ROLE OF PHARMACOGENETICS FOR THE IMPROVEMENT OF CANCER TREATMENT

VLOGA FARMAKOGENETIKE PRI UČINKOVITEJŠEM ZDRAVLJENJU RAKA

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Abstract

Background *Cancer chemotherapy is associated with a great heterogeneity in patient response that makes the prediction of tumor response and/or drug toxicity very difficult. Many factors such as tumor biology, patient's age, sex and organ function, are known to affect the therapeutic effects of drugs. Although these factors may be considered in decisions on the treatment regimens, the same regimen may result in undertreatment and insufficient therapeutic efficacy in some patients, while it can lead to overtreatment and increased toxicity in other patients.*

There is increasing evidence that genetic variability in drug metabolizing enzymes and/or drug targets influences drug response and may have a great impact on treatment outcome. This may be especially important for drugs with a narrow therapetic window and high level of toxicity, such as anticancer drugs. Many studies have shown that genetic variation of drug metabolizing enzymes such as cytochromes P450 (CYPs), UDP-glucuronosyltransferase (UGT), glutathione transferases (GSTs), thiopurine methyltransferase (TPMT) or dihydropyrimidine dehydrogenase (DPYD), drug transporters such as p-glycoprotein (MDR1) or reduced folate carrier (RFC1) and drug targets such as thymidylate syntahse (TYMS) or 5,10-methylenetetrahydrofolate reductase (MTHFR) influence not only the pharmacokinetics and/or pharmacodynamics of anticancer drugs but also the outcome of cancer treatment. In addition, somatic variations in the cancer cells can also have an impact on the anti-cancer drug efficacy, such as epidermal growth factor receptor 2 protein (HER2) overexpression and epidermal growth factor receptor (EGFR) mutations.

Conclusions *By studying the genetic variability of drug response, pharmacogenetics offers the possibility to identify patients that could benefit most from a particular treatment as well as those at increased risk for severe toxicity thus holding promises for a more individualized cancer treatment.*

Key words *cancer treatment; genetic variability; pharmacogenetics*

Izvleček

Izhodišča *Izbira načina zdravljenja, pa tudi učinkovitost zdravljenja raka s kemoterapijo se močno razlikuje med bolniki. Medtem, ko je izbira načina zdravljenja odvisna je predvsem od bioloških značilnosti in razsežnosti tumorja, pa na učinkovitost zdravljenja vplivajo tudi dejavniki, kot so starost, spol in splošno stanje bolnika, funkcija organa, ki ga zdravimo in podobno. Poleg teh dejavnikov pa na učinkovitost zdravljenja lahko vpliva tudi genetska variabilnost encimov, ki sodelujejo v presnovi ali bioaktivaciji zdravila in molekulskih tarč, na katera zdravila delujejo. Še zlasti so genetski polimorfizmi, ki vplivajo na odziv na zdravila, pomembni za napovedovanja odziva na kemoterapijo, saj je za večino kemoterapevtikov značilna visoka stopnja toksičnosti in ozka terapevtska širina. Številne študije*

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so pokazale, da encimi, ki presnavljajo zdravila, kot na primer citokromi P450 (CYP), UDP-glukuronozil-transferaze (UGT), glutation-transferaze (GST), tiopurin-metiltransferaza (TPMT) ali dihidropirimidin-dehidrogenaza (DPYD), prenašalci zdravil, kot na primer p-glikoprotein (MDR1) ali folatni prenašalec (RFC1) ter tarče zdravil, kot sta na primer timidilat sintaza (TS) ali 5,10-methiletetrahidrofol-reduktaza (MTHFR) vplivajo tako na farmakokinetiko kemoterapevtikov, pa tudi na odziv tumorja in bolnika na zdravljenje. Na odziv nekaterih tumorjev na kemoterapijo pa vplivajo tudi pridobljene somatske mutacije tumorskih celic, kot na primer prekomerna ekspresija receptorja za epidermalni rastni faktor 2 (HER2) in mutacije v genu za epidermalni rastni faktor (EGFR).

Zaključki *Določanje farmakogenetske variabilnosti dejavnikov, ki vplivajo na presnovo in učinkovitost kemoterapevtikov lahko pomaga pri individualizaciji zdravljenja raka, saj omogoča napovedati odziv posameznika na zdravljenje ter temu ustrezno prilagajanje sheme zdravljenja posameznemu bolniku.*

Ključne besede *zdravljenje raka; genetska variabilnost; farmakogenetika*

Introduction

Cancer chemotherapy is associated with a great heterogeneity in patient response and the prediction of tumor response and/or drug toxicity is very difficult. Many factors such as tumor biology, patient's age, sex and organ function, are known to affect the therapeutic effects of drugs. Although these factors may be considered in decisions on the treatment regimens, the same regimen may result in undertreatment and insufficient therapeutic efficacy in some patients, or lead to overtreatment and increased toxicity in other patients.

There is increasing evidence that genetic variability in drug metabolizing enzymes, drug transporters and/or drug targets influences the pharmacokinetics and/or pharmacodynamics of anticancer drugs and may thus have a great impact on treatment outcome. The main goal of pharmacogenetics is to identify genetic variants that could be used to predict the outcome of therapeutic agents and, thus, involved in the inter-individual variation in the response to cancer treatments. Clinically significant examples of the role of genetic factors in cancer treatment are presented in Tables 1 and 2 and discussed below.

Table 1. *Cytochromes P450 involved in the metabolism of anticancer drugs.*

Razpr. 1. *Citokromi P450, ki presnavljajo zdravila za zdravljenje raka.*

P450 involved Citokrom P450	Prodrug/drug Učinkovina/zdravilo
CYP1A1/2	decarbazine, etoposide, flutamide, procarbazine, tegafur, toremifene
CYP1B1	docetaxel, mitoxantrone
CYP2A6	Tegafur, letrozole
CYP2B6	cyclophosphamide, ifosfamide, procarbazine, thiotepa
CYP2C8	paclitaxel, tegafur
CYP2C19	cyclophosphamide, tamoxifen
CYP2D6	tamoxiefn, gefitinib, idarubicin
CYP2E1	etoposide, decarbazine
CYP3A	cyclophosphamide, docetaxel, etoposide, exemestane, fulvestrant, gefitinib, ifosfamide, imatinib, irinotecan, letrozole, mitoxantrone, paclitaxel, tamoxifen, teniposide, thiotepa, topotecan, toremifene, vinblastine, vincristine, vindesine, vinorelbine

Cytochrome *P*450 pharmacogenetics and cancer treatment

The cytochrome P450 (CYP) superfamily of enzymes catalyses the biotranformation, mainly oxidations but occassionaly also reductions, of many structurally diverse endogenous and exogenous compounds to more soluble hydrophylic forms that are more easily excreted from the body. In particular, the CYP families 1–3 are involved in the phase I-dependent metab-

Table 2. *Enzymes involved in the metabolism of the most commonly used anticancer drugs, drug targets and DNA repair capacity.*

Razpr. 2. *Ostali encimi, ki presnavljajo pogosto uporabljane citostatike, so tarče zdravljenja ali sodelujejo pri popravljanju DNA.*

	Metabolism / Presnova	
Prodrug/drug	Activating enzyme/ drug target	Inactivating enzyme/ DNA repair capacity
Učinkovina/ zdravilo	Encim, ki aktivira zdravilo/tarča zdravljenja	Encim, ki inaktivira zdravilo/mehanizmi za popravljanje DNA
Thiopurines	hypoxantine phosphoribosyl transferase (HPRT)	xantine oxidase (XO), tiopurine methyltransferase (TPMP)
5-Fluorouracil	thymidylate synthase (TS)	dihydropyrimidine dehydrogenase (DPD)
Irinotecan	carboxylesterase	UDP-glucuronosyl-transferase 1A1 (UGT1A1)
Methotrexate	thymidylate synthase (TS), dihydropyrimidine dehydrogenase (DHFR)	
Platinum agents		Xeroderma Pigmentosus Group D (XPD), X-ray cross-complementing group 1 (XRCC1), glutathione transferase (GSTP1)

olism of drugs and other xenobiotics. Because of the important role of the CYPs in the bioactivation and inactivation of carcinogens and their participation in the activation and inactivation of anticancer drugs, they play an important role both in the aetiology of cancer diseases and as determinants of cancer therapy.[1, 2]

Enzymes from CYP1A subfamily activate and detoxify numerous environmental polycyclic aromatic hydrocarbons (PAHs) and aromatic and heterocyclic amines present in combustion products such as cigarette smoke and charcoal-grilled foods and were implicated in environmental carcinogenesis,[3] while CYP1B1 metabolizes steroid hormones and may play a role in susceptibility to hormone-dependent cancers such as those from the breast and prostate.[4]

Many enzymes from CYP2 and CYP3 families are involved in the metabolism of cytotoxic drugs: some catalyse inactivation of the drugs leading to increased elimination, while others activate the prodrugs, rendering them cytotoxic and effective in cancer chemotherapy.[5]

CYP2A6 and tegafur

CYP2A6 metabolizes a number of tobacco-related precarcinogens, as well as clinically important drugs such as nicotine, coumarin, methoxyflurane, halothane, valproic acid and disulfiram. Considering anticancer drugs, CYP2A6 catalyses the activation of tegafur to 5-fluorouracil, a drug commonly used in colorectal cancer. There is increasing evidence that individuals who are homozygous or heterozygous for *CYP2A6* defective alleles are poor (slow) metabolizers of CYP2A6 substrates.[6–8]

The CYP2A6 gene is highly polymorphic[9] and the variant genes of highest importance are *CYP2A6*4,* the allele with a whole gene deleted, *CYP2A6*9,* with a mutation in the TATA box which causes a decreased expression of the enzyme, and *CYP2A6*1B* resulting in higher *in vivo* metabolism of nicotine.[6, 8, 10] The most important functionally altered allele, *CYP2A6*4,* has a 7–22 % allele frequency in Asians, but only 0.5–1 % in Caucasians.[11] Another defective allele in Caucasians is the *CYP2A6*2,* but it is very rare. The higher expression of the CYP2A6 enzyme among carriers of *CYP2A6*1B* apparently affects smoking behavior.[12] In Japan, where the defective *CYP2A6*4* allele is very common, carriers of this genotype have been shown to have lower risk of tobacco-induced lung cancer.[13] In one study, a patient with a slower tegafur metabolism was found to be heterozygous for *CYP2A6*4* and *CYP2A6*11*.[14] However, also CYP2C8 and CYP1A2 catalyse the activation of tegafur[15] and further investigations are needed to clarify the impact of CYP2A6 polymorphisms on anticancer drug metabolism.

CYP2B6, CYP2C19 and cyclophosphamide

CYP2B6 participates in the metabolism of a few precarcinogens and some important therapeutic drugs such as artemisinin, ketamine, propofol, bupropion and the HIV-1 reverse transcriptase inhibitors nevirapine and efavirenz. Several potent and specific inhibitors have been described, including the anticancer agent N,N0,N00-triethylene thiophosphoramide (thiotepa).[16] CYP2B6 is involved in the metabolic activation of the cytotoxic prodrugs cyclophosphamide, ifosfamide, thiotepa and procarbazine. Although CYP2B6 contributes only 20 % to ifosfamide activation (compared to 40 % of the prodrug activated by CYP3A4), it contributes more than 80 % to cyclophosphamide activation (compared to the 4 % share of CYP3A4).[17, 18] Phosphoramide mustard is the activated form that acts as DNA alkylating agent. Acrolein may be produced as a toxic byproduct that causes hemorrhagic cystitis. Owing to the important role of CYP2B6 for cyclophosphamide activation, polymorphisms of this enzyme would likely affect cyclophosphamide pharmacokinetics.

CYP2B6 is mainly expressed in liver, where it constitutes about 3–5 % of the total microsomal P450 pool[19, 20] but it is also detected at lower levels in extrahepatic tissues, including intestine, kidney, lung, skin and the brain.[20–22] CYP2B6 expression is induced via constitutive androstane receptor (CAR) and CYP2B6 activity in liver microsomes varies more than 100-fold.[23, 24] CYP2B6 is highly polymorphic and presently more than 48 variant alleles have been described (http://www.imm.ki.se/CYPalleles/cyp2b6.htm). The role of the different CYP2B6 alleles for the *in vivo* metabolism of drugs is still largely unknown. The CYP2B6*6 allele (Q172H and K262R) has been associated with a decreased protein expression, but higher activity using cyclophosphamide as substrate.[25] On the other hand, two studies showed that *CYP2B6*6* carriers have a reduced *in vivo* capacity to metabolize efavirenz[26, 27] and a lower activity using bupropion as probe drug.[28] *CYP2B6*16* with K262R and I328T substitutions has a decreased stability that influences the *in vivo* rate of efavirenz metabolism.[26]

Some studies have identified other polymorphic CYPs involved in cyclophosphamide and ifosfamide activation. CYP3A4 and the human CYP2C enzymes, but to a major extent CYP2C19, are implicated in these reactions.[17] In fact, it was shown that the presence of the inactive *CYP2C19*2* caused a reduction in the metabolic activation of cyclophosphamide, thereby lowering the risk of toxicity but worsening the therapeutic response.[29, 30] Similarly, it could be envisioned that the rapid *CYP2C19*17* allele with an allele frequency of about 18 % in Caucasians[31] would cause a more efficient treatment with cyclophosphamide.

More clinical studies are needed in order to characterize the impact of polymorphic CYPs on the outcome of treatment with cyclophosphamide.

CYP2C8 and paclitaxel

CYP2C8 plays an important role in about 5 % of used drugs, especially antidiabetics and antimalarials. CYP2C8 is involved in the metabolism of paclitaxel to the 30 times less toxic 6a-hydroxypaclitaxel.[32]

CYP2C8 belongs to the family of four CYP2C genes that have been mapped to chromosome 10q24 in the order 2C18–2C19–2C9–2C8 and is primarily expressed in the liver. CYP2C9 and CYP2C19 enzymes are involved in the metabolism of many frequently used drugs and their polymorphism are clinically very important.[31, 33, 34] As for the clinical relevance of the CYP2C8 polymorphisms, it is not clear yet. CYP2C8*3 (R139K and K399R) is mainly present in Caucasians (13 % frequency) and has been shown to exhibit a lower paclitaxel 6a-hydroxylase activity in heterologous expression systems. The influence of the CYP2C8*2, CYP2C8*3 and CYP2C8*4 variant alleles on therapy outcome needs further investigation.

CYP2D6 and tamoxifen

CYP2D6 is involved in the metabolism of 20–25 % of all drugs in clinical use and it has a special impact on the treatment of psychiatric and cardiovascular diseases. CYP2D6 plays a crucial role in the metabolism of tamoxifen, which is an estrogen receptor modulator widely used for the endocrine treatment of all stages of hormone receptorpositive breast cancer.

The *CYP2D6* gene is one of the best studied human *P*450 genes and correlations between the phenotype and genotype have been extensively studied for various drugs, providing a rather well-understood molecular basis for the variation in CYP2D6 activity.[35] The polymorphisms can result in defective or increased enzyme activity and CYP2D6 genotypes usually exhibit large inter-ethnic differences. Increased activity results from gene duplication/ amplification and individuals carrying up to 13 functional CYP2D6 copies in one allele have been found.[36, 37] Defective CYP2D6 allelic variants carry gene deletions, stop codons or splicing defects, and the most common functionally altered variants are: the null *CYP2D6*4* (15–21 % in Caucasians) and *CYP2D6*5* (about 3–6 % in the different populations), and the decreased activity *CYP2D6*10* (38–70 % in Asians, 3–9 % in Africans) and *CYP2D6*17* (20–34 % in Africans).

It has been shown that the active metabolite of tamoxifen is endoxifen, which requires CYP2D6 oxidation. In a study of 80 patients with breast cancer starting tamoxifen adjuvant therapy, the plasma concentrations of endoxifen after 4 months of therapy were significantly lower in patients being homozygous or heterozygous for defective *CYP2D6* genes as compared to those with two functional alleles.[38] A subsequent study validated these results and it was estimated that CYP2D6 PM women had the most significant risk of breast cancer relapse (HR 3.12, p = 0.007) and that CYP2D6 metabolism, as measured by genetic variation and enzyme inhibition, was found to be an independent predictor of breast cancer outcome in post-menopausal women receiving tamoxifen for early breast cancer.[39] Medications which decrease CYP2D6 activity such as the anti-depressants selective serotonin reuptake inhibitors and the serotonin and norepinephrine reuptake inhibitors, should be taken into account when prescribing tamoxifen and it appears that should even be avoided. Thus, the determination of CYP2D6 genotype may be of value in selecting adjuvant hormonal therapy.

The CYP2D6 genotype is also relevant for cancer patients with respect to the action of the antiemetic drugs tropisetron and ondasetron. Lower plasma levels and higher frequency and intensity of vomiting were found in subjects carrying a higher number of active CYP2D6 gene copies.[40]

CYP3A4/5, docetaxel and etoposide

With respect to the action of anticancer drugs, the variability of CYP3A4 is expected to influence the outcome of several different treatments. In cancer patients treated with docetaxel in combination with the potent CYP3A4 inhibitor ketoconazole, a 49 % decrease in docetaxel clearance was found.[41] Further on, in patients with low CYP3A4 activity docetaxel treatment resulted in greater toxicity.[42, 43] Similarly to docetaxel, irinotecan is inactivated by CYP3A4 and induction of CYP3A4 in patients receiving irinotecan resulted in a significant decrease in the formation of the toxic metabolite of this drug.[44, 45]

The human CYP3A locus carries four genes, but only CYP3A4, CYP3A5 and CYP3A7 encode active enzymes relevant for drug metabolism. The expression of these enzymes is highest in the liver and gastrointestinal tract, and they are involved in the metabolism of 50 % of all drugs currently on the market and participate in the metabolic activation of several carcinogens. The substrate specificities of the CYP3A enzymes are overlapping, but CYP3A4 usually exhibits a higher specific activity towards many CYP3A substrates when compared to CYP3A5 and CYP3A7.[46] Generally, a five-fold interindividual variability in clearance of CYP3A substrates *in vivo* has been found that can be caused by environmental factors, inducers and inhibitors or by genetic factors.[47] Important genetic polymorphisms that severely decrease the expression of CYP3A5 protein have been described, that is, *CYP3A5*3, CYP3A5*6* and *CYP3A5*7*.[48, 49] However, this is not true for CYP3A4 since no major functionally variant allele has been found at an allele frequency higher than 0.1 %. The only allele that appears to influence the CYP3A4 expression is *CYP3A4*1B,* common in Africans and present at 5 % frequency in Caucasians[50] and has been associated to prostate and lung cancer.[51–55]

Tiopurine methyltransferase and thiopurines

Thiopurines are a family of cytostatic agents that include mercaptopurine, thioguanine and azathioprine. Mercaptopurine and thioguanine are mainly in use for treatment of childhood acute lymphoblastic leukemia and myeloblastic leukemia, respectively, while azathioprine is an immunosuppresant prescribed mostly for the treatment of rheumatic diseases, inflammatory bowel disease and solid organ transplants. Thiopurines are inactive prodrugs that require activation to thioguanine nucleotides that are incorporated into DNA and inhibit normal DNA replication.

Several enzymes are involved in their activation including hypoxantine phosphoribosyl transferase (HPRT) and their inactivation include xantine oxidase (XO) and tiopurine methyltransferase (TPMP). Genetic polymorphisms in the latter enzymes alter enzymatic activity and may influence treatment outcome.[56-59] Variants *TPMT*2, TPMT*3A* and *TPMT*3C* have been associated with low enzyme activity that occurs in approximately 0.3 % of individuals, while 10 % of individuals have an intermediate enzyme activity.[60-61] Patients with intermediate TPMT activity treated with mercaptopurine have a greater incidence of treatment toxicity, while patients with low or absent TPMT activity may suffer severe or even fatal toxicity if treated with conventional doses of mercaptopurine.[62, 63] Furtermore, these individals require lower thiopurine dose than individuals with the wild-type allele to achieve similar therapeutic effect.

Thymidylate synthase, dihydropyrimidine dehydrogenase and 5-fluorouracil

5-Flourouracil (5-FU) is a uracil analogue that is most commonly prescribed in combination treatment of colorectal cancer, head and neck cancer and other solid tumours. Like thiopurines 5-FU is an inactive prodrug that requires activation in order to inhibit tumour cell replication. The active substance 5-fluoro-2-deoxyuridine monophosphate (5-FdUMP) inhibits its enzyme thymidylate synthase (TS) that catalyses the conversion of deoxyuridine monophosphate (dUMP) to deoxythymidine monophosphate (dTMP) and plays an important role in the supply of nucleotide precursors for the DNA synthesis. The promoter enhancer region of the *TS* gene contains a double (2R), a triple (3R) or more 28-base pairs (bp) tandem repeats.[64] *TS* 3R is associated with a higher *TS* gene expression and therefore with an increased dTMP synthesis and a reduced uracil incorporation into DNA.[65] Hence, patients with *TS* 3R require higher dose of 5-FU than patients with *TS* 2R genotype to achieve similar therapeutic effect.

On the other hand, 5-FU is inactivated by enzyme dihydropyrimidine dehydrogenase (DPD) which is also polymorphic. Single nucleotide polymorphisms (SNPs) in *DPYD* gene have been described that have been associated with severe 5-FU toxicity.[66-68] The most common *DPYD*2A* abolish the splicing site in exon 14 thus, leading to the production of nonfunctional protein.[69-71] Other polymorphisms in *DPYD* gene have also been described. To date, 20 SNPs have been associated with reduced DPD activity. In general population, 3–5 % of individuals are heterozygous carriers of SNPs that lower DPD activity and 0.1 % of individuals are homozygous for these SNPs.[69-72] Therefore, both *TS* and *DPYD* polymorphisms influence the outcome of 5-FU treatment. To prevent toxicity and to achieve good efficacy of the treatment patients should be genotyped for both genes prior to the treatment and the dose of the drug should be adjusted accordingly.

UDP-glucuronosyl-transferase 1A1 and irinotecan

Irinotecan is a inhibitor of topoisomerase I used to treat various solid tumors including colorectal and lung cancer. Irinotecan is an inactive prodrug that requires activation by carboxylesterase to form active metabolite SN-38. In order to be eliminated from the body SN-38 needs to be glucuronidated by hepatic enzyme UDP-glucuronosyl-transferase 1A1 (UGT1A1). A polymorphism arising from a variable number of tandem repreats (VNTR) in the promoter region of the *UGT1A1* gene has been described that results in altered UGT1A1 activity. *UGT1A1* promoter contains 5 to 8 TA repeats in healthy population, the six-repeat allele being the most common.[73, 74] The presence of seven repeats in *UGT1A1* promoter occurs in 33 % of Caucasians and results in the variant allele *(UGT1A1*28)* that has been associated with reduced *UGT1A1* expression and hence, reduced SN-38 elimination.[75] Individuals with lower UGT1A1 activity had a higher risk of severe toxicity of the irinotecan treatment, including diarrhea and leukopenia.[76]

Folate metabolic pathway and methotrexate

Methotrexate (MTX) is a folic acid antagonist that is commonly used in high doses to treat leukemias, lymphomas and breast cancer. Although MTX has been in use for the treatment of cancer for decades its precise mechanism of action is not completely understood. However, it is known that MTX enters the cells through reduce folate carrier (RFC1) and directly inhibits several enzymes of the folate metabolic pathway. Intracellular folate metabolism is complex and involves enzymes such as 5,10-methylenetetrahydrofolate reductase (MTHFR). MTHFR catalyses the reduction of 5,10-methylenetetrahydrofolate (5,10-methylene THF) to 5-methyltetrahydrofolate (5-methyl THF) and thus defines the ratio between the two forms of THF that are directed into distinct pathways. 5, 10-methylene THF is required for a conversion of dUMP to dTMP, an essential precursor of the DNA synthesis. Likewise 5-methyl THF is needed for the synthesis of methionine, the precursor of S-adenosyl methionine (SAM), a principal methyl donor in DNA and protein methylation. Two common SNPs C677T and A1298C are known in the *MTHFR* gene that occur in approximately 10 % of the population.[77, 78] *MTHFR* SNPs result in a lower enzyme activity and have been associated with an increased risk of MTX toxicity, such as MTX-induced oral mucositis.[79]

Glutathione transferases, DNA excision repair and platinum agents

Platinum agents such as cisplatin, carboplatin in oxaliplatin are intercalating agents that are used in combination with 5-FU for the treatment of advanced lung and colorectal cancer. They form crosslinks in the

DNA, produce DNA adducts and inhibit cellular replication. DNA adducts can be removed by DNA excision repair mechanisms or by conjugation. Proteins of the Xeroderma Pigmentosus Group D (XPD) and the X-ray cross-complementing group 1 (XRCC1) are enzymes involved in DNA excision repair. SNPs that have been associated with platinum agent response have been described in *XPD* and *XRCC1* genes. *XPD* A751C was observed in 10 % of colorectal cancer patients who received 5-FU/oxoliplation therapy and was associated with shorter medial survival time and worse treatment response.[80] A SNP in XRCC1 protein (Arg399Gln) resulted in a decreased DNA repair capacity and was more frequent in patients that did not respond to the 5-FU/oxoliplation treatment.[81] The presence of polymorphic variants in DNA-repair genes such as ERCC1, XPD and XRCC1 has also been shown to be powerful prognosis factors and response predictors to cisplatin-based chemotherapy in patients with non-small-cell lung cancer and squamous cell carcinoma of the head and neck.[82, 83]

Platinum agents are also detoxified by a pi-class of glutathione transferases (GSTP1) that catalyse the conjugation of glutathione (GSH) to platinum agents forming less toxic compounds which are excreted from the body. Two frequent SNPs in *GSTP1* gene resulting in an amino acid substitution have been reported. A313G polymorphism results in Ile to Val change at position 105 (Ile105Val) close to the active site and decreases the enzymatic activity. The functional role of C341T polymorphism (Ala114Val) is not clear but enzymes with both polymorphisms present had reduced conjugation capacity.[84, 85] GSTP1 105 Val/Val genotype was associated with longer median survival in colorectal cancer patients who received 5-FU/oxoliplatin therapy.[86]

Somatic variation and therapy efficacy: Herceptin, EGFR inhibitors and breast cancer gene expression profiling

The monoclonal antibody Herceptin (trastuzumab) targets the human epidermal growth factor receptor 2 HER2 protein receptors on the surface of the cancer cells and it is used to treat advanced breast cancer. By binding to the cells, Herceptin slows the growth and spread of tumors that have an overexpression of HER2. However, not all breast cancer patients are candidates for Herceptin, since the drug only appears to work for women whose breast cancer cells carry extra copies of HER2. Thus, prior to the use of Hercepin it is necesary to determine the HER2 expression in the tumoral cells, and select the patients with HER2 protein over-expression, which will likely benefit from this treatment. This is the case in approximately 30 % of the breast cancer patients.

Epidermal growth factor receptor (EGFR)-targeted therapies have improved lung cancer treatment, but only in a small number of patients. Approximately 10 % of patients with non-small-cell lung cancer, experience a relative response from these drugs. Several studies have shown that the patients that benefit from these therapies are mostly those with an increased activity of the EGFR pathway in the cancer cells.[87, 88] Both EGFR somatic activating mutations and EGFR amplifications have shown to influence the sensitivity to both small molecule inhibitors Gefitinib (Iressa) and erlotinib (Tarceva) and specific monoclonal antibodies (Cetuximab).

The *in vitro* diagnostic multivariate index assay for breast cancer patients (*MammaPrint,* by Agendia BV, approved by the European Union and United States) helps to determine the likelihood of recurrence within 5 to 10 years of initial surgery. The product consists of a microarray with 70 genes that assess the tumoral gene expression: the mRNA is extracted from the tumor sample, labeled with fluorescent dye and hybridized to the DNA microarray. The resulting signature is then compared to a specific signature that is strongly prognostic for the development of distant metastasis. The result can be used with clinical information and other laboratory tests to help determine appropriate patient treatment and follow-up. The 70 genes analyzed in the test predict 10-year survival at a significance level more than 3 times greater than existing methods (St. Gallen criteria) and with an accuracy level of 96.7 %.

Conclusions

Genetic polymorphisms have been identified in the majority of genes coding for drug metabolizing enzymes, drug transporters and/or drug targets. Many of these polymorphisms were shown to influence the pharmacokinetics and/or pharmacodynamics of anticancer drugs and there is increasing evidence they may also influence patients' response to cancer treatment. In addition, the cancer cells carry multiple genetic alterations (somatic mutations) that can contain clinically relevant genetic information affecting drug treatment response. Although more clinical studies are needed to asses if analysis of candidate genes can improve the outcome of anticancer drug therapy in the clinical practice, it seems that pharmacogenetic testing of candidate genes could help to identify patients who are at increased risk for severe toxicity as well as those that could benefit most from a particular treatment thus holding promises for a more individualized cancer treatment.

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