

# Pharmacogenomic tests in Oncology – finding the right dose

Testes farmacogenômicos em oncologia: encontrando a dose certa

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## ABSTRACT

A pharmacogenetics/genomics (PGx) anticancer drug testing program is being developed by Kurtz and his group at the Brazilian National Cancer Institute (INCA). Drug -gene pairs were selected for PGx testing based on the presence of clinically validated PGx associations and the availability of international guidelines with PGx-informed dosing recommendations. Fluoropyrimidines-DPYD, irinotecan-UGT1A1 and thiopurines-TPMT/NUDT15 were initially included. The current estimation of anticancer therapy doses usually does not reflect the complexities of metabolism. Therefore, efforts should be made in order to refine the ways we prescribe these drugs, being conventional cytotoxic or newer ones. This program is extremely welcome and may lead to more multi-institutional partnerships and should bring a broader discussion on the use of PGx and pharmacokinetics in routine oncology practice.

Keywords: DPYD, fluoropyrimidines, irinotecan, NUDT15, pharmacogenomics, thiopurines, TPMT, UGT1A1

#### RESUMO

Um programa farmacogenética / genômica (PGx) para drogas anticancer está sendo desenvolvido por Kurtz e seu grupo no Instituto Nacional do Câncer (INCA). Pares de droga-gene foram selecionados para o teste de PGx com base na presença de associações de PGx clinicamente validadas e na disponibilidade de diretrizes internacionais com recomendações de dosagem informadas por PGx. Fluoropirimidinas-DPYD, irinotecano-UGT1A1 e tiopurinas-TPMT / NUDT15 foram inicialmente incluídos. A estimativa atual de dosagem da terapia anticâncer geralmente não reflete as complexidades do metabolismo. Portanto, esforços devem ser feitos para refinar as formas de prescrever esses medicamentos, sejam os citotóxicos convencionais como os mais novos. Este programa é extremamente bem-vindo e pode levar a mais parcerias multi-institucionais e deve trazer uma discussão mais ampla sobre o uso de PGx e farmacocinética na prática oncológica.

Descritores: DPYD, fluoropirimidinas, irinotecano, NUDT15, farmacogenômica, tiopurinas, TPMT, UGT1A1

1. Programa de Pós-Graduação em Medicina: Ciências Médicas, Faculdade de Medicina da Universidade Federal do Rio Grande do Sul, Porto Alegre, Brasil

Financial support: none to declare.

Conflicts of interest: The authors declare no conflict of interest relevant to this manuscript.

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Received on: December 9, 2020 | Accepted on: December 19, 2020 | Published on: May 14, 2021 DOI: https://doi.org/10.5935/2526-8732.20210003

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# INTRODUCTION

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In this issue of Brazilian Journal of Oncology (BJO), Suarez-Kurtz discusses the pharmacogenetics/ genomics (PGx) anticancer drug-testing program developed by his group at the Brazilian National Cancer Institute (INCA).<sup>(1)</sup> Drug -gene pairs were selected for PGx testing based on the presence of clinicallyvalidatedPGx associations and the availability of international guidelines with PGx-informed dosing recommendations. Fluoropyrimidines-DPYD, irinotecan-UGT1A1, and thiopurines-TPMT/NUDT15 were initially included in the evaluation. Tamoxifen PGx testing was also briefly mentioned.

The goal of this type of study is to identify genome variants that influence drug effects, usually through alterations in drug pharmacokinetics (i.e., absorption, distribution, metabolism or elimination) or pharmacodynamics, meaning changes in drug targets or in biological pathways that alter sensitivity to its pharmacological effects. In cancer patients, genome variations, as well as somatically acquired genome variants, can influence the antitumor and/ or toxic effect of therapeutic agents.<sup>(1)</sup>

Most human disorders, including cancer, may be influenced by different genes and genetic variants. Likewise, pharmacokinetics and pharmacological effects of therapeutic agents can be determined by genes encoding drug-metabolizing enzymes, transporters, targets, and disease-modifying genes. The genetic polymorphism in thiopurine methyltransferase (TPMT) and its effects on the risk of bone marrow toxicity from therapeutic agents, such as mercaptopurine or azathioprine illustrates this situation. As an example, adjustments are recommended on the dosage of mercaptopurine, based on TPMT genetic test results.<sup>(2)</sup>

However, when clinicians decide to prescribe the agents mentioned above, polymorphisms in other relevant genes seem as important, such as ITPA, and polymorphisms in other genes along the same pathway may be also relevant, as shown for inherited variants of NUDT15 for the occurrence of thiopurine toxicity. NUDT15 variants appear to be rare in Caucasian patients and individuals of African ancestry, but more common among those of Asian origin.<sup>(3)</sup>

The higher frequency of thiopurine intolerance – due to distinct causes – can explain differences in drug tolerability between these two patient populations, revealing that TPMT variants are the major determinant of tolerated dose in European and African patients, whereas NUDT15 is the major genetic determinant in Asian and Native Americans. As metabolism and effects of thiopurines can be affected by both germline and somatic genome variation, this can add to the complexity of interpreting cancer pharmacogenomics in this context.<sup>(4)</sup>

5-fluorouracil (5-FU) is widely used in the treatment of solid malignancies and is the backbone cytotoxic agent in anticancer drug regimens against gastrointestinal neoplasms. Despite advances in its management, up to a third of patients treated with fluoropyrimidines as monotherapy develops significant treatment-related toxicity, with 0.5 to 1% deaths. The most well-known reason for 5-FU intolerance is the deficiency of dihydropyrimidine dehydrogenase (DPD) activity, the key enzyme for its metabolism. Complete DPD deficiency is observed in 0.1 to 0.5% of the population, whereas partial DPD deficiency occurs in up to 15% of the population. Furthermore, DPD deficiency is observed in about half of patients exhibiting severe toxicity.<sup>(5)</sup>

Polymorphisms in the gene encoding DPD (DPYD), as a predictor of fluoropyrimidine-related toxicity is receiving more attention. Several sequence variations in the DPYD gene have been identified, DPYD\*2A being the most prevalent. The Clinical Pharmacogenetics Implementation Consortium established that 5-FU and analogs should undergo dose reductions based on clinical DPYD genotype tests. An initial dose reduction of at least 50% is proposed for individuals who are heterozygous for DPYD\*2A, DPYD\*13, and c.2846 A>T, who appear to show intermediate or partial DPD enzyme activity. The use of alternative drugs, however, is strongly recommended for patients with complete DPD deficiency.(6)

It is important to consider that DPD activity is regulated not only at the level of DPYD gene, but at the transcriptional and at the post-transcriptional levels as well. DPYD genotyping fails to identify severe DPD deficiency in a significant percentage of cases. In our laboratory, we have focused on functional studies, such as the measurements of UH2/U metabolic ratios in plasma or saliva. These tests showed enough sensitivity and specificity to deserve further evaluation. Other strategies for assessing DPD activity, such as DPD phenotyping, are therefore critical to be pursued.<sup>(7)</sup>

DPD converts uracil, its endogenous substrate, into dihydrouracil, and the pretreatment dihydrouracil (UH2)/uracil (U) ratio or uracil concentrations (U) alone have the potential to identify patients at fluoropyrimidine-associated severe toxicity risk. The UH2/U ratio correlates with 5-FU clearance and risk of toxicity. However, despite strong evidence on its clinical validity, the use of the UH2/U ratio in daily clinical practice has not been routinely applied. Further studies are needed to validate the strategies mentioned above of DPD function measurements in a larger patient population.<sup>(8)</sup>

Irinotecan (IRI) is a prodrug converted in the liver by carboxylesterases (CES) to 7-ethyl-10hydroxycamptothecin (SN-38), which is much more active and cytotoxic than its parent drug. Treatment with irinotecan is usually associated with doselimiting toxicities, mainly diarrhea and neutropenia/ leukopenia. The wide interindividual variability intolerability with the occurrence of severe toxicity is partially related to interindividual pharmacokinetic and pharmacogenetic differences, especially in the glucuronidation of the active metabolite through the action of  ${\rm UGT}^{(9)}$ 

Carriers of the UGT1A1\*28 allele have consistently shown lower glucuronidation ratio, with decreased SN-38G to SN-38 ratio. Due to the higher systemic exposure to SN-38 metabolite, patients with impaired UGT metabolism are at a higher risk of developing drug-induced toxicity. Several studies have found a significant association between the UGT1A1\*28 polymorphism and severe neutropenia and/or diarrhea. Similar results were found for the exon encoding UGT1A1\*6 polymorphism in Asians, indicating a central role of the variant allele in this ethnic population.<sup>(10)</sup>

Since SN-38 is much more cytotoxic than irinotecan, plasma levels of SN-38, clearance of SN-38, and/or polymorphism of UGT1A1 have clinical relevance. The clearance ability of SN-38 can be predicted by determining SN-38G/SN-38 plasma concentration ratios. It was suggested a one-point plasma SN-38G/SN-38 concentration ratio to define IRI induced neutropenia and to guide IRI dose adjustments. We have recently reviewed the pharmacokinetic and pharmacogenetic markers of irinotecan toxicity, pointing out that the most straightforward approach for IRI dose individualization should be UGT1A1 genotyping.<sup>(11)</sup>

However, this strategy is still sub-optimal due to several other genetic and environmental contributions to the variable pharmacokinetics of IRI and its active metabolites. The quantification of IRI and its active metabolite SN-38 in dried blood spots may be an alternative to individualize the drug dose through a minimally invasive collection method. Our research group and others are researching such alternative sampling strategy that eventually could allow larger studies to evaluate the relationship between exposure to IRI and its metabolites to toxicity and clinical responses, also supporting the establishment of exposure targets.<sup>(12)</sup>

As briefly mentioned by Suarez-Kurtz, the metabolism of tamoxifen (TAM) is also important to be considered when prescribing this drug. Cytochrome P450 plays an essential role on TAM metabolic activation. The major metabolite N-desmethyltamoxifen (NDT) is produced by CYP3A4/5, with minor contributions by CYPs 2D6, 1A, 1A2, 2C19, and 2B6. NDT undergoes further 4-hydroxylation by CYP2D6 being converted to 4-hydroxy-N-desmethyltamoxifen or (Z)-endoxifen (EDF).<sup>(13)</sup> Using plasma samples obtained from breast cancer patients who attended our clinic, we have also reported that CYP3A4 contributes to the bioactivation of TAM and becomes increasingly important in case of reduced or absent CYP2D6 activity.<sup>(14)</sup>

The PGx anticancer drug-testing program developed by Suarez-Kurtz and his group at the Brazilian National Cancer Institute is extremely welcome and must be discussed and implemented in other institutions of the region. This may lead to more multi-institutional partnerships and should bring a broader discussion on the use of pharmacogenomics and pharmacokinetics in routine oncology practice. Although we have great perspectives for the introduction of new immunotherapies and targeted agents, chemotherapy will continue to be administered in cancer patients in the years to come.

We should bear in mind that pharmacodynamic effects of new classes of anticancer agents would probably be influenced by different genes and genetic variants as well. In short, the current estimation of anticancer therapy doses usually does not reflect the complexities of metabolism. Therefore, efforts should be made in order to refine the ways we prescribe these drugs, being conventional cytotoxic or newer ones. Maximizing benefits, while minimizing side effects, should be our therapeutic goals.

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