 3 out of 16 warfarin studies). No societal perspectives was used in evaluating other medications. This suggests that the current knowledge base on PGx-guided treatment in CVD management is primarily from the payer's perspective but there is not sufficient evidence for the impact of the adoption of PGx tests at the societal level. Only a few studies from the societal perspective were found on clopidogrel and warfarin. Evidence for other medications were still needed.

1.5.1.2 Cost-effectiveness analysis

The fundamental goal of any healthcare system is to improve health and life span of the population it serves, and resources are used to achieve this goal are not unlimited. Ideally, the decision-making process in health care system should override the economic considerations and focus on health outcomes. However, the scarce societal resources put limits on this decision-making rationale and requires choosing interventions that maximize benefits (in terms of health outcomes) with acceptable costs, or the ones that need the least amount of resources to achieve the same outcome benefit to meet with the requirement of beneficence.

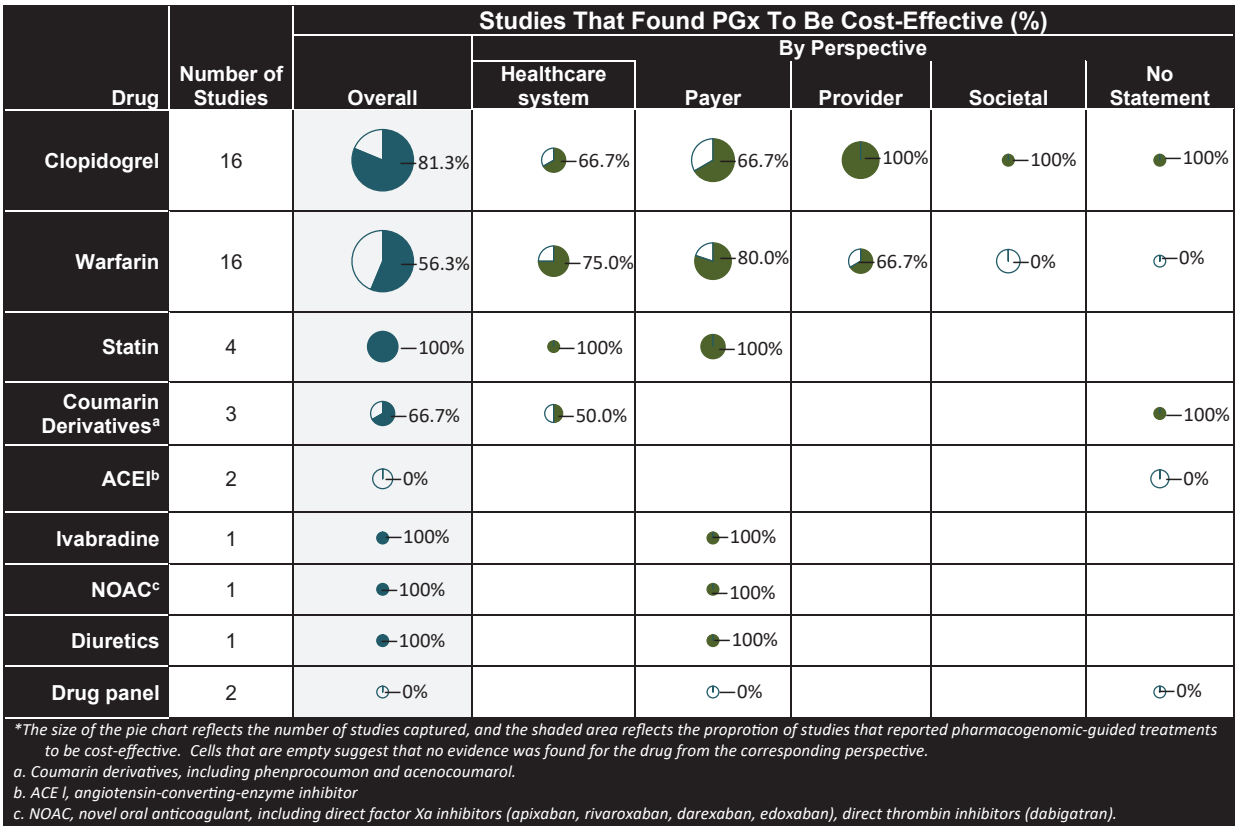

Fig. 2 Evidence map of study conclusions regarding the cost-effectiveness of PGx-guided testing. From Zhu Y, Swanson KM, Rojas RL, Wang Z, St Sauver JL, Visscher SL, Prokop LJ, Bielinski SJ, Wang L, Weinshilboum R, and Borah BJ (2020) Systematic review of the evidence on the cost-effectiveness of pharmacogenomics-guided treatment for cardiovascular diseases. *Genetics in Medicine* 22(3): 475–486.

Cost-effectiveness analysis is one of the most commonly used methods to evaluate the trade-off between different health interventions. It is used to compare the economic efficiency between two or more interventions when or with the presumption that the effectiveness of these interventions are comparable. It uses incremental cost-effectiveness ratio or ICER, which is incremental or the difference in costs between two interventions divided by the difference in effectiveness between the two interventions. ICER represents the costs per unit of effectiveness. Effectiveness, as already indicated earlier, is typically measured in terms of QALY, which offers a generic measure of effectiveness that facilitates comparison of QALY gain or loss across different diseases. Innovative and improved technologies are usually more expensive than the existing ones. Spending a maximum of $100,000 on health care to achieve one QALY has been widely adopted in economic evaluation in the US health system, which is also known as cost-effective threshold. This means that it is acceptable to adopt the healthcare intervention being compared (e.g., advanced technology such as drug or device, new healthcare delivery model) if it cost no more than $100,000 to extend patients life by a year. This value rooted in the cost of dialysis for patients with chronic kidney disease patients and was considered to be the best society can afford to help these groups of patients (Neumann et al., 2014). This rationale was extended to other diseases through the application of cost-effectiveness analysis. For example, one recent impactful cost-effectiveness PGx study in CVD management was conducted by Kazi et al. comparing standard statin treatment and addition of proprotein convertase subtilisin/kexin type 9 (PCSK9) inhibitors in lipid-lowering therapies (Kazi et al., 2017). The study built on the extant evidence that statin has been the first-line pharmacotherapy for hypercholesteremia management; however, statin can be ineffective in some patients. Statin therapy has side effects of muscle toxicity and therefore constrain its adoption by some of the patients. PCSK9 inhibitors are found to have equivalent or better effectiveness in hyperlipidemia management. Based on the FOURIER trial, Kazi et al. found that adding PCSK9 inhibitor can improve patients' health-adjusted quality of life by 0.62 years and can be cost-effective (spending less than $100,000 to achieve 1 year QALY) if the cost can be $4536 per year (2015 USD).

In CVD management, studies have emerged that examined PGx-guided treatments for their cost-effectiveness. Medications evaluated not only included FDA approved in the Table of Pharmacogenetic Associations, but also extended into new areas where the potential outcomes were simulated. Fig. 2 listed the evidence that variety of PGx-guided pharmacotherapies have been examined so far. Majority of studies focused on warfarin and other coumarin derivatives, clopidogrel and statin, which are the earliest ones on the FDA's approval list, while other medications were studied using more simulating components. Majority of the

warfarin studies suggested implementing PGx-guided therapy was cost-effective from health sector's perspective, while numerous studies from societal perspective suggested that it was not cost-effective (Fig. 2) (Eckman et al., 2009). This evidence suggested that implementing PGx in anticoagulant therapy was still not affordable to the society as a whole but may be economically attractive if only direct (payer) costs are considered. Clopidogrel was found to be economically attractive from both health care sector and societal perspective with evidence from both sides suggesting that PGx-guide therapy was cost-effective, which costs less than $100,000–50,000 per QALY gained. Statin was suggested to be cost-effective from health sector's perspective, but the body of evidence is still in a nascent stage; no evidence could be found from societal perspective, which suggested that PGx implementation in this area is still immature. Overall, the existing economic evaluations have generated mixed evidence on the cost-effectiveness of PGx-guided therapies, and they tend to be neutral in supporting PGx implementation in CVD management.

1.5.2 Legal

There are two critical steps associated with the FDA approval for a PGx-guided drug. FDA provides advice on the pharmacogenomic drug information and requires it to be included in the drug labeling. The other issue is that genetic testing should be approved by FDA as a reliable genetic testing method to ensure that the patient receives accurate PGx information and consequently appropriate pharmacotherapy. In order to further clarify this issue, FDA lists out the currently approved nucleic acid-based tests (updated 2/9/2021, https://www.fda.gov/medical-devices/vitro-diagnostics/nucleic-acid-based-tests). There are 133 types of genetic tests for 47 diseases/uses from approximately 60 manufacturers that received FDA approval so far.

FDA has been cautious on approving genetic tests to be used in direct-to-consumer tests and approval process follows the table of drug labeling rigorously—for drugs without approved relationships from genotypes to phenotype, no pharmacogenetic testing and reporting should be provided to patients.

In 2018, FDA has issued a safety communication stating that "clinical evidence is not currently available for these genetic tests or software programs and, therefore, these claims are not supported for most medications" (FDA, 2019a). This communication was issued regarding the unapproved gene-drug interaction information available in the market by test manufacturers and developers. In addition to the safety communication, in 2019, FDA issued a warning letter to Virginia-based Inova Genomics Laboratory over its tests and reporting of the unapproved gene-drug responses. An example of violation pointed out by FDA was that the relationship between CYP2C19 genotype and the drug responses to escitalopram and sertraline was not established (FDA, 2019b). The company should not have included this pharmacogenomic information in the testing and reporting in order to avoid inappropriate treatment and adverse outcomes for patients (FDA, 2019b). In this case, it is important to note that the relationship between CYP2C19 phenotype (e.g., poor metabolizers) and response to citalopram has been already approved by FDA in the Table of Pharmacogenomic Biomarkers in Drug Labeling. However, the phenotypes were defined by pharmacokinetics (e.g., drug serum level). FDA hasn't reached any conclusion on how to define each of the phenotypes using genotype information. Therefore, test manufacturers are prohibited to report any information regarding the phenotypes of the patients using the unapproved algorithms. FDA is concerned about misclassification of patients' phenotypes and lead to inappropriate treatment advice from medical providers.

In CVD related medications, only clopidogrel and warfarin are provided genotype related information (Table 1). In drug labeling, clopidogrel poor metabolizers are defined as homozygous of loss-of-function alleles, while no allele information was provided. Warfarin was the one with most complete information that allele information was provided as part of the genotype-based advice. As of March 2021, FDA hasn't approved any genetic testing that can be adopted in clinical decision making. With that being said, FDA cleared genetic tests that provide results with sequencing and allele information. The details were listed in Table 8. Only warfarin-CYP2C9/VKORC1 genotyping can be translated into phenotypes, which was consistent with FDA's table of drug labeling.

Although PGx-guided CVD therapies has been gaining more attention recently with evidence merging to report the relationship between genotype, phenotype and patients' drug responses, majority of the therapies still need regulatory approval. The only therapy ready to be implemented in clinical practice is warfarin-CYP2C9/VKORC1. There are nine medications that have been cleared for their pharmacogenomic associations. Once genotype to phenotype algorithm is established, then only PGx-guided implementation of these medications will be ready for roll-out in clinical practice. As discussed previously, PGx-guided therapies can be potentially attractive with regard to their cost-effectiveness, and once their cost-effectiveness is established, they can be implemented in large scale patient population (Fig. 3).

1.5.3 PGx test reporting inconsistency

A major component of PGx implementation is reporting of PGx test results and translating them into clinically actionable information. The current test reporting system could be very different across clinical laboratories. First, there are different calling systems (nomenclatures) for the variants. The most widely used system is the star (*) system in which the star (*) that refers to different allele variants. For example, CYP2C19-clopidogrel was reported in the star system. CYP2C19*1 refers to the most common allele carried by most people and is considered as a wild type status with full function of CYP2C19 enzyme, and therefore is used as "default" reference. The variant alleles are reported as *2, *3 and others. These star alleles are further categorized into levels of metabolism accordingly (e.g., ultrarapid metabolizer, poor metabolizer, etc.).

Another widely used nomenclature pertains to reporting the SNP, and the phenotype classifications are based on the SNP information. For example, VKORC1-warfarin doesn't have star system nomenclature, and is often reported using a single SNP rs9923231. The wild type is rs99-1639 GG, and if any of the G mutated into A (-1639 G>A), then the function of VKOR enzyme is impaired, which can be reported as abnormal metabolizer and need to adjust warfarin dosage. Some variants of genes, especially the

Table 8 FDA approved nucleic acid based tests in cardiovascular disease management.

Test biomarkers	*Trade name*	*Manufacturer*	*Phenotype report*
CYP2D6 deletion (*5) and duplication	xTAG CYP2D6 Kit v3	Luminex Molecular Diagnostics, Inc.	No
CYP2D6	xTAG CYP2D6 Kit v3	Luminex Molecular Diagnostics, Inc.	No
CYP2C19	Spartan RX CYP2C19 Test System	Akonni Biosystems Inc.	Yes
CYP2C9 and VKORC1	TruDiagnosis System	Akonni Biosystems Inc	Yes
–	Verigene CYP2C 19 Nucleic Acid Test	Nanosphere, Inc.	Recalled by FDA
CYP450 2C19 gene product, specifically *2, *3, *17	INFINITI CYP2C19 Assay	AutoGenomics, Inc.	No
UGT1A1 *1 (TA6) and *28 (TA7) alleles	Invader UGT1A1 Molecular Assay	Third Wave Technologies Inc.	No
CYP2C19	Roche AmpliChip CYP450 microarray	Roche Molecular Systems, Inc.	No
CYP2C9*2 and *3 alleles and VKORC1 - 1639G>A	eSensor Warfarin Sensitivity Saliva Test	GenMark Diagnostics	Yes
CYP2C9*2 and *3 alleles and VKORC1 - 1639G>A	eQ-PCR LC Warfarin Genotyping kit	TrimGen Corporation	Yes
CYP2C9*2 and *3 alleles and VKORC1 - 1639G>A	eSensor Warfarin Sensitivity Test and XT-8 Instrument	Osmetech Molecular Diagnostics	Yes
CYP2C9 *2 and *3 and VKORC1 1173C>T	Gentris Rapid Genotyping Assay—CYP2C9 & VKORCl	ParagonDx, LLC	Yes
CYP2C9*2 and *3 alleles and VKORC1 - 1639G>A	INFINITI 2C9 & VKORC1 Assay for Warfarin	AutoGenomics, Inc.	No
–	Verigene Warfarin Metabolism Nucleic Acid Test and Verigene System	Nanosphere, Inc.	Recalled by FDA

FDA Nucleic Acid Based Tests. https://www.fda.gov/medical-devices/in-vitro-diagnostics/nucleic-acid-based-tests.

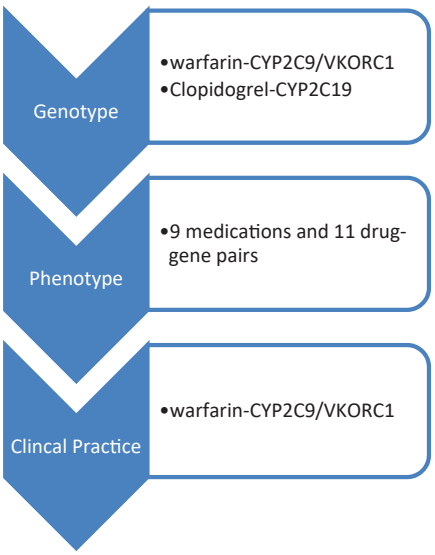

Fig. 3 Approved pharmacogenomic information.

newly discovered ones, are often reported in the SNP form as well. For example, CYP2C9 rs12777823 A is categorized as abnormal metabolizer biomarker, while *2, *3, *5 and *6 alleles are found to be abnormal as well (Table 4).

Although limited number of tests have been approved by FDA due to the limited knowledge of patient outcomes, genetic testing and methodologies to identify the variants have been developed by laboratories across the country. NIH Genetic Test Registry (https://www.ncbi.nlm.nih.gov/gtr/) documented the current genetic tests, laboratories, and the methods used to identify variants. The test information for genes relevant to CVD management and listed on FDA Table of Pharmacogenomic Biomarkers in Drug Labeling are captured in Table 9. These tests are submitted by laboratories and NIH claims to hold no responsibility and no endorsement of these tests. There are 40–244 different types of tests developed for each gene from 26 to 89 laboratories. For each gene, up to seven molecular genetic methods were used, which could lead to results that are different from each other. For example,

Table 9 Test information for genes relevant to cardiovascular disease management in NIH Genetic Testing Registry.

Drug	*Biomarker*	*Tests*	*Laboratories*	*Methods*	*Molecular genetics methods (number of unique methods)*
Carvedilol Metoprolol Propafenone Propranolol Quinidine	CYP2D6	95	51	7	Deletion/duplication analysis (16) Microsatellite instability testing (MSI) (1) Mutation scanning of select exons (1) RNA analysis (2) Sequence analysis of select exons (3) Sequence analysis of the entire coding region (14) Targeted variant analysis (71)
Clopidogrel	CYP2C19	86	49	5	Deletion/duplication analysis (17) Mutation scanning of select exons (1) Sequence analysis of select exons (4) Sequence analysis of the entire coding region (15) Targeted variant analysis (60)
Prasugrel Warfarin	CYP2C9	75	46	5	Deletion/duplication analysis (19) Mutation scanning of select exons (1) Sequence analysis of select exons (4) Sequence analysis of the entire coding region (15) Targeted variant analysis (50)
Isosorbide dinitrate Isosorbide mononitrate	CYB5R	40	26	5	Deletion/duplication analysis (20) Mutation scanning of the entire coding region (1) Sequence analysis of select exons (2) Sequence analysis of the entire coding region (36) Targeted variant analysis (2)
Rivaroxaban	F5 (Factor V Leiden)	191	89	7	Biochemical genetics: analyte (2) Deletion/duplication analysis (43) Mutation scanning of select exons (4) Mutation scanning of the entire coding region (2) Sequence analysis of select exons (7) Sequence analysis of the entire coding region (73) Targeted variant analysis (104)
Tafamidis	TTR	244	63	6	Deletion/duplication analysis (97) Mutation scanning of select exons (1) Mutation scanning of the entire coding region (5) Sequence analysis of select exons (7) Sequence analysis of the entire coding region (218) Targeted variant analysis (23)
Warfarin	VKORC1	60	43	6	Deletion/duplication analysis (16) Mutation scanning of select exons (1) Mutation scanning of the entire coding region (1) Sequence analysis of select exons (3) Sequence analysis of the entire coding region (23) Targeted variant analysis (31)
Warfarin	PROS1	44	32	4	Deletion/duplication analysis (25) Sequence analysis of select exons (1) Sequence analysis of the entire coding region (38) Targeted variant analysis (5)
Warfarin	PROC	173	35	7	Cytogenetics: FISH-interphase (1) Deletion/duplication analysis (46) Mutation scanning of select exons (2) Mutation scanning of the entire coding region (2) Sequence analysis of select exons (4) Sequence analysis of the entire coding region (74) Targeted variant analysis (87)

Information source: NIH Genetic Test Registry (https://www.ncbi.nlm.nih.gov/gtr/).

RNA analysis was used in CYP2D6 analysis, while other methods are testing of DNA. Results from RNA analysis can include the information of gene regulation, which could include the interaction with promoters or other genes and could also include the transit status of patient (i.e., RNA can change anytime). Clinical adoption of these tests is hindered by test interpretation, genotype translation and phenotype definition.

1.5.4 Ethics

Although PGx implementation is not much different from other clinical tests regarding the test ethics, there are some unique aspects in PGx implementation that are attributed by CVD management.

First, regarding the access to PGx tests, if PGx tests were to be implemented at the general population level, then the potential beneficiaries should be targeted early, starting from early to middle-ages. Besides, if DNA sequencies are used in defining genotype, the patient needs to be tested only one time during his or her lifetime as the genotypes are valid for the lifetime. Therefore, PGx test in CVD management can potentially facilitate patient management at the point-of-care and can be beneficial both from patients' and providers' perspectives. An economic evaluation suggested that the PGx test in a panel form should be implemented in wide range of population since the economic outcomes are not likely to change with race and gender; however, relatively young age (>40 years old) was recommended compared to patients with older age (>65 years old) for such PGx implementation (Zhu et al., 2020a). This strategy is regarding the actual usefulness of the treatment and the tests. American College of Medical Genetics and Genomics (ACMG) noted that programs and organizations implementing PGx testing should ensure access to such tests for the entire population in order to eliminate health care disparities (Murray et al., 2021).

Second issue with substantial ethical concern surrounding PGx testing implementation, especially test results from whole exome sequencing involves incidental findings. PGx tests, especially when exomes are sequenced, may unintentionally reveal variants that are not relevant to the intended diagnostic goal but the PGx test results might suggest higher risks for other diseases (Green et al., 2013). These variants are defined as incidental findings. Ethically, patients have the rights to choose to be informed about the incidental findings. According to the principle of beneficence, the ordering clinicians have the responsibility to provide counseling regarding any positive findings, which include both diagnostic test results and incidental findings which could potentially play a role in impacting patient's quality of life (Green et al., 2013). However, it could potentially bring burden and unnecessary medical tests and interventions to the patients, especially when the incidental finding does pose immediate risks.

Last but not the least, patient-provider shared decision making plays a significant role in test ordering, revealing and further investigation on positive test results. Patients have the right to decide ordering or not ordering a specific PGx test, which can provide a comprehensive picture of their risk profile. A recent study found that patients had favorable attitudes toward PGx testing and felt that PGx information was very important (Mukherjee et al., 2017). Some patients found PGx test to provide value as it empowers them in communication with providers (Meagher et al., 2021). Some patients found PGx testing had value to their family especially the ones with family history (Meagher et al., 2021). Some patients preferred not to know about irrelevant results (Christenhusz et al., 2013). In this case, ACMG recommended that provider should warn patients of the potential risk and the clinical concern (Green et al., 2013). There are other approaches to respect patient's autonomy with respect to not knowing about potential incidental findings, such as a selection of analyzed genes that only focused on relevant genes (Saelaert et al., 2020). In some circumstances, patients prefer not to pursue further action even though they acknowledge the genetically positive findings. In CVD management, PGx testing aims to proactively prevent side effects from drugs and improve treatment effectiveness. There is evidence supporting that patient with no genetic information receives no worse effective care in this disease group. This finding seem to suggest that PGx test plays a role that add benefits to patient's care for those who has actionable findings (Pereira et al., 2020). Therefore, autonomy is the major principle in disease management and patient's preference should be fully respected if they opt to receive usual care, with or without PGx information.

1.5.5 Professional training or preparation

Currently, there is no specific PGx training for providers in CVD management. American College of Medical Genetics and Genomics offers a Continuing Certification Program to encourage competency of genetic information in participant's specialty area, but this program is neither PGx-specific nor tailored to medical providers wanting to implement PGx (American College of Medical Genetics and Genomics) (ACMG, 2021b). A study interviewed providers in internal medicine and cardiology regarding PGx implementation, and found that clinicians were self-studying PGx-related knowledge through various sources (Unertl et al., 2015). This implied that clinicians were interested in adopting PGx-guided treatments. However, this raises the concern that clinicians may have various levels of understanding of PGx testing and results, which may lead to practice heterogeneity.

As listed in Table 9, there are multiple genetic tests with different methods offered by different laboratories across the United States, which may be used for CVD management, and are registered in NIH Genetic Testing Registry, while only a limited number of medications have been approved by FDA. It's can be challenging for practicing clinicians to navigate in ordering process for different PGx tests. Therefore, genetic clinics have been established in the United States, where clinical geneticists and genetic counselors work together to handle patient's genetic information and to help create an individualized treatment plan. Regarding the wide use of these medications, preemptive screening strategy will be most scalable and likely to be increasingly used, with which patient's individualized treatment options are ready at the point-of-care (Zhu et al., 2020a; Bean et al., 2021). There are combined medical genetics and genomics training programs integrated in pediatrics, internal medicine and other departments implying that the professional preparation for PGx implementation is slowly expanding in recent times (ACMG, 2021a). However, such PGx expansion will need to largely involve cardiologists, primary care physicians and others who are involved in direct patient care (Pulley et al., 2012), and programs aimed to expand PGx implementation need to address potential barriers in care coordination, including the participation of the clinicians engaged in CVD management.

1.6 Barriers

With the growing of PGx testing in clinical practice, especially cardiovascular medicine, there are barriers and challenges that hinder its application. Stakeholders including providers and patients hold unique perspectives and are critical to successful uptake of PGx-guided treatment, while institutions, payers, legal teams play critical roles in supporting the smooth adoption and facilitate wide application of PGx in patient population. In this section, we are going to discuss the barriers and challenges in each of these groups and the issues that need to be addressed in expanding PGx implementation (Fig. 4).

1.6.1 Physician perspective

Health professionals in cardiovascular medicines have evinced interests in adopting PGx in practice. They are the key group in PGx implementation but unfortunately not the most involved group in the implementation (McLaughlin and McLaughlin, 2009).

Health professionals play major role in clinical care, however, they do not play major role in policy-making. Implementation is usually a top-down process in which actual care providers don't usually get to involve in policy formulation regarding PGx implementation. This can potentially hinder the providers' understanding of the intervention, incentives to get involved and adopting PGx testing in their daily practice. However, as we have mentioned in Section 1.4.2, cardiology professional societies are making efforts in contributing to the PGx implementation initiatives and providing voices from the providers in cardiovascular medicine.

Second, despite of the increasing interests in PGx testing, providers look for additional support in terms of education, evidence, and knowledge support in this area. Studies revealed that providers have difficulties in adopting PGx in daily practice due to the lack of evidence, knowledge, and confidence in ordering tests, interpreting test results and further investigations on the positive results (Unertl et al., 2015; Deininger et al., 2019; Murray et al., 2021; Hachad et al., 2019). In addition, providers may be interested in learning specific knowledge regarding different types of tests they could order, the process of ordering, accessing the test reports and other practice logistics. If these problems could be addressed, more providers would come forward to adopt PGx-guided patient care (Deininger et al., 2019). It is worth noting that medical education is not only limited in continuous medical education for licensed providers, but also need to be incorporated into medical schools curriculum (Ta et al., 2019).

Third, some of the providers are actively involved in PGx-guided treatment, due to lack of consistent training and education resources, quality of care might vary substantially. Educating current providers is still a barrier since there is a lack of universally-agreed-upon knowledge source or guidelines that can fill in the gaps in the current practitioners' knowledge. Studies found that clinicians prefer to learn from the current infrastructure of grand rounds and group meetings (Unertl et al., 2015). However, these meetings could not provide a systematic view of the PGx knowledge. Additionally, providers might have over expectations on the PGx testing and looked for solutions that were not yet available to them (Unertl et al., 2015).

Fourth, the established genetic clinics within different health systems or health care provider networks can release the burden on providers in other specialties and facilitate PGx testing ordering and reporting. Some providers expressed the difficulties in ordering and interpreting PGx results, and expressed interests in referring patients directly to genetic clinics for the PGx related problems and this model was recognized as a part of opportunities in promoting PGx implementation (Bean et al., 2021). This is similar as other care model where multiple specialties work together on patient care. Studies found that collaborations among internist and specialists improve patient outcomes in complicated patient cases (McLaughlin and McLaughlin, 2009). However, efforts are needed for determining when to refer the patients and how to incorporate care coordination among various specialties, especially a care coordination model that is tailored for PGx-guided patient management is needed.

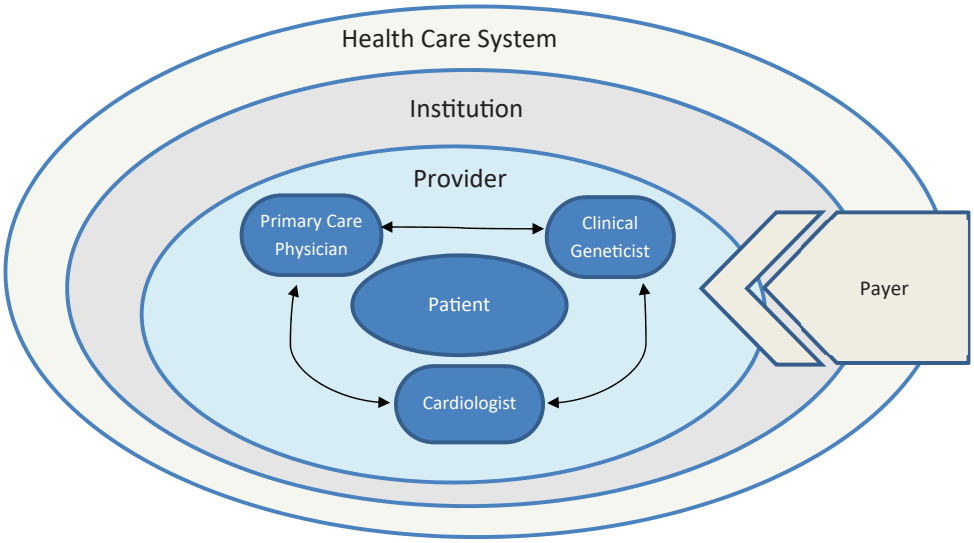

Fig. 4 The stakeholders of PGx implementation in cardiovascular disease management.

Fifth, another important issue for PGx implementation from the provider side is that the new intervention should keep the disruption to the current workflow as small as possible (Unertl et al., 2015). Programs such as RIGHT 10K project ensured that PGx information was incorporated in the electronic health system. When a patient is prescribed a PGx actionable medication, the system alerts notifying the clinician about patient's genetic risk. This requires institutional level support and human factorial design of alert message. Some providers expressed annoyance and fatigue on receiving the alert message and some selected to ignore such messages (Footracer, 2015).

Last but not the least, some resistance come from the concerns providers raised regarding the benefit vs harm on patients from PGx testing (Deininger et al., 2019; Hachad et al., 2019). Providers' concern about the long test turn-around time could delay the treatment. Some are concerned about financial burden on patients when PGx tests are added as diagnostic tools or determining appropriateness of PGx-guided drugs. Others are concerned about discrimination issues from health insurance companies which might cherry pick individuals into the plan or increase premiums for individuals with positive findings. These concerns might decrease the providers incentives to order PGx tests for the patients.

1.6.2 Patient perspective

Generally, patients have welcoming attitude toward PGx testing but have not fully prepared themselves for PGx implementation due to lack of knowledge and ineffective communication of PGx results to patients (Meagher et al., 2021). Patients are generally interested in genetic results and understand the general concept of DNA testing, but they often find it difficult to locate the test results and the relevance to their care (Boardman and Hale, 2018; Meagher et al., 2021). This creates barriers in communication and education, and could potentially be harmful to patients since they could develop misunderstanding toward PGx results. This could lead to trust issue with providers, and non-adherence to treatment regimens.

Unlike providers whose primary goal is to provide better care when ordering PGx tests, patients generally agree to conduct genetic testing for variety of reasons. Some of them have the same goal of getting better care by knowing themselves better, while some of them were curiosity based and had low incentives to pursue further interventions with the genetic results (Boardman and Hale, 2018). The mismatch in testing goal brings questions to the value of genetic tests. If patients are not interested in PGx-guided therapies, or the patients don't have trust in this new form of testing, the benefit of PGx testing brought to the patients will not maximized. These barriers in patient education and the shared decision-making process will need to be addressed in order to maximize the potential of PGx implementation.

Regarding the incidental findings, patients often have different attitudes toward them. The genetic information is generally a new concept to overwhelmingly majority. A study interviewing participants in 100,000 Genomes Project in United Kingdom found that majority of them were willing to accept health-related findings, while a small group refused to know all incidental findings (Boardman and Hale, 2018). Therefore, when and what to report for these incidental findings still to be determined and how to report them need to be resolved since this is an import part of the genetic testing.

1.6.3 Institutional efforts

The PGx implementation needs support from institutional level. First of all, PGx implementation was assumed to be incorporated into the existing electronic health record system. However, there are very limited knowledge on how the genetic information should be incorporated. Should only the relevant and identified high-risk variant findings be included, or other "normal" genotypes be included too? Should the incidental findings be included and listed? Should genotype and/or phenotype be included? To what extent the providers should have access to the genetic information of the patient?

Second, many of the current electronic health record system are locally developed and suited only for a specific institution or one health system. Many of the systems lack interoperability, which makes data sharing very difficult (Peterson et al., 2013). Most of the patients' PGx test results don't change over their lifetime, which makes record sharing more important and economically efficient. It takes not only local efforts within the institutions, but also collaborations between institutions to develop a standardized EHR system for PGx implementation.

Finally, PGx implementation requires upfront financial investment for health care staff and equipment. It was estimated that total expense for establishing PGx routine logistics was s proximately 5 million for the first 2 years in 2012 (Pulley et al., 2012). Therefore, it could be a barrier for relatively small practices or practices with limited resources (e.g., safety-net hospitals, or practices located in underserved areas).

1.6.4 Technology

One big barrier to PGx implementation is the inconsistency and heterogeneity of test methods and reporting templates for PGx test results (Hong et al., 2012; Miclaus et al., 2010). Further guidance and standardization of the nomenclatures of variants are needed in order to facilitate clinical implementation (Kalman et al., 2016). Sequencing errors leading to false negative results is a concern (Kim et al., 2019). The CDC held GeT-RM study revealed that even with consistent results of the common variants, there were discrepancies in findings in which no assays found the same set of genotypes (Pratt et al., 2016). For example, in CYP2C19 analysis sample, some assays found eight haplotypes, while some assays found up to 41 haplotypes. This creates challenge in standard reporting in relevant or incidental findings. There were also discrepancies in the nomenclature system. Some laboratories reported haplotype using star system, while others reported using locations. This creates barriers in identifying phenotypes and potentially lead to misclassification of patients and inappropriate treatments.

Even though genotypes are derived from DNA sequences, some studies have found that patient's phenotype could change over time (Nachtomy et al., 2007; Ismail et al., 2014). Genes are regulated by various mechanisms (e.g., promotor, methylation, etc.) as well as drug-drug interaction. There are numerous medications that can interfere with CYP enzymes' functions and change the phenotype of patients. These changes create barriers in genotype to phenotype conversion. Phenotype is the ultimate factor that impact patient's drug response, while genotype is used as an approximation. Therefore, the barriers that lie downstream of genotype need to be addressed with a prediction models that incorporate other factors (e.g., age, disease status, social factors, lifestyle).

1.6.5 Payer perspective

The main barrier from payer's perspective is the economic viability as well as the clinical utility of an intervention. The payer's primary concerns about the value of the PGx implementation are costs and uncertainties surrounding PGx-guided therapies under different circumstances, typically including patient's characteristics, disease types, treatment categories, etc. These issues can be answered by cost-effectiveness analysis. However, while cost-effectiveness analyses are usually based inputs from clinical trials with or somewhat restricted patient population, in order for the payers to make decisions on whether to payer for PGx-guided treatments or not, cost-effectiveness analyses must be based on real-world patient populations through clinical trials so that the true effect of the intervention could be evaluated. Then results from real-world study at population levels need to be conducted since such studies are readily generalizable.

Another issues about the economic viability of PGx-guided CV therapy is whether there is alternative therapy that could be used with PGx test results. The value of PGx not only lies in predicting drug responses, but also in patients getting alternative therapies so as to improve the treatment effectiveness as well as to minimize adverse drug side effects. Currently, most of the alternative treatments are relatively new medications, which cost much higher than the treatment under consideration. If costs of alternative treatment decreases, the cost-effectiveness of PGx implementation decreases. For example, the cost-effectiveness of hyperlipidemia treatment decreased with the cost of PCSK9 inhibitor, which makes payers more willing to pay for the new treatment (Kazi et al., 2017; Dangi-Garimella, 2019). Therefore, the PGx implementation could be hindered by high costs of the newly developed medication from payer's perspective.

1.6.6 Privacy

The major barrier to expansion and adoption of PGx-guided therapy for both providers and patients is the concern about confidentiality of the PGx results. A few providers agreed that PGx information is similar to other medical record, which are protected under HIPAA. Some are concerned about the security issue and think that PGx results need to have an additional layers of protection (Deininger et al., 2019). PGx tests are different from other standard diagnostic tests in that the latter are necessary for diagnosis and determining standard treatment plans. On the other hand, PGx tests are usually optional in most of the cases in CVD management. Providers are still able to develop a standard treatment plan without PGx information. With the concerns about confidentiality of PGx test results, patients would be hesitant to receive PGx tests, or would not like to share the results with providers, therefore, hindering PGx implementation.

2 Future application

The PGx implementation requires a more integrated system that allows various specialties work together with genetic testing. The integration process will continue to evolve, and new knowledge of genomic sciences will emerge. It can be anticipated that the case for PGx tests will grow stronger with more evidence supporting PGx-guided therapy and thereby proactively prevent harm to patients. With the advancing of data science (e.g., artificial intelligence), the integrating process will also extend into intersection of these fields.

2.1 Test frequency and population selection

Currently there are no well established guidelines for PGx initiation in CVD management. There has been evidence in single drug and model predictions for economic evaluation that suggested preemptive PGx implementation in general population can benefit patients and improve patient outcomes (Zhu et al., 2020a; Dong et al., 2020). Regulators are still waiting for more evidence to support the details (Shuren, 2020). More studies will emerge regarding the mechanisms of conducting PGx tests.

2.2 Novel treatments discovery

New drugs are emerging as alternative therapies for CVD management. For example, anti-inflammatory drugs have been studied as potential therapies for hyperlipidemia, in addition to statin and PCSK9 inhibitors (Nelson and Erridge, 2019; Boekholdt et al., 2003). With these novel treatments, more therapy options will be available for the patients with PGx positive results and tailored toward specific patient characteristics.

2.3 Novel gene-drug discovery

Novel variants and gene-drug pairs are discovered at a faster speed and new variants for additional disease categories are under investigations. For example, more genes were found to treat hypertension and ACEI therapy (Zhu et al., 2020b). The PGx studies are targeting more specific population, especially for minorities (Kaye et al., 2017; CPIC, 2019). With the advancing of Genomic science, more genome side associations will be discovered and can be used to develop new PGx tests.

2.4 Models of care coordination

American College of Medical Genetics and Genomics (ACMG) provided a points to consider (PTC) for individuals and healthcare providers to guide the future DNA-based test implementations in clinical practice (Bean et al., 2021). This PTC explores the challenges of genetic tests, specifically exome and genome sequencing (ES/GS), and aims to promote effective use of genetic and genomic tests.

This PTC adopted three models of care (Bean et al., 2021): (1) traditional genetic health-care model in which geneticists works in the similar mechanism as other specialties and as a part of multidisciplinary team in patient care; (2) nontraditional genetic health-care model in which primary care physician and direct patient care physician order the genetic tests and hold the responsibility to communicate results with patients; (3) consumer-directed genetic health-care model in which individuals can order genetic tests without their providers' direction and the providers do not hold the responsibility to follow up and investigate the genetic results (Battista et al., 2012; Bean et al., 2021). There are unique opportunities and challenges for each of these models in the preanalytical, analytical and postanalytical stages, and all the models will increase in use with the PGx implementation, while non-traditional and consumer-directed model will be expected to expand with more focus on continuous medical education Further investigations are needed to understand the impact of personal and family factors under these models.

3 Summary and conclusion

PGx implementation will continue to grow as health care is transitioning into individualized care and precision medicine. Cardiovascular medicine, as one of the frontiers of PGx implementation, will lead the way to be one of the specialties that adopts PGx-guided patient care in clinical practice. This process requires individual level, institutional level, and societal level efforts, which will open up opportunities to improve patient outcomes and bring the patient care in a new era. Challenges and opportunities both lie ahead, which require better understanding in this area and efforts from all the stakeholders.

References

Ademi Z, Watts GF, Pang J, Sijbrands EJ, Van Bockxmeer FM, O'Leary P, Geelhoed E, and Liew D (2014) Cascade screening based on genetic testing is cost-effective: Evidence for the implementation of models of care for familial hypercholesterolemia. *Journal of Clinical Lipidology* 8: 390–400.

Alhazzani AA, Munisamy M, and Karunakaran G (2017) Pharmacogenetics of CYP2C19 genetic polymorphism on clopidogrel response in patients with ischemic stroke from Saudi Arabia. *Neurosciences* 22: 31–37.

American College Of Medical Genetics And Genomics (2021a) Careers in Medical Genetics. [Online]. Available from: https://www.acmg.net/ACMG/ACMG/Education/Student/Careers_in_Medical_Genetics.aspx. Accessed 11 April 2021.

American College Of Medical Genetics And Genomics (2021b) *Continuing Certification Program*. American College of Medical Genetics and Genomics. [Online]. Available from: https://www.acmg.net/ACMG/Education/Continuing%20Certification%20Program. Accessed 10 April 2021.

American Heart Assocation (2017) Cardiovascular Disease: A Costly Burden for America, Projections Through 2035. CVD Burden Report. [Online]. Available from: https://www.heart.org/en/get-involved/advocate/federal-priorities/cardiovascular-disease-burden-report. [Accessed].

Battista R, Blancquaert I, Laberge A-M, Van Schendel N, and Leduc N (2012) Genetics in health care: An overview of current and emerging models. *Public Health Genomics* 15: 34–45.

Bean LJ, Scheuner MT, Murray MF, Biesecker LG, Green RC, Monaghan KG, Palomaki GE, Sharp RR, Trotter TL, and Watson MS (2021) DNA-based screening and personal health: A points to consider statement for individuals and health-care providers from the American College of Medical Genetics and Genomics (ACMG). *Genetics in Medicine* 23: 979–988.

Bednar EM, Sun CC, McCurdy S, and Vernon SW (2020) Assessing relatives' readiness for hereditary cancer cascade genetic testing. *Genetics in Medicine* 22: 719–726.

Bellcross CA, Kolor K, Goddard KA, Coates RJ, Reyes M, and Khoury MJ (2011) Awareness and utilization of BRCA1/2 testing among U.S. primary care physicians. *American Journal of Preventive Medicine* 40: 61–66.

Benjamin EJ, Virani SS, Callaway CW, Chamberlain AM, Chang AR, Cheng S, Chiuve SE, Cushman M, Delling FN, and Deo R (2018) Heart disease and stroke statistics—2018 update: A report from the American Heart Association. *Circulation* 137: e67–e492.

Bielinski SJ, Olson JE, Pathak J, Weinshilboum RM, Wang L, Lyke KJ, Ryu E, Targonski PV, Van Norstrand MD, and Hathcock MA (2014) Preemptive genotyping for personalized medicine: Design of the right drug, right dose, right time—Using genomic data to individualize treatment protocol. *Mayo Clinic Proceedings* 89: 25–33.. Elsevier.

Boardman F and Hale R (2018) Responsibility, identity, and genomic sequencing: A comparison of published recommendations and patient perspectives on accepting or declining incidental findings. *Molecular Genetics and Genomic Medicine* 6: 1079–1096.

Boekholdt SM, Agema WRP, Peters RJG, Zwinderman AH, Van Der Wall EE, Reitsma PH, Kastelein JJP, Jukema JW, and Group, R. E. G. E. S. S (2003) Variants of toll-like receptor 4 modify the efficacy of statin therapy and the risk of cardiovascular events. *Circulation* 107: 2416–2421.

Bush WS, Oetjens MT, and Crawford DC (2016) Unravelling the human genome–phenome relationship using phenome-wide association studies. *Nature Reviews Genetics* 17: 129.

Caraballo PJ, Bielinski SJ, St Sauver JL, and Weinshilboum RM (2017) Electronic medical record-integrated pharmacogenomics and related clinical decision support concepts. *Clinical Pharmacology and Therapeutics* 102: 254–264.

Caudle KE, Klein TE, Hoffman JM, Muller DJ, Whirl-Carrillo M, Gong L, Mcdonagh EM, Sangkuhl K, Thorn CF, and Schwab M (2014) Incorporation of pharmacogenomics into routine clinical practice: The Clinical Pharmacogenetics Implementation Consortium (CPIC) guideline development process. *Current Drug Metabolism* 15: 209–217.

Caudle KE, Gammal RS, Whirl-Carrillo M, Hoffman JM, Relling MV, and Klein TE (2016) Evidence and resources to implement pharmacogenetic knowledge for precision medicine. *American Journal of Health-System Pharmacy* 73: 1977–1985.

Chang EMS, Christopher S, and Raldow AC (2020) Explaining health state utility assessment. *JAMA* 323: 1085–1086.

Christenhusz GM, Devriendt K, and Dierickx K (2013) Disclosing incidental findings in genetics contexts: A review of the empirical ethical research. *European Journal of Medical Genetics* 56: 529–540.

CPIC (2019) Clinical Pharmacogenetics Implementation Consortium (CPIC®). Guidelines. [Online]. Available from: https://cpicpgx.org/guidelines/. Accessed 25 April 2019.

Dangi-Garimella S (2019) Amgen Announces 60% Reduction in List Price of PCSK9 Inhibitor Evolocumab. [Online]. Available from: https://www.ajmc.com/newsroom/amgen-announces-60-reduction-in-list-price-of-pcsk9-inhibitor-evolocumab. Accessed 7 August 2019.

Deininger KM, Page RL, Lee YM, Kauffman YS, Johnson SG, Oreschak K, and Aquilante CL (2019) Non-interventional cardiologists' perspectives on the role of pharmacogenomic testing in cardiovascular medicine. *Personalized Medicine* 16: 123–132.

Dong OM, Wheeler SB, Cruden G, Lee CR, Voora D, Dusetzina SB, and Wiltshire T (2020) Cost-effectiveness of multigene pharmacogenetic testing in patients with acute coronary syndrome after percutaneous coronary intervention. *Value in Health* 23: 61–73.

Dunbar SB, Khavjou OA, Bakas T, Hunt G, Kirch RA, Leib AR, Morrison RS, Poehler DC, Roger VL, and Whitsel LP (2018) Projected costs of informal caregiving for cardiovascular disease: 2015 to 2035: A policy statement from the American Heart Association. *Circulation* 137: e558–e577.

Eckman MH, Rosand J, Greenberg SM, and Gage BF (2009) Cost-effectiveness of using pharmacogenetic information in warfarin dosing for patients with nonvalvular atrial fibrillation. *Annals of Internal Medicine* 150: 73–83.

Evans WE and McLeod HL (2003) Pharmacogenomics—Drug disposition, drug targets, and side effects. *New England Journal of Medicine* 348: 538–549.

Evans O, Gaba F, and Manchanda R (2020) Population-based genetic testing for Women's cancer prevention. *Best Practice and Research. Clinical Obstetrics and Gynaecology* 65: 139–153.

FDA (2019a) The FDA Warns Against the Use of Many Genetic Tests with Unapproved Claims to Predict Patient Response to Specific Medications: FDA Safety Communication. [Online]. Available from: https://www.fda.gov/medical-devices/safety-communications/fda-warns-against-use-many-genetic-tests-unapproved-claims-predict-patient-response-specific. [Accessed].

FDA (2019b) *Warning Letter*. Inova Genomics Laboratory.

Franconi F and Campesi I (2014) Pharmacogenomics, pharmacokinetics and pharmacodynamics: Interaction with biological differences between men and women. *British Journal of Pharmacology* 171: 580–594.

Footracer KG (2015) Alert fatigue in electronic health records. *Journal of the American Academy of Physician Assistants* 28: 41–42.

Gage BF and Lesko LJ (2008) Pharmacogenetics of warfarin: Regulatory, scientific, and clinical issues. *Journal of Thrombosis and Thrombolysis* 25: 45–51.

Green RC, Berg JS, Grody WW, Kalia SS, Korf BR, Martin CL, McGuire AL, Nussbaum RL, O'Daniel JM, and Ormond KE (2013) ACMG recommendations for reporting of incidental findings in clinical exome and genome sequencing. *Genetics in Medicine* 15: 565–574.

Hachad H, Ramsey LB, and Scott SA (2019) Interpreting and implementing clinical pharmacogenetic tests: Perspectives from service providers. *Clinical Pharmacology and Therapeutics* 106: 298–301.

Holmes MV, Perel P, Shah T, Hingorani AD, and Casas JP (2011) CYP2C19 genotype, clopidogrel metabolism, platelet function, and cardiovascular events: A systematic review and meta-analysis. *JAMA* 306: 2704–2714.

Hong H, Xu L, Su Z, Liu J, Ge W, Shen J, Fang H, Perkins R, Shi L, and Tong W (2012) Pitfall of genome-wide association studies: Sources of inconsistency in genotypes and their effects. *Journal of Biomedical Science and Engineering* 5: 557–573.

Ismail S, Lee YM, Patel M, Duarte JD, and Ardati AK (2014) Genotype- and phenotype-directed antiplatelet therapy selection in patients with acute coronary syndromes. *Expert Review of Cardiovascular Therapy* 12: 1289–1303.

January CT, Wann LS, Calkins H, Chen LY, Cigarroa JE, Cleveland JC Jr., Ellinor PT, Ezekowitz MD, Field ME, Furie KL, Heidenreich PA, Murray KT, Shea JB, Tracy CM, and Yancy CW (2019) 2019 AHA/ACC/HRS focused update of the 2014 AHA/ACC/HRS guideline for the management of patients with atrial fibrillation: A report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines and the Heart Rhythm Society. *Journal of the American College of Cardiology* 74: 104–132.

Ji Y, Skierka JM, Blommel JH, Moore BE, Vancuyk DL, Bruflat JK, Peterson LM, Veldhuizen TL, Fadra N, and Peterson SE (2016) Preemptive pharmacogenomic testing for precision medicine: A comprehensive analysis of five actionable pharmacogenomic genes using next-generation DNA sequencing and a customized CYP2D6 genotyping cascade. *Journal of Molecular Diagnostics* 18: 438–445.

Jia D-M, Chen Z-B, Zhang M-J, Yang W-J, Jin J-L, Xia Y-Q, Zhang C-L, Shao Y, Chen C, and Xu Y (2013) CYP2C19 polymorphisms and antiplatelet effects of clopidogrel in acute ischemic stroke in China. *Stroke* 44: 1717–1719.

Johansen Taber KA and Dickinson BD (2014) Pharmacogenomic knowledge gaps and educational resource needs among physicians in selected specialties. *Pharmacogenomics and Personalized Medicine* 7: 145–162.

Kalman LV, Agúndez JA, Appell ML, Black JL, Bell GC, Boukouvala S, Bruckner C, Bruford E, Caudle K, and Coulthard S (2016) Pharmacogenetic allele nomenclature: International workgroup recommendations for test result reporting. *Clinical Pharmacology and Therapeutics* 99: 172–185.

Karimi M and Brazier J (2016) Health, health-related quality of life, and quality of life: What is the difference? *PharmacoEconomics* 34: 645–649.

Kaye JB, Schultz LE, Steiner HE, Kittles RA, Cavallari LH, and Karnes JH (2017) Warfarin pharmacogenomics in diverse populations. *Pharmacotherapy: The Journal of Human Pharmacology and Drug Therapy* 37: 1150–1163.

Kazi DS, Penko J, Coxson PG, Moran AE, Ollendorf DA, Tice JA, and Bibbins-Domingo K (2017) Updated cost-effectiveness analysis of PCSK9 inhibitors based on the results of the FOURIER trial. *JAMA* 318: 748–750.

Kearon C, Akl EA, Ornelas J, Blaivas A, Jimenez D, Bounameaux H, Huisman M, King CS, Morris TA, Sood N, Stevens SM, Vintch JRE, Wells P, Woller SC, and Moores L (2016) Antithrombotic therapy for VTE disease: CHEST guideline and expert panel report. *Chest* 149: 315–352.

Kim Y-H, Song Y, Kim J-K, Kim T-M, Sim HW, Kim H-L, Jang H, Kim Y-W, and Hong K-M (2019) False-negative errors in next-generation sequencing contribute substantially to inconsistency of mutation databases. *PLoS One* 14: e0222535.

Levine GN, Bates ER, Bittl JA, Brindis RG, Fihn SD, Fleisher LA, Granger CB, Lange RA, Mack MJ, and Mauri L (2016a) 2016 ACC/AHA guideline focused update on duration of dual antiplatelet therapy in patients with coronary artery disease: A report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *Journal of the American College of Cardiology* 68: 1082–1115.

Levine GN, Bates ER, Bittl JA, Brindis RG, Fihn SD, Fleisher LA, Granger CB, Lange RA, Mack MJ, Mauri L, Mehran R, Mukherjee D, Newby LK, O'gara PT, Sabatine MS, Smith PK, and Smith SC Jr. (2016b) 2016 ACC/AHA guideline focused update on duration of dual antiplatelet therapy in patients with coronary artery disease: A report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *Journal of the American College of Cardiology* 68: 1082–1115.

Lewis JP, Backman JD, Reny J-L, Bergmeijer TO, Mitchell BD, Ritchie MD, Dery J-P, Pakyz RE, Gong L, Ryan K, Kim E-Y, Aradi D, Fernandez-Cadenas I, Lee MTM, Whaley RM, Montaner J, Gensini GF, Cleator JH, Chang K, Holmvang L, Hochholzer W, Roden DM, Winter S, Altman RB, Alexopoulos D, Kim H-S, Gawaz M, Bliden KP, Valgimigli M, Marcucci R, Campo G, Schaeffeler E, Dridi NP, Wen M-S, Shin JG, Fontana P, Giusti B, Geisler T, Kubo M, Trenk D, Siller-Matula JM, Ten Berg JM, Gurbel PA, Schwab M, Klein TE, Shuldiner AR, and Investigators I (2020) Pharmacogenomic polygenic response score predicts ischaemic events and cardiovascular mortality in clopidogrel-treated patients. *European Heart Journal. Cardiovascular pharmacotherapy* 6: 203–210.

McLaughlin CP and McLaughlin CD (2009) *Health Policy Analysis: An Interdisciplinary Approach*. Jones & Bartlett Publishers.

Meagher KM, Curtis SH, Borucki S, Beck A, Srinivasan T, Cheema A, and Sharp RR (2021) Communicating unexpected pharmacogenomic results to biobank contributors: A focus group study. *Patient Education and Counseling* 104: 242–249.

Miclaus K, Chierici M, Lambert C, Zhang L, Vega S, Hong H, Yin S, Furlanello C, Wolfinger R, and Goodsaid F (2010) Variability in GWAS analysis: The impact of genotype calling algorithm inconsistencies. *Pharmacogenomics Journal* 10: 324–335.

Mukerjee G, Huston A, Kabakchiev B, Piquette-Miller M, Van Schaik R, and Dorfman R (2018) User considerations in assessing pharmacogenomic tests and their clinical support tools. *NPJ Genomic Medicine* 3: 26.

Mukherjee C, Sweet KM, Luzum JA, Abdel-Rasoul M, Christman MF, and Kitzmiller JP (2017) Clinical pharmacogenomics: Patient perspectives of pharmacogenomic testing and the incidence of actionable test results in a chronic disease cohort. *Personalized Medicine* 14: 383–388.

Murray MF, Giovanni MA, Doyle DL, Harrison SM, Lyon E, Manickam K, Monaghan KG, Rasmussen SA, Scheuner MT, and Palomaki GE (2021) DNA-based screening and population health: A points to consider statement for programs and sponsoring organizations from the American College of Medical Genetics and Genomics (ACMG). *Genetics in Medicine* 23: 989–995.

Musunuru K, Roden DM, Boineau R, Bristow MR, McCaffrey TA, Newton-Cheh C, Paltoo DN, Rosenberg Y, Wohlgemuth JG, and Zineh I (2012) Cardiovascular pharmacogenomics: Current status and future directions—Report of a National Heart, Lung, and Blood Institute Working Group. *Journal of the American Heart Association* 1: , e000554.

Nachtomy O, Shavit A, and Yakhini Z (2007) Gene expression and the concept of the phenotype. *Studies in History and Philosophy of Science Part C: Studies in History and Philosophy of Biological and Biomedical Sciences* 38: 238–254.

Nelson CP and Erridge C (2019) Are toll-like receptors potential drug targets for atherosclerosis? Evidence from genetic studies to date. *Immunogenetics* 71: 1–11.

Neumann PJ, Cohen JT, and Weinstein MC (2014) Updating cost-effectiveness—The curious resilience of the $50,000-per-QALY threshold. *New England Journal of Medicine* 371: 796–797.

Patel PD, Vimalathas P, Niu X, Shannon CN, Denny JC, Peterson JF, Chitale RV, and Fusco MR (2021) CYP2C19 loss-of-function is associated with increased risk of ischemic stroke after transient ischemic attack in intracranial atherosclerotic disease. *Journal of Stroke and Cerebrovascular Diseases* 30: , 105464.

Pereira NL, Farkouh ME, So D, Lennon R, Geller N, Mathew V, Bell M, Bae J-H, Jeong MH, and Chavez I (2020) Effect of genotype-guided oral P2Y12 inhibitor selection vs conventional clopidogrel therapy on ischemic outcomes after percutaneous coronary intervention: The TAILOR-PCI randomized clinical trial. *JAMA* 324: 761–771.

Peterson JF, Bowton E, Field JR, Beller M, Mitchell J, Schildcrout J, Gregg W, Johnson K, Jirjis JN, and Roden DM (2013) Electronic health record design and implementation for pharmacogenomics: A local perspective. *Genetics in Medicine* 15: 833–841.

Peterson JF, Field JR, Shi Y, Schildcrout JS, Denny JC, McGregor TL, Van Driest SL, Pulley JM, Lubin IM, Laposata M, Roden DM, and Clayton EW (2016) Attitudes of clinicians following large-scale pharmacogenomics implementation. *Pharmacogenomics Journal* 16: 393–398.

Pratt VM, Everts RE, Aggarwal P, Beyer BN, Broeckel U, Epstein-Baak R, Hujsak P, Kornreich R, Liao J, and Lorier R (2016) Characterization of 137 genomic DNA reference materials for 28 pharmacogenetic genes: A GeT-RM collaborative project. *Journal of Molecular Diagnostics* 18: 109–123.

Pulley JM, Denny JC, Peterson JF, Bernard GR, Vnencak-Jones CL, Ramirez AH, Delaney JT, Bowton E, Brothers K, and Johnson K (2012) Operational implementation of prospective genotyping for personalized medicine: The design of the Vanderbilt PREDICT project. *Clinical Pharmacology and Therapeutics* 92: 87–95.

Rettie AE, Wienkers LC, Gonzalez FJ, Trager WF, and Korzekwa KR (1994) Impaired (S)-warfarin metabolism catalysed by the R144C allelic variant of CYP2C9. *Pharmacogenetics and Genomics* 4: 39–42.

Roosan D, Hwang A, and Roosan MR (2021) Pharmacogenomics cascade testing (PhaCT): A novel approach for preemptive pharmacogenomics testing to optimize medication therapy. *Pharmacogenomics Journal* 21: 1–7.

Saelaert M, Mertes H, Moerenhout T, De Baere E, and Devisch I (2020) Ethical values supporting the disclosure of incidental and secondary findings in clinical genomic testing: A qualitative study. *BMC Medical Ethics* 21: 9.

Sanders GD, Neumann PJ, Basu A, Brock DW, Feeny D, Krahn M, Kuntz KM, Meltzer DO, Owens DK, and Prosser LA (2016) Recommendations for conduct, methodological practices, and reporting of cost-effectiveness analyses: Second panel on cost-effectiveness in health and medicine. *JAMA* 316: 1093–1103.

Scordo MG, Pengo V, Spina E, Dahl ML, Gusella M, and Padrini R (2002) Influence of CYP2C9 and CYP2C19 genetic polymorphisms on warfarin maintenance dose and metabolic clearance. *Clinical Pharmacology and Therapeutics* 72: 702–710.

Shaw K, Amstutz U, Kim RB, Lesko LJ, Turgeon J, Michaud V, Hwang S, Ito S, Ross C, Carleton BC, and Group, T. C. C. R (2015) Clinical practice recommendations on genetic testing of CYP2C9 and VKORC1 variants in warfarin therapy. *Therapeutic Drug Monitoring* 37: 428–436.

Shuren JE (2020) *FDA Announces Collaborative Review of Scientific Evidence to Support Associations Between Genetic Information and Specific Medications*. U.S Food & Drug Administration. [Online]. Available from: https://www.fda.gov/news-events/press-announcements/fda-announces-collaborative-review-scientific-evidence-support-associations-between-genetic. Accessed 5 May 2021.

Singh M, Sporn ZA, Schaff HV, and Pellikka PA (2019) ACC/AHA versus ESC guidelines on prosthetic heart valve management: JACC guideline comparison. *Journal of the American College of Cardiology* 73: 1707–1718.

Sturm AC (2016) Cardiovascular cascade genetic testing: Exploring the role of direct contact and technology. *Frontiers in Cardiovascular Medicine* 3: 11.

Swen J, Wilting I, De Goede A, Grandia L, Mulder H, Touw D, De Boer A, Conemans J, Egberts T, and Klungel O (2008) Pharmacogenetics: From bench to byte. *Clinical Pharmacology and Therapeutics* 83: 781–787.

Swen J, Nijenhuis M, De Boer A, Grandia L, Maitland-Van Der Zee A-H, Mulder H, Rongen G, Van Schaik R, Schalekamp T, and Touw D (2011) Pharmacogenetics: From bench to byte—An update of guidelines. *Clinical Pharmacology and Therapeutics* 89: 662–673.

Ta R, Cayabyab MA, and Coloso R (2019) Precision medicine: A call for increased pharmacogenomic education. *Personalized Medicine* 16: 233–245.

Tikkanen R and Abrams MK (2020) *U.S. Health Care from a Global Perspective, 2019: Higher Spending, Worse Outcomes?* Commonwealth Fund.

U.S Food And Drug Administration (2019) Table of Pharmacogenomic Biomarkers in Drug Labeling. Available from: https://www.fda.gov/drugs/science-research-drugs/table-pharmacogenomic-biomarkers-drug-labeling. Accessed 20 March 2021.

Unertl KM, Jaffa H, Field JR, Price L, and Peterson JF (2015) Clinician perspectives on using pharmacogenomics in clinical practice. *Personalized Medicine* 12: 339–347.

Van Schie RM, El Khedr N, Verhoef TI, Teichert M, Stricker BH, Hofman A, Buhre PN, Wessels JA, Schalekamp T, and Le Cessie S (2012) Validation of the acenocoumarol EU-PACT algorithms: Similar performance in the Rotterdam Study cohort as in the original study. *Pharmacogenomics* 13: 1239–1245.

Wang T, Pan Y, Lin J, Anand R, Wang D, Johnston SC, Meng X, Li H, Zhao X, Liu L, Wang Y, Wang Y, and Investigators C (2019) Influence of smoking on CYP2C19 genetic variants and clopidogrel efficacy in patients with minor stroke or transient ischaemic attack. *European Journal of Neurology* 26: 1175–1182.

Weinshilboum R (2003) Inheritance and drug response. *New England Journal of Medicine* 348: 529–537.

Weinshilboum R and Wang L (2004) Pharmacogenomics: Bench to bedside. *Nature Reviews Drug Discovery* 3: 739–748.

Weinshilboum RM and Wang L (2017) Pharmacogenomics: Precision medicine and drug response. *Mayo Clinic Proceedings* 92: 1711–1722.

Wetterstrand KA (2020) *The Cost of Sequencing A Human Genome*. National Human Genome Research Institute. [Online]. Available from: https://www.genome.gov/about-genomics/fact-sheets/Sequencing-Human-Genome-cost. [Accessed].

Zhu Y, Moriarty JP, Swanson KM, Takahashi PY, Bielinski SJ, Weinshilboum R, Wang L, and Borah BJ (2020a) A model-based cost-effectiveness analysis of pharmacogenomic panel testing in cardiovascular disease management: Preemptive, reactive, or none? *Genetics in Medicine* 12: 12.

Zhu Y, Swanson KM, Rojas RL, Wang Z, St Sauver JL, Visscher SL, Prokop LJ, Bielinski SJ, Wang L, Weinshilboum R, and Borah BJ (2020b) Systematic review of the evidence on the cost-effectiveness of pharmacogenomics-guided treatment for cardiovascular diseases. *Genetics in Medicine* 22: 475–486.