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Impact of germline *RAD51D* mutations on breast cancer: Susceptibility to DNA-damaging agents

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Germline mutations in homologous recombination-related genes significantly increase the lifetime risk of breast, ovarian, and other cancers; thus, the concept of BRCAness has been extensively studied. In this study, we aimed to investigate the association between germline *RAD51D* mutations and breast cancer in clinical and experimental settings. Germline pathogenic or likely pathogenic variants in *RAD51D* were identified through multigene panel testing in the PLEASANT studies. Fourteen cases of breast cancer with germline pathogenic or likely pathogenic variants in *RAD51D* were identified. Compared to 1,446 wild-type cases of breast cancer, they appeared to be more aggressive. The proportion of patients with triple-negative breast cancer (TNBC) was higher and larger tumors (>2 cm) were more common. RAD51D-deficient tumors showed better pathological complete responses, defined as ypT0 ypN0, to neoadjuvant chemotherapy. Based on clinical findings, we analyzed the impact of RAD51D-deficiency on drug sensitivity *in vitro* to identify targetable vulnerabilities in RAD51D-deficient tumors. These TNBC cells were significantly more sensitive to cisplatin than wild-type TNBC cells, consistent with our clinical data. In conclusion, RAD51D-deficient tumors were shown to have a phenotype similar to that of BRCA1-deficient tumors, and further investigation of responses to DNA-damaging agents, particularly platinum-based chemotherapy, is warranted.

INTRODUCTION

The DNA damage response (DDR) is essential for protecting cells from genotoxic stress and preventing potentially mutagenic genomes from transforming into oncogenes.[1] Specifically, when DNA double-strand breaks (DSBs) occur, cells employ the homologous recombination (HR), non-homologous end joining (NHEJ), or microhomology-mediated end joining (MMEJ) pathways to maintain genomic stability.[2–4] HR is the most reliable repair pathway because it uses a homologous template to accurately repair DNA. In contrast to HR, NHEJ, which predominately occurs in the G0 or G1 cell cycle, can rapidly ligate DSB ends and introduce genomic insertions, deletions, and rearrangements.[5] In other words, DNA damage in HR-deficient cells is repaired by error-prone mechanisms such as NHEJ and MMEJ, which can cause oncogenic transformation. Therefore, individuals with HR deficiency are predisposed to develop several cancers, including breast, ovarian, and pancreatic cancers.[6]

BRCA1 and BRCA2 play key roles in HR to protect cells from DNA damage. The concept of BRCAness, which represents tumors with defective HR, has been expanded to provide appropriate therapeutic approaches for tumors with a phenotype similar to BRCA-deficient tumors.[7] Recent studies have shown that RAD51D plays an important role in maintaining genomic integrity.[8] RAD51D suppresses tumorigenesis by mediating HR to prevent genetic alterations.[9,10] RAD51D is one of six paralogs that shares a conserved domain

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with RAD51 in humans.[11] The BCDX2 complex, which contains RAD51B, RAD51C, RAD51D, and XRCC2, promotes RAD51 filament assembly to stimulate HR-mediated DNA repair.[12] Disruption of RAD51 paralogs impairs RAD51 foci formation and leads to HR deficiency.[13,14] These paralogs are also involved in replication fork protection and restart.[15,16] Although all of these paralogs are required for HR, germline pathogenic variants of *RAD51C* and *RAD51D* have been consistently associated with a predisposition to ovarian cancer and with breast cancer (BC) risk that is enriched in estrogen receptor (ER)-negative or triple-negative breast cancer (TNBC) disease across multiple cohorts.[17–20] However, population-based data on germline *RAD51D* variants in the Republic of Korea remain limited, and the association between germline *RAD51D* variants and BC, including clinicopathological features, has not been well characterized.

Previous studies have revealed that genomic instability induced by HR defects can confer sensitivity to DNA-damaging agents.[21–23] Platinum-based chemotherapy causes DNA damage in proliferating tumor cells by forming crosslinks with DNA, resulting in apoptosis. Poly (ADP-ribose) polymerase (PARP) inhibitors have been developed to specifically target DDR-deficient tumors based on synthetic lethality, which can minimize the off-target toxicity of conventional chemotherapeutic agents.[24–26] PARP inhibitors block the catalytic activity of PARP family members, causing PARP-trapping and inhibition of single-strand break repair.[27,28] In BC, olaparib and talazoparib have been approved for treatment of germline *BRCA* mutation carriers with metastatic HER2-negative BC.[29,30] PARP inhibitor monotherapy resulted in a significant improvement in progression-free survival compared with standard chemotherapy. These DNA-damaging agents effectively act on cancer cells, which commonly lack DNA repair capacity.[31] Nevertheless, the relatively low frequency of *BRCA1/2* mutations limits their applicability in patients.[32] Therefore, further research is needed to identify potential biomarkers that can increase their application.

In the current study, we determined the clinicopathological characteristics of patients with germline *RAD51D*-mutated BC in the Republic of Korea. Furthermore, we demonstrated that RAD51D-deficient tumors could benefit from platinum-based chemotherapy or PARP inhibitors *in vitro*. This can help to understand the association between germline *RAD51D*-mutation and BC and establish more appropriate treatment strategies for patients with *RAD51D* mutations, beyond *BRCA1/2*.

RESULTS

Differences in clinicopathological characteristics in germline *RAD51D* mutation carriers and non-carriers

To characterize BC with *RAD51D* mutations, patients with germline *RAD51D* mutations were identified using comprehensive multigene panel tests. We identified 14 patients with BC harboring a germline *RAD51D* variant in our cohort, with a prevalence of 0.97%. When comparing the clinical data, the *RAD51D* mutation carrier group showed more aggressive tumor phenotypes than the non-carrier group (Table 1). TNBC was more prevalent in the mutation carrier group (50%, $n = 7$) than in the non-carrier group (20.5%, $n = 297$) ($p = 0.014$). Mutation carriers were more likely to have ER-negative ($p = 0.001$) and progesterone receptor (PR)-negative tumors ($p = 0.003$) than non-carriers. Cell proliferation was increased in tumors with *RAD51D* mutations, as reflected by Ki-67 expression levels ($p = 0.048$). A tumor size >2 cm was more common in the carrier (64.3%, $n = 9$) than the non-carrier group (33.2%, $n = 480$) ($p = 0.003$). In the mutation carrier group, a higher proportion of patients received neoadjuvant chemotherapy (NAC) ($p = 0.006$), and notably, they achieved better pathological complete response (pCR) than the non-carriers who received NAC ($p = 0.03$) (Figure 1). Although the proportion of TNBC in each group receiving NAC was not significantly different, the pCR rate was 66.7% ($n = 6$) in the mutation carrier group, whereas it was only 30.5% ($n = 125$) in the non-carrier group. Most patients were diagnosed with BC before the age of 50 years, and there was no significant difference in age at diagnosis between the two groups because the patients enrolled in this study were at a high risk of hereditary BC.

Germline *RAD51D* variants identified in patients with BC

A total of 14 variants in *RAD51D* (GenBank: NM_002878.3) were identified in the present study (Figure 2). The effects of the variants on the protein were frameshift (42.9%, $n = 6$), splicing (28.6%, $n = 4$), nonsense (21.4%, $n = 3$), or exon deletion (7.1%, $n = 1$). The most frequent variant, p.Lys91fs, was located in exon 4 and accounted for nearly 50% of cases. The second most frequent variant in intron 9 disrupts RNA splicing. A novel deletion of exons 7–10 was observed in our cohort. All the variants are described in Table 2.

Depletion of RAD51D reduces RAD51 expression, leading to HR deficiency

Defects in RAD51 paralogs cause impairment of RAD51 foci formation and HR deficiency.[13,15,40] To evaluate the effect of *RAD51D*-inactivation on RAD51 expression, we used siRNA to silence the *RAD51D* gene in TNBC cell lines. To minimize confounding from canonical *BRCA1*-mutant models with well-established, severe HR deficiency, we did not use HCC1937 or HCC1395. Instead, we selected two widely used TNBC models (MDA-MB-231 and MDA-MB-468) to assess the incremental effect of RAD51D depletion across TNBC cellular contexts. Proteins from siRNA-transfected TNBC cell lines were analyzed by western blotting. Successful *RAD51D* silencing was confirmed using quantitative reverse transcription PCR (RT-qPCR) and western blotting. Treatment with siRNA reduced *RAD51D* mRNA expression by approximately 80% (Figure 3A) and downregulated protein expression by more than 70% (Figure 3B). Decreased RAD51 expression was observed in TNBC cells transfected with siRAD51D compared to that in cells transfected with siControl (Figure 3C). Because RAD51 mediates strand exchange to accurately repair DNA in HR, decreased RAD51 expression may be associated with HR deficiency.

Table 1. Clinicopathological characteristics between germline *RAD51D* mutation carriers and non-carriers

Status	WT (n = 1,446)	P/LP (n = 14)	p value
Age			
median	47.0 [39.0;56.0]	42.0 [38.0;52.0]	0.587
≤50	907 (62.7%)	10 (71.4%)	0.694
>50	539 (37.3%)	4 (28.6%)	–
Subtype			0.003
Luminal A	610 (42.2%)	1 (7.1%)	–
Luminal B	413 (28.6%)	3 (21.4%)	–
HER2	126 (8.7%)	3 (21.4%)	–
TNBC	297 (20.5%)	7 (50.0%)	–
TNBC			0.014
no	1,149 (79.5%)	7 (50.0%)	–
yes	297 (20.5%)	7 (50.0%)	–
ER			0.001
negative	424 (29.3%)	10 (71.4%)	–
positive	1022 (70.7%)	4 (28.6%)	–
PR			0.003
negative	610 (42.2%)	12 (85.7%)	–
positive	836 (57.8%)	2 (14.3%)	–
HER2			0.378
negative	1,181 (81.7%)	10 (71.4%)	–
positive	251 (17.4%)	4 (28.6%)	–
unknown	14 (1.0%)	0 (0.0%)	–
Ki67			
median	12.7 [5.0;32.0]	48.1 [6.3;72.1]	0.048
<14	752 (52.0%)	4 (28.6%)	0.196
≥14	683 (47.2%)	10 (71.4%)	–
unknown	11 (0.8%)	0 (0.0%)	–
Tumor size			
median	1.5 [0.8; 2.3]	2.2 [1.5; 3.4]	0.023
<2	960 (66.4%)	4 (28.6%)	0.003
≥2	480 (33.2%)	9 (64.3%)	–
unknown	6 (0.5%)	1 (7.1%)	–
Histologic grade			0.229
1	354 (24.5%)	1 (7.1%)	–
2	561 (38.8%)	7 (50.0%)	–
3	300 (20.7%)	5 (35.7%)	–
unknown	231 (16.0%)	1 (7.1%)	–
NAC			0.006
no	1,036 (71.6%)	5 (35.7%)	–
yes	410 (28.4%)	9 (64.3%)	–
Metastasis			0.216
no	1,117 (77.2%)	9 (64.3%)	–
lymph node	298 (20.6%)	4 (28.6%)	–
distant	31 (2.1%)	1 (7.1%)	–
Recurrence			1
no	1,432 (99.0%)	14 (100.0%)	–
yes	14 (1.0%)	0 (0.0%)	–

WT; wild-type, P/LP; pathogenic/likely pathogenic.
Subtype represents an IHC-based classification derived from ER/PR/HER2 status and Ki-67; therefore, subtype comparisons are not independent of these component variables and are presented for descriptive purposes.
Given multiple comparisons in Table 1, p values should be interpreted cautiously.

RAD51D-deficiency sensitizes TNBC cells to DNA-damaging agents

Previous studies have shown that patients with *BRCA*-mutated BC achieved a higher pCR rate than non-carriers after NAC.[22,41] Thus, as with our clinical data, we sought to determine the impact of RAD51D-deficiency on drug sensitivity to confirm whether RAD51D-deficient tumors can also benefit from DNA-damaging agents (Figure 4). The cell proliferation assay revealed that RAD51D-deficiency sensitized TNBC cells to cisplatin and olaparib, which induce genomic instability. Cisplatin was shown to be more effective than olaparib. Cisplatin was more toxic at low concentrations to BC cells treated with siRNA targeting *RAD51D* than to cells treated with siControl (Figure 5A). Mild reduction in cell viability was observed following olaparib treatment (Figure 5B). Olaparib treatment resulted in slightly greater differences in the viability of MDA-MB-231 cells compared to MDA-MB-468 cells. These data indicate that RAD51D contributes to the protection of genomic integrity, consistent with clinical data.

DISCUSSION

In this study, we characterized *RAD51D*-mutated BCs and found that RAD51D-deficiency contributes to genomic instability. We demonstrated that RAD51D-deficiency sensitizes BC cells to DNA-damaging agents using clinical data and *in vitro* experiments. Patients with *RAD51D*-mutated BC achieved a higher pCR rate than wild-type patients after receiving NAC. Platinum-based NAC is associated with significantly increased pCR rates in patients with TNBC[42]; however, the proportion of TNBC in each group was not significantly different in our study. Higher pCR rates may be due to the intrinsic properties of RAD51D contributing to DNA repair rather than the effect of TNBC. There is supporting evidence that breast cancers exhibiting BRCAness show enhanced sensitivity to chemotherapy.[43,44] BRCA status is associated with sensitivity to platinum-based chemotherapy and PARP inhibitors.[45–47] *BRCA* mutation carriers have higher pCR rates than non-carriers, and pCR is associated with a better prognosis, regardless of BRCA status.[45] In addition to BRCA1/2, RAD51-deficiency can be a predictive biomarker of response to platinum-based chemotherapy, and RAD51 overexpression is associated with resistance to chemotherapy.[48,49] Huang et al. revealed that regulation of

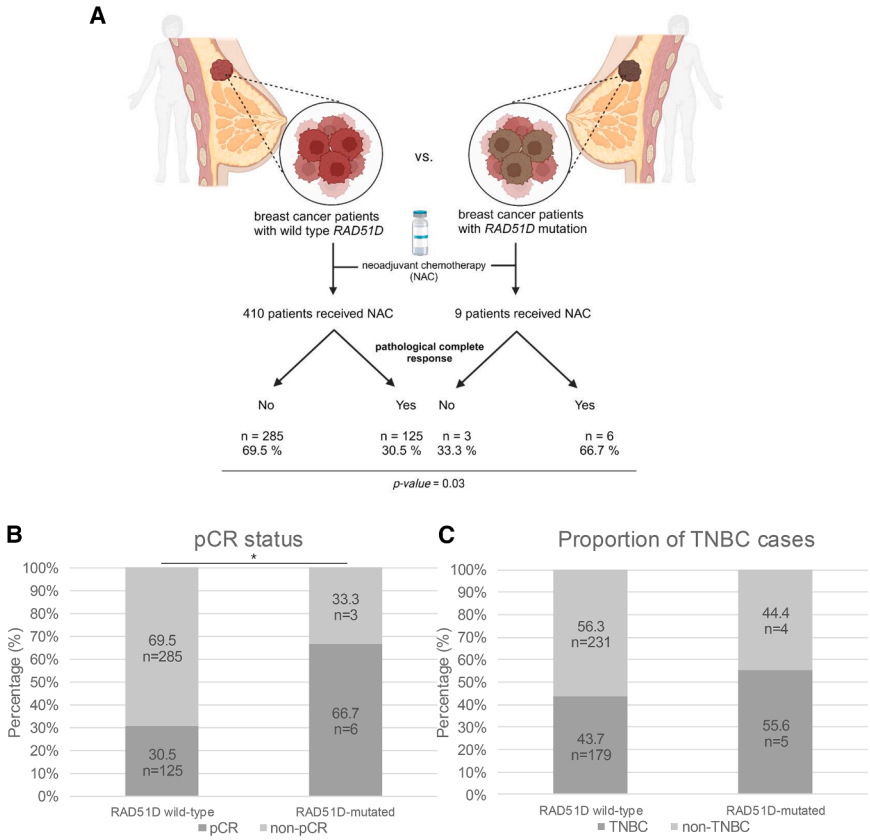

Figure 1. Pathological complete response according to *RAD51D* mutation status

(A) Mutation carriers achieved a better pCR than non-carriers after receiving NAC. (B) Bar graph showing the pCR status in the two groups (p = 0.03). (C) Bar graph showing the proportion of TNBC cases among patients receiving NAC.

RAD51 and RAD51D by microRNAs promotes chemosensitivity to DNA-damaging agents, mediated by HR defects.[50] The better efficacy of platinum-based chemotherapy and PARP inhibitors in tumors with BRCAness is related to defects in the DNA repair capacity.[51,52] These findings are consistent with those of the present study.

Based on these, we determined whether *RAD51D* inactivation affects the expression of RAD51, which plays a crucial role in strand exchange with homologous DNA templates in HR.[2,53] RAD51 expression was downregulated as a result of *RAD51D* silencing. This result is consistent with a previous study showing that disruption of RAD51 paralogs leads to reduced HR activity, with disruption of RAD51C, and RAD51D causing the greatest reduction.[13] RAD51 nuclear foci formation, as assessed by RAD51 expression at DNA damage sites, was lower in RAD51 paralog-deficient cells than in wild-type cells under both spontaneous and irradiation-induced conditions.[13] As RAD51D is involved in HR-mediated DNA repair, we hypothesized that nonfunctional RAD51D induces HR deficiency, resulting in vulnerability to DNA-damaging agents such as platinum-based chemotherapy or PARP inhibitors.

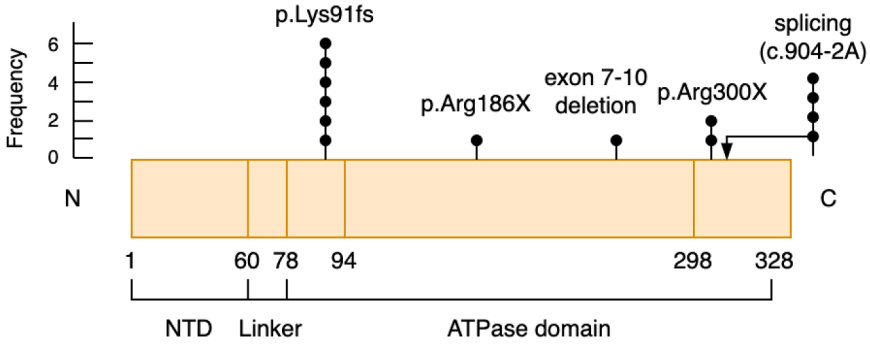

Figure 2. Distribution of deleterious variants in *RAD51D* identified in this study

The diagram describes the distribution of the 14 variants in *RAD51D*. (*N, N terminus; NTD, N-terminal domain; C, C terminus).

Table 2. Pathogenic variants in *RAD51D* identified in this study

Location	Nucleotide	Protein	Molecular consequence	Genotype	N (%)	Reference
Exon 4	c.270_271dup	p.Lys91IlefsTer13	frameshift	hetero	6 (42.9%)	Loveday et al.[33]; Thompson et al.[34]
Exon 6	c.556C>T	p.Arg186Ter	nonsense	hetero	1 (7.1%)	Sun et al.[35]; Baker et al.[36]
Exon 7–10	exon 7–10 deletion	NA	gross deletion	hetero	1 (7.1%)	Related exon deletions were reported by Susswein et al.[37]
Exon 9	c.898C>T	p.Arg300Ter	nonsense	hetero	2 (14.3%)	Sun et al.[35]; Konstanta et al.[38]
Intron 9	c.904-2A>T	NA	splicing	hetero	4 (28.6%)	Eoh et al.[39]

NA; not applicable.

Remarkably, RAD51D-deficient TNBC cells were significantly more sensitive to DNA-damaging agents *in vitro*, consistent with clinical data. DNA-damaging agents significantly reduced tumor cell proliferation in RAD51D-deficient TNBC cells, which was similar to the trend observed in *BRCA*-mutated cancer cells, albeit to different degrees.[54] Cisplatin was shown to be more effective against these cells than olaparib in this study, whereas there was only a slight difference in sensitivity to olaparib between siRAD51D-transfected BC cells and siControl-transfected cells. Overall, our experimental data suggest that RAD51D contributes to protecting genomic integrity and conferring sensitivity to cisplatin but appears to be insufficient to induce significant synthetic lethality with PARP inhibitors alone. This study is noteworthy because there are few studies evaluating drug sensitivity in RAD51D-deficient TNBC cells and reporting on patients with *RAD51D*-mutated BC.

RAD51D-mutated BCs were shown to have phenotypes similar to *BRCA1*-mutated BCs. The mutation carrier group had a high incidence of TNBC, which is the most aggressive subtype of BC with a poor prognosis. *RAD51D*-mutated BCs were more likely to have ER-negative and PR-negative tumors than wild-type BCs. The cell proliferation rate was significantly higher in *RAD51D*-mutated BCs than in wild-type BCs. The median Ki67 expression level in the mutation carrier group was 48.1, whereas that in the non-carrier group was 12.7. These data indicated that *RAD51D*-mutated BCs were similar to *BRCA1*-mutated BCs rather than to *BRCA2*-mutated BCs.[55] While it has been reported that the mean age at diagnosis of patients with *RAD51D*-mutated BC is significantly younger than that of non-carriers, the age at diagnosis of the mutation carrier and non-carrier groups did not differ significantly in our study.[56] This is because the PLEASANT study group only included high-risk patients, and one of the criteria was a diagnosis of BC before the age of 40 years. This selection bias, targeting patients with a high risk for hereditary BC syndrome, may have resulted in a relatively high frequency of deleterious *RAD51D* mutations in our study compared to previous studies, which ranged from 0.07% to 0.38%.[56–59]

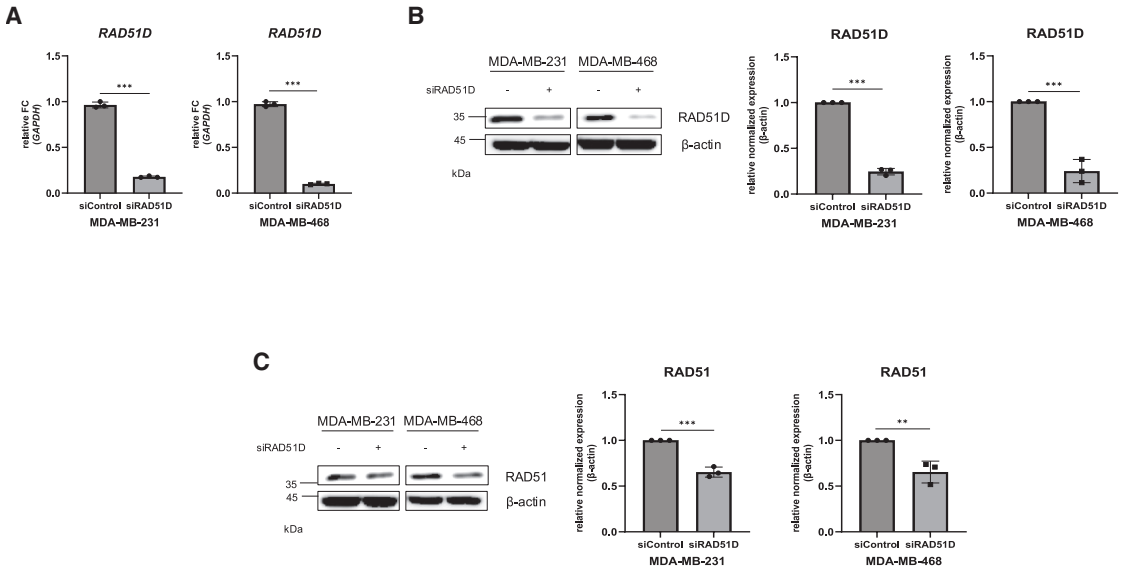

Figure 3. RAD51D depletion reduces RAD51 expression in TNBC cells

(A) Relative *RAD51D* mRNA expression levels in TNBC cells treated with siControl or siRAD51D. (B) Relative RAD51D protein expression levels in TNBC cell lines treated with siControl or siRAD51D. (C) *RAD51D* silencing reduces RAD51 protein expression. Data are presented as mean with SEM of $n = 3$ independent experiments (*$p < 0.05$, **$p < 0.01$, ***$p < 0.005$).

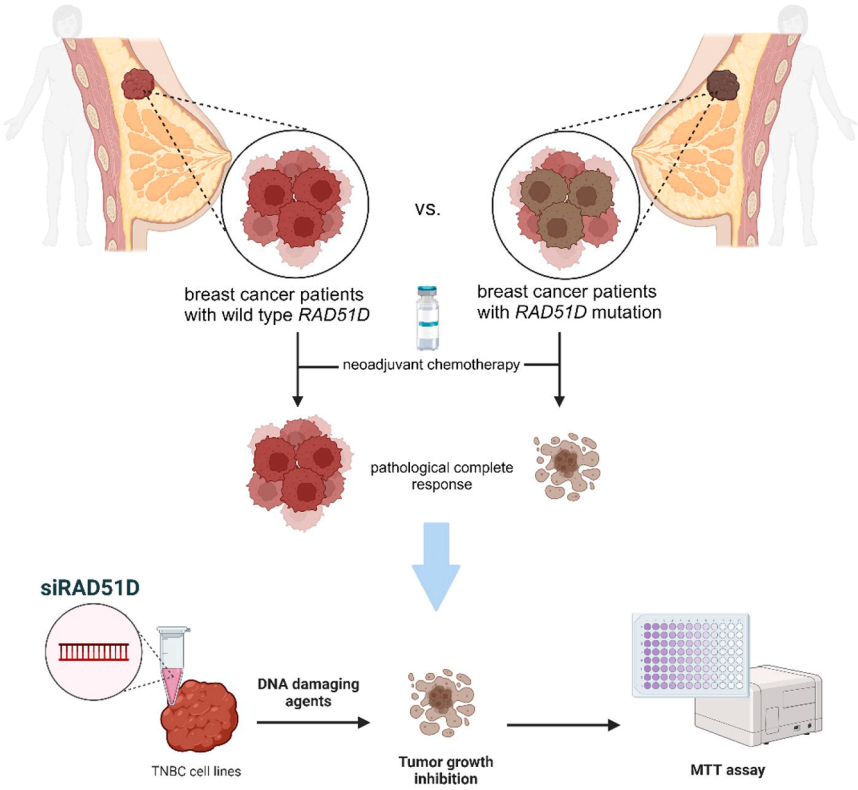

Figure 4. Study scheme showing that nonfunctional RAD51D sensitizes cancer cells to DNA-damaging agents

The scheme of the hypothesis states that nonfunctional RAD51D confers sensitivity to DNA-damaging agents in experimental and clinical data.

Among the 14 cases of *RAD51D* variants, we identified six cases of the p.K91fs variant. This variant was previously found in UK and Chinese populations.[33,34,56] The frequency of the p.K91fs mutation in our study was 42.9%, which was higher than that in the UK population, but similar to the Chinese study by Chen et al. in that it accounted for the largest proportion of mutation cases. Although gross exon deletions in *RAD51D* have been reported previously, a novel deletion of exons 7–10 was observed in our cohort.[37]

This study had some limitations. The first limitation was the small number of patients with germline *RAD51D* mutations, reflecting the rarity of these variants. This limited sample size restricts statistical power for analyses; therefore, our clinical associations should be validated in larger cohorts. Another point to consider is that experiments were not performed to accurately assess HR deficiency in RAD51D-deficient TNBC cells. Moreover, while we avoided canonical *BRCA*-mutant TNBC models by using MDA-MB-231 and MDA-MB-468, baseline functional HR status was not systematically profiled in these cell lines prior to RAD51D depletion. Although we observed a slight reduction in total RAD51 protein levels, which may be consistent with impaired HR, functional assays such as RAD51 foci formation will be required in future studies to directly assess HR capacity and to mechanistically link RAD51D-deficiency to the observed drug responses. In parallel, HR deficiency profiling of patient tumors, using HRDetect and assessment of loss of heterozygosity at the *RAD51D* locus, could enable a clearer interpretation of chemosensitivity driven by *RAD51D*-associated HR defects. While our siRNA-based experiments provide functional evidence that reduced RAD51D activity is associated with altered susceptibility to DNA-damaging agents, our current approach models *RAD51D* loss of function rather than the effects of individual patient-derived variants. Accordingly, further variant-specific functional validation using CRISPR-Cas9 knock-in models will help refine allele-specific consequences of the individual pathogenic variants in our cohort. Despite these limitations, our clinical observations together with the *in vitro* findings support a role for RAD51D in maintaining genomic stability.

Our study underlines the association between germline *RAD51D* mutations and BC and provides insights into the clinicopathological features of germline *RAD51D*-mutated BC. Notably, a significantly better response to DNA-damaging agents was observed in RAD51D-deficient BC cells in both clinical and *in vitro* data. Taken together, these results indicate that RAD51D contributes to the protection of genomic integrity.

MATERIALS AND METHODS

Clinical data collection

In a previous study called the PLEASANT study, comprehensive multigene panel tests were performed for patients at high risk of hereditary breast and ovarian cancer syndrome at Yonsei Cancer Center.[60] Patients with at least one of the following risk factors were considered to be at high risk for hereditary breast cancer syndrome: at least one case of breast or ovarian cancer in first- or second-degree relatives, a first diagnosis of BC before the age of 40, or bilateral BC. This study aimed to evaluate the clinical implications of multigene panel testing of genes other than *BRCA* in Korean patients with *BRCA1/2* mutation-negative BC. Comprehensive multigene panel tests targeting 65 cancer predisposition genes were performed on genomic DNA extracted from peripheral blood samples. Genetic variants were classified in accordance with the American College of Medical Genetics and Genomics guidelines. Mutation carriers were referred to as patients with pathogenic or likely pathogenic variants.

A total of 1,872 patients with high risk factors were enrolled in the PLEASANT study. In our study, patients with carcinomas other than BC, mutations detected in genes other than *RAD51D*, or missing clinical information were excluded. There were 14 patients

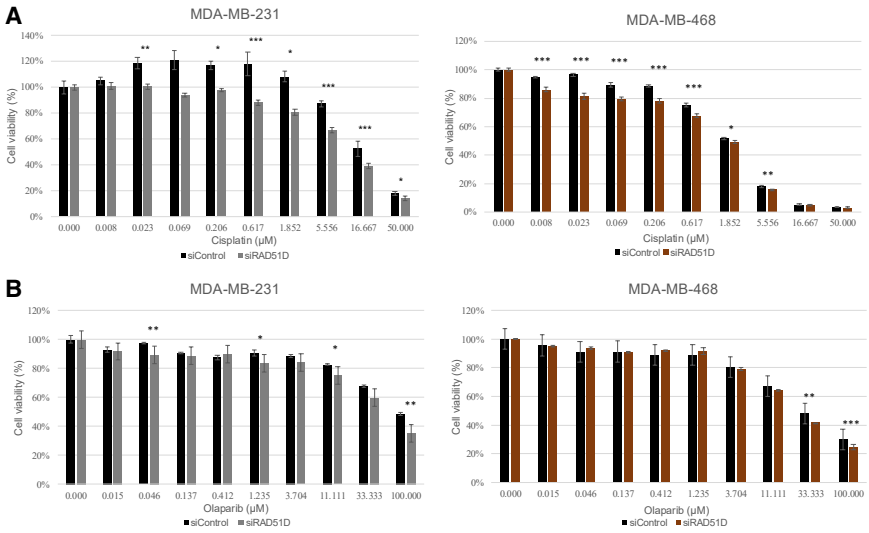

Figure 5. Efficacy of DNA-damaging agents in RAD51D-deficient TNBC cells

(A) RAD51D-deficiency induces TNBC cells to be more sensitive to cisplatin. (B) RAD51D-deficiency confers mild sensitivity to olaparib in TNBC cells. Data are presented as mean with SEM of $n = 6$ independent experiments (*$p < 0.05$, **$p < 0.01$, ***$p < 0.005$).

with BC carrying germline pathogenic or likely pathogenic *RAD51D* variants, and 1,446 BC patients with wild-type *RAD51D* were included as the control group. A detailed flowchart of the study is presented in Figure 6. The institutional review board at Severance Hospital, Seoul, Korea reviewed and approved this study (IRB approval number: 4-2023-1682). As this was a post-hoc analysis of the PLEASANT study (IRB approval numbers: 4-2015-0819 and 4-2018-0259), the institutional review board waived the need for informed consent.

The following clinicopathological characteristics of BC were obtained by reviewing the electronic medical records: age at diagnosis, next-generation sequencing results, molecular subtype, hormone receptor status, human epidermal growth factor receptor 2 (HER2) status, Ki67 expression level, tumor size, histologic grade, NAC, metastasis, and recurrence.

Definition of pathological complete response and molecular subtypes

pCR was defined as the absence of residual invasive and *in situ* cancer in the breast and axillary lymph nodes, ypT0 ypN0, after NAC.[61] Patients with TNBC were offered anthracycline-, taxane-, nitrogen mustard-, or platinum-containing NAC. Patients with HER2-positive BC were offered a taxane- or platinum-containing regimen in combination with trastuzumab or pertuzumab. Molecular subtypes of BC were determined based on hormone receptor status, HER2 status, and Ki67 expression level, which were assessed by immunohistochemistry of tumor tissues.

Cell culture

The human TNBC cell lines MDA-MB-231 (ATCC, HTB-26) and MDA-MB-468 (ATCC, HTB-132) were cultured in RPMI 1640 medium (Thermo Fisher Scientific, Waltham, MA, USA) and Dulbecco's modified Eagle's medium (Thermo Fisher Scientific), supplemented with 10% fetal bovine serum (Thermo Fisher Scientific) and 1% penicillin and streptomycin (Thermo Fisher Scientific). All cells were maintained at 37°C and 5% CO_2 in a humidified incubator.

Cell proliferation assay

Cisplatin (Cayman Chemical, Ann Arbor, MI, USA) was dissolved in phosphate buffered saline containing 140 mM NaCl prior to culture treatment. Olaparib (Selleckchem, Houston, TX, USA) was dissolved in dimethyl sulfoxide (Duchefa Biochemie, Amsterdam, Netherlands) to yield a 10 mM stock solution, and aliquots were stored at −80°C for drug treatment. Cells were seeded in 96-well plates (Corning, Corning, NY, USA) and transfected with small interfering RNA (siRNA) for 4 h according to the manufacturer's instructions. Transfection conditions were pre-optimized to avoid cytotoxicity. Drug-containing medium was added (top-up) and cells were treated for 48 h prior to viability measurement using an MTT assay. Olaparib was serially diluted by 1/3 from 100 μM, and cisplatin was serially diluted by 1/3 from 50 μM, and used to evaluate the sensitivity of RAD51D-deficient BC cells. After 48 h of treatment, the medium was replaced with fresh medium containing diluted MTT (10%) and incubated for 2 h at 37°C. Formazan crystals were dissolved in methanol and the absorbance was measured at 570 nm using a VersaMax Microplate Reader (Molecular Devices, San Jose, CA, USA). siRNA-only controls without drugs were included, and drug effects were normalized to the corresponding siRNA-only condition. Cell proliferation assays were performed in at least three independent replicates.

siRNA transfection

RAD51D gene silencing was achieved by siRNA transfection using the RNAiMAX Transfection Reagent (Thermo Fisher Scientific) according to the manufacturer's instructions. Cells were seeded in six-well plates at a density of 5×10^5 cells/well and transfected with siRNA after 24 h incubation. Pre-designed siRNA purchased from Bioneer (AccuTarget Negative Control siRNA, Bioneer Corporation, Daejeon, Republic of Korea) and Thermo Fisher Scientific (SilencerSelect siRNA against *RAD51D*, assay ID: S11743) was used to induce RAD51D-deficiency in BC cells. The samples were analyzed 48 h after transfection.

RT-qPCR

Total RNA was extracted using TRIzol reagent (Thermo Fisher Scientific) according to the manufacturer's instructions, and the RNA

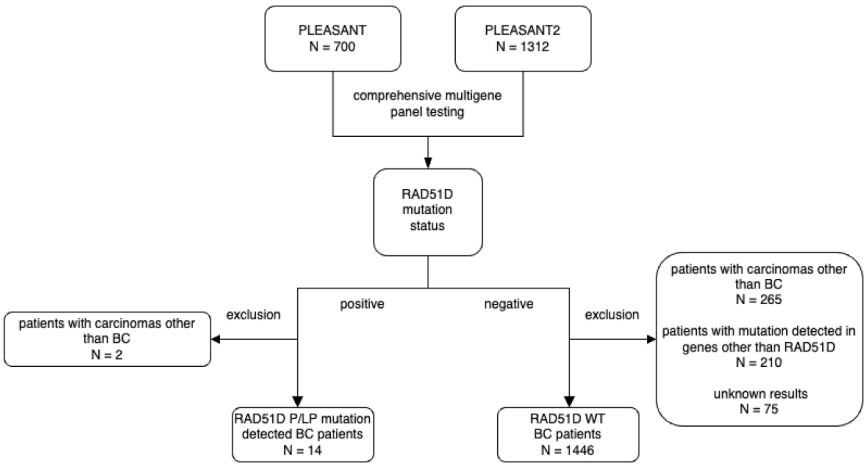

Figure 6. Flowchart of the study population
The flowchart shows the patient selection process.

concentration was determined using a Nanodrop 2000/2000× spectrophotometer (Thermo Fisher Scientific). Reverse transcription was performed using PrimeScript RT Master Mix (Takara, Kyoto, Japan) in an Applied Biosystems Veriti 96-well Thermal Cycler (Thermo Fisher Scientific). Real-time PCR was performed with SsoAdvanced Universal SYBR Green Supermix (Bio-Rad, Hercules, CA, USA) in a CFX96 Touch Real-Time PCR Detection System (Bio-Rad). Target gene expression levels were determined by normalizing to housekeeping gene levels. The following primers were used in this study: *RAD51D* (forward-AATGGCGCTGATCT CTACGA, reverse-TGTCAGCCCTCCATTGGAAT) and *GAPDH* (forward-CAACGGATTTGGTCGTATTGG, reverse-GCAACAAT ATCCACTTTACCAGAGTTAA).

Western blot analysis

Parental TNBC cell lines and their transfected cells were lysed in a cell extraction buffer (Thermo Fisher Scientific) containing a protease inhibitor cocktail (Roche, Basel, Switzerland). The protein concentration in each sample was measured using a QuickStart Bradford protein assay kit (Bio-Rad). Sodium dodecyl sulfate (SDS, 5×) sample buffer was added to the cell lysates and the samples were heated at 100°C for 7 min. From each sample, 10 μg of protein was loaded onto 10% SDS-polyacrylamide gel electrophoresis gels and then transferred to a nitrocellulose membrane (Bio-Rad) for 2 h. The membranes were blocked with 5% skim milk in Tris-buffered saline-Tween 20 (TBST) for 1 h and then incubated with primary antibodies diluted in 5% skim milk overnight at 4°C. The membranes were washed with TBST and incubated with secondary antibodies in 5% skim milk for 1 h at room temperature. After washing the membranes, the proteins were visualized using enhanced chemiluminescence substrate (Thermo Fisher Scientific) and imaged by an Amersham ImageQuant 800 (Cytiva, Marlborough, MA, USA). The following primary and secondary antibodies were used: RAD51D rabbit (ab202063, Abcam, 1:1000), RAD51 rabbit (8875S, Cell Signaling Technology, 1:1000), β-actin rabbit (AbC-2002, Abclonal, 1:5000), and horseradish peroxidase-conjugated anti-rabbit IgG (ADI-SAB-300, Enzo Life Sciences, 1:5,000). Quantification of each protein expression level was performed using ImageJ.[62] The uncropped blot images are included in the supplemental information.

Statistical analysis and illustration

Statistical analyses were conducted to compare the clinicopathological features of germline *RAD51D* mutation carriers and non-carriers using R version 4.3.1 (Posit, Boston, MA, USA). Categorical variables were compared using the Fisher's exact test. Wilcoxon rank-sum tests were used to compare the continuous variables. The experimental data were statistically analyzed using Microsoft Excel (Microsoft Corporation, Redmond, WA, USA). A p value of <0.05 was considered statistically significant ($^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.005$). The workflow used in this study was created using BioRender (Toronto, Canada).

DATA AND CODE AVAILABILITY

The datasets generated and analyzed during the current study are not publicly available because of the privacy of the study participants but are available from the corresponding author upon reasonable request.

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AUTHOR CONTRIBUTIONS

Conceptualization, H.S.P., and J.S.P.; methodology, S.J., W.-J.R., Y.H., and M.W.K.; resources, J.S.P., Y.J.L., S.H.L., D.W., E.J.N., J.W.H., T.I.K., J.H.A., and H.S.P.; investigation, S.J. and W.C.S., writing – original draft, S.J., writing, review, and editing, S.P., T.K., and H.S.P., visualization, S.J., supervision, H.S.P., funding acquisition, H.S.P.

DECLARATION OF INTERESTS

The authors declare that they have no competing interests.

SUPPLEMENTAL INFORMATION

Supplemental information can be found online at https://doi.org/10.1016/j.omton.2026.201242.

REFERENCES

1. Jackson, S.P., and Bartek, J. (2009). The DNA-damage response in human biology and disease. Nature *461*, 1071–1078. https://doi.org/10.1038/nature08467.
2. Krejci, L., Altmannova, V., Spirek, M., and Zhao, X. (2012). Homologous recombination and its regulation. Nucleic Acids Res. *40*, 5795–5818. https://doi.org/10.1093/nar/gks270.
3. Stinson, B.M., and Loparo, J.J. (2021). Repair of DNA Double-Strand Breaks by the Nonhomologous End Joining Pathway. Annu. Rev. Biochem. *90*, 137–164. https://doi.org/10.1146/annurev-biochem-080320-110356.
4. Black, S.J., Ozdemir, A.Y., Kashkina, E., Kent, T., Rusanov, T., Ristic, D., Shin, Y., Suma, A., Hoang, T., Chandramouly, G., et al. (2019). Molecular basis of microhomology-mediated end-joining by purified full-length Poltheta. Nat. Commun. *10*, 4423. https://doi.org/10.1038/s41467-019-12272-9.
5. Zhao, B., Rothenberg, E., Ramsden, D.A., and Lieber, M.R. (2020). The molecular basis and disease relevance of non-homologous DNA end joining. Nat. Rev. Mol. Cell Biol. *21*, 765–781. https://doi.org/10.1038/s41580-020-00297-8.
6. Venkitaraman, A.R. (2002). Cancer susceptibility and the functions of BRCA1 and BRCA2. Cell *108*, 171–182. https://doi.org/10.1016/s0092-8674(02)00615-3.
7. Lord, C.J., and Ashworth, A. (2016). BRCAness revisited. Nat. Rev. Cancer *16*, 110–120. https://doi.org/10.1038/nrc.2015.21.
8. Reh, W.A., Nairn, R.S., Lowery, M.P., and Vasquez, K.M. (2017). The homologous recombination protein RAD51D protects the genome from large deletions. Nucleic Acids Res. *45*, 1835–1847. https://doi.org/10.1093/nar/gkw1204.
9. Hinz, J.M., Tebbs, R.S., Wilson, P.F., Nham, P.B., Salazar, E.P., Nagasawa, H., Urbin, S.S., Bedford, J.S., and Thompson, L.H. (2006). Repression of mutagenesis by Rad51D-mediated homologous recombination. Nucleic Acids Res. *34*, 1358–1368. https://doi.org/10.1093/nar/gkl020.
10. Takata, M., Sasaki, M.S., Tachiiri, S., Fukushima, T., Sonoda, E., Schild, D., Thompson, L.H., and Takeda, S. (2001). Chromosome instability and defective recombinational repair in knockout mutants of the five Rad51 paralogs. Mol. Cell Biol. *21*, 2858–2866. https://doi.org/10.1128/MCB.21.8.2858-2866.2001.
11. Sullivan, M.R., and Bernstein, K.A. (2018). RAD-ical New Insights into RAD51 Regulation. Genes *9*, 629. https://doi.org/10.3390/genes9120629.
12. Rawal, Y., Jia, L., Meir, A., Zhou, S., Kaur, H., Ruben, E.A., Kwon, Y., Bernstein, K.A., Jasin, M., Taylor, A.B., et al. (2023). Structural insights into BCDX2 complex function in homologous recombination. Nature *619*, 640–649. https://doi.org/10.1038/s41586-023-06219-w.
13. Garcin, E.B., Gon, S., Sullivan, M.R., Brunette, G.J., Cian, A.D., Concordet, J.P., Giovannangeli, C., Dirks, W.G., Eberth, S., Bernstein, K.A., et al. (2019). Differential Requirements for the RAD51 Paralogs in Genome Repair and Maintenance in Human Cells. PLoS Genet. *15*, e1008355. https://doi.org/10.1371/journal.pgen.1008355.
14. Yonetani, Y., Hochegger, H., Sonoda, E., Shinya, S., Yoshikawa, H., Takeda, S., and Yamazoe, M. (2005). Differential and collaborative actions of Rad51 paralog proteins in cellular response to DNA damage. Nucleic Acids Res. *33*, 4544–4552. https://doi.org/10.1093/nar/gki766.
15. Berti, M., Teloni, F., Mijic, S., Ursich, S., Fuchs, J., Palumbieri, M.D., Krietsch, J., Schmid, J.A., Garcin, E.B., Gon, S., et al. (2020). Sequential role of RAD51 paralog complexes in replication fork remodeling and restart. Nat. Commun. *11*, 3531. https://doi.org/10.1038/s41467-020-17324-z.
16. Somyajit, K., Saxena, S., Babu, S., Mishra, A., and Nagaraju, G. (2015). Mammalian RAD51 paralogs protect nascent DNA at stalled forks and mediate replication restart. Nucleic Acids Res. *43*. gkv880-9855. https://doi.org/10.1093/nar/gkv880.
17. Breast Cancer Association, C., Dorling, L., Carvalho, S., Allen, J., Gonzalez-Neira, A., Luccarini, C., Wahlstrom, C., Pooley, K.A., Parsons, M.T., Fortuno, C., et al. (2021). Breast Cancer Risk Genes - Association Analysis in More than 113,000 Women. N. Engl. J. Med. *384*, 428–439. https://doi.org/10.1056/NEJMoa1913948.
18. Yang, X., Song, H., Leslie, G., Engel, C., Hahnen, E., Auber, B., Horváth, J., Kast, K., Niederacher, D., Turnbull, C., et al. (2020). Ovarian and Breast Cancer Risks Associated With Pathogenic Variants in RAD51C and RAD51D. J. Natl. Cancer Inst. *112*, 1242–1250. https://doi.org/10.1093/jnci/djaa030.
19. Southey, M.C., Dowty, J.G., Riaz, M., Steen, J.A., Renault, A.L., Tucker, K., Kirk, J., James, P., Winship, I., Pachter, N., et al. (2021). Population-based estimates of breast cancer risk for carriers of pathogenic variants identified by gene-panel testing. NPJ Breast Cancer *7*, 153. https://doi.org/10.1038/s41523-021-00360-3.
20. Rowlands, C.F., Allen, S., Balmaña, J., Domchek, S.M., Evans, D.G., Hanson, H., Hoogerbrugge, N., James, P.A., Nathanson, K.L., Robson, M., et al. (2024). Population-based germline breast cancer gene association studies and meta-analysis to inform wider mainstream testing. Ann. Oncol. *35*, 892–901. https://doi.org/10.1016/j.annonc.2024.07.244.
21. Kennedy, R.D., Quinn, J.E., Mullan, P.B., Johnston, P.G., and Harkin, D.P. (2004). The role of BRCA1 in the cellular response to chemotherapy. J. Natl. Cancer Inst. *96*, 1659–1668. https://doi.org/10.1093/jnci/djh312.
22. Byrski, T., Huzarski, T., Dent, R., Marczyk, E., Jasiowka, M., Gronwald, J., Jakubowicz, J., Cybulski, C., Wisniowski, R., Godlewski, D., et al. (2014). Pathologic complete response to neoadjuvant cisplatin in BRCA1-positive breast cancer patients. Breast Cancer Res. Treat. *147*, 401–405. https://doi.org/10.1007/s10549-014-3100-x.
23. Pennington, K.P., Walsh, T., Harrell, M.I., Lee, M.K., Pennil, C.C., Rendi, M.H., Thornton, A., Norquist, B.M., Casadei, S., Nord, A.S., et al. (2014). Germline and somatic mutations in homologous recombination genes predict platinum response and survival in ovarian, fallopian tube, and peritoneal carcinomas. Clin. Cancer Res. *20*, 764–775. https://doi.org/10.1158/1078-0432.CCR-13-2287.
24. Lorusso, D., Bria, E., Costantini, A., Di Maio, M., Rosti, G., and Mancuso, A. (2017). Patients' perception of chemotherapy side effects: Expectations, doctor-patient communication and impact on quality of life - An Italian survey. Eur. J. Cancer Care *26*, e12618. https://doi.org/10.1111/ecc.12618.
25. Fong, P.C., Boss, D.S., Yap, T.A., Tutt, A., Wu, P., Mergui-Roelvink, M., Mortimer, P., Swaisland, H., Lau, A., O'Connor, M.J., et al. (2009). Inhibition of poly(ADP-ribose) polymerase in tumors from BRCA mutation carriers. N. Engl. J. Med. *361*, 123–134. https://doi.org/10.1056/NEJMoa0900212.
26. Ashworth, A. (2008). A synthetic lethal therapeutic approach: poly(ADP) ribose polymerase inhibitors for the treatment of cancers deficient in DNA double-strand break repair. J. Clin. Oncol. *26*, 3785–3790. https://doi.org/10.1200/JCO.2008.16.0812.
27. Ray Chaudhuri, A., and Nussenzweig, A. (2017). The multifaceted roles of PARP1 in DNA repair and chromatin remodelling. Nat. Rev. Mol. Cell Biol. *18*, 610–621. https://doi.org/10.1038/nrm.2017.53.
28. Murai, J., Huang, S.y.N., Das, B.B., Renaud, A., Zhang, Y., Doroshow, J.H., Ji, J., Takeda, S., and Pommier, Y. (2012). Trapping of PARP1 and PARP2 by Clinical PARP Inhibitors. Cancer Res. *72*, 5588–5599. https://doi.org/10.1158/0008-5472.CAN-12-2753.
29. Robson, M., Goessl, C., and Domchek, S. (2017). Olaparib for Metastatic Germline BRCA-Mutated Breast Cancer. N. Engl. J. Med. *377*, 1792–1793. https://doi.org/10.1056/NEJMc1711644.
30. Litton, J.K., Rugo, H.S., Ettl, J., Hurvitz, S.A., Gonçalves, A., Lee, K.H., Fehrenbacher, L., Yerushalmi, R., Mina, L.A., Martin, M., et al. (2018). Talazoparib in Patients with Advanced Breast Cancer and a Germline BRCA Mutation. N. Engl. J. Med. *379*, 753–763. https://doi.org/10.1056/NEJMoa1802905.
31. Kelland, L. (2007). The resurgence of platinum-based cancer chemotherapy. Nat. Rev. Cancer *7*, 573–584. https://doi.org/10.1038/nrc2167.
32. Armstrong, N., Ryder, S., Forbes, C., Ross, J., and Quek, R.G. (2019). A systematic review of the international prevalence of BRCA mutation in breast cancer. Clin. Epidemiol. *11*, 543–561. https://doi.org/10.2147/CLEP.S206949.
33. Loveday, C., Turnbull, C., Ramsay, E., Hughes, D., Ruark, E., Frankum, J.R., Bowden, G., Kalmyrzaev, B., Warren-Perry, M., Snape, K., et al. (2011). Germline mutations in RAD51D confer susceptibility to ovarian cancer. Nat. Genet. *43*, 879–882. https://doi.org/10.1038/ng.893.

34. Thompson, E.R., Rowley, S.M., Sawyer, S., Campbell, I.G., Eccles, D.M., Eccles, D.M., Trainer, A.H., Mitchell, G., and James, P.A. (2013). Analysis of RAD51D in ovarian cancer patients and families with a history of ovarian or breast cancer. PLoS One *8*, e54772. https://doi.org/10.1371/journal.pone.0054772.

35. Sun, J., Meng, H., Yao, L., Lv, M., Bai, J., Zhang, J., Wang, L., Ouyang, T., Li, J., Wang, T., et al. (2017). Germline Mutations in Cancer Susceptibility Genes in a Large Series of Unselected Breast Cancer Patients. Clin. Cancer Res. *23*, 6113–6119. https://doi.org/10.1158/1078-0432.CCR-16-3227.

36. Baker, J.L., Schwab, R.B., Wallace, A.M., and Madlensky, L. (2015). Breast cancer in a RAD51D mutation carrier: case report and review of the literature. Clin. Breast Cancer *15*, e71–e75. https://doi.org/10.1016/j.clbc.2014.08.005.

37. Susswein, L.R., Marshall, M.L., Nusbaum, R., Vogel Postula, K.J., Weissman, S.M., Yackowski, L., Vaccari, E.M., Bissonnette, J., Booker, J.K., Cremona, M.L., et al. (2016). Pathogenic and likely pathogenic variant prevalence among the first 10,000 patients referred for next-generation cancer panel testing. Genet. Med. *18*, 823–832. https://doi.org/10.1038/gim.2015.166.

38. Konstanta, I., Fostira, F., Apostolou, P., Stratikos, E., Kalfakakou, D., Pampanos, A., Kollia, P., Papadimitriou, C., Konstantopoulou, I., and Yannoukakos, D. (2018). Contribution of RAD51D germline mutations in breast and ovarian cancer in Greece. J. Hum. Genet. *63*, 1149–1158. https://doi.org/10.1038/s10038-018-0498-8.

39. Eoh, K.J., Kim, J.E., Park, H.S., Lee, S.T., Park, J.S., Han, J.W., Lee, J.Y., Kim, S., Kim, S.W., Kim, J.H., et al. (2018). Detection of Germline Mutations in Patients with Epithelial Ovarian Cancer Using Multi-gene Panels: Beyond BRCA1/2. Cancer Res. Treat. *50*, 917–925. https://doi.org/10.4143/crt.2017.220.

40. Greenhough, L.A., Liang, C.C., Belan, O., Kunzelmann, S., Maslen, S., Rodrigo-Brenni, M.C., Anand, R., Skehel, M., Boulton, S.J., and West, S.C. (2023). Structure and function of the RAD51B-RAD51C-RAD51D-XRCC2 tumour suppressor. Nature *619*, 650–657. https://doi.org/10.1038/s41586-023-06179-1.

41. Byrski, T., Gronwald, J., Huzarski, T., Grzybowska, E., Budryk, M., Stawicka, M., Mierzwa, T., Szwiec, M., Wiśniowski, R., Siolek, M., et al. (2010). Pathologic complete response rates in young women with BRCA1-positive breast cancers after neoadjuvant chemotherapy. J. Clin. Oncol. *28*, 375–379. https://doi.org/10.1200/JCO.2008.20.7019.

42. Li, Z.Y., Zhang, Z., Cao, X.Z., Feng, Y., and Ren, S.S. (2020). Platinum-based neoadjuvant chemotherapy for triple-negative breast cancer: a systematic review and meta-analysis. J. Int. Med. Res. *48*, 0300060520964340. https://doi.org/10.1177/0300060520964340.

43. Evers, B., Helleday, T., and Jonkers, J. (2010). Targeting homologous recombination repair defects in cancer. Trends Pharmacol. Sci. *31*, 372–380. https://doi.org/10.1016/j.tips.2010.06.001.

44. Chalasani, P., and Livingston, R. (2013). Differential chemotherapeutic sensitivity for breast tumors with "BRCAness": a review. Oncologist *18*, 909–916. https://doi.org/10.1634/theoncologist.2013-0039.

45. Wunderle, M., Gass, P., Häberle, L., Flesch, V.M., Rauh, C., Bani, M.R., Hack, C.C., Schrauder, M.G., Jud, S.M., Emons, J., et al. (2018). BRCA mutations and their influence on pathological complete response and prognosis in a clinical cohort of neoadjuvantly treated breast cancer patients. Breast Cancer Res. Treat. *171*, 85–94. https://doi.org/10.1007/s10549-018-4797-8.

46. Mateo, J., Carreira, S., Sandhu, S., Miranda, S., Mossop, H., Perez-Lopez, R., Nava Rodrigues, D., Robinson, D., Omlin, A., Tunariu, N., et al. (2015). DNA-Repair Defects and Olaparib in Metastatic Prostate Cancer. N. Engl. J. Med. *373*, 1697–1708. https://doi.org/10.1056/NEJMoa1506859.

47. Hodgson, D.R., Dougherty, B.A., Lai, Z., Fielding, A., Grinsted, L., Spencer, S., O'Connor, M.J., Ho, T.W., Robertson, J.D., Lanchbury, J.S., et al. (2018). Candidate biomarkers of PARP inhibitor sensitivity in ovarian cancer beyond the BRCA genes. Br. J. Cancer *119*, 1401–1409. https://doi.org/10.1038/s41416-018-0274-8.

48. Llop-Guevara, A., Loibl, S., Villacampa, G., Vladimirova, V., Schneeweiss, A., Karn, T., Zahm, D.M., Herencia-Ropero, A., Jank, P., van Mackelenbergh, M., et al. (2021). Association of RAD51 with homologous recombination deficiency (HRD) and clinical outcomes in untreated triple-negative breast cancer (TNBC): analysis of the GeparSixto randomized clinical trial. Ann. Oncol. *32*, 1590–1596. https://doi.org/10.1016/j.annonc.2021.09.003.

49. Grundy, M.K., Buckanovich, R.J., and Bernstein, K.A. (2020). Regulation and pharmacological targeting of RAD51 in cancer. NAR Cancer *2*, zcaa024. https://doi.org/10.1093/narcan/zcaa024.

50. Huang, J.W., Wang, Y., Dhillon, K.K., Calses, P., Villegas, E., Mitchell, P.S., Tewari, M., Kemp, C.J., and Taniguchi, T. (2013). Systematic screen identifies miRNAs that target RAD51 and RAD51D to enhance chemosensitivity. Mol. Cancer Res. *11*, 1564–1573. https://doi.org/10.1158/1541-7786.MCR-13-0292.

51. Farmer, H., McCabe, N., Lord, C.J., Tutt, A.N.J., Johnson, D.A., Richardson, T.B., Santarosa, M., Dillon, K.J., Hickson, I., Knights, C., et al. (2005). Targeting the DNA repair defect in BRCA mutant cells as a therapeutic strategy. Nature *434*, 917–921. https://doi.org/10.1038/nature03445.

52. Timms, K.M., Abkevich, V., Hughes, E., Neff, C., Reid, J., Morris, B., Kalva, S., Potter, J., Tran, T.V., Chen, J., et al. (2014). Association of BRCA1/2 defects with genomic scores predictive of DNA damage repair deficiency among breast cancer subtypes. Breast Cancer Res. *16*, 475. https://doi.org/10.1186/s13058-014-0475-x.

53. Baumann, P., and West, S.C. (1998). Role of the human RAD51 protein in homologous recombination and double-stranded-break repair. Trends Biochem. Sci. *23*, 247–251. https://doi.org/10.1016/s0968-0004(98)01232-8.

54. Teraoka, S., Muguruma, M., Takano, N., Miyahara, K., Kawate, T., Kaise, H., Yamada, K., Miyazawa, K., and Ishikawa, T. (2020). Association of BRCA Mutations and BRCAness Status With Anticancer Drug Sensitivities in Triple-Negative Breast Cancer Cell Lines. J. Surg. Res. *250*, 200–208. https://doi.org/10.1016/j.jss.2019.12.040.

55. Krammer, J., Pinker-Domenig, K., Robson, M.E., Gönen, M., Bernard-Davila, B., Morris, E.A., Mangino, D.A., and Jochelson, M.S. (2017). Breast cancer detection and tumor characteristics in BRCA1 and BRCA2 mutation carriers. Breast Cancer Res. Treat. *163*, 565–571. https://doi.org/10.1007/s10549-017-4198-4.

56. Chen, X., Li, Y., Ouyang, T., Li, J., Wang, T., Fan, Z., Fan, T., Lin, B., and Xie, Y. (2018). Associations between RAD51D germline mutations and breast cancer risk and survival in BRCA1/2-negative breast cancers. Ann. Oncol. *29*, 2046–2051. https://doi.org/10.1093/annonc/mdy338.

57. Golmard, L., Castéra, L., Krieger, S., Moncoutier, V., Abidallah, K., Tenreiro, H., Laugé, A., Tarabeux, J., Millot, G.A., Nicolas, A., et al. (2017). Contribution of germline deleterious variants in the RAD51 paralogs to breast and ovarian cancers. Eur. J. Hum. Genet. *25*, 1345–1353. https://doi.org/10.1038/s41431-017-0021-2.

58. Couch, F.J., Shimelis, H., Hu, C., Hart, S.N., Polley, E.C., Na, J., Hallberg, E., Moore, R., Thomas, A., Lilyquist, J., et al. (2017). Associations Between Cancer Predisposition Testing Panel Genes and Breast Cancer. JAMA Oncol. *3*, 1190–1196. https://doi.org/10.1001/jamaoncol.2017.0424.

59. Hauke, J., Horvath, J., Groß, E., Gehrig, A., Honisch, E., Hackmann, K., Schmidt, G., Arnold, N., Faust, U., Sutter, C., et al. (2018). Gene panel testing of 5589 BRCA1/2-negative index patients with breast cancer in a routine diagnostic setting: results of the German Consortium for Hereditary Breast and Ovarian Cancer. Cancer Med. *7*, 1349–1358. https://doi.org/10.1002/cam4.1376.

60. Park, J.S., Shin, S., Lee, Y.J., Lee, S.T., Nam, E.J., Han, J.W., Lee, S.H., Kim, T.I., and Park, H.S. (2022). Implication and Influence of Multigene Panel Testing with Genetic Counseling in Korean Patients with BRCA1/2 Mutation-Negative Breast Cancer. Cancer Res. Treat. *54*, 1099–1110. https://doi.org/10.4143/crt.2021.978.

61. von Minckwitz, G., Untch, M., Blohmer, J.U., Costa, S.D., Eidtmann, H., Fasching, P.A., Gerber, B., Eiermann, W., Hilfrich, J., Huober, J., et al. (2012). Definition and impact of pathologic complete response on prognosis after neoadjuvant chemotherapy in various intrinsic breast cancer subtypes. J. Clin. Oncol. *30*, 1796–1804. https://doi.org/10.1200/JCO.2011.38.8595.

62. Schneider, C.A., Rasband, W.S., and Eliceiri, K.W. (2012). NIH Image to ImageJ: 25 years of image analysis. Nat. Methods *9*, 671–675. https://doi.org/10.1038/nmeth.2089.