REVIEW

Development of precision medicine approaches based on inter-individual variability of BCRP/ABCG2

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Abstract  Precision medicine is a rapidly-developing modality of medicine in human healthcare. Based on each patient’s unique characteristics, more accurate dosages and drug selection can be made to achieve better therapeutic efficacy and less adverse reactions in precision medicine. A patient’s individual parameters that affect drug transporter action can be used to develop a precision medicine guidance, due to the fact that therapeutic efficacy and adverse reactions of drugs can both be affected by expression and function of drug transporters on the cell membrane surface. The purpose of this review is to summarize unique characteristics of human breast cancer resistant protein (BCRP) and the genetic variability in the BCRP encoded gene ABCG2 in the development of precision medicine. Inter-individual variability of BCRP/ABCG2 can impact choices and outcomes of drug treatment for several diseases, including cancer chemotherapy. Several factors have been implicated in expression and function of BCRP, including genetic, epigenetic, physiologic, pathologic, and environmental factors. Understanding the roles of these factors in controlling expression and function of BCRP is critical for the development of precision medicine based on BCRP-mediated drug transport.

Abbreviations: 3'-UTR, 3'-untranslated region; 5-aza-C, 5-aza-2'-deoxycytidine; ABCG2, ATP-binding cassette subfamily G member 2; ALL, acute lymphocytic leukemia; AUC, area under curve; BCRP, breast cancer resistant protein; miRNA, microRNA; FTC, fumitremorgin C; H3K4me3, histone H3 lysine 4 trimethylation; H3K9me3, histone H3 lysine 9 trimethylation; H3S10P, histone H3 serine 10 phosphorylation; HDAC, histone deacetylase; HIF-1α, hypoxia inducible factor 1 subunit alpha; HIV-1, human immunodeficiency virus type-1; HMG-CoA, β-hydroxy-β-methyl-glutaryl-coenzyme A; MDR, multidrug resistance; MDR1, multidrug resistance 1; NBD, nucleotide binding domain; P-gp, P-glycoprotein; RISC, RNA-induced silencing complex; siRNA, small RNA interference; SNP, Single nucleotide polymorphism; Tat, transactivator protein; TKI, tyrosine kinase inhibitor

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1. Introduction

Precision medicine is a rapidly-developing field in human healthcare that is influencing the practice of medicine. Based on each patient’s unique characteristics that affect responses to drugs, more accurate doses or drug treatments can be given to the patient to achieve better therapeutic benefits and less adverse reactions using a precision medicine approach.

A patient’s individual characteristics that affect drug transporter action are an important aspect of precision medicine. Drug transporters are a class of proteins located on the membrane surface of cells that allow drugs to enter and exit cells via carrier-mediated mechanisms. Therapeutic efficacy and adverse reactions of drugs can be affected by the expression and function of drug transporters located in biological membrane barriers of a multitude of target tissues. Human breast cancer resistant protein (BCRP) is a key transporter involved in a patient’s idiosyncratic functionality with respect to drug transport. BCRP is so called because it was first identified in breast cancer cells that exhibited drug resistance to mitoxantrone, doxorubicin, and daunorubicin. The human BCRP protein is encoded by the ATP-binding cassette subfamily G member 2 (ABCG2) gene, which is a member of the ATP-binding cassette transporter superfamily. Human BCRP is apically localized in the cell membranes of multiple organs, including epithelial cells of the small intestine, the canalicular domain of liver hepatocytes, renal tubule cells, brain capillary endothelial cells in blood-brain barrier, and placental syncytiotrophoblasts (Fig. 1). BCRP utilizes the energy generated from ATP hydrolysis to drive the efflux of diverse chemicals across cell membranes, including both endogenous substrates, such as folic acid and uric acid, and xenobiotics, as well as environmental toxins, such as phophororide A. Detailed information about BCRP substrates are discussed elsewhere.

BCRP has been extensively studied for its role as an efflux transporter of drugs, leading to drug resistance in target cells and decreased pharmacological effects of substrate drugs. Overexpression of BCRP has been regarded as one of the causes of multi drug resistance (MDR) in diseases, especially cancer. In cell based studies, over expression of BCRP has been found to correlate with MDR in cells derived from several cancers, including breast cancer, ovarian cancer, colon cancer, small cell lung cancer, and myeloma. In addition to cellular models, BCRP overexpression has been correlated with MDR and cancer treatment outcomes in clinical settings. BCRP has also been found to be overexpressed in hematopoietic malignancies and solid tumors after chemotherapy treatment. In studies of acute myeloid leukemia (AML), high BCRP levels were reported in 33% of AML blast cells, which also correlated with disease prognosis and overall survival. However, no correlation between BCRP expression and overall AML incidence has been found. In acute lymphocytic leukemia (ALL), another hematopoietic malignancy, correlations between BCRP expression and drug response differed between studies. The roles of BCRP have also been assessed in multiple solid tumors with conflicting conclusions about overall survival or prognosis, including breast carcinoma, non-small cell lung cancer, melanoma, and retinoblastoma. The lack of consensus concerning the role of BCRP in these studies might stem from multiple sources, including BCRP detection methods, sample sizes, or patient composition. Despite the fact that the role of BCRP is still not completely understood, inhibition of BCRP has been considered as a pharmacological strategy to improve outcomes of drug treatment.

Differential expression and altered function of BCRP lead to dramatic inter-individual differences in drug response and disease progression. Several factors have been reported to be responsible for variations of BCRP expression and function. In this review, we summarize some well-documented factors, including genetic, epigenetic, physiologic, pathologic, and environmental factors in the regulation of BCRP expression and alterations of BCRP function. Combinations of these factors form each patient’s unique characteristics that impact BCRP-mediated drug transport and can then be used to develop precision medicine-based treatments to achieve better therapeutic outcomes.

2. Genetic polymorphisms of the ABCG2 gene

The human ABCG2 gene encodes the BCRP protein and is fully-annotated in the Human Genome build GRCh38/h38 in the UCSC Genome Browser. The ABCG2 gene is located on human chromosome at the locus 4q22.1 and consists of approximately 141 kb of genomic sequence with 16 exons and 15 introns (Fig. 2A). The BCRP protein contains two major domains: a cytoplasmic hydrophilic nucleotide binding domain (NBD), where ATP binds and is hydrolyzed, and a hydrophobic membrane-spanning domain, consisting of six transmembrane domains (Fig. 2B). Unlike other “full
transporters”, such as multidrug resistance 1 (MDR1) protein, which has two halves, BCRP is regarded as a “half transporter”.

Usually, two BCRP “half transporters” form a homodimer to perform its transporting function as illustrated in Fig. 3. Common genetic polymorphisms exist in the *ABCG2* gene in various populations across the world. Single nucleotide polymorphisms (SNPs) are the most common genetic polymorphisms, responsible for more than 90% of all interindividual genetic variations in *ABCG2*. The detection of SNPs in the human genome has been carried out utilizing recent advanced DNA sequencing technologies in several genomic projects, including protein-coding genetic variations, the Exome Aggregation Consortium, and the 1000 Genomes Project. Over 100 SNPs associated with the *ABCG2* gene are retrievable from the ExAC Browser in the Exome Aggregation Consortium database (http://exac.broadinstitute.org/).

### Table 1

| SNPs rs number | cDNA and protein variant | Minor allele frequency |
|----------------|--------------------------|-----------------------|
| rs2231142      | C421A (Q141K)            | 0.1180                |
| rs2231137      | G34A (V12M)              | 0.1076                |
| rs34783571     | G1858A (D620N)           | 0.0056                |
| rs138606116    | G1060A (G354R)           | 0.0026                |
| rs45605536     | G1582A (A528T)           | 0.0014                |
| rs192169063    | T1465C (F489L)           | 0.0010                |
| rs148475733    | A211G (M71V)             | 0.0009                |
| rs147547385    | A1787G (N596S)           | 0.0005                |
| rs72552713     | C376T (Q126Stop)         | 0.0005                |
| rs34264773     | A1768T (N590Y)           | 0.00003               |

Minor allele frequency is derived from the Exome Aggregation Consortium ExAC Browser (http://exac.broadinstitute.org/).

The global population consists of East Asian, Latino, European (Finnish and non-Finish), South Asian, African, and others.

### Table 2

| Ethnic groups          | rs2231142 | rs2231137 |
|------------------------|-----------|-----------|
| Caucasian               | 11%–17%   | 2%–6%     |
| African American        | 5%        | 4%–9%     |
| Chinese                | 34%–45%   | 29%–38%   |
| Korean                  | 40%       | 20%–27%   |
| Japanese                | 36%–41%   | 12%–22%   |

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and Korean\(^{40}\) and lower (2%–10%) in Caucasians\(^{36–38,41}\) and Africans\(^{35–39}\). The minor allele frequencies of these two \(ABCG2\) variants in the 1000 Genomes Project (http://www.internationalgenome.org/) populations show a similar global distribution. The minor allele of rs2231137 A has a frequency of 15.8% in the global populations of the 1000 Genomes Project, but varied significantly in different ethnic groups with 6.3% in Africans, 23.8% in Native Americans, 6.1% in Europeans, 15.4% in South Asians, and 32.6% in East Asians (Fig. 2A, rs2231137 and Table 3). Similarly, the minor allele of rs2231142 A has a frequency of 11.9% in the global populations of the 1000 Genomes Project, but varied with 1.3% in Africans, 14.1% in Native Americans, 9.4% in Europeans, 9.7% in South Asians, and 29.1% in East Asians (Fig. 2A, rs2231142 and Table 3). Higher frequencies in the East Asian populations may indicate higher variability on BCRP-mediated drug transport in these populations. The minor alleles of the two most common \(ABCG2\) variants, rs2231137 G34A (V12M) and rs2231142 C421A (Q141K), may cause a reduced level of BCRP expression. The rs2231137 G34A (V12M) variant was first identified in mitoxantrone-resistant cell lines\(^{42}\). BCRP protein expression levels in placentas are significantly lower in rs2231137 A/A samples compared to rs2231137 C/C\(^{39}\). The rs2231142 C421A (Q141K) variant was also first identified in mitoxantrone-resistant cell lines\(^{42}\). A functional analysis of SNPs of the \(ABCG2\) gene was achieved by transfecting cDNA plasmids containing different variants into LLC-PK1 cells, which showed a significantly lower expression level of BCRP in rs2231142 A construct as compared to rs2231142 C construct\(^{43}\).

The rs2231142 A SNP also yielded a reduced amount of cell membrane expression of BCRP in K562 cells as compared to rs2231142 C\(^{44}\). The protein expression level from the rs2231142 A construct was about half that of rs2231142 C construct in Flp-in-293 cells\(^{45}\). The reduced protein expression of rs2231142 A construct is due to the reduced stability of the BCRP protein in the endoplasmic reticulum and increased susceptibility to ubiquitin-mediated proteasomal degradation\(^{55}\). These cell-based mechanistic studies showed the ability to alter BCRP expression by the specific \(ABCG2\) SNPs, which might further cause altered BCRP function in these SNP carriers.

The minor alleles of the two most common \(ABCG2\) variants, rs2231137 G34A (V12M) and rs2231142 C421A (Q141K) may also result in decreased transport of BCRP substrates. Drug resistance profiles of Flp-In-293 cells expressing the two major \(SNP\) variants of rs2231137 G34A and rs2231142 C421A showed approximately a 50% reduction in transport of the drug SN-38 in cells expressing the mutated proteins as compared to wild type\(^{46}\). In HEK293 cells transfected with rs2231142 C421A constructs, the A allele construct had an 80% higher intracellular accumulation of the carcinogenic heterocyclic amine 2-amino-1-methyl-6-phenylimidazo[4,5-b]pyridine as compared to the C allele construct\(^{47}\). The reduced transport activity caused by the rs2231142 A allele resulted in increased cytotoxicity when K562 cells were treated with the anticancer drugs imatinib, dasatinib, and nilotinib\(^{48}\). The results generated from these studies could explain the BCRP dysfunction in patients carrying the same SNPs, which might assist in the development of guidance for drug prescription.

Minor alleles of less common \(ABCG2\) variants may also lead to decreased drug transport via BCRP. Several variants, such as F208S and S441N were expressed at markedly low levels in Flp-In-293 cells\(^{48}\). These mutations affect intracellular distribution and ubiquitin-mediated proteasomal degradation of the mutant BCRP protein\(^{59}\). Several variants were also defective in porphyrin transport in insect cells transfected with constructs made by site-directed mutagenesis\(^{60}\). Due to the low variant frequency in human populations, it is difficult to determine if the data from cell models can be extrapolated to humans. However, these studies could potentially be utilized to develop dosing guidelines for patients with rare \(ABCG2\) variants.

### 2.1. The common \(ABCG2\) variants in prediction and treatment of cancer

The common genetic variants of the \(ABCG2\) gene, rs2231137 G34A and rs2231142 C421A, have been reported to be associated with disease risk, reduced efficacy of drug treatments, and increased adverse reactions in different human diseases. Below we summarize recent findings on how the \(ABCG2\) genetic variants play roles in carcinogenesis, efficacy of chemotherapy treatment, and drug-induced cytotoxicity.

Relationship between carcinogenesis and common \(ABCG2\) variants is controversial in population-based association studies in various types of cancer. Association of \(ABCG2\) rs2231142 C421A with the development of breast cancer was examined in 100 Kurdish patients and 200 healthy controls\(^{62}\). Patients with AA genotype of rs2231142 were at a higher risk of breast cancer.
progression as compared to patients with rs2231142 CC. Similarly, association of the genetic variations rs2231137 G34A and rs2231142 C421A with breast cancer susceptibility was determined in 1,230 Chinese breast cancer patients. The study found that rs2231137 GA/AA and rs2231142 AA genotypes as well as rs2231137 A-rs2231142 C and rs2231137 G-rs2231142 A haplotypes were associated with a significantly increased risk for developing breast cancer. Associations of the common ABCG2 variants with cancer susceptibility have also been examined in other types of cancer. A meta-analysis based on 10 studies involving a total of 3593 cases and 5875 controls was performed to evaluate the contribution of the ABCG2 rs2231142 C421A with cancer susceptibility. The analysis found that the rs2231142 A allele is associated with a lower risk for the development of multiple cancer types, including leukemia and colorectal cancer. Based on these data, the authors conclude that the A allele C421A in BCRP might be a protective factor against cancer development. In a Danish prospective case-cohort study, rs2231137 A allele carriers with the high fiber intake exhibited a statistically-significantly decreased risk of colorectal cancer as compared to patients harboring the rs2231137 G allele. However, in a case-controlled study in a Turkish population, rs2231142 A allele carriers had a significantly higher risk of colorectal cancer, while the rs2231137 A allele was not associated with the risk of colorectal cancer. A study including 170 patients with androgen-independent prostate cancer and 141 healthy control subjects showed no significant difference in the prevalence of prostate cancer in subjects with rs2231142 C421A genetic variants, but another study reported that patients carrying rs2231142 A allele had a significantly shorter time to prostate cancer recurrence post-prostatectomy than patients carrying the rs2231142 C allele. From the above results, we can see that the relationship between the common ABCG2 variants and cancer risk is complex and may be different in divergent human populations and variable across cancer types. Further studies are needed to clarify the impacts of BCRP transporter genotypes upon carcinogenesis.

The common ABCG2 variants are contributors to variations in the efficacy of cancer drug treatments since the primary function of BCRP is to mediate the active efflux of drugs out of cancer cells. In a study with Kurdish breast cancer patients, rs2231142 AA carriers had a better response rate to the chemotherapeutic agents anthracycline and paclitaxel. In a study of breast cancer with a Chinese patient population, rs2231142 AA homozygotes had a significantly enhanced therapeutic response in patients treated with neoadjuvant anthracycline-based chemotherapy. Moreover, rs2231137 AA genotype carriers had improved response in patients with cancers positive for the estrogen or progesterone receptors treated with anthracycline-based chemotherapy. A correlation of rs2231137 A and rs2231142 A alleles with altered sorafenib plasma levels was found in a study with 47 French patients with advanced hepatocellular carcinoma, where the heterozygous group showed lower sorafenib plasma concentrations with a tendency toward better clinical response. Variants of ABCG2 and dysfunction of BCRP were also reported to alter non-BCRP substrate drugs through indirect mechanisms. Prostate cancer patients carrying the rs2231142 A allele showed increased survival rates as compared to patients carrying the rs2231142 C allele after treatment with docetaxel, which is thought not to be a BCRP substrate. One study showed the involvement of serine/threonine kinase Pim-1 (Pim-1L), which co-localizes and phosphorylates BCRP at the threonine 362 position, as being functionally involved in BCRP mediated docetaxel resistance. This study also showed that transfection of Pim-1L cDNA directly caused docetaxel resistance in LNCaP cells, suggesting that BCRP might be able to alter the transport of a broader range of substrates than previously thought. In summary, based on genotypes of the common ABCG2 variants, patients with the minor alleles of rs2231137 G34A and rs2231142 C421A may, in general, have a better therapeutic response to anti-cancer drugs. These genetic SNP markers may therefore serve as predictors for efficacious outcomes of chemotherapies.

However, the common ABCG2 variants may also be related to severe drug-induced adverse reactions to chemotherapy. A study with 219 Japanese renal cell carcinoma patients showed a significant association between the rs2231142 C421A genetic variant and severe thrombocytopenia after treatment with sunitinib. Dose adjustments of sunitinib are needed for patients with the rs2231142 A variant. A prospective clinical study with 83 Japanese non-small-cell lung cancer patients found a statistically-significant association between the rs2231137 A allele and an increased occurrence of skin rash after gefitinib treatment. The results suggest that rs2231137 G34A might be a useful predictor of grade 2 or worse gefitinib-induced skin toxicity. Knowing the relationship between the common ABCG2 variants and predisposition to drug-induced toxicity may serve to guide a better and more precise prescription of drugs for patients carrying specific ABCG2 variants.

2.2. The role of common ABCG2 variants in prediction and treatment of diseases other than cancer

Considering the fact that BCRP also transports endogenous compounds and non-chemotherapeutic drugs, dysregulation of BCRP expression or function can also impact the progression of a variety of diseases in addition to cancer. BCRP involvement in gout and statin-based hyperlipidemia treatment are two well-studied cases where BCRP function is critical and can be affected by specific ABCG2 variants. In this section, we will summarize and discuss the overall impact of BCRP in disease and the efficacy of certain drugs for gout and hyperlipidemia treatment.

The common ABCG2 variants are associated with the development of gout. Gout is a common inflammatory disease characterized by hyperuricemia and arthritis. Since BCRP plays a critical role in the elimination of uric acid, disrupted BCRP function might lead to reduced urate excretion and higher levels of blood uric acid, which may result in gout development. Recent studies have shown the relationship between the common ABCG2 variants and the etiology of gout. A study with 39,853 subjects consisting of European Americans, African Americans, Mexican Americans, and American Indians found that the rs2231142 A allele was significantly associated with higher serum levels of uric acid and a higher risk of gout. Similarly, a study with a Chinese Han male population of 352 cases and 350 controls also show a significant association of the rs2231142 A allele with an increased risk of gout. In addition, a meta-analysis including 9 case-control studies involving 17,942 individuals clearly showed an increased gout risk in patients with the rs2231142 A allele as opposed to the rs2231142 C allele. Gender and ethnicity were found to be additional co-factors affecting the association with susceptibility to gout. The rs2231142 C421A variant was also associated with poor response to gout treatment with allopurinol, suggesting this SNP might be a robust genetic marker for the prediction of gout treatment success. Another study with a Chinese Han population
of 143 cases and 310 controls identified four additional susceptible ABCG2 SNPs associated with occurrence of gout, including rs2622621, rs3114018, rs17731799, and rs3114020. An ABCG2 haplotype containing these SNPs is associated with a decreased risk of development of gout.

The common ABCG2 variants are associated with efficacy of lowering cholesterol by statin drugs. Statins are a class of drugs widely used in the treatment of hyperlipidemia through their ability to reduce plasma low-density lipoprotein cholesterol by inhibiting β-hydroxy-β-methyl-glutaryl-coenzyme A (HMG-CoA) reductase, a rate-controlling enzyme for cholesterol biosynthesis in liver. The biological target of statins is the hepatocyte and full drug efficacy requires their retention in liver. Rosuvastatin has been shown to be a substrate of BCRP, indicating that its pharmacokinetics can be affected by common ABCG2 variants. A study with 14 healthy Chinese male individuals showed that rosuvastatin has a higher Cmax and a lower clearance in individuals with rs2231142 CA or AA genotypes as compared to individuals with rs2231142 CC. Another study also reported that individuals with the ABCG2 rs2231142 A variant had a lower clearance of rosuvastatin. The ABCG2 rs2231142 C421A variant also showed a similar impact on the pharmacokinetics of atorvastatin and simvastatin in Caucasian and Asian subjects. Furthermore, a study focusing on adverse drug reactions induced by atorvastatin showed that patients with rs2231142 CA or AA had a higher risk of developing dose-dependent atorvastatin adverse effects as compared to rs2231142 CC patients. These studies showed the role of BCRP in the transport of several statin drugs, where specific BCRP alleles were able to alter drug therapeutic efficacy and even toxicity. In precision medication, carriers of these ABCG2 variants should receive modified doses of drugs to improve therapeutic efficacy and avoid unnecessary adverse reactions).

In summary, genetic polymorphisms are common in the ABCG2 gene with more than 100 SNPs identified in the human genome. The two most common ABCG2 variants have minor allele frequencies of over 10% in global populations, with higher frequencies in East Asians. The minor alleles of both variants result in amino acid substitutions in the BCRP NBD domain, which have been proven to reduce the expression and function of BCRP and its ability to transport its substrates. Although population-based association studies revealed a potential effect on cancer progression, the minor alleles of these two common ABCG2 variants are associated with a significant better response rate to chemotherapy with various anticancer drugs on a range of cancers. Because of the reduced efflux of substrates out of target cells, minor alleles of these two common ABCG2 variants also have the potential to trigger adverse events during chemotherapy. In addition to cancer, the common ABCG2 variants also have a proven impact on progression of other diseases, such as gout, and therapeutic efficacy of additional drugs, such as statins.

The current literature does not yet give a complete picture of all the genetic polymorphisms of the ABCG2 gene. Current cell-based and population-based studies mainly focused on the two most common ABCG2 variants, rs2231137 G34A (V12M) and rs2231142 C421A (Q141K), while knowledge of other ABCG2 SNPs, particularly for SNPs in the promoter, untranslated regions, and introns, is very limited. Several studies have shown that altered pharmacokinetic behavior of drugs in patients carrying the common ABCG2 variants exist, but there are no dosing guidance or drug labels on how to best use substrate drugs in individuals harboring genetic variants of the ABCG2 gene.

However, sufficient data are now in hand to develop precision medicine approaches based on ABCG2 genetic polymorphisms. As illustrated in Fig. 4, before a patient is prescribed a drug,

![Figure 4](https://example.com/figure4.png)
sequencing of the \textit{ABCG2} gene should be performed and this information should be annotated. A genotype associated with decreased expression and function of BCRP protein can be predicted to have decreased drug efflux mediated by BCRP accompanied with increased efficacy, but with risk of increased toxicity. Deviation from standard doses with either an increased dose for better efficacy or a decreased dose for less toxicity could be made to achieve a more precise treatment plan. More accurate associations of \textit{ABCG2} genotypes with expression and function of BCRP protein and more precise dose adjustments are the areas that warrant future study in order to develop precision medicine strategies related to \textit{ABCG2} genotypes.

### 3. Alterations of BCRP expression by epigenetic mechanisms

In addition to the common genetic polymorphisms of the \textit{ABCG2} gene, expression and function of BCRP protein can be altered by epigenetic mechanisms. Epigenetic mechanisms refer to the molecular events that can affect gene expression without changes of the DNA sequence \textit{per se}. DNA methylation, histone modifications, and non-coding RNAs are among the most highly studied epigenetic pathways. Expression of BCRP protein has been shown to be regulated by each of these epigenetic mechanisms.

#### 3.1. Alteration of BCRP expression by DNA methylation

DNA methylation usually occurs at the 5' carbon position of cytosine residues located in “CpG” islands. DNA methylation at the promoter region of a gene can inhibit transcriptional initiation and leads to repression of transcription.

The DNA methylation status of the \textit{ABCG2} promoter contributes to BCRP-mediated chemotherapeutic drug resistance in various cancer cell types. The lung cancer cell line PC-6 showed increased BCRP expression at both mRNA and protein levels after treatment with a DNA methyltransferase inhibitor, 5-aza-2'-deoxycytidine (5-aza-C)\textsuperscript{70}. An inverse correlation between methylation status of the CpG sites in the \textit{ABCG2} promoter and BCRP expression levels was found in several small cell and non-small cell lung cancer cells\textsuperscript{70}. The study indicated that higher levels of demethylation of the CpG sites in the \textit{ABCG2} promoter may serve as a mechanism leading to higher levels of BCRP expression and resistance to SN-38 for anti-cancer therapy in lung cancer cells. A correlation between the DNA methylation status of the \textit{ABCG2} promoter and drug resistance was assessed in 32 colorectal cancer cell lines\textsuperscript{71}. Lower levels of BCRP expression were significantly correlated with higher levels of DNA methylation. After treatment with 5-aza-C, enhanced expression of BCRP and decreased sensitivity of the colon cancer cells to the anticancer drugs of 5-fluorouracil, oxaliplatin, and irinotecan were observed\textsuperscript{71}. A high level of DNA methylation at the CpG sites in the \textit{ABCG2} promoter and a low level of BCRP expression are also common in T-cell acute lymphoblastic leukemia cells\textsuperscript{72}. Treatment with the chemotherapeutic drugs sulfasalazine and topotecan resulted in demethylation of the CpG sites in the \textit{ABCG2} promoter in the T-cell model and a dramatic induction of BCRP mRNA levels as well as consequent acquisition of a BCRP-dependent drug resistant phenotype\textsuperscript{72}. Combinational treatment of malignant glioma cells with a chemotherapeutic drug temozolomide together with melatonin has shown a synergistic toxic effect on the cancer cells. Melatonin-induced DNA methylation at the \textit{ABCG2} promoter and a downregulation of the expression and function of BCRP have been suggested to be a potential mechanism for the synergistic toxic effect of melatonin and chemotherapeutic drugs\textsuperscript{73}, therefore, melatonin can be viewed as a promising drug to overcome drug resistance in the treatment of glioblastomas and improve the efficacy of chemotherapy.

![Figure 5](image.png) A strategy for the development of a precision medicine approach based on DNA methylation in the \textit{ABCG2} promoter in a mixed population.
A strategy to develop a precision medication approach based on DNA methylation of the \textit{ABCG2} promoter can be developed. As illustrated in Fig. 5, inhibition of DNA demethylases or activation of DNA methyltransferases can serve as a potential strategy for decreasing BCRP expression, whereas inhibition of DNA methyltransferases or activation of DNA demethylases might serve as a potential strategy for increasing BCRP expression and decreasing drug-induced toxicity. Evaluation of existing drugs and new drugs as inhibitors of DNA methyltransferases or demethylases to modulate BCRP-mediated drug transport are areas of future studies for the development of precision medicine strategies.

3.2. Alteration of BCRP expression by histone modifications

Histone modification is another well studied epigenetic influence that affects the regulation of gene expression at the transcriptional level. Some amino acid residues (especially lysine and arginine) of the histone tails can be modified by acetylation, methylation, phosphorylation, citrullination, and ubiquitination. Some modifications specifically enhance gene transcription, but others repress.

The histone modification status near the \textit{ABCG2} promoter has a great impact on BCRP expression and multidrug-resistant phenotypes. Overexpression of BCRP and subsequent drug resistance are associated with increased acetylated histone H3 at the \textit{ABCG2} promoter\cite{74}. Permissive histone modifications, including histone H3 lysine 4 trimethylation (H3K4me3) and histone H3 serine 10 phosphorylation (H3S10P), were observed to accompany the development of drug resistant phenotypes. The repressive histone H3 lysine 9 trimethylation (H3K9me3) mark has an inhibitory effect on BCRP expression. Histone deacetylase (HDAC) inhibitors have been demonstrated to increase BCRP expression. Transcription of BCRP in acute AML cells was induced by treatment of phenylbutyrate, an HDAC inhibitor\cite{75}. KG-1a cells treated with phenylbutyrate developed resistance to multiple anticancer drugs, including daunorubicin, mitoxantrone, etoposide, and 5-fluorouracil. The HDAC inhibitors romidepsin, panobinostat, and vorinostat were able to restore normal levels of efflux transport mediated by BCRP in cells bearing the common \textit{ABCG2} variant Q141K by enhancing synthesis of BCRP protein and preventing aggresome targeting of the mutated protein\cite{76}. These results indicate that the involvement of epigenetic regulation in drug resistance transporters and treatment with HDAC inhibitors might increase drug resistance when combined with other anticancer drugs.

A strategy to develop precision medication approaches based on histone modifications of the \textit{ABCG2} gene can be potentially be established. As illustrated in Fig. 6 using histone acetylation as an example for histone modifications, inhibition of histone acetyltransferases or activation of histone deacetylases can serve as a potential strategy for decreasing BCRP expression, thus increasing drug efficacy, whereas inhibition of histone deacetylases or activation of histone acetyltransferases can serve as a potential strategy for increasing BCRP expression, thus enhancing drug disposition and decreasing toxic potential. Evaluation of existing drugs and new drugs as inhibitors of histone acetyltransferases or histone deacetylases for BCRP-mediated drug transport are areas of future study for the development of precision medicine approaches. A similar strategy can be developed for other histone modifications, such as histone methylation and phosphorylation, which have been shown to have a clear implication for the regulation of BCRP expression.

3.3. Alteration of BCRP expression by microRNAs

MicroRNAs (miRNA) are small non-coding RNAs transcribed from miRNA genes. Mature miRNAs are able to bind with...
Argonaut protein to form the RNA-induced silencing complex (RISC). An interaction between RISC and target mRNA causes mRNA degradation and/or decreased translational efficiency, which may further affect protein expression and function. The roles of miRNAs in anti-cancer therapy and multidrug resistance have been studied extensively in recent years, where regulation of drug transporters by miRNAs being one of the major mechanisms.

Expression of BCRP at the post-transcriptional level is regulated in part by miRNAs. Several studies have shown that miRNAs were able to regulate BCRP mRNA or protein expression and alter pharmacokinetic characteristics of BCRP substrate drugs. By targeting the 3'-untranslated region (3'-UTR) of BCRP mRNA, miR-328-3p was able to regulate BCRP expression level and restore the chemosensitivity of MCF7/MX100 cells to Mitoxantrone, a topoisomerase inhibitor used in cancer treatment. MiR-487a is another miRNAs that binds to 3'-UTR of BCRP mRNA. By targeting BCRP mRNA, miR-487a also restored the susceptibility of breast cancer cells to mitoxantrone. Further, inhibition of miR-487a reverses this phenotype. By comparing the differential expression patterns of miRNAs between the mitoxantrone-resistant breast cancer cell line MCF7/MX and its parental mitoxantrone-sensitive cell line MCF-7, another miRNA (miR-181a) was found to be the most down-regulated miRNA in MCF-7/MX cells. Luciferase assays proved that miRNA-181a targets the 3'-UTR of the BCRP mRNA. Overexpression of miR-181a down-regulated BCRP expression and caused mitoxantrone-resistant MCF-7/MX cells to become responsive to mitoxantrone. In addition, miRNAs belonging to the miR-302 family (including miR-302a, miR-302b, miR-302c, and miR-302d) were also able to inhibit BCRP expression by targeting the 3'-UTR of the BCRP mRNA. Consistent with similar targeting by the other miRNAs described here, miRNA-302 also increases the sensitivity of breast cancer cells to mitoxantrone. Other than directly targeting the 3'-UTR of the BCRP mRNA, a novel mechanism on the regulation of BCRP expression by miRNAs was found for miR-519c. MiR-519c was able to bind HuR, an mRNA binding protein, and regulate BCRP expression. The indirect regulation of BCRP through miR-519c-HuR regulatory pathway in colorectal cancer tissue was able to cause down-regulation of HuR or up-regulation of miR-519c, which repressed BCRP expression. These studies indicate that several miRNAs may serve as potential targets for preventing and reversing drug resistance in cancer treatment.

A strategy to develop a precision medication strategy based on miRNA binding to the ABCG2 3'-UTR can be established. As illustrated in Fig. 7, either increased miRNA expression or enhancement of miRNA binding by adding miRNA mimics can serve as a potential strategy for decreasing BCRP expression, whereas decreased miRNA expression or use of small RNA interference (siRNAs) for miRNA inhibition can serve as potential strategies for increasing BCRP expression with associated decreases in toxicity.

In summary, the epigenetic mechanisms that control BCRP expression are attractive areas for future study. Numerous studies confirmed that epigenetic pathways, including DNA methylation, histone modification, and non-coding RNA action, are able to regulate the transcription or translation of the ABCG2 gene and

**Figure 7** A strategy for the development of a precision medicine approach based on miRNA binding in the BCRP mRNA 3'-UTR in a mixed population.
mRNA, and thus affect function of the BCRP transporter. However, most studies on the epigenetic regulation of BCRP expression and function were performed in cell-based assays. Since those actions of epigenetic modifications are highly specific in different organs, collecting biological specimens and building up an epigenetic database are much more complicated as compared to genotyping approaches. Whether alterations of the epigenetic mechanisms in the target organs can be well represented in surrogate specimens is an area of active research. Until biomarkers representing alterations of the epigenetic mechanisms are discovered in clinical specimens, a precision medicine model for dose justification on drugs based on the epigenetic modifications is difficult at best, and more studies are needed in these areas.

4. Impact of physiologic factors upon BCRP expression and function

The expression of BCRP protein can be impacted by seemingly normal physiological factors. Normal human physiological and existential conditions such as gender, age, circadian rhythms, and oxygenation states have impacts on the expression and function of BCRP.

4.1. Impact of gender upon expression of BCRP

Whether or not BCRP expression differs between males and females is a current topic of debate. In one study, the quantification of BCRP protein expression in 65 human liver samples by mass spectrometry revealed no difference between males and females. Similarly, no differences in BCRP mRNA and protein were observed between males and females in a cohort of 87 human liver samples. In another study, the quantification of BCRP protein levels in 32 intestinal samples by Western blotting identified a slightly higher amount of BCRP protein in females compared to males. The sample sizes are perhaps not sufficient in some of these studies and only certain organs have been studied. Therefore, the question of whether or not gender is a significant contributor to differential BCRP expression requires further investigation.

4.2. Impact of age upon expression and function of BCRP

It is also unclear whether age should be considered as a physiologic factor that can impact BCRP function, but provocative studies exist. In one study, BCRP expression in 65 human liver samples was not correlated with ages examined (7 and 70 years). In a contrary study, BCRP protein levels in 67 human liver samples showed a correlation to age with significantly lower levels in elderly samples (age over 65 years, n=13) as compared to adults of 19–64 years, n=46 and neonates and children (0.02–18 years, n=8). Current human studies are not adequately to support any relationship between age and BCRP function. However, other factors such as health condition of tested subject might also lead to inter-individual difference in BCRP function, which covers up the impact of age in BCRP function. Further experiments and larger testing population are still needed to study how BCRP changes with human ages.

Age-related differences in the expression of BCRP have also been assessed in various animal models. BCRP mRNA expression in mouse liver is highest before birth, markedly decreases immediately after birth, stays a consistent level in immature ages, and gradually increases in young adults. A significantly lower level of BCRP mRNA was found in liver of older female rats (30 and 50 weeks of age) compared to young female rats (10 weeks of age). BCRP mRNA expression level in rat kidney was low in fetal samples, increased gradually following birth, and markedly increased on adult. More detailed studies in both humans and rodents are needed to determine whether or not age should have a significant impact on BCRP expression.

4.3. Impact on expression and function of BCRP by circadian rhythms

Expression of BCRP may be controlled by circadian rhythms. Circadian rhythms are molecular clocks regulating a variety of biological processes across the daily cycle. Circadian clocks have been shown to exert a strong impact on the pharmacokinetics of drug response, which may result in alterations of therapeutic efficacy or adverse reactions through taking drugs at different times of day. A circadian time-dependent expression pattern of BCRP was found in mouse small intestinal cells, which was controlled by circadian clock-activating transcription factor-4 pathway. This finding indicates that examinations on whether expression and function of BCRP are regulated by circadian rhythms in various human organs should be investigated to inform future precision medicine approaches to determine the best times of day for drug administration. In addition, since circadian disruption occurs in certain occupations such as shift work and in air travel, these too should be considered for their impact on BCRP function and for pharmacological treatments of shift workers and frequent air travelers. Future studies focused on circadian alterations of BCRP expression are needed in the development of precision medicine for specific populations with potential circadian disruption and desynchrony.

In summary, implications on alterations of BCRP expression by several physiologic factors, such as gender, age, and circadian, have been studied, but limitations still exist in the current research. The results from the studies examining the relationship of BCRP expression with age and gender are controversial upon different sample pools and no solid conclusions are made. For circadian effects, due to difficulty in collecting human sample and data, the studies were only performed on animals. Overall, the question of whether or not physiologic factors can be used as guidance for precision medicine approaches is unclear.

5. Impact of pathologic factors upon BCRP expression and function

5.1. Impact of diabetes upon expression and function of BCRP

The expression and functionality of BCRP may be altered by chronic disease conditions such as diabetes. Alterations of BCRP expression and function at blood–brain barrier under diabetes and the clinical significances have been discussed. In a streptozotocin-induced diabetic rat model, a lower level of insulin in diabetic rats is correlated with a lower level of BCRP expression and function in the brain cortex, and this is accompanied by increases in the distribution of BCRP substrates, such as prazosin and cimetidine, in the brain. Insulin treatment attenuated the impairment of BCRP function in the brain cortex induced by diabetes. In a high fat diet and streptozotocin-induced diabetic rat model, BCRP protein levels were induced in the kidneys, while
unchanged in the liver\textsuperscript{96}. These findings in animal models suggest that similar changes might take place in type 2 diabetic patients, and that the pharmacokinetics of drug that are substrates for BCRP may be altered with a potential of increased risk for drug toxicity and decreased therapeutic efficacy. So far, large-scale clinical investigations on a potential correlation between diabetes and altered expression and function of BCRP have not been conducted. However, a positive correlation was observed between levels of BCRP expression in placenta and levels of plasma hemoglobin A1c, a marker of the state of diabetes, in pregnant diabetic women, indicating that placenta BCRP might be affected by poorly managed diabetes\textsuperscript{97}. Even the population-based clinical studies on correlation between diabetes and BCRP expression and function are still limited. The studies above including both animal and human data suggested that diabetic should be considered as a factor in BCRP regulation. Prescription of BCRP substrate drugs to diabetic patients should be adjusted based on patients’ BCRP function.

5.2. Impact of Alzheimer’s disease on the expression and function of BCRP

The expression of BCRP may be altered in the brain of Alzheimer’s patients. A major pathological hallmark of Alzheimer’s disease is the accumulation of beta amyloid peptide plaques ($A\beta$ peptide) in the brain. BCRP has been referred as a gatekeeper of the blood–brain barrier that acts to prevent blood $A\beta$ peptide from entering the brain, based on an observation of a significantly higher accumulation of $A\beta$ peptide in the brain of Abcg2-null mice as compared to wild type mice\textsuperscript{98}. This study also found a significantly higher level of BCRP expression in the brains of Alzheimer’s disease patients as compared to age-matched controls. The up-regulation of BCRP expression is considered as a compensatory response in an attempt to reduce the $A\beta$ peptide burden by enhancing efflux of $A\beta$ peptide across the blood brain barrier\textsuperscript{99}. Alternatively, up-regulation of BCRP expression can protect the brains of patients with Alzheimer’s disease by reducing oxidative stress and inflammatory responses\textsuperscript{100}. Of therapeutic importance, high levels of BCRP expression in the brains of Alzheimer’s patients may lead to reduced brain drug penetration and efficacy for other neurological disorders.

5.3. Effect of viral infection expression and function of BCRP

The expression and function of BCRP can be altered during viral infection. The transactivator protein (Tat) from human immuno-deficiency virus type-1 (HIV-1) has been shown to alter the expression of various multidrug resistant transporters, including BCRP. In a T cell leukemia-derived cell line, overexpression of HIV-1 Tat upregulated BCRP expression, enhanced the BCRP-dependent efflux of doxorubicin, and reduced doxorubicin-induced cytotoxicity\textsuperscript{101}. This strongly suggested that decreased anti-viral drug efficacy may be caused by increase expression of BCRP by Tat. In summary, BCRP expression and function have been shown to be altered under different disease conditions, where the altered BCRP function might change the efficacy or toxicity of substrate drugs if given to patients. In this case, patients’ health condition and disease history might be important for prescription, especially BCRP substrate drugs. However, current study on how pathologic factors alter BCRP function is more focus on pre-clinical mechanistic studies with scarce clinical data available. For the development of a precision medicine approach, implications on clinical outcomes mediated by BCRP drug transport by pathologic factors need to be examined in future studies.

6. Impact on BCRP expression and function by environmental factors

6.1. Impact on expression and function of BCRP by diet

Nutritional factors have been shown to impact BCRP expression and function. Oleic acid is present in fat-rich meals. Human epithelial colorectal adenocarcinoma Caco-2 cells treated with oleic acid showed significant intracellular accumulation of the BCRP substrate mitoxantrone\textsuperscript{102}. This accumulation was due to decreased BCRP-mediated efflux caused by oleic acid treatment. Similarly, oleic acid treatment increased intestinal accumulation of mitoxantrone in mice, which was also the result of decreased BCRP-mediated efflux\textsuperscript{103}. Curcumin, an ingredient supplement widely used in food, has been shown to increase the sensitivity of breast cancer cells to several anticancer drugs, including paclitaxel, cisplatin, doxorubicin, and mitomycin C. The combination treatment of curcumin with mitomycin C significantly suppressed tumor growth derived from breast cancer stem cells in mice\textsuperscript{104}. Curcumin treatment renders breast cancer stem cells susceptible to mitomycin C by reducing BCRP expression. These studies show that diet and nutrition can affect BCRP function with obvious implications for drug efficacy.

Flavonoids, common polyphenolic compounds found in foods and herbal products, have been shown to inhibit BCRP function\textsuperscript{105}. Hesperetin, quercetin, daidzein, and stilbene resveratrol were also able to alter BCRP function for the transport of intracellular BCRP substrates\textsuperscript{106}. Considering the possibly of high intake of these natural products, altered BCRP substrate absorption and distribution might happen due to transporter dysfunction.

The fruit extract ellagitannin (containing ellagic acid) can be converted to urolithins by gut microbiota, which have been shown to possess anti-inflammatory and chemo preventive properties. Urolithins are proven BCRP substrates, which also showed an inhibitory effect on BCRP-mediated efflux of mitoxantrone in MDCKII cells\textsuperscript{106}. The results suggest that physiologically relevant concentrations of the gut microbiota-derived metabolites from fruit extracts could modulate BCRP-mediated transport processes of anticancer drugs and result in increased chemotherapy efficacy.

6.2. Impact on expression and function of BCRP by hypoxic conditions

BCRP expression is altered by hypoxic environments in organs. The alteration of BCRP expression by hypoxic conditions can occur either in tissues exposed to low oxygen environments\textsuperscript{107} or in persons at a high altitude\textsuperscript{108}. BCRP function in the apical membranes of the placenta syncytiotrophoblasts of pregnant women has a direct impact on protection of the fetus from exposure to various endogenous and exogenous substrates. An adequate supply of oxygen is critical for fetal development during pregnancy. A hypoxic state may have an impact on fetal development through modulation of BCRP expression in the placenta. Using human BeWo choriocarcinoma cells, a study revealed decreased expression of BCRP mRNA and protein under hypoxic conditions (3% oxygen), which is accompanied by
increased hypoxia inducible factor 1 subunit alpha (HIF-1α) signaling\textsuperscript{[108].} Hypoxia-induced functional impairment of BCRP was also demonstrated by accumulation of the BCRP substrate Hoechst 33342. The same study also found that women who gave birth at a high altitude (3100 m above the sea level) exhibited signs of chronic placental hypoxia in comparison with women at a moderate altitude (1600 m above the sea level)\textsuperscript{[108].} The women exposed to high altitude hypoxic conditions had enhanced expression of the HIF-1α protein and reduced BCRP protein in microvillus membranes. This study indicates that the regulation of placental BCRP expression by hypoxia may be an important factor for the protection of the fetus from exposure to chemicals during early development and in hypoxia-related pregnancy disorders. In addition, the fetus under hypoxic conditions may be more sensitive to drug-induced toxicity caused by BCRP-mediated efflux drugs.

BCRP expression in cancer can also be regulated by oxygen levels. Exposure of human para-carcinoma tissues and the pancreatic cancer cell line Capan-2 to hypoxic conditions induced BCRP expression\textsuperscript{[109].} The induction was due to increased HIF-1α binding of the ABCG2 promoter and consequent increased transcription. This hypoxia-induced chemo-resistance \textit{via} BCRP overexpression is indicative of a therapeutic potential for cancer treatment through an inhibition on the HIF-1α signaling. Hypoxic conditions or doxorubicin exposure individually or in combination could also result in induced BCRP expression in a colon cancer cell line, causing drug resistance\textsuperscript{[10].} All these results indicate that oxygen levels play an important role in regulating BCRP expression with tissue-dependent impact. Controlling oxygen levels or related signaling pathways could be a potential target for the improvement of cancer treatments by modulating BCRP-mediated drug efflux transport. However, the effect of hypoxia on BCRP function has been studied only in cell culture models, and clinical data of BCRP expression in pregnant women has not been assessed.

### 6.3. Impact on expression and function of BCRP by drugs

The expression or function of BCRP can be altered by drug treatment, potentially triggering drug–drug interactions. Modulation of BCRP function with drugs influences both cancer and non-cancer drug treatment outcomes.

With the potential role of BCRP in MDR, inhibition of BCRP function has been regarded as an attractive strategy to re-sensitize BCRP-overexpressing cancer cells to chemotherapy drugs. To date, several types of BCRP inhibitors have been discovered and reported. Fumitremorgin C (FTC), the first identified BCRP inhibitor, can reverse the resistance of several BCRP substrate anticancer drugs \textit{in vitro}\textsuperscript{[111,112].} Ko143, a tetracyclique analogue of FTC, also showed specific and potent inhibition of BCRP both \textit{in vitro} and \textit{in vivo}, possibly through an ATPase inhibitory mechanism\textsuperscript{[113].} There are also several other reported FTC-type BCRP inhibitors, including diketopiperazines and tryprostatin A\textsuperscript{[114,115].} PZ-39 is another identified BCRP inhibitor with the ability to cause BCRP inhibition by both binding to BCRP as well as acceleration of transporter degradation\textsuperscript{[116–118].} Several MDR inhibitors targeting other transporters were later found to inhibit BCRP function. Elacridar (GF120918), a P-glycoprotein (P-gp) inhibitor, was reported to re-sensitize cell to the BCRP substrate drug topotecan by increasing its oral bioavailability in a pharmacokinetic study\textsuperscript{[119].} Other than selective BCRP inhibitors, tyrosine kinase inhibitors (TKIs) are a group of drugs that display effective inhibition of MDR-associated ABC transporters. By targeting the ATP-binding pocket, TKIs block the hydrolysis of ATP and the energy-dependent transport function of ABC transporters. Several TKIs have shown BCRP inhibition and resensitization of cells to BCRP substrate drugs, including imatinib, dasatinib, ponatinib, lapatinib, sunitinib, and olmutinib\textsuperscript{[120–126].} Detailed information about the TKI mechanism is reviewed elsewhere\textsuperscript{[127,128].}

BCRP also has been reported to play an important role in the absorption and retention of statin drugs. Fostamatinib is a proven BCRP inhibitor. A cell-based study using polarized Caco-2 cells demonstrated a strong drug–drug interaction on accumulation of rosuvastatin influenced by inhibition of BCRP function by fostamatinib\textsuperscript{[129].} The study concludes that inhibition of intestinal BCRP function by fostamatinib can result in an up to 2-fold accumulation of rosuvastatin, which suggests that a reduced dose of rosuvastatin could be indicated in clinical practice. Co-administration of rosuvastatin with eltrombopag, an orally thrombopoietin receptor agonist, in MDCKII cells caused a strong cellular accumulation of rosuvastatin. The interaction of eltrombopag with rosuvastatin can also be explained by inhibition of BCRP-mediated efflux of rosuvastatin by eltrombopag\textsuperscript{[130].} In support of this, concomitant administration of eltrombopag with rosuvastatin in 42 healthy adult volunteers proved an increased plasma rosuvastatin area under curve (AUC) in the overall study population\textsuperscript{[131].}

These studies indicate that BCRP might be responsible for drug-drug interactions in clinical treatments. Drugs, which can alter BCRP function, should be paid with extra attention when given together with a BCRP substrate drug. However, there is only limited knowledge about which chemical or drugs can alter BCRP function and to avoid BCRP-related drug–drug interaction, further studies are still needed.

### 7. Conclusions

Interindividual differences in BCRP expression can alter pharmacokinetic properties of substrate drugs and cause variations in drug response and disease risk. In this review, we summarized several factors that might contribute to individual variability of BCRP function for drug transport.

Utilizing advanced sequencing technologies, BCRP genetic polymorphisms and single nucleotide polymorphisms have been identified and their clinical significance has been studied extensively. Several of the most common \textit{ABCG2} SNPs were summarized here and their roles in cancer prediction or treatment have been discussed using data from multiple clinical experiments. Epigenetic regulations, including DNA methylation, histone modifications, and non-coding RNAs are also shown to be involved in the regulation of BCRP expression. Other factors, including dietary factors or environmental toxicants might also impact BCRP function and influence BCRP substrate drug treatment. Physiologic conditions of patients, including age, gender, and health conditions, may have implications in the regulation of BCRP expression, but further studies are needed to determine their impacts on the development of precision medicine. Especially, considering the expression of BCRP in placenta, pregnant women should be carefully prescribed and the function of BCRP should be taken into consideration.

In summary, expression and function of BCRP protein in various human tissues are controlled by multiple individual
Development of precision medicine approaches based on inter-individual variability of BCRP/ABCG2

factors, leading to significant variability of BCRP function for drug transport. Understanding these factors and their impacts on BCRP function is critical for the development of precision medicine approaches for better therapeutic efficacy and less adverse toxicity.

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Development of precision medicine approaches based on inter-individual variability of BCRP/ABCG2

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