RESEARCH ARTICLE

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Genetic Polymorphisms in Glutathione S-Transferase (*GST*) Gene and Their Correlation with Toxicity of Chemotherapy in Breast Cancer Patients

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Abstract

Background: Glutathione S-Transferase (*GST*) is a family of phase II metabolizing enzymes contribute to detoxification and elimination of variety of endogenous as well as exogenous xenobiotics including chemotherapeutic agents. The comprehensive knowledge on the impact of genetic polymorphisms in GST) enzyme coding gene will help to understand the clinical outcomes in breast cancer patients treated with either Adriamycin or paclitaxel or combination of both. In this study we attempted to assess the genetic polymorphisms in *GSTM1, GSTT1, GSTP1* and their association with Adriamycin and Paclitaxel induced toxicity reactions in breast cancer patients. **Methods:** Two hundred BC patients receiving Adriamycin and Paclitaxel chemotherapy were enrolled in this study and chemotherapy induced hematological and non-hematological toxicity reactions were noted. The polymorphisms in *GSTM1, GSTP1* and *GSTT1* gene were studied by PCR and RFLP analysis. **Results:** After the univariate analysis of the genetic polymorphisms of *GSTM1*, *GSTP1* and *GSTT1* showed that *GSTT1* null genotype showed significant association with neutropenia (OR=2.84, 95% CI: 1.06-7.56; p=0.036) in breast cancer patients treated with Adriamycin and *GSTT1* null genotype in patients with >1 CINV toxicity confirmed significant correlation (OR=3.75, 95% CI: 1.46-9.59; p=0.005). The genetic polymorphisms of *GSTP1* (exon 5) A/G heterozygous genotype was significant in grade >1 toxicity reactions of mucositis (OR=3.22, 95% CI: 1.06-9.71; p=0.037) in breast cancer patients administered with Paclitaxel chemotherapy. **Conclusion:** The findings obtained from this study proposed significant involvement of *GSTT1*-null genotype in hematological neutropenia toxicity in response to Adriamycin and *GSTM1*-null genotype showed negative association with non-hematological toxicity (bodyache) in response to Paclitaxel.

Keywords: Breast cancer- Genetic polymorphism- *GSTM1- GSTP1- GSTT1-* Chemotherapy

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Introduction

Breast cancer (BC) is the most common cancer among women, increasing alarmingly in developing as well as developed countries. It is also the second biggest cause of cancer related deaths in women worldwide. Breast cancer is a heterogenous disease with different cellular origins and multiple distinct molecular subtypes [1, 2]. The rising incidence of BC may be attributed to multiple factors. Overall outcomes in the disease are better because of improved knowledge of disease biology and advances in treatment modalities. The current treatment of BC includes combinations of surgery, systemic therapy and radiotherapy. Systemic therapy in turn is an important component of treatment and includes various combinations of chemotherapy, hormonal therapy, targeted therapy and now immunotherapy. The modalities of treatment used in each patient are carefully chosen based on the type of BC, stage, patient characters, molecular features, etc. Usually, early stages of BC are treated with surgery followed by adjuvant therapy, whereas locally advanced disease requires neoadjuvant therapy to downstage the tumor followed by surgery and further systemic therapy. Metastatic BC on the other hand requires chemotherapy, hormonal therapy and targeted therapy in various combinations and sequences. As can be seen from these, it is evident that chemotherapy plays a significant role in treatment of most cases of BC. Many chemotherapy drugs are used in the protocols used with Anthracyclines (Doxorubicin, Epirubicin) and Taxanes (Paclitaxel, Docetaxel) forming the backbone of most schedules. Despite all the advances that have happened in the recent times, the outcome predictions cannot be generalized for all patients. Both the treatment

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responses and the toxicities experienced are varied and unpredictable in each patient. This unpredictability can largely be explained by the genetic variations of drug metabolizing enzymes of every patient. Therefore, it is important to understand pharmacogenetic susceptibility of each individual towards the efficacy and toxicity of chemotherapy drugs. The genetic diversity with inherited variations of individual determines the treatment outcomes where genetic variations of genes involved in drug metabolism may influence the efficacy and altered responses to the treatment [3]. The toxicities experienced can be acute occurring during chemotherapy or can be late, which appear after completion of treatment course. Acute toxicities are very important as they directly affect the treatment compliance and hence the outcomes. Acute toxicities can be haematological (anemia, thrombocytopenia, neutropenia and febrile neutropenia) or Non-hematologic (nausea, vomiting, fatigue, peripheral neuropathy, etc). Liver or kidney dysfunction can also be seen as a consequence of chemotherapy [4].

Glutathione S-Transferase (GST) is a family of phase II metabolizing enzymes contribute to detoxification and elimination of variety of endogenous as well as exogenous xenobiotics including environmental carcinogens and chemotherapeutic agents [5-7]. GSTs are grouped into *GSTM1*, *GSTT1* and *GSTP1*which play an important role in cellular protection and cellular resistance to drugs [8]. The GSTs are highly polymorphic in nature. The *GSTM1* and *GSTT1* exhibit a polymorphism where complete deletion of gene (Null genotype) which resulted into no enzyme activity and ultimately cells lose their ability to detoxify the drugs. The *GSTP1* polymorphism with single nucleotide substitutions in exon 5 with Ile105Val and exon 6 with Ala114Val amino acid substitution are well known. Thus, individuals with polymorphic GSTs with reduced or no enzymatic activity might be at higher risk of developing cancer due to reduced detoxification of carcinogenic compounds. The homozygous deletion of *GSTM1* (Null genotype) has been associated to influence clinical response and treatment outcomes with anticancer drugs [9, 10], however the results were inconsistent. Similarly, several chemotherapeutic drugs (doxorubicin, cisplatin) are substrate for *GSTP1*. The polymorphism of enzyme coding gene reduce the activity of *GSTP1* as compared to the wild-type form of *GSTP1*. Various reports on the polymorphisms showed that genetic variations in *GSTP1* (A313G; rs1695; Ile105Val and C341T;rs1338272; Ala114Val) and null genotypes of *GSTM1* and *GSTT1* have impact on treatment outcome and toxicities in different cancers treated with platinum based chemotherapy [5,11,12]. The detailed understanding of the plausible impact of genetic polymorphisms in drug detoxification or metabolizing enzyme genes helps in predicting clinical outcomes in patients receiving chemotherapy treatment [12-14]. Some evidences supported the hypothesis that the toxicity of chemotherapy drugs associated with genetic susceptibility of individual. However, the impact of genetic polymorphisms in GSTs genes (*GSTM1*, *GSTT1*, *GSTP1*) is not clear in predicting the BC outcomes in response to chemotherapy treatment in clinical settings. Also, the focus on studying the aspects of chemotherapy toxicity and their association with genetic makeup of the patient is lacking in Indian background. This knowledge will be of great benefit in therapeutic decisions of BC in future. Therefore in this study we assessed the polymorphisms in *GSTM1*, *GSTP1* and *GSTT1* genes and their possible association with both hematological and non-hematological toxicities in BC patients treated with both adjuvant and neo-adjuvant chemotherapy.

Materials and Methods

Patient enrollment and Clinical data

A total of two hundred (200) breast cancer patients seeking treatment at Department of Oncology of Krishna Hospital & Medical Research Center, Karad were enrolled in this study based on predefined inclusion and exclusion criteria.

Inclusion critera

i) Patients with 27 to 78 years age range, histopathologically confirmed, no metastasis at diagnosis, clinically localised or locally advanced tumors according to standard staging system were included in this study. ii) Patients diagnosed with Carcinoma Breast who were planned to receive Adjuvant chemotherapy postoperatively with standard Anthracycline and Cyclophosphamide chemotherapy followed by Paclitaxel chemotherapy. iii) Locally advanced Ca Breast patients receiving neoadjuvant chemotherapy for downstaging. iv) Patients with metastatic breast cancer receiving palliative chemotherapy with any of the drugs mentioned.

Exclusion criteria

i) Male breast cancer patients. ii) No pathological diagnosis, relapsed disease or metastasis, No associated co-morbidities, incomplete treatment taken, incomplete follow-up, missing or incomplete data were excluded from the study. iii) Patients with abnormal renal or liver function tests at the time of enrollment. iv) Performance score of Estern Cooperative Oncology Group (ECOG) >/= 2. The detailed clinical information with all examination findings were recorded in predefined proforma. The detailed clinicopathological and demographic characteristics and follow-up information of the patients was recorded and depicted in Table 2.

Chemotherapy Treatment Regimen Follow-up and Toxicity assessment

Once patient got enrolled in to the protocol after fulfilling inclusion and exclusion criteria,written iformed consent was be taken. Chemotherapy was planned as per the stage of the patient. Peripheral blood was collected for genetic polymorphism studies. Other labaratory studies which include complete blood count, renal function test and liver function test were done and noted in the proforma. Patients received 4 cycles of combination chemotherapy with Doxorubicin and Cyclophsphamide, followed by 4 cycles of 3 weekly Paclitaxel. After receiving 1st cycle of chemotherapy in each schedule, patient was followed again between Day10 to Day14 postchemotherapy for assessing chemotherapy related toxicities. Patient

was explained regarding possible adverse effects and advised to report back in case of serious side effects or report during scheduled followupand details were noted and graded as per NCI-CTC 4.03 criteria.Additionally patients with locally advanced breast cancer receiving neoadjuvant chemotherapy were assessed for response at the end of planned Adriamycin and Cyclophosphamide chemotherapy and again at end of Paclitaxel chemotherapy. The goal was to assess if specific genetic polymorphisms were correlating with chemotherapy related toxicities in a statistically significant manner.

Sample collection and Genomic DNA isolation

Five milliliter (mL) of intravenous blood from BC patients was collected in sterile EDTA containing vacutainer after receiving informed consent. Genomic DNA extraction was carried out from the peripheral blood sample using HipurA®Blood genomic DNA miniprep purification kit. (Cat no. MB504-250PR; HiMedia Laboratories) following the manufacturer's instructions. This genomic DNA was used for genotyping assays.

Genotyping assays

The genotyping of *GSTM1* and *GSTT1* were performed by polymerase chain reaction (PCR). The PCR amplification of *GSTM1* and *GSTT1* were carried out separately in 20 micro liter (µL) reaction mixtures containing 1X PCR buffer 0.2 mM each dNTP, 10 picomole (pmol) of each primers (IDT technologies) , 1U Taq DNA polymerase (GeNei, Merck Bioscience) and 100 nanogram (ng) of purified genomic DNA. The prime sequence used to amplify the *GSTM1* and *GSTT1* are shown in Table 1. The PCR conditions for amplification of 625 bp fragment of *GSTM1*: Initial denaturation at 95°C for 5 minutes (min) followed by 30 cycles of 95°C- 30 seconds (sec) , 56°C- 30 sec, 72°C- 30 sec and final extension at 72°C for 10 min. The conditions for *GSTT1* of 480 bp: Initial denaturation at 95°C for 5 min followed by 30 cycles of 95°C- 30 sec, 60°C- 30 sec, 72°C- 30 sec and final extension at 72°C- 10 min. The nonfunctional allele homozygous-null for *GSTM1* and *GSTT1* was evidenced by the absence of gene fragment, and presence of gene was indicated by amplification gene fragment in the PCR. The *GSTP1* Ile/Val of exon 5 and Ala/Val of exon 6 polymorphism was determined by PCR followed by restriction fragment length polymorphism (PCR-RFLP). The exon 5 and 6 of *GSTP1* were amplified by using specific primers mentioned in Table 1. The PCR cycling conditions for the amplification of 433 bp fragment of *GSTP1* Ile105Val: Initial denaturation at 95°C for 5 minutes (min) followed by 30 cycles of 95°C- 20 seconds (sec) , 55°C- 20 sec, 72°C- 20 sec and final extension at 72°C for 10 min) and 420 bp of *GSTP1* Ala114Val : Initial denaturation at 95°C for 5 minutes (min) followed by 30 cycles of 95°C- 30 seconds (sec) , 57°C- 20 sec, 72°C- 30 sec and final extension at 72°C for 10 min) respectively. The PCR amplicons were subjected to restriction digestion using restriction enzymes with digestion conditions are detailed in Table 1. The PCR products and restriction digestion reactions were checked by agarose gel electrophoresis in Tris-Acetate-EDTA (TAE) buffer thereafter stained with ethidium bromide (10 mg/mL) and visualized under UV-transilluminator and photographed in gel documentation system (BioRad Laboratories).

Statistical Analysis

Allele and genotype frequencies for each polymorphism were calculated and tested for their distribution according to the Hardy-Weinberg equilibrium. A univariate logistic regression model was used to assess the effect of the polymorphisms on toxicity (grade: 0-1 grade vs. grade: 2-4), expressing results as odds ratios (OR) and relative 95% confidence intervals (95% CIs). The OR estimated whether any association exists between the grade >1toxicity caused by chemotherapy and selected gene polymorphisms. The association of each polymorphism and clinical-pathological and demographic information of the patients was compared by means of a chi-square test. The occurrence of clinical severity of post chemotherapy adverse effects are defined as hematological and non-hematological toxicity reactions with >1grade. Statistical significance was set at $p < 0.05$. All statistical analyses were carried out using SPSS (Version 21.0).

Results

Demographic and Clinical characteristics of study population

Two hundred (200) patients of age range 27-78 years with median age 48 years were enrolled in the study and distribution of patients based on clinical characteristics, demographic information, histopathological grading and follow up details are presented in Table 2. 78.50 % women were more than 40 years age. Total of 61% women enrolled in the study were with ≤25 body mass index. 54.50 % patients were tobacco users as compared to non-smokers (45.50%). Majority of the women were economically poor and less educated. A total of 156 patients were treated with adjuvant chemotherapy and 44 patients were administered neo-adjuvant chemotherapy. Only 81 patients were exposed to adjuvant radiotherapy. 104 patients were administered Adriamycin and 96 patients were treated with Paclitaxel. A total of 47.50 % patients showed tumor size of more than 2 cm size and remaining 52.50% women were with ≤ 2 cm tumor size. Out of 200 patients 178 (89.0%), patients showed II and III TNM stage.

Genotype distribution of GSTM1, GSTT1, GSTP1 gene polymorphisms and chemotherapy toxicity in BC patients

The univariate analysis of polymorphism of phase II detoxification i.e. glutathione S- transferase gene family with *GSTM1*, *GSTP1* and *GSTT1* and their association with chemotherapy induced severe toxicity of hematological and non-hematological reactions are presented in Table 3 and Table 4. The hematological toxicities were grouped into anemia, neutropenia and thrombocytopenia. The severity of toxicities were grouped into grade ≤1 or >1 for each hematological reactions based on CTC criteria. Out of 200 patients, 104 patients were treated with adjuvant chemotherapy with Adriamycin

Table 2. Details of Demographic and Clinico-Pathological Characteristics of Breast Cancer Patients Enrolled in the Study

Variables	Number/Percentage (%)	
Total Number of patients	200	
Age (Mean ± SD) years	50.24 ±10.93 (Range:27-78) Median:48	
≤ 40	43	21.5
>40	157	78.5
BMI Kg/m^2		
<25	122	61
25-30	62	31
>30	16	8
Tobacco smoking Status		
Tobacco users	109	54.5
Tobacco no users	91	45.5
Diet		
Vegetarian	42	21
Mixed	158	79
Education		
High School	33	16.5
High School graduate (12 y)	26	13
College /Graduate	11	5.5
No School	130	65
Economic status		
Poor	148	74
Middle	52	26
Family history of Cancer		
Yes	50	25
No	150	75
Tumor localization		
Left breast	102	51
Right breast	98	49
Tumor size in cm		
≤ 2	105	52.5
> 2	95	47.5
Histological Grade		
I, II	107	53.5
III, IV	93	46.5
Clinical TNM Stage		
I	4	2
II	98	49
III	80	40
IV	18	9
Histopathological TNM Stage		
I	2	1
II	88	44
III	90	45
IV	20	10
Hormone Receptor Status		
ER/ PR+ve	83	41.5
ER/ PR-ve	109	54.5
ER/PR/Her2+ve	6	3
ER/PR/Her2-ve	85	42.5
ER/ PR+ve Her2-ve	78	39

Table 1. The List of Candidate *GST* Genes Selected in the Present Study with Details of PCR and RFLP Procedures Including Primers and Restriction Enzymes and Expected Products of Selected Genes.

Gene Genotype	rs number	Amino acid/ nucleotide change	Primer Sequence Forward/Reverse	PCR product size	Enzyme / Digestion conditions	Dominant (Wild type)	Heterozygous	Recessive (Mutant)
GSTM1		*GSTM1*	FP: 5'-CAA ATT CTG GAT TGT AGC AGA TCA TGC-3'			625 bp Gene Present		No Amplification
GSTM1		NULL	RP: : 5'-CAC AGC TCC TGA TTA TGA CAG AAG CC-3'	625 bp	NIL		NA	Null Genotype
								No Amplification
GSTT1		*GSTT1*	FP: 5'-TTC CTT ACT GGT CCT CAC ATC TC-3'	480 bp	NIL	480 bp	NA	Null Genotype
		NULL	RP: 5'-TCA CCG GAT CAT GGC CAG CA-3'			Gene Present		
GSTP1 exon-5 codon-105 (A313G)	rs1695	Ile105Val (A>G)	FP: 5'-GTA GTT TGC CCA AGG TCA AG-3' RP: 5'-AGC CAC CTG AGG GGT AAG -3'	433 bp	1 U of BsmA1 37°C for 16h	328 bp 105 bp	328 bp 222 bp 106 bp 105 bp	222 bp 106 bp 105 bp
GSTP1 codon-114 exon-6 (C341T)	rs1838272	Ala114Val (C>T)	FP: 5'-GGG AGC AAG CAG AGG AGA AT-3' RP: 5'-CAG GTT GTA GTC AGC GAA GGA G-3'	420 bp	1 U of AciI 37°C for 16h	246 bp 116 bp 58 bp	362 bp 246 bp 116 bp 58 bp	362 bp 58 bp

Table 2. Continued

Variables	Number/Percentage (%)	
Total Number of patients	200	
Hormone Receptor Status		
ER/ PR-ve Her2+ve	24	12
Chemotherapy		
Adjuvant chemotherapy	155	77.5
Neo-Adjuvant chemotherapy	31	15.5
Palliative chemotherapy	14	7
Radiotherapy		
Adjuvant RT	81	40.5
No Adjuvant RT	119	59.5

drug, severe hematological toxicity reactions (grade >1) including anemia in 23 patients, neutropenia in 25 patients and thrombocytopenia in 7 patients. Similarly the non-hematological toxicity reactions with grade>1 were observed with mucositis in 16 patients, CINV in 34 patients, fatigue in 37 patients, body ache in 15 patients and peripheral neuropathy in 5 patients. The associations between the genetic polymorphisms of *GSTM1*, *GSTT1* and *GSTP1* gene and severe hematological and non-hematological toxicity reaction in patients treated with Adriamycin and paclitaxel chemotherapy are studied. The hematological toxicities in response to Adriamycin chemotherapy and distribution of *GSTM1*, *GSTT1*, *GSTP1*

Table 3. Univariate Analysis of Polymorphisms of Phase II Detoxification (*GST*) Genes and Risk of Adriamycin Chemotherapy Induced Severe Toxicity of Hematological Reactions in Breast Cancer Patients

		Anemia			
Gene Name SNP	Genotype	Grade ≤1 (n=81)	Grade >1 (n=23)	OR (95% CI)	p value
GSTM1	*GSTM1*	47	16	1 (Reference)	
	NULL	34	7	0.60 (0.22-1.63)	0.32
GSTT1	*GSTT1*	65	14	1 (Reference)	
	NULL	16	9	2.61 (0.96-7.10	0.06
GSTP1	A/A	52	12	1 (Reference)	
rs1695	A/G	24	10	1.80 (0.68-4.75	0.231
	G/G	5	1	0.86 (0.09-8.11)	0.9
	A/G +G/G	29	11	1.64 (0.64-4.19	0.298
GSTP1	C/C	67	20	1 (Reference)	
rs1838272	C/T	14	2	0.47 (0.10-2.28)	0.355
	T/T	0	1	9.87 (0.38-251.87)	0.165
	C/T+T/T	14	3	0.71 (0.18-2.75)	0.628
		Neutropenia			
Gene Name SNP	Genotype	Grade ≤1 (n=79)	Grade >1 (n=25)	OR (95% CI)	p value
GSTM1	*GSTM1*	46	17	1 (Reference)	
	NULL	33	8	0.65 (0.25-1.69)	0.385
GSTT1	*GSTT1*	64	15	1 (Reference)	
	NULL	15	10	2.84 (1.06-7.56)	0.036*
GSTP1	A/A	52	12	1 (Reference)	
rs1695	A/G	23	11	2.07 (0.79-5.38)	0.134
	G/G	4	2	2.16 (0.35 (13.23)	0.402
	A/G +G/G	27	13	2.08 (0.83-5.19)	0.114
GSTP1	C/C	66	21	1 (Reference)	
rs1838272	C/T	13	3	0.72 (0.18-2.79)	0.64
	T/T	0	1	9.27 (0.36-0.236)	0.177
	C/T+T/T	13	4	0.96 (0.28-3.28)	0.957
		Febrile Neutropenia			
Gene Name SNP	Genotype	Grade ≤1 (n=80)	Grade >1 (n=24)	OR (95% CI)	p value
GSTM1	*GSTM1*	55	15	1 (Reference)	
	NULL	25	9	1.32 (0.50-3.42)	0.567
GSTT1	*GSTT1*	64	15	1 (Reference)	
	NULL	16	9	2.40 (0.89-6.45)	0.083

Table 3. Continued

Gene Name SNP	Genotype	Grade ≤1 (n=80)	Grade >1 (n=24)	OR (95% CI)	p value
		Febrile Neutropenia			
GSTP1	A/A	52	12	1 (Reference)	
rs1695	A/G	24	10	1.80 (0.68-4.75)	0.231
	G/G	4	2	2.16 (0.35-13.23)	0.402
	A/G +G/G	28	12	1.85 (0.73-4.67)	0.188
GSTP1	C/C	68	19	1 (Reference)	
rs1838272	C/T	12	4	1.19 (0.34-4.12)	0.78
	T/T	0	1	10.53 (0.41-269.07)	0.154
	C/T+T/T	12	5	1.49 (0.46-4.76)	0.499
		Thrombocytopenia			
Gene Name SNP	Genotype	Grade ≤1 (n=97)	Grade >1 (n=7)	OR (95% CI)	p value
GSTM1	*GSTM1*	57	6	1 (Reference)	
	NULL	40	1	0.23 (0.02-2.04)	0.191
GSTT1	*GSTT1*	75	4	1 (Reference)	
	NULL	22	3	2.55 (0.53-12.29)	0.241
GSTP1	A/A	62	2	1 (Reference)	
rs1695	A/G	29	5	5.34 (0.97-29.20)	0.053
	G/G	6	0	1.92 (0.08-44.52)	0.683
	A/G +G/G	35	5	4.42 (0.81-24.03)	0.084
GSTP1	C/C	81	6	1 (Reference)	
rs1838272	C/T	16	0	0.38 (0.02-7.07)	0.516
	T/T	0	1	37.31 (1.38-101.86)	0.031
	C/T+T/T	16	1	0.84 (0.09-7.49)	0.878

SNP, Single nucleotide polymorphism; OR, Odds ratio, CI, Confidence interval; Significance p< 0.05; *, Indicates significant Odds Ratio (p<0.05), p value determined based on χ^2

Table 4. Univariate Analysis of Polymorphisms of Phase II Detoxification (*GST*) Genes and Risk of Adriamycin Chemotherapy Induced Severe Toxicity of Chemotherapy Induced Non-Hematological Reactions in Breast Cancer Patients.

Gene Name SNP	Genotype	Grade ≤1 (n=88)	Grade >1 (n=16)	OR (95% CI	p value
		(Mucositis)			
GSTM1	*GSTM1*	51	12	1 (Reference)	
	NULL	37	4	0.45 (0.13-1.53)	0.207
GSTT1	*GSTT1*	69	10	1 (Reference)	
	NULL	19	6	2.17 (0.70-6.76)	0.177
GSTP1	A/A	58	6	1 (Reference)	
rs1695	A/G	25	9	3.48 (1.11-10.82)	0.031*
	G/G	5	1	1.93 (0.19-19.39)	0.575
	A/G+ G/G	30	10	3.22 (1.06-9.71)	0.037*
GSTP1	C/C	74	13	1 (Reference)	
rs1838272	C/T	14	2	0.81 (0.16-4.00)	0.799
	T/T	0	1	16.55(0.64-428.15)	0.09
	C/T+ T/T	14	3	1.21 (0.30-4.84)	0.777
		CINV			
Gene Name SNP	Genotype	Grade ≤1 (n=70)	Grade >1 (n=34)	OR (95% CI)	p value
GSTM1	*GSTM1*	40	23	1 (Reference)	
	NULL	30	11	0.636 (0.26-1.50)	0.305

Table 4. Continued

		CINV			
Gene Name SNP	Genotype	Grade ≤1 (n=70)	Grade >1 (n=34)	OR (95% CI)	p value
GSTT1	*GSTT1*	59	20	1 (Reference)	
	NULL	11	14	3.75 (1.46-9.59)	0.005*
GSTP1	A/A	44	20	1 (Reference)	
rs1695	A/G	22	12	1.20 (0.49-2.89)	0.684
	G/G	4	2	1.10 (0.18-6.50)	0.916
	A/G+ G/G	16	14	1.92 (0.78-4.69)	0.149
GSTP1	C/C	56	31	1 (Reference)	
rs1838272	C/T	13	3	0.41 (0.11-1.57)	0.197
	T/T	1	0	0.59 (0.02-15.11)	0.755
	C/T+ T/T	14	3	0.38 (0.10-1.45)	0.159
		Fatigue			
Gene Name SNP	Genotype	Grade ≤1 (n=67)	Grade >1 (n=37)	OR (95% CI)	p value
GSTM1	GSTM1	37	26	1 (Reference)	
	NULL	30	11	0.52 (0.22-1.22)	0.135
GSTT1	GSTT1	54	25	1 (Reference)	
	NULL	13	12	1.99 (0.79-4.98)	0.14
GSTP1	A/A	44	20	1 (Reference)	
rs1695	A/G	19	15	1.73 (0.73-4.09)	0.207
	G/G	4	2	1.10 (0.18-6.50)	0.916
	A/G+ G/G	23	17	1.62 (0.71-3.69)	0.245
GSTP1	C/C	57	30	1 (Reference)	
rs1838272	C/T	10	6	1.14 (0.37-3.44)	0.816
	T/T	0	1	5.65 (0.22-143.06)	0.293
	C/T+ T/T	10	7	1.33 (0.45-3.84)	0.598
		Bodyache			
Gene Name SNP	Genotype	Grade ≤1 (n=89)	Grade >1 (n=15)	OR (95% CI)	p value
GSTM1	*GSTM1*	56	7	1 (Reference)	
	NULL	33	8	1.93 (0.64-5.83)	0.238
GSTT1	*GSTT1*	70	9	1 (Reference)	
	NULL	19	6	2.45 (0.77-7.76)	0.125
GSTP1	A/A	57	7	1 (Reference)	
rs1695	A/G	27	7	2.11 (0.67-6.62)	0.2
	G/G	5	1	1.62 (0.16-16.01)	0.675
	A/G+ G/G	32	8	2.03 (0.67-6.13)	0.206
GSTP1	C/C	75	12	1 (Reference)	
rs1838272	C/T	14	2	0.89 (0.17-4.43)	0.889
	T/T	0	1	18.12 (0.69-470.20)	0.081
	C/T+ T/T	14	3	1.33 (0.33-5.36)	0.679
		Peripheral neuropathy			
Gene Name SNP	Genotype	Grade ≤1 (n=99)	Grade >1 (n=5)	OR (95% CI)	p value
GSTM1	*GSTM1*	58	5	1 (Reference)	
	NULL	41	0	0.12 (0.006-2.38)	0.168
GSTT1	*GSTT1*	73	3	1 (Reference)	
	NULL	23	2	2.11 (0.33-13.45)	0.427

Table 4. Continued

		Peripheral neuropathy			
Gene Name SNP	Genotype	Grade ≤1 (n=99)	Grade >1 (n=5)	OR (95% CI)	p value
GSTP1	A/A	63	1	1 (Reference)	
rs1695	A/G	30	4	8.40 (0.89-78.43)	0.061
	G/G	6	0	3.25 (0.12-88.36)	0.483
	A/G+ G/G	36	4	7.00 (0.75-65.05)	0.087
GSTP1	C/C	82	5	1 (Reference)	
rs1838272	C/T	16	0	0.45 (0.02-8.62)	0.599
	T/T	1	0	5.00 (0.18-137.61)	0.341
	C/T+ T/T	17	0	0.42 (0.02-8.11)	0.572

SNP, Single nucleotide polymorphism; OR, Odds ratio; CI, Confidence interval; Significance p< 0.05; *, Indicates significant Odds Ratio (p<0.05); p value determined based on χ^2

Table 5. Univariate Analysis of Polymorphisms of Phase II Detoxification (*GST*) Genes and Risk of Paclitaxel Chemotherapy Induced Severe Toxicity of Hematological Reactions in Breast Cancer Patients

		Anemia			
Gene Name SNP	Genotype	Grade ≤1 (n=80)	Grade >1 (n=16)	OR (95% CI)	p value
GSTM1	*GSTM1*	49	9	1 (Reference)	
	NULL	31	7	1.22 (0.41-3.63)	0.709
GSTT1	*GSTT1*	55	9	1 (Reference)	
	NULL	25	7	1.71 (0.57-5.11)	0.336
GSTP1	A/A	49	8	1 (Reference)	
rs1695	A/G	28	8	1.75 (0.59-5.17)	0.311
	G/G	3	0	0.83 (0.03-17.58)	0.905
	A/G +G/G	31	8	1.58 (0.53-4.64)	0.405
GSTP1	C/C	67	13	1 (Reference)	
rs1838272	C/T	12	3	1.28 (0.31-5.21)	0.722
	T/T	1	0	1.66 (0.06-43.13)	0.758
	C/T+T/T	13	3	1.18 (0.29-4.76)	0.806
		Neutropenia			
Gene Name SNP	Genotype	Grade ≤1 (n=81)	Grade >1 (n=15)	OR (95% CI)	p value
GSTM1	*GSTM1*	49	9	1 (Reference)	
	NULL	32	6	1.02 (0.33-3.14)	0.971
GSTT1	*GSTT1*	56	8	1 (Reference)	
	NULL	25	7	1.96 (0.64-5.99)	0.238
GSTP1	A/A	49	8	1 (Reference)	
rs1695	A/G	30	6	1.22 (0.38-3.87)	0.729
	G/G	2	1	3.06 (0.24-37.84)	0.382
	A/G +G/G	32	7	1.33 (0.44-4.05)	0.604
GSTP1	C/C	66	14	1 (Reference)	
rs1838272	C/T	14	1	0.33 (0.04-2.77)	0.311
	T/T	1	0	1.52 (0.05-39.45)	0.798
	C/T+T/T	15	1	0.31 (0.03-2.57)	0.281
		Febrile Neutropenia			
Gene Name SNP	Genotype	Grade ≤1 (n=82)	Grade >1 (n=14)	OR (95% CI)	p value
GSTM1	*GSTM1*	49	9	1 (Reference)	
	NULL	33	5	0.82 (0.25-2.68)	0.749

Table 5. Continued

		Febrile Neutropenia			
Gene Name SNP	Genotype	Grade ≤1 (n=82)	Grade >1 (n=14)	OR (95% CI)	p value
GSTT1	*GSTT1*	57	7	1 (Reference)	
	NULL	25	7	2.28 (0.72-7.18)	0.159
GSTP1	A/A	49	8	1 (Reference)	
rs1695	A/G	31	5	0.98 (0.29-3.29)	0.984
	G/G	2	1	3.06 (0.24-37.84)	0.382
	A/G +G/G	33	6	1.11 (0.35-3.50)	0.854
GSTP1	C/C	68	12	1 (Reference)	
rs1838272	C/T	13	2	0.87 (0.17-4.36)	0.867
	T/T	1	0	1.82 (0.07-47.43)	0.716
	C/T+T/T	14	2	0.80 (0.16-4.02)	0.796
		Thrombocytopenia			
Gene Name SNP	Genotype	Grade ≤1 (n=94)	Grade >1 (n=2)	OR (95% CI)	p value
GSTM1	*GSTM1*	56	2	1 (Reference)	
	NULL	38	0	0.29 (0.01-6.28)	0.432
GSTT1	*GSTT1*	62	2	1 (Reference)	
	NULL	32	0	0.38 (0.01-8.25)	0.541
GSTP1	A/A	57	0	1 (Reference)	
rs1695	A/G	34	2	8.33 (0.38-178.73)	0.175
	G/G	3	0	16.42 (0.28-957.98)	0.177
	A/G +G/G	37	2	7.66 (0.35-164.19)	0.192
GSTP1	C/C	78	2	1 (Reference)	
rs1838272	C/T	15	0	1.01 (0.04-22.14)	0.993
	T/T	1	0	10.46 (0.33-326.24)	0.18
	C/T+T/T	16	0	0.95 (0.04-20.75)	0.974

SNP, Single nucleotide polymorphism; OR, Odds ratio; CI, Confidence interval; Significance p< 0.05; *, Indicates significant Odds Ratio (p<0.05), p value determined based on χ^2

genotypes are summarized in Table 3. When we studied, *GSTM1*, *GSTT1* null genotypes and *GSTP1* (A/G and C/T) polymorphisms, the univariate analysis showed that none of the variant genotypes were associated with anemic reactions and thrombocytopenia in BC patients. The *GSTT1* null genotype when compared to the patients with *GSTT1* genotype, showed significant correlation with neutropenia (OR=2.84, 95% CI: 1.06-7.56; p=0.036). The non-hematological toxicities in response to Adriamycin chemotherapy and distribution of *GSTM1*, *GSTT1*, *GSTP1* genotypes are represented in Table 4. The univariate logistic regression analysis showed that none of the polymorphisms in *GSTM1*, *GSTT1* or *GSTP1* confirmed correlation with fatigue, body ache and peripheral neuropathy. The *GSTT1* null genotype in patients with >1 CINV toxicity confirmed significant correlation (OR=3.75, 95% CI: 1.46-9.59; p=0.005). The great difference was detected when mucositis reactions were compared with GST polymorphisms where, *GSTP1* (exon 5) A/G heterozygous genotype was significant in grade >1 toxicity reactions (OR=3.22, 95% CI: 1.06-9.71; p=0.037). The univariate analysis of *GSTM1*, *GSTT1* and *GSTP1* polymorphism and their relation with risk of paclitaxel chemotherapy induced hematological (Table 5) and non-hematological toxicity reactions (Supplementry Table 6) in BC patients showed non-significant association.

Association of GSTM1, GSTT1, GSTP1 polymorphisms with demographic and clinicopathological factors of BC patients

The association between genetic polymorphisms of *GSTM1*, *GSTT1*, *GSTP1* and the patients clinicopathological features are depicted in Supplementry Table 7. When demographic factors including age and BMI and clinicopathological of BC patients were considered, the univariate logistic regression analysis showed that none of the *GSTM1* and *GSTT1* genotype showed significant association. The *GSTP1* heterozygous (A/G) genotype showed negative association with >40 age of BC patients (OR=0.48 95% CI: 0.24-0.95; p=0.036). The results in present study found no association *GSTM1*, *GSTT1* and *GSTP1* polymorphisms with clinicopathological factors except histopathological TNM grade >II which was negatively associated with heterozygous A/G genotype of *GSTP1* (exon-5) (OR=0.53 95% CI: 0.29-0.94; p=0.031). There was no statistically significant association among

genotype distributions of *GSTM1 GSTT1*, *GSTP1* and Clinical TNM grading, ER, PR and HER2 status.

Discussion

Breast cancer is treated with adjuvant or neo-adjuvant chemotherapy along with surgery and radiotherapy. The response of patients towards chemotherapy is unlike because of diverse genetic susceptibility of each individual towards the treatment response. However, number of studies has been carried out on the genetic polymorphisms of different pathway genes and their association with carcinogenesis, but the information on treatment response and outcomes is inadequate. The drug and xenobiotic detoxification genes including phase I and phase II detoxification enzyme coding genes (Cytochrome P450 (CYP) and Glutathione S- transferase (GSTs) are important components in detoxification and elimination of chemotherapy drugs. Along with CYPs, the GSTs are also contemplated as potential modifiers of the adverse effects of both radiotherapy and chemotherapy [15, 16]. Earlier, polymorphisms of *GSTM1*, *GSTP1* and *GSTT1* are reported for their association with platinum based chemotherapy treatment outcomes in ovarian [17], breast cancer [18] and leukemia [19] (where as other reported converse opinion with no association in colorectal cancer [20]. However, neoadjuvant chemotherapy with adriamycin and paclitaxel drugs are not assessed by for their response and toxicity effects in any clinical settings. In present study, we attempted to address the association of GST genes with chemotherapy induced toxicity reactions and observed positive association of *GSTT1*-Null genotypes with hematological toxicity reactions in women where GSTT1- null genotype lower the risk of severe toxicity (grade >1) for neutropenia. The results obtained in current study also showed positive correlation of *GSTT1*-null genotype with non-hematological chemotherapy induced nausea and vomiting toxicity and *GSTT1*-null genotype when exposed to Adriamycin chemotherapy. Similarly, the heterozygous genotype of *GSTP1* exon-5 also significantly associated with mucositis reactions in BC patients treated with Adryamycin. The negative association of *GSTM1*-Null was noted with bodyace in patients administered paclitaxel, however polymorphisms in *GSTT1* and *GSTP1* did not show any significant association with hematological or non-hematological toxicities. These results were corroborated with the hypothesis that the successful treatment of patients with *GSTT1*-null genotype is associated with absence of GST enzyme activity [21-23], and showed no effect of *GSTT1*–null genotype on BC patients who had received chemotherapy with adreamycin and paclitaxel. These results are in agreement with the findings other researchers who showed no effect of *GSTM1*–null genotype in BC patients treated with chemotherapy [24, 25].

The hematological toxicity especially neutropenia was a major toxicity associated with polymorphisms of *GSTT1* and *GSTP1* observed in BC patients treated with adriamycin and paclitaxel drugs. Earlier reports demonstrateed the association of *GSTP1* with A313G polymorphism with chemotherapy induced severe hematological or nonhematological toxicities in cancer patients treated with paclitaxel [26] but our results verified no association of *GSTP1* with either hematological or non-hematological toxicities. Number of studies also revealed association between *GSTP1* 313 A>G polymorphism with platinum based therapy induced hematological toxicities in cancer patients [27, 28] including prostate [29] and hepatocellolar carcinoma [30], ovarian [31] and colorectal cancer [32]; however other proved inconsistent conclusions. In this study we observed that the genetic variability in *GSTT1* genotype was significantly associated with the treatment response and chemotherapy toxicity. The *GSTT1*-null genotype contributed significantly with severe toxicity i.e., neutropenia in BC patients treated with adjuvant chemotherapy. This results support the statement that the genetic variation in drug detoxification enzyme gene have a significant role in chemotherapy efficacy in breast cancer. Previous studies also demonstrated correlation of polymorphism between *GSTM1* and *GSTP1* and its association with treatment outcomes [33] or no association with platinum based chemotherapy in colorectal cancer or gastric cancer [34, 35]. However, to the best of our knowledge there are no reports from Indian clinical settings on association between polymorphisms of drug metabolizing enzyme coding genes and their clinical response and toxicity during BC treatment. In summary our study provides information about the significance of *GSTT1* and *GSTP1* polymorphisms in treatment outcomes after adjuvant and neoadjuvant chemotherapy in breast cancer.

In conclusion, the results obtained in this study suggested that the *GSTT1*-null genotype was significantly associated with neutropenia and heterozygous A/G genotype of *GSTP1* (rs1695) was associated with mucositis in Adriamycin treated breast cancer patients. The *GSTM1*-null genotype showed negative association with non-hematological (bodyace) toxicity in response to paclitaxel. This is the first study of the kind in this geographic and ethnic background and hence may serve as a benchmark for further evaluations to know clinical correlations of various treatment modalities.

Author Contribution Statement

Concept: RAG, SJB Design: RAG; KDD, AKG, Experimental Studies: KDD Clinical studies: RAG, AKG, Data analysis: KDD, RAG, Statistical analysis: KDD, Manuscript preparation: RAG, SJB, KDD, All authors read and approved the final manuscript.

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Approval

The study protocol was approved by protocol committee of Krishna Vishwa Vidyapeeth (Deemed to be University)

Declaration of Conflict of interest

The authors declare that they have no competing financial or any other conflict of interests that could have appeared to influence the work reported in this paper.

Ethics Committee Approval

The study protocol was approved by Institutional Ethics Committee of Krishna Vishwa Vidyapeeth 'Deemed to be University', Karad.

Abbreviations

BC: Breast Cancer
BMI: Body Mass Index
GST: Glutathione S-Transferase
PCR-RFLP: Polymerase Chain Reaction-Restriction Fragment Length Polymorphism
DNA: Deoxyribose Nucleic Acid
CINV: Chemotherapy Induced Nausea and Vomiting
ECOG: Estern Cooperative Oncology Group
CTC: Common Toxicity Criteria
OR: Odds Ratio
CI: Confidence Interval

References

1. Anderson WF, Matsuno R. Breast cancer heterogeneity: a mixture of at least two main types? J Natl Cancer Inst. 2006;98(14):948-51. https://doi.org/10.1093/jnci/djj295.
2. Danova M, Delfanti S, Manzoni M, Mariucci S. Tissue and soluble biomarkers in breast cancer and their applications: ready to use? J Natl Cancer Inst Monogr. 2011; 2011(43):75-8. https://doi.org/10.1093/jncimonographs/lgr023.
3. Ruiz C, Tolnay M, Bubendorf L. Application of personalized medicine to solid tumors: opportunities and challenges. Swiss Med Wkly. 2012;142:w13587. https://doi.org/10.4414/smw.2012.13587.
4. Yao S, Barlow WE, Albain KS, Choi JY, Zhao H, Livingston RB, et al. Gene polymorphisms in cyclophosphamide metabolism pathway,treatment-related toxicity, and disease-free survival in SWOG 8897 clinical trial for breast cancer. Clin Cancer Res. 2010;16(24):6169-76. https://doi.org/10.1158/1078-0432.CCR-10-0281.
5. Jiao L, Bondy ML, Hassan MM, Chang DZ, Abbruzzese JL, Evans DB, et al. Glutathione S-transferase gene polymorphisms and risk and survival of pancreatic cancer. Cancer. 2007;109(5):840-8. https://doi.org/10.1002/cncr.22468.
6. Ekhart C, Rodenhuis S, Smits PH, Beijnen JH, Huitema AD. An overview of the relations between polymorphisms in drug metabolising enzymes and drug transporters and survival after cancer drug treatment. Cancer Treat Rev. 2009;35(1):18-31. https://doi.org/ 10.1016/j.ctrv.2008.07.003.
7. Tulsyan S, Chaturvedi P, Agarwal G, Lal P, Agrawal S, Mittal RD, Mittal B. Pharmacogenetic influence of GST polymorphisms on anthracycline-based chemotherapy responses and toxicity in breast cancer patients: a multi-analytical approach. Mol Diagn Ther. 2013;17(6):371-9. https://doi.org/10.1007/s40291-013-0045-4.
8. Daly AK. Pharmacogenetics of the major polymorphic metabolizing enzymes. Fundam Clin Pharmacol. 2003;17(1):27-41. https://doi.org/10.1046/j.1472-8206.2003.00119.x.
9. Stanulla M, Schrappe M, Brechlin AM, Zimmermann M, Welte K. Polymorphisms within glutathione S-transferase genes (*GSTM1*, *GSTT1*, *GSTP1*) and risk of relapse in childhood B-cell precursor acute lymphoblastic leukemia: a case-control study. Blood. 2000;95(4):1222-8.
10. nderer G, Schrappe M, Brechlin AM, Lauten M, Muti P, Welte K, Stanulla M. Polymorphisms within glutathione S-transferase genes and initial response to glucocorticoids in childhood acute lymphoblastic leukaemia. Pharmacogenetics. 2000;10(8):715-26. https://doi.org/ 10.1097/00008571-200011000-00006.
11. Okcu MF, Selvan M, Wang LE, Stout L, Erana R, Airewele G, et al. Glutathione S-transferase polymorphisms and survival in primary malignant glioma. Clin Cancer Res. 2004;10(8):2618-25. https://doi.org/10.1158/1078-0432.ccr-03-0053.
12. Rednam S, Scheurer ME, Adesina A, Lau CC, Okcu MF. Glutathione S-transferase P1 single nucleotide polymorphism predicts permanent ototoxicity in children with medulloblastoma. Pediatr Blood Cancer.2013;60(4):593-8. https://doi.org/10.1002/pbc.24366.
13. Ulrich CM, Robien K, McLeod HL. Cancer pharmacogenetics: polymorphisms, pathways and beyond. Nat Rev Cancer.2003;3(12):912-20. https://doi.org/10.1038/nrc1233.
14. Scripture CD, Figg WD, Sparreboom A. The role of drug-metabolising enzymes in clinical responses to chemotherapy. Expert Opin Drug Metab Toxicol. 2006;2(1):17-25. https://doi.org/ 10.1517/17425255.2.1.17.
15. Sweeney C, Nazar-Stewart V, Stapleton PL, Eaton DL, Vaughan TL. Glutathione S-transferase M1, T1, and P1 polymorphisms and survival among lung cancer patients. Cancer Epidemiol Biomarkers Prev. 2003;12(6):527-33.
16. Duggan C, Ballard-Barbash R, Baumgartner RN, Baumgartner KB, Bernstein L, McTiernan A. Associations between null mutations in *GSTT1* and *GSTM1*, the GSTP1 Ile(105)Val polymorphism, and mortality in breast cancer survivors. Springerplus. 2013;2:450. https://doi.org/10.1186/2193-1801-2-450.
17. Lallas TA, McClain SK, Shahin MS, Buller RE. The glutathione S-transferase M1 genotype in ovarian cancer. Cancer Epidemiol Biomarkers Prev. 2000;9(6):587-90.
18. Ambrosone CB, Sweeney C, Coles BF, Thompson PA, McClure GY, Korourian S, et al. Polymorphisms in glutathione S-transferases (*GSTM1* and *GSTT1*) and survival after treatment for breast cancer. Cancer Res. 2001;61(19):7130-5.
19. Davies SM, Robison LL, Buckley JD, Tjoa T, Woods WG, Radloff GA, et al. Glutathione S-transferase polymorphisms and outcome of chemotherapy in childhood acute myeloid leukemia. J Clin Oncol. 2001;19(5):1279-87. https://doi.org/10.1200/JCO.2001.19.5.1279.
20. Stoehlmacher J, Park DJ, Zhang W, Groshen S, Tsao-Wei DD, Yu MC, Lenz HJ. Association between glutathione S-transferase P1, T1, and M1 genetic polymorphism and survival of patients with metastatic colorectal cancer. J Natl Cancer Inst. 2002;94(12):936-42. https://doi.org/10.1093/jnci/94.12.936.
21. Gao LB, Pan XM, Li LJ, Liang WB, Bai P, Rao L, et al. Null genotypes of *GSTM1* and *GSTT1* contribute to risk of cervical neoplasia: an evidence-based meta-analysis. PLoS One. 2011;6(5):e20157. https://doi.org/10.1371/journal.pone.0020157.
22. McIlwain CC, Townsend DM, Tew KD. Glutathione S-transferase polymorphisms: cancer incidence and therapy. Oncogene. 2006;25(11):1639-48. doi: 10.1038/sj.onc.1209373.
23. Abbas M, Kushwaha VS, Srivastava K, Banerjee M. Glutathione S-Transferase Gene Polymorphisms

and Treatment Outcome in Cervical Cancer Patients under Concomitant Chemoradiation. PLoS One. 2015;10(11):e0142501. https://doi.org/10.1371/journal.pone.0142501.
24. Yang G, Shu XO, Ruan ZX, Cai QY, Jin F, Gao YT, Zheng W. Genetic polymorphisms in glutathione-S-transferase genes (*GSTM1*, *GSTT1*, GSTP1) and survival after chemotherapy for invasive breast carcinoma. Cancer. 2005;103(1):52-8. https://doi.org/10.1002/cncr.20729.
25. Ludovini V, Antognelli C, Rulli A, Foglietta J, Pistola L, Eliana R, et al. Influence of chemotherapeutic drug-related gene polymorphisms on toxicity and survival of early breast cancer patients receiving adjuvant chemotherapy. BMC Cancer. 2017;17(1):502. https://doi.org/10.1186/s12885-017-3483-2.
26. Yoshihama T, Fukunaga K, Hirasawa A, Nomura H, Akahane T, Kataoka F, et al. GSTP1 rs1695 is associated with both hematological toxicity and prognosis of ovarian cancer treated with paclitaxel plus carboplatin combination chemotherapy: a comprehensive analysis using targeted resequencing of 100 pharmacogenes. Oncotarget. 2018;9(51):29789-800. https://doi.org/10.18632/oncotarget.25712.
27. Peng Z, Wang Q, Gao J, Ji Z, Yuan J, Tian Y, Shen L. Association between GSTP1 Ile105Val polymorphism and oxaliplatin-induced neuropathy: a systematic review and meta-analysis. Cancer Chemother Pharmacol. 2013;72(2):305-14. https://doi.org/10.1007/s00280-013-2194-x.
28. Lv F, Ma Y, Zhang Y, Li Z. Relationship between GSTP1 rs1695 gene polymorphism and myelosuppression induced by platinum-based drugs: a meta-analysis. Int J Biol Markers. 2018;33(4):364-71. https://doi.org/10.1177/1724600818792897.
29. Santric V, Djokic M, Suvakov S, Pljesa-Ercegovac M, Nikitovic M, Radic T, Acimovic M, et al. GSTP1 rs1138272 Polymorphism Affects Prostate Cancer Risk. Medicina (Kaunas). 2020;56(3):128. https://doi.org/10.3390/medicina56030128.
30. Sophonnithiprasert T, Saelee P, Pongtheerat T. Glutathione S-Transferase P1 Polymorphism on Exon 6 and Risk of Hepatocellular Carcinoma in Thai Male Patients. Oncology. 2020;98(4):243-7. https://doi.org/10.1159/000505213.
31. Deng X, Hou J, Deng Q, Zhong Z. Predictive value of clinical toxicities of chemotherapy with fluoropyrimidines and oxaliplatin in colorectal cancer by DPYD and GSTP1 gene polymorphisms. World J Surg Oncol. 2020;18(1):321. https://doi.org/10.1186/s12957-020-02103-3.
32. Ferracini AC, Lopes-Aguiar L, Lourenço GJ, Yoshida A, Lima CSP, Sarian LO, et al. GSTP1 and ABCB1 Polymorphisms Predicting Toxicities and Clinical Management on Carboplatin and Paclitaxel-Based Chemotherapy in Ovarian Cancer. Clin Transl Sci. 2021;14(2):720-8. https://doi.org/10.1111/cts.12937.
33. Yang Y, Xian L. The association between the GSTP1 A313G and *GSTM1* null/present polymorphisms and the treatment response of the platinum-based chemotherapy in non-small cell lung cancer (NSCLC) patients: a meta-analysis. Tumour Biol. 2014;35(7):6791-9. https://doi.org/10.1007/s13277-014-1866-4.
34. Ye F, Liu Z, Tan A, Liao M, Mo Z, Yang X. XRCC1 and GSTP1 polymorphisms and prognosis of oxaliplatin-based chemotherapy in colorectal cancer: a meta-analysis. Cancer Chemother Pharmacol. 2013;71(3):733-40. https://doi.org/10.1007/s00280-012-2067-8.
35. Wang Z, Chen JQ, Liu JL, Qin XG, Huang Y. Polymorphisms in ERCC1, GSTs, TS and MTHFR predict clinical outcomes of gastric cancer patients treated with platinum/5-Fu-based chemotherapy: a systematic review. BMC Gastroenterol. 2012;12:137. https://doi.org/10.1186/1471-230X-12-137.