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Original Article

Genetics Section

Breast Cancer Susceptibility Gene in Base Excision Repair Pathway in a Southern Indian Population

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ABSTRACT

Introduction: Breast cancer, the most complex multifactorial disease, one of the most common malignancies seen in females over the world. Single-Nucleotide Polymorphisms (SNPs) that occur in DNA repair genes are the contributors to cancer development as they lead to alteration in protein function, impair DNA damage responses, and result in loss of efficiency of DNA repair pathways.

Aim: To determine the association of the two polymorphisms (rs25487 and rs1799782) of the gene *XRCC1* involved in the Base Excision Repair (BER) pathway with breast cancer risk.

Materials and Methods: In the present study, 200 breast cancer cases and 200 healthy age-matched controls were analysed with regard to the genotype distribution of *XRCC1*rs1799782 and rs25487 polymorphism using Taqman allelic discrimination assay by Real time PCR. Chi-square test was the statistical method used, and compliance of the genotype frequencies with Hardy-Weinberg equilibrium was verified. The relative risk was assessed by determining the Odds Ratios (OR) and 95% Confidence Intervals (CI). Insilico studies were carried out for identifying the possible functional and deleterious nsSNPs in *XRCC1* gene.

Results: With regard to SNPs involved in the genes that play a role in the BER pathway, a significant association was found for the G/A heterozygous genotype of the *XRCC1* (rs25487) (OR; 1.79; 95% CI, 1.17-2.73; p-value: 0.006) with the risk of breast carcinoma. Also, an association was observed between the A/A homozygous genotype of the *XRCC1* (rs25487) (OR; 2.08; 95% CI, 1.08-4; p-value 0.02) and breast carcinoma risk. There was a lack of association of the CT/TT genotype of the *XRCC1* (rs1799782) (OR, 1.12; 95% CI, 0.69-1.80; P 0.63) with breast carcinoma risk. The insilco studies revealed structural variation in *XRCC1* gene with respect to rs25487.

Conclusion: The variant rs25487 of *XRCC1* gene was associated with the risk of breast carcinoma, but no association was found with regard to frequency of the rs1799782 variant in the *XRCC1* gene. The stability prediction and pathogenicity analysis with computational tools revealed the nsSNPrs25487 interfering with function and structure of the *XRCC1* protein. The data suggest that the variant of *XRCC1* (rs25487) may contribute to breast cancer susceptibility, extensive study linking DNA repair, environmental factors and ethnicity are needed.

Keywords: rs1799782, rs25487, *XRCC1*

INTRODUCTION

Worldwide, breast cancer research is a challenging one due to various reasons. The heterogeneous nature of breast cancer, is further complicated because of the differences in histopathological characteristics, stage of the cancer, and molecular subtypes [1-4]. Despite the fact of available treatment modalities, the mortality and incidence of breast cancer is on the rapid rise [5]. The DNA damage that is caused by exogenous and endogenous mutagenic substances is one of the reasons for the disease. If this damage progresses unrepaired, cell death may ensue on one hand, and on the other dysregulated cell growth can be a consequence, which would probably lead to cancer.

The body has several repair mechanisms in place, to repair the damaged DNA and among these the base excision repair pathway which is involved in mending the breaks in the DNA that are caused by oxidation damage [6]. *XRCC1* is a gene that plays a vital role in this pathway by interacting with the DNA pol-β polynucleotide kinase enzyme, DNA ligase IIIα, and PARP1, DNA repair machinery enzymes, to fulfill its role [7,8]. The SNP rs25487 of *XRCC1* is a functional polymorphism that is characterised by a base change of G to A, which leads to the substitution of glutamine for arginine at exon 10 codon 399 [9,10]. This SNP is present in the polymerase binding domain (poly ADP-ribose) of *XRCC1*, which is in the conserved region. The SNP rs1799782 is the conserved region that acts as a linker (hydrophobic) between the polymerase-interacting domains and polymerase β domain; thus, the substitution of tryptophan for arginine will lead to a change in the mechanism of interaction with the proteins that are involved in repair of DNA damages [11]. DNA repair processes are an essential part in a cell that helps to maintain genomic stability. A reduction in the repair capacity could lead to accumulation of damages, further mutations, and the development of diseases such as cancer [12]. Therefore, understanding the mechanisms of mutation and polymorphism, either somatic or inherited, of the DNA repair pathway may possibly lead to the development of new antitumour agents and chemopreventive targets, thus making this a critical area of research [12, 13]. The current study aimed to determine the risk attributed by the two polymorphisms in the *XRCC1* gene (rs25487 and rs1799782) polymorphisms which plays a major role in base excision repair pathway for breast cancer and to determine the structural and protein modification of the variant *XRCC1* by insilico tools.

MATERIALS AND METHODS

Study Design

The present case control study involved 200 patients with histologically confirmed breast cancer from Southern India. The patients were recruited from the Department of Oncology,

Sri Ramachandra Medical College and Research Institute, Chennai, India, from January 2013 to December 2017. The DNA was isolated from the collected samples. The quality and quantity of the DNA is checked by agarose gel electrophoresis and nanodrop. Genotyping was determined by Real time PCR Taqman allelic discrimination assay for rs1799782 and rs25487 of the *XRCC1* gene. All statistical analysis was carried by SPSS statistical software ver. 9.0. For the *XRCC1* gene computational tools are used to predict the protein stability for the variant *XRCC1* protein.

Study Subjects

For all the patients, relevant clinical and pathological data were obtained while obtaining the informed consent [Table/Fig-1]. For healthy volunteers, inclusion criteria were as follows: lack of a previous diagnosis of benign breast disease of any kind; no history of mastectomy, hysterectomy or oophorectomy; absence of a family history of ovarian, breast, endometrial and/or prostate cancer; and no mental or physical disability. With regard to ethnicity, the patients and controls were well matched, and they were also age matched assessed while obtaining consent for participation. The criteria for breast cancer patients to be included in the study were as follows: no previous treatment for cancer and confirmation of breast malignancy with histological diagnosis. Informed consent was obtained from all the subjects involved in the study. Ethical Clearance was obtained from the institutional ethical committee of Sri Ramachandra Medical College and Research Institute (Deemed to be University) (Ref No: IEC-NI/13/APR/33/28).

Variables	Breast cancer patients (Number/percentage)
Menopausal status	
Premenopausal	86
Post Menopausal	114
Familial/Sporadic	
Sporadic	171
Familial	29
Number of Births	
<2 births	151
>2 births	49
Parous/Nulliparous	
Parous	178
Nulliparous	22
Stage of Cancer	
Stage I	16
Stage II	60
Stage III	96
Stage IV	28
Receptor Status	
ER+ve	46%
PR+ve	42%
Her 2 +ve	34%
Triple Negative	20%

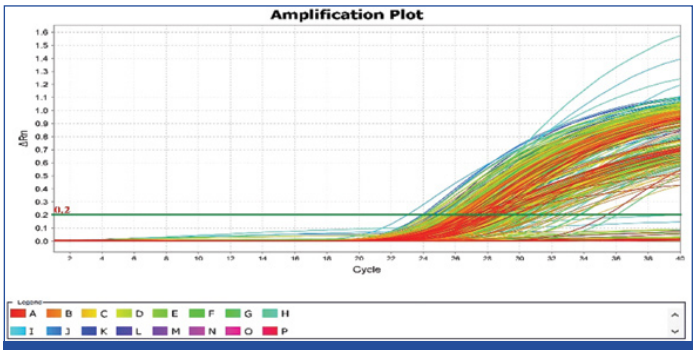
[Table/Fig-1]: Demographic features of breast cancer patients in the study.

Molecular Analysis by Taqman Allelic Discrimination Assay

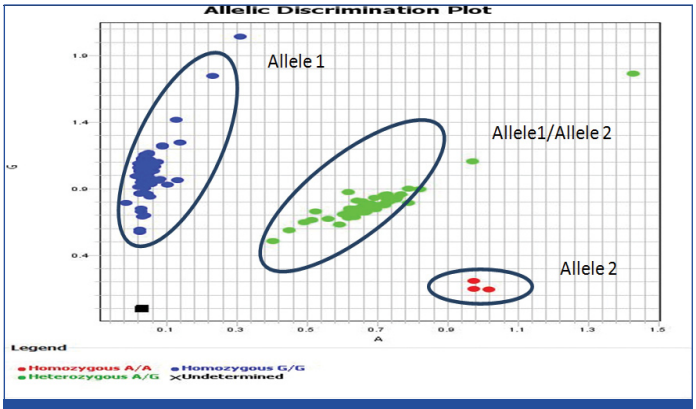
Peripheral blood lymphocyte samples about 3 mL was obtained by veinpuncture and collected into K2-EDTA vacutainers. Genotyping of the SNPs was carried out with real-time polymerase chain reaction technology (Taqman SNP Genotyping Assay, Applied Biosystems, Carlsbad, USA). The isolated DNA was amplified using Taq Gold Polymerase of Taqman PCR master mix in ABI machine Quant studio 6 Flex using sequence specific primers [Table/Fig-2]. The reaction volume was set to 5 µL, consisting of 2.50 µL of (2X) Taqman genotyping master mix, 0.25 µL of (20X) Taqman mutation assay mix and 2.25 µL of the genomic DNA of 10 ng concentration obtained on diluting the DNA with distilled water. Thermal cycle reaction condition is programmed to initial denaturation temperature and timing set at 95°C for 10 minutes followed by further denaturation at temperature set at 95°C for 15 seconds for 40 cycles followed by annealing and extension temperature and timing set at 60°C for one minute in 384 wells in Quant studio 6 Flex machine. Fluorescent signals are obtained for the amplification of each allele by the Taqman probes [Table/Fig-3]. After the amplification by PCR, an end plate read will be performed in ABI machine. The fluorescent measurement taken during the plate read will be used by the sequence detection system and fluorescence the (Rn) values are plotted with the signals obtained from each well [Table/Fig-4,5]. Each fluorescent detector will be a perfect match for the allele 1 and the other for allele 2. The VIC and FAM dye are used for labeling the primers [Table/Fig-2].

rsid	Primer Sequence
rs1799782	[VIC/FAM]:TCACCTGGGGATGTCTTGTTGATCC[A/G] GCTGAAGAAGAGAGCCCCCGGCCTC
rs25487	[VIC/FAM]:GGGTTGGCGTGTGAGGCCTTACCTC[C/T] GGGAGGGCAGCCGCCGACGCATGCG

[Table/Fig-2]: Primer Sequences for Taqman allelic discrimination assay real time PCR *XRCC1* gene regions.
rsid: Reference SNP cluster ID



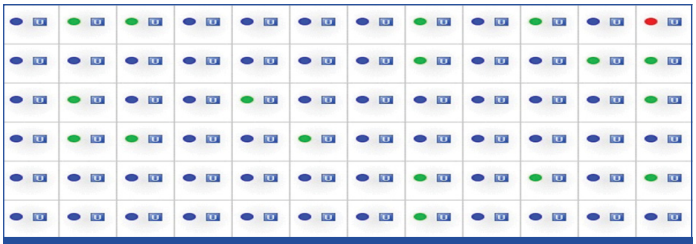
[Table/Fig-3]: Representative amplification plot of *XRCC1* rs1799782 for the by Taqman allelic discrimination assay by real time PCR.



[Table/Fig-4]: Representation of genotype detection by end point fluorescence using labelled primers.
The scatter plot is depicted. Taqman allelic discrimination assay by real time PCR Allele1- labelled with VIC and allele 2- labelled with FAM

STATISTICAL ANALYSIS

The SPSS statistical software version 9.0 was used for statistical analysis. Chi-square goodness-of-fit test carried out for comparison of expected and observed genotype frequencies for performing Hardy Weinberg equilibrium. Chi-square test



[Table/Fig-5]: Result grid in 384 well late for Taqman allelic discrimination assay real time PCR representation of rs1799782 polymorphism of *XRCC1* gene.
Taqman allelic discrimination assay by real time PCR Allele1- labelled with VIC and allele 2-labelled with FAM

was done for comparing the frequencies of the genotype of the polymorphisms between the controls and subjects with breast cancer. Using the genotypes of the wild type as the reference groups the OR and 95% CI was determined. p<0.05 level was considered to be significant.

Prediction of deleterious variants: The protein sequence for *XRCC1* gene and its SNP (rs25487) and (rs1799782) was obtained from SWISS-Protein database and from National Centre for Biotechnology Information (NCBI) database respectively. The 3D structure of *XRCC1*was obtained from PDB database. Five computational tools (I-Mutant Suite, iStable, PolyPhen2, SNAP, and PROVEAN) were used for insilico predictive analysis on the implication of the mutation on the function and structure of the variant *XRCC1* protein [Table/Fig-6]. The I-Mutant Suite predicts the protein the variant based on the stability of the protein in to three classes on the basis of the DDG value as neutral stability, large decrease in stability and large increase in stability. SVM based is table integrates the result obtained from the five-prediction analysis with the information of the sequence to analyse the stability of the protein. PolyPhen2 classifies the variant in to three types as benign, probably damaging and possibly damaging. SNAP predicts the variant effect on the function of the protein as neutral and non-neutral based on structural and evolutionary sequence information. The PROVEAN predicts the impact of the variation in the protein sequence due to amino acid substitution as neutral or deleterious [14-18].

Deleterious effects prediction of rs25487 using in silico tools	
Prediction tool	**Interpretation**
I-MUTANT 3	Decrease (-0.94)
I-STABLE	Decrease (0.774155)
PolyPhen 2	Probably damaging (0.979)
SNAP 2	Non-neutral
PROVEAN	Deleterious (-3.655)
Deleterious effects prediction of rs1799782 using in silico tools	
Prediction tool	**Interpretation**
I-MUTANT 3	Decrease (-0.31)
I-STABLE	Decrease (0.674266)
PolyPhen 2	Possibly damaging (0.899)
SNAP 2	Non-neutral
PROVEAN	Deleterious (-4.418)

[Table/Fig-6]: Deleterious effects prediction of rs25487 using in silico tools and (XRCC1; and rs1799782 using in silico tools. (XRCC1; 580C>T; Arg194Trp.
Five computational tools (I-Mutant Suite, iStable, PolyPhen2, SNAP, and PROVEAN) were used for insilico predictive analysis on the implication of the mutation on the function and structure of the variant XRCC1 protein

RESULTS

Hardy-Weinberg Equilibrium

The frequency distribution of the genotype of the polymorphism in the control and of the breast cancer patients was found to be consistent with Hardy Weinberg equilibrium [Table/Fig-7,8].

Genotyping and Breast Cancer Risk

The genotype frequency of distributions of the *XRCC1* rs1799782 and rs25487 polymorphisms between cases affected with breast cancer and healthy controls shown in the [Table/Fig-7,8]. For *XRCC1*rs1799782 C to T polymorphism, the frequency of homozygous wild type CC was 79% in controls and 77% in cases, heretozygous variant CT was 19.5% and 21.50% between controls and cases and Homozygous mutant TT was 3% in the control and cases. Thus, the distribution frequency is similar in cases and control groups for rs1799782. For rs25487 of *XRCC1* gene the homozygous GG; found to be 51.5% and 36.5% in controls and cases. The heterozygous GA; 39% and 49.5% in controls and cases with OR of 1.79 p=0.006. The homozygous AA genotype was 9.5% in controls and 14% in cases with OR 2.08 and p-value=0.02. A significant increase of GA and AA alleles of the gene *XRCC1*rs25487 was found in patients in comparison with that of controls. The polymorphism rs25487 and rs1799782 the genotype frequencies were compared with clinical parameters and tumor grade [Table/Fig-9,10]. For rs25487 of *XRCC1* gene the heterozygous genotype GA was found to be associated with premenopausal females odds ratio (OR) of 2.3, p-value=0.01. There was no association found for rs1799782 with regard to tumour grade and menopausal status [Table/Fig-9,10].

Gene	Genotype	Control	Case	OR (95% CI)	p-value
XRCC1	CC	158 (79%)	154 (77%)	Ref	
	CT	39 (19.50%)	43 (21.50%)	1.13 (0.69-1.84)	0.62
	TT	3 (1.50%)	3 (1.500%)	1.03 (0.20-5.16)	0.97
	CT+TT	42 (21)	46 (23)	1.12 (0.69-1.80)	0.63
	C	355 (88.75)	351 (87.75)	Ref	
	T	45 (11.25)	49 (12.25)	1.10 (0.71-1.69)	0.66
	MAF	0.11	0.12		
	HWP	0.81	0.99		

[Table/Fig-7]: Allele and genotype frequency distribution of *XRCC1* (rs1799782) in Breast cancer affected and control groups.
Association between breast cancer cases and SNP was evaluated by χ^2 test. HW compliance was verified for the genotypes. The relative risk was accessed by calculating the odds ratio(OR) and 95% confidence Interval (CI), p<0.05 level was considered to be significant

Gene	Genotype	Control	Case	OR (95% CI)	p-value
XRCC1	GG	103 (51.5%)	73 (36.5%)	Ref	
	GA	78 (39%)	99 (49.5%)	1.79 (1.17-2.73)	0.006**
	AA	19 (9.5%)	28 (14%)	2.08 (1.08-4.00)	0.02*
	GA+AA	97 (48.5)	127 (63.5)	1.85 (1.24-2.75)	0.002**
	G	284 (71)	245 (61.25)	Ref	
	A	116 (29)	155 (38.75)	1.55 (1.15-2.08)	0.003**
	MAF	0.29	0.39		
	HWP	0.45	0.54		

[Table/Fig-8]: Allele and genotype frequency distribution of *XRCC1* (rs25487) in Breast cancer affected and control groups.
Association between breast cancer cases and SNP was evaluated by χ^2 test. HW compliance was verified for the genotypes. The relative risk was accessed by calculating the odds ratio(OR) and 95% confidence Interval (CI), p<0.05 level was considered to be significant; p<0.05 level was considered to be significant, *p<0.05, **p<0.01

In Silico Predictive Functional Analysis

The pathogenicity prediction and stability was analysed using five important tools (I-Mutant Suite, iStable, SNAP, PolyPhen2, and PROVEAN). The analysis showed the variant Arg399Gln to be affecting the structure of the protein as the structure is different from the native protein, affecting the stability of the protein structure [Table/Fig-11,12]. The stability of the structure analysed by I-Mutant suite revealed decrease, largely in the stableness of the variant protein due to the free energy change (DDG value-0.94). The iStable analysis also revealed a decrease

in the structure stability due to the variant form. PolyPhen2, analysis by the score of 0.979 revealed the variant Arg399Gln as probably damaging the functional analysis performed through PolyPhen2 classified the mutant (Arg399Gln) as "Probably damaging", by the score of 0.979. The SNAP and PROVEAN analysis predicted the variant (Arg399Gln) to be deleterious [Table/Fig-6].

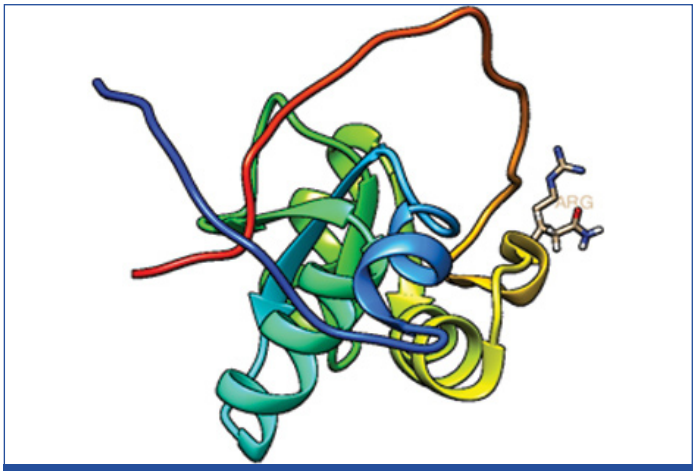
For rs1799782, the stability of the structure analysed by I-Mutant suite revealed decrease largely in the stableness of the variant protein due to the free energy change DDG value -0.31. The iStable analysis also revealed a decrease in the structure stability due to the variant form. PolyPhen2 analysis by the score of 0.899 revealed the variant Arg194Trp as possibly damaging. The SNAP and PROVEAN analysis predicted the variant Arg194Trp to be deleterious [Table/Fig-6].

XRCC1 (rs1799782) genotype among breast cancer patients and control stratified by age at diagnosis					
Genotype (rs1799782)	**Controls (%)**	**Patients (%)**	**OR**	**95% CI**	**p-value**
Premenopausal	N=86	N=86			
CC	68 (79%)	70 (81.3%)			
CT	17 (19.7%)	14 (16.2%)	0.8	0.36-1.74	0.57
TT	1 (1.16%)	2 (2.3%)	1.94	0.17-21.9	0.59
Menopausal	N=114	N=114			
CC	90 (78.9%)	84 (73.6%)			
CT	22 (19.2%)	29 (25.4%)	1.41	0.75-2.64	0.28
TT	2 (1.75%)	1 (0.87%)	0.53	0.04- 6.01	0.61
XRCC1 (*rs25487*) genotype among breast cancer patients and control stratified by age at diagnosis					
Genotype (rs25487)	**Controls (%)**	**Patients (%)**	**OR**	**95% CI**	**p-value**
Premenopausal	N=86	N=86			
GG	52 (60.4%)	34 (39.5%)			
GA	24 (27.9%)	36 (41.8%)	2.3	1.16-4.49	0.015**
AA	10 (11.6%)	16 (18.6%)	2.4	0.99-6.02	0.05
Menopausal	N=114	N=114			
GG	51 (44.7%)	39 (34.2%)			
GA	54 (47.3%)	63 (55.2%)	1.52	0.87- 2.65	0.13
AA	9 (7.8%)	12 (10.5%)	1.74	0.66-4.55	0.256

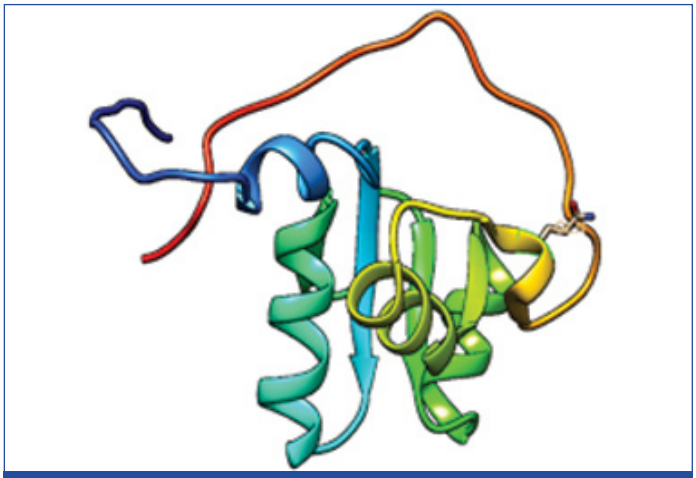
[Table/Fig-9]: *XRCC1* (rs1799782) and (rs25487) genotype among breast cancer patients and control stratified by age at diagnosis.
Association between premenopausal , menopausal and SNP was evaluated. The relative risk was accessed by calculating the odds ratio(OR) and 95% confidence Interval (CI), p<0.05 level was considered to be significant.

Association between *XRCC1* (rs1799782) and tumor grade					
Genotype (rs179982)	**Patients %**		**OR**	**95% CI**	**p-value**
	Low grade Tumors	**High Grade Tumors**			
CC	55 (72.3%)	99 (79.8%)			
CT	18 (23.6%)	25 (20.1%)	0.77	0.38-1.5	0.46
TT	3 (96%)	0 (0)	3.9	0.19-76.9	0.37
Association between *XRCC1* (rs25487) and tumor grade					
Genotype (rs25487)	**Patients %**		**OR**	**95% CI**	**p-value**
	Low grade Tumors	**High Grade Tumors**			
GG	27 (35.5%)	46 (37.08%)			
GA	34 (44.7%)	65 (52.4%)	1.12	0.59-2.10	0.72
AA	15 (19.7%)	13 (10.4%)	0.50	0.21-1.22	0.133

[Table/Fig-10]: Association between *XRCC1* (rs1799782), (rs25487) and tumor grade.
Association between tumor grade and SNP was evaluated. The relative risk was accessed by calculating the odds ratio (OR) and 95% confidence Interval (CI), p<0.05 level was considered to be significant



[Table/Fig-11]: *XRCC1* 399-ARG (NATIVE).
Native structure showing arginine at positions 399 position.



[Table/Fig-12]: *XRCC1* 399 - GLN (MUTANT).
Variant structure showing Glutamine at positions 399 position

DISCUSSION

XRCC1 plays a key role as a scaffold protein in the BER pathway and it helps to assemble the protein machineries of the DNA repair complex. *XRCC1* has been reported as a candidate gene influencing the risk of cancer. The present study aimed at assessing the association of polymorphisms in *XRCC1*, rs25487 (Arg399Gln) and rs1799782 (Arg194Trp), with breast cancer risk in a Southern Indian population and found rs25487 to be significantly associated with breast cancer risk. The reactive oxygen species that is generated during normal metabolic processes in the cell and also in response to exogenous genotoxins is one of the main causes of DNA damage [19,20]. The presence of adducts in the DNA has been found in breast ductal epithelial cells and even in tissue of human breast [21-23]. An increased risk of breast cancer is seen with the presence of DNA adducts [24,25]. BER pathway is the one that helps to remove the oxidised and alkylated bases. In addition to *XRCC1*, PARP1 and OGG1 enzymes are also involved in BER pathway. Polymorphisms in any of these genes may contribute to the risk of developing malignancy.

The *XRCC1* polymorphism and its association with other various cancers namely breast cancer, head and neck cancer and lung cancer have been studied [26-28]. The result obtained from the previous studies are not consistent [7,29]. The same is also true for the studies determining the association between *XRCC1* polymorphism and breast cancer, where studies have shown conflicting results [10,26,30,31]. Thus, the association between *XRCC1* genetic polymorphisms at codons 194 and 399 with regard to the susceptibility to breast cancer is still an open-ended question [32].

The 194Trp codon location is at a special hydrophobic linker region which is the highly conserved region of *XRCC1* gene. This region serves as a linker between the polymerase interacting domain

(ADP-ribose) Poly ADP ribose and the β domain of the DNA polymerase the tryptophan substitution in place of arginine, this change brings about the alteration in the binding efficiency either to both the domains or to one of domain [11]. In a study conducted by Przybylowska-Sygut K et al., and Moullan N et al., in polish population Arg194Trp was reported to be associated with breast cancer risk [11,26]. In a study conducted in white women found the Arg194Trp to be weakly associated with breast cancer risk, it was found to be associated with benign breast disease showing the heritability of the component [33]. Polymorphisms in *XRCC1* have also been shown to lead to reduced capacity of DNA repair [34,35]. Fan X et al., Studies suggested that the Arg194Trp polymorphism in *XRCC1* gene also has a key role in maintaining the efficiency of platinum-based chemotherapy [36,37]. Sanjari Moghaddam A et al., have propagated the idea that polymorphisms in *XRCC1* are associated with a risk for breast cancer [38]. However, Al Mutairi FM et al., study on Saudi patients, observed that *XRCC1* polymorphism, rs1799782, had a significant association with breast cancer risk [39]. A study from North eastern region of India reported significant association of Arg194Trp gene polymorphism with premenopausal breast cancer patients [40]. Th study conducted in southern India reported both the polymorphism to be associated with increased breast cancer risk [31]. Studies have also suggested the association of the polymorphism rs1799782 and rs25487 and the resistance capacity to cisplatin treatment in cancer patients [41,42]. The rs1799782 variant of *XRCC1* did not show a significant association when comparing genotype distribution in healthy controls with breast cancer patients though the insilico analysis revealed deleterious effect of the protein and decreased activity, this may be attributed to other factors influencing the risk [Table/Fig-5].

A functional SNP rs25487 caused due to single base change from G to A results in substitution of glutamine aminoacid in place of arginine [9,10]. The A allele transition was found to be highly associated with increased levels of glycophorin mutation, elevated frequency of sister chromatid exchanges, higher DNA adduct formation and more sensitive towards ionising radiation exposures all of which are the responses towards decreased efficiency of base excision repair pathway. Several mutation in the *XRCC1* gene have been associated with interruption in protein functionality, thereby the catalytic domain or the protein binding domain region is altered [43]. The functionally relevant polymorphism in *XRCC1* gene rs25487 has been studied in various cancers. It has also been studied in breast cancer owing to its importance as it is located in COOH-terminal end of the PARP-binding BRCT-domain [7,44,45]. This single base change variation causes the substitution of alternate amino acid in the BRCT domain resulting in complete disturbance in the functioning of *XRCC1* gene with low capability of DNA repair. Owing to its part in DNA repair machinery the variation due to SNP may modulate the susceptibility to breast cancer. Thus, when DNA repair proteins become deficient, probably due to genetic variants, this can lead to the initiation or can further aggravate breast cancer development. Despite the fact that many epidemiological studies have been performed in different populations on the association between DNA repair genes and breast cancer risk, only inconsistent and inconclusive results have been obtained. The *XRCC1* (rs25487) did not show any significant association with breast cancer susceptibility in Jordanian population [46]. Recent meta-analysis conducted on the results obtained on 69 studies provided no evidence of association for breast cancer risk for the polymorphism rs25487 and rs179982 of the *XRCC1* gene [47]. The results of the current study are also in agreement with those conducted by Wu K et al., a recent meta-analysis on 44 independent case-control studies [48]. The *XRCC1*rs25487 showed a significant association with hereditary and young breast cancer patients in Siberia [49]. The Non synonymous SNPrs25487 in the coding region of the *XRCC1* gene revealed variations in the phenotype that affected the stability of the protein, function and structure that prohibit the protein from performing its function in terms of forming complex with DNA repair partners [Table/Fig-4,5]. Based on the insilico tools the extent of deletriousness of the variant protein is predicted. The study evaluated the risk attributed by the polymorphisms rs25487 and rs179982. The study suggests the involvement of rs25487 polymorphism as a susceptible marker for breast cancer in South Indian women. Genotype profiling enables individualised therapy further extensive study with large samples linking the DNA repair pathway, environment factors; ethnicity would shed more light on breast cancer susceptibility.

LIMITATION

The limitation of the present study was the its sample size consideration, a study with a larger sample size should be conducted in the future for the individual population.

CONCLUSION

The results obtained on the genotyping analysis for rs25487 was also similar in effect determined by stability prediction and pathogenicity analysis with computational tools revealing the nsSNP rs25487 interfering with function and structure of the protein. The rs1799782 variant of *XRCC1* did not show a significant association with breast cancer risk. The *XRCC1* gene has been implicated in breast cancer susceptibility in Southern Indian Women.

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