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Pharmacogenomics as a tool to prevent drug-related hospitalization of elderly cardiology-oncology patients receiving chemotherapeutic agents and multiple symptomatic treatments: a pilot study planned for the Italian health system

R. DI FRANCIA[1], V. DE LUCIA[2], A. GIORDANO[3], G. BENINCASA[3], M. MIGNANO[3], M. BERRETTA[4]

[1]Italian Association of Pharmacogenomics and Molecular Diagnostics (IAPharmagen), Ancona, Italy
[2]Memory Clinic, System Medicine Department, Università di Roma Tor Vergata, Rome, Italy
[3]Pineta Grande Hospital Group, Castel Volturno (CE), Italy
[4]Department of Medical Oncology, Centro di Riferimento Oncologico, CRO IRCCS, Aviano (PN), Italy

Abstract. – OBJECTIVE: Current precision medicine approaches offer powerful tools to optimize medication regimens; however, the potential impact of these tools in cancer patients with multiple drug treatments has not fully appreciated yet. Here we describe a planning project scheduled to start in the next six months.

PATIENTS AND METHODS: The overall endpoint of this project is to explore the potential association between the presence of individual genetic profile and severe toxicity rates in so-called "frail" cancer patients, using a nested case-control study design. The pilot study includes the detection of the individual pharmacogenetic profile of 150 (cases), prospect enrolled cancer "frail" patients, and 150 (control) retrospectively paired enrolled individuals. Methods for addressing the primary endpoint include: (a) Evaluation of cost-effectiveness analysis by recording QALY criteria; (b) Data recording by a brief self-administered questionnaire used to evaluate the adherence of a patient's tests and the impact of this genotyping on the patient's adverse drug reactions (ADR); (c) A sample size of paired (for age, gender, education, social status, geriatric syndromes, number of medications and comorbidities) 150 (cases) and 150 (controls); (d) Genotyping method choice by current widely diffuse platforms.

RESULTS: The investigators believe that genotype screening and the management of the overall cost of health care personalized therapy has the potential to reduce the health care costs of the Italian national health system (SSN).

CONCLUSIONS: Finally, the innovative issue of this project is to advocate the creation of a new model of the co-operative team (Physicians, pharmacist, geneticist and lab manager) that join for planning the most appropriated personalized therapy for their patient.

Key Words:
"Frail" patients, Chemotherapy, Comorbidity, Personalized medicine, Dose adjustment, Cytochrome P450.

Introduction

In the 15 years, advances in re-sequencing technology have led to an exponential decrease in genotyping costs (more than 20 thousand-fold) due to the development of new targeted molecules whose now include pharmacogenomic information. Only 67 of them have a precise label (Level 1a, 1b and 2a, see Figure 1 below) about the dosage adjustments based on the pharmacogenomic test, as formalized by guidelines from Clinical Pharmacogenomics Implementation Consortium (CPIC)[1].

Though not every test is related to a therapeutic option, a genetic diagnosis often permits targeted prevention or justification strategies (i.e., lifestyle

Corresponding Author: Raffaele Di Francia, Ph.D; e-mail: rdifrancia@iapharmagen.org

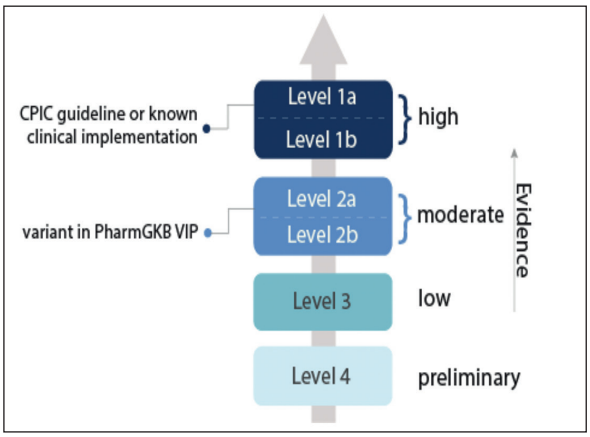

Figure 1. Schematic representation of the clinical importance of the test. **1a. Testing required**. The label states that some class of protein, gene or chromosomal testing, (i.e functional protein assays, cytogenetic studies, etc.), should be assayed before administering the tagged drug. This prerequisite may only be for a particular subset of patients. PharmGKB considers labels that state the variant is an indication for the drug, as implying a test requirement. If the label states a test "should be" performed, this is also interpreted as a requirement. **1b. Testing recommended.** The label states that some sort of test is recommended before using the tagged drug.**2a. Testing actionable.** The label does not discuss genetically or another testing for gene/protein/chromosomal variants, but does contain information about changes in efficacy, dosage or toxicity due to such variants. The label may mention contraindication of the drug in a particular subset of patients but does not require or recommend gene, protein or chromosomal testing. **2b. Testing informative.** The label comments either a gene or protein that is involved in the metabolism or transports of the drug, but there is no evidence (the literature is still scary) to suggest that variation in these genes/proteins leads to different response.

in cardiovascular disease). Further, it helps eliminate the need for further costly diagnostic testing[2].

According to several studies, about 5.3% of all hospital admissions were associated with adverse drug reactions (ADRs) and its results from variations in genes that code for drug transporters, and drug-metabolizing enzymes, such as cytochrome P450 (CYP450)[3]. These variants are single nucleotide polymorphisms (SNP) that cause influences drugs metabolization. In fact, between 2002 and 2016, the number of adverse events recorded by the Food and Drug Administration (FDA) was nearly tripled[4].

Various patients do not benefit from the first treatment they are planned. For example, 38% of depression patients, 50% of arthritis patients, 40% of asthma patients, 30% of diabetics and 75% of cancer patients will not respond to initial treatment[5]. To date the literature is still low about the prevention of drug-drug interactions in a patient with cancer and cardiopathy (C&C) comorbidities[6].

Cancer "Frail" patients with polypharmacy exhibit higher rates of hospitalizations and adverse drug reactions (ADRs)[7]. In a cohort of 887,165 of over 55 years old patients, 39.4% had polypharmacy by as the simultaneous use of ≥5 medications[8]. A topical forthcoming cohort study established that older adults with polypharmacy had over 80% higher risk for 1-year hospitalizations than older adults without polypharmacy[9]. Italian "OsMED" 2014 report 9534 (18,6%) re-hospitalizations and 276 of them died because of ADR[10]. However, the economic impact of this re-hospitalization has not fully appreciated yet. The US health care system disbursed about $300 billion on prescription medicines in 2014 (about $20 billion in Italy) with an estimated incremental increase of 6.3% spending annually over the next decade[8]. Also, this economic problem is exacerbated by a growing aging population accompanied by relatively disjointed prescribing behaviors and poor patient monitoring[9].

In another study, it was found that high rates of hospitalization in the frail patients with polypharmacy exhibited a genotype risk profile, whereas no polymorphic profile was found in older adults with polypharmacy who were rarely hospitalized[11]. This report is consistent with foremost studies that demonstrated that hospitalization rates could diverge broadly in over 55 years old even though they have an analogous profile of disease severity. However, no systematic comparison has been performed about the individual genotype in C&C patients with polypharmacy as related to their hospitalization rate[11]. The current research is designed to address this gap in Italy. The main hypothesis to be tested in this study is: frequently hospitalized cancer patients with cardiopathy underwent to multiple drug treatments have a higher frequency of pharmacogenetic variants as compared to paired patients with multi-drugs who are infrequently admitted to the hospital?

A huge problem is imposed on health care because of prescribing inappropriate medication, particularly in the context of C&C frail patients and polypharmacy.

The authors believe that genotype screening and the management of the overall cost of health care personalized therapy have the potential to reduce the health care costs of the Italian National Health System (SSN). Thus, the primary endpoint

of this research is the evaluation of the cost-effectiveness of pharmacogenomics test in C&C patients[12].

Other targets include:

- Increasing patient adherence to treatment, avoiding adverse drug reactions based on the individual metabolic profile.
- Evaluation of the hypothesis of adequate Governative (SSN) reimbursement for pharmacogenomics test for cancer patients who receive Tailored Therapy.
- Create a new model of the co-operative team (oncologist, Pharmacist, genetics and lab manager) for scheduling the most appropriated personalized therapy.

Patients and Methods

Study Design

Participants in this study will be genotyping by Cytochrome P450 variants and other metabolic genes (Table I). Based on the genetic metabolic profile, physicians and their team (i.e pharmacist, geneticist) will provide with an interpretation of genomic results, for scheduling the most appropriated personalized therapy[11].

Pipeline description (Figure 2):

a The physicians select "C&C patients" (see above in "inclusion criteria) in paired profile (matching with age, race, comorbidity and sociodemographic profile).
b Recording patient anamnesis, current therapy (cardiovascular) and planned anticancer therapy. In order to connect all unit participants, a simple algorithm software will be developed (data not shown).
c Sampling: buccal swab.
d Sending the sample to a centralized genetic laboratory.
e The laboratory retrieves the individual genotype profile to an oncologist in 7 working day.
f The oncologist, eventually, adopts correction to planned therapy, based on the individual metabolic profile of their patient.
g Data of outcomes, adverse drug reactions are recorded for statistical analysis.
h Publishing results in international contests.

Study Population

In our pilot study, the enrolment of 150 patients can be enough for statistical analysis. The similar number of a non-genotyped cohort of patients (150) could be enrolled retrospectively for

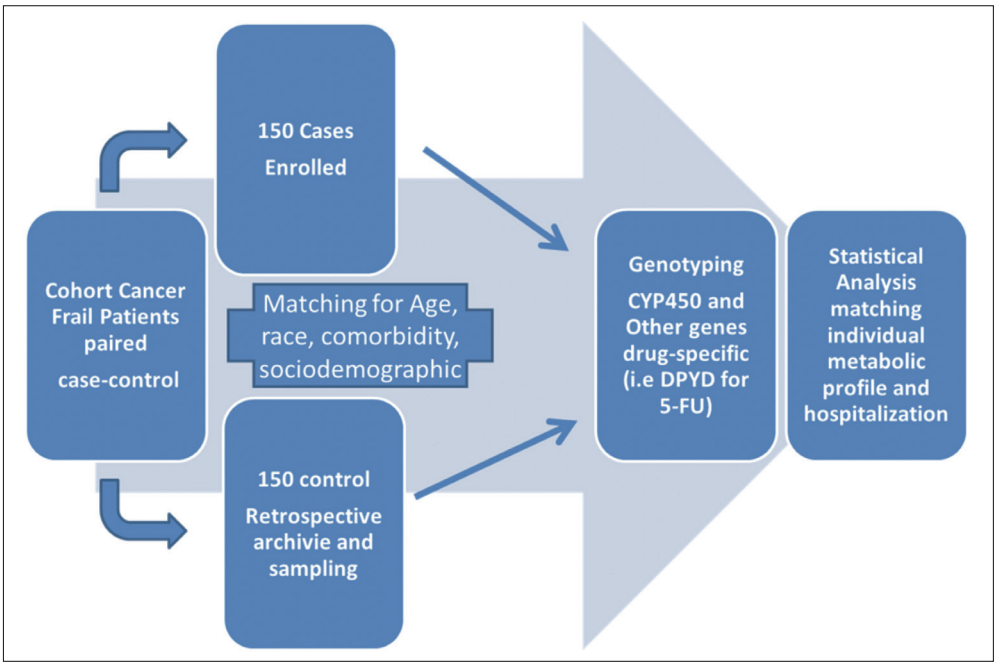

Figure 2. Study design. The pilot study includes the detection of the individual pharmacogenetic profile of 150 (cases) prospect enrolled cancer "frail" patients and 150 (control) retrospectively paired enrolled individuals.

control. The control cohort, must be selected including the same clinical and behavioural specificity (i.e chronic disease, medication adherence, smoking, Body Mass Index, number of comorbidity, use of Complementary and Alternative Medicines), and sociodemographic profile (i.e. age, gender, race, ethnicity, scholarly, work, salaries etc).

Inclusion criteria

Inclusion criteria are as follows: (1) Diagnosis of histologically proven cancer; (2) Frail cardiac and cancer patients: frail is an arbitrary definition including either elderly patients with geriatric evaluation (CGA) and patients carrying several comorbidities with HIV, HCV and HBV infections, immunodeficiency, thrombophilic risk, chronic metabolic disease etc.[13]; (3) All patients who developed an unpredictable adverse drug reaction including toxicity and or resistance to therapy; (4) Ethnicity/race: only Caucasian.

Exclusion criteria

Exclusion criteria are as follows: (1) Severe hepatic dysfunctions, (2) Undergone surgery within the last six months; (3) Documented psychiatric condition that would interfere with compliance to study participation; (4) Organ transplantation (i.e Liver); (5) A diagnosis of current malabsorption syndrome.

Genotyping Methods

Each participant/patients will genotype for the specific metabolic variants depending on the drug scheduled in according to guidelines of CPIC and recommendation of pharmacogenomics Knowledge basis[14].

Applied methods to detect polymorphism in CYP450, DPYD, TYMS, MTHFR and GSTP1, have been described for assessment of the SNP status of patients, without defining a gold standard for the daily diagnostic routine so far[15].

In our case the detection assay will be planned on FRET-based platforms using a CE/IVD labeled kit, currently available (AmpliCyp450, Diachem-srl, Naples, Italy). In each case, subsequent re-sequencing represents the unique method for the definitive confirmation of results. Participants will also be genotyped for variants to define the individual metabolic profile (Table I). Each patient's data will be stratified based on genetic metabolic profile and statistically correlated to their follow up.

Discussion

Pharmacogenomic and pharmacogenetics (PGx) testing have been increasingly used to identify genetic biomarkers that predict individual sensitivity to particular drugs. Since CYP450 enzymes are involved in the metabolism of >70% of all prescribed drugs, SNP of these has been the main target of pharmacogenomic tests. Particular consideration has been devoted to SNP of CYP2D6, CYP2C19, CYP2C9, and CYP3A4/5 genes because they are highly polymorphic. Individual drug-metabolizing activity varies from little or no activity defined poor metabolizer phenotype (PM), reduced function likely intermediate metabolizer phenotype (IM), to "wild-type" activity extensive metabolizer phenotype (EM), and increased enzyme expression in the case of gene duplication causing ultrarapid metabolizer phenotype (UM). Genotyping cancer patients who receive concomitant oncological and cardiological treatments and other drugs for comorbidities lead to toxicity health effects and increased global health care costs[16]. When personalized therapies show more effective or present fewer side effects, patients may be more likely to comply with their drug therapy[17].

Recent changes to reimbursement policies for genetic tests demonstrate how poorly conceived policies could be a negative impact on personalized care. In the so-called "Decreto Lorenzin" (DL 09/12/2015), the prescribing of genetic test undergo the high constraint policy that it doesn't encourage the private investments (i.e upgrading technology in the genetic laboratory) in this field. However, for example, to use "gap fill" methodology, which allowed regional contractors to set prices for laboratory and molecular diagnostic tests, coupled with other payment decisions, unfortunately caused a near-complete cessation of federal payments for genomic tests[18].

The highest impact should be for the treatment of C&C patients carrying chronic diseases, such as asthma and diabetes, in which non-compliance commonly exacerbates the condition. For example, inherited forms of hypercholesterolemia should increase the risk of myocardial dyscrasia before the age of 45 by more than 125-fold in women and 50-fold in men. Knowledge of a genetic predisposition for hypercholesterolemia provides patients with a powerful incentive to make lifestyle changes and manage their condition. Cancer patients genotyped to confirm the diagnosis

Table I. Report of the major polymorphisms known to influence the pharmacokinetics of the drugs.

CYP450 family	Exon/ intron	*MAF (%)	Rs code	Nucleotide	Codon	Effect enzymatic activity	Note	Pharmacogenomic correlation to drugs
CYP1A2*1F	Intr 1	0.40	762551	-163C>A	5'UTR	High inducibility		High clearance to olanzapine
CYP2B6*6		0.20	3745274 2279343		Q172H K262R	Low expression	Linkage disequilibrium	Toxicity to Efavirenz nevirapine
CYP2C8*3		0.65	11572080 10509681		R139K K399R	Low expression	Linkage disequilibrium	Increased risk of acute gastrointestinal bleeding during the use of NSAIDs and Paclitaxel .
CYP2C9*2	3	0.1	1799853	430C>T	R144C	Low activity	PM	Warfarin Phenitoin
CYP2C9*3	7	0.06	1057910	1075A>C	I359L	Low activity	PM	Warfarin Phenitoin
CYP2C19*2	5	0.09	4244285	681G>A	Splicing d.	Null Allele	PM	Low clearance clopidrogel
CYP2C19*3	4	0.01	4986893	636G>A	W212X	Null Allele	PM	Antidepressant, voriconazole
CYP2C19*17	Intr 1	0.22	12248560	-806C>T	5'UTR	High activity and expression	UM	Subtherapeutic for clopidrogel bleeding risk
CYP2D6*3		0.01	35742686	2459delA	Frameshift	Null Allele	PM	Low clearance
CYP2D6*4		0.20	3892097	1846G>A	Splicing d.	Null Allele	PM	Tamoxifen inefficacy
CYP2D6*5	Recombination	0.05			No Gene		No metabolizing	
CYP2D6*10		0.02	1065852	100C>T	P34S	Low activity and expression	IM	
CYP2D6*41		0.09	28371725	2988G>A	Splicing d.	Low activity and expression	IM	
CYP2D6 XN	Gene duplication	0.09			Copy number variation	High activity and expression	UM	Risk toxicity opioids
CYP 3A4*22	Intr 6	0.08	35599367	15389C>T	5'UTR	Low activity and expression	PM	Taxanes Nifedipine
CYP3A5*3	Intr 3	0.97	776746	6986A>G	Splicing d.	Low activity and expression	PM	Tacrolimus

PM: poor metabolizer; UM: Ultra rapid metabolizer; Intermediate Metabolizer. Source: http://www.imm.ki.se/CYPalleles.htm.

have more adherence to their statin-based treatment program, compared to the subject without genotyping[19]. The research demonstrated that genetic testing to target dosing of the blood thinner drug warfarin resulted in 31% fewer hospitalizations overall for patients and up to 48% fewer hospitalizations for bleeding or thromboembolism. The Mayo Clinic put the model to the test in a 3,600-subject prospective study. Hospitalization rates for heart patients were reduced by about 30% when genetic information was available to physicians prescribing the drug[17].

Conclusions

In our pilot study, the enrolment of 150 patients can be enough for statistical analysis. The similar number of a non-genotyped cohort of patients (150) could be enrolled retrospectively for control. The control cohort must be selected including the same clinical and behavioral specificity (i.e chronic disease, medication adherence, smoking, Body Mass Index, number of comorbidities), and sociodemographic profile (i.e. age, gender, race, ethnicity, scholarly, work, salaries etc.)[20].

The current study has some limitations including small sample size and restriction to the unique racial population. Small sample size precluded us from estimating relative risks and contributions of different factors to hospitalization risk using conditional logistic regression[21]. These two tests were used for small sample comparisons in previous studies; however, the difference between samples should be very substantial to achieve an acceptable level of statistical significance.

Although same studies concluded that there are no principal objections to using appropriate statistical tests for groups with small sample sizes, no definitive conclusions can be adequately drawn from our proof-of-concept study due to its limited sample size[22]. However, even with small sample size, this pilot study will be able to demonstrate the potential difference in the frequency of pharmacogenetic variants in older adults with polypharmacy who are frequently hospitalized *vs.* those who rarely need to hospital care. Moreover, with the decreasing of the cost for genotyping and the increasing of the methodology, the early diagnosis and the identification of the genotype profile is cost-effective[23,24].

Finally, physicians, pharmacist, geneticist and lab manager join together to provide an interpretation of individual genotype results, for planning the best therapy currently available for their patient.

Conflict of Interest

The Authors declare that they have no conflict of interests.

Authors' Contribution

The authors contributed solely to the article. Di Francia R, Berretta M. made substantial contributions to conception and design of the study.

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