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The effect of *BRCA1/2* mutation on breast cancer and its impact on PARP inhibitor treatments: Differential expression genes approach

Rafika Indah Paramita[1,2,3], Sonar Soni Panigoro[4,5*], Septelia Inawati Wanandi[4,6,7*], Fadilah Fadilah[2,3,4]

[1]Doctoral Program in Biomedical Sciences, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia.
[2]Department of Medical Chemistry, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia.
[3]Bioinformatics Core Facilities-IMERI, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia.
[4]Master's Programme in Biomedical Sciences, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia.
[5]Surgical Oncology Division, Department of Surgery, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia.
[6]Department of Biochemistry and Molecular Biology, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia.
[7]Molecular Biology and Proteomics Core Facilities-IMERI, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia.

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ABSTRACT

The occurrence of germline mutations within the *BRCA1/2* genes has been linked to an elevated vulnerability toward the onset of breast cancer (BC). At present, ongoing clinical trials are being undertaken to evaluate the efficacy of poly(ADP-ribose) polymerase (PARP) inhibitors as a therapeutic intervention for BC, with particular emphasis on their application in the management of BC patients harboring *BRCA1/2* gene mutations. The objective of this research was to investigate the presence of different expression genes in BC with *BRCA1/2* mutations compared to the wild type and to evaluate the impact of PARP inhibitor therapy on the DEGs. This study utilized two distinct datasets sourced from the Gene Expression Omnibus (GEO) database. The initial datasets utilized in this study were GSE25835 and GSE40115. These datasets were employed to conduct a comparative analysis of differentially expressed genes (DEGs) in BC cases with *BRCA1/2* mutations and those with wild-type status. Whereas in the GSE55399 dataset, the DEGs were compared between PARP inhibitor treatment and no PARP inhibitor treatment. The interactions among DEGs were assessed utilizing the search tool for the retrieval of interacting genes/proteins tool and subsequently displayed through the use of Cytoscape software. The molecular complex detection technique was employed for the identification of gene clusters within the interaction network. The DEGs that were discovered were further analyzed for gene ontology (GO) enrichment using Enrichr and CLueGO. Furthermore, the biological pathways linked to these DEGs were examined using REACTOME. We got significant DEGs by using parameter *p*-value of 0.05; log2FC > 1 and log2FC < −1. The GO analysis conducted on the DEGs revealed their significant involvement in crucial biological processes and molecular pathways. For datasets GSE25835 and GSE40115, it showed the effect on *BRCA1/2* mutations was upregulating cell cycle response and downregulating mRNA splicing. For dataset GSE55399, the impact of PARP inhibitor treatments was upregulating the interferon signaling and downregulating the cytokine signaling. Our study identified hub genes of cell cycle response (CDK1 and BIRC5) that are strongly linked to *BRCA1/2* mutation and hub genes of interferon signaling interferon-induced transmembrane 1 (IFITM1) and cytokine signaling (IL11) that are strongly linked to PARP inhibitor treatments in *BRCA1/2* mutant carriers. We identified hub genes of cell cycle response (CDK1 and BIRC5) that are strongly linked to *BRCA1/2* mutation. PARP inhibitor treatments in *BRCA1/2* mutant carriers are strongly related to the upregulation of IFITM1 (interferon signaling) and the downregulation of IL11 (cytokine signaling). Therefore, PARP inhibitors may improve the treatment by activation/modulation the immune system and attenuating the inflammatory response. However, the dataset used to analyze the DEGs of PARP inhibitor treatments in *BRCA1/2* mutant carriers still used BC cell lines, forthcoming research may be able to use clinical patients as the subjects. Moreover, functional studies are further needed to validate this finding.

**Corresponding Author*
Sonar Soni Panigoro, Master's Programme in Biomedical Sciences, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia. E-mail: sonar.soni @ ui.ac.id
Septelia Inawati Wanandi, Master's Programme in Biomedical Sciences, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia. E-mail: septelia.inawati @ ui.ac.id

INTRODUCTION

Breast cancer (BC) is the most prevalent malignancy in women worldwide. In 2020, the World Health Organization (WHO) projects that 2.3 million new cases of BC will be diagnosed. The incidence rates in various regions and countries vary considerably. Certain mutations in *BRCA1/2* have been found to be more prevalent in Asian populations, despite the fact that mutations can vary between populations. For instance, the *BRCA1* c.3331_3334delCAAG mutation has been observed more frequently in Asians. The clinical consequences of germline mutations in *BRCA1/2* among BC patients of Asian descent are of considerable importance [1]. According to existing literature, these mutations have been found to be correlated with a heightened susceptibility to breast and ovarian malignancies, just like in other demographic groups [2–4].

The proteins *BRCA1* and *BRCA2* play crucial roles in the homologous recombination (HR) pathway, which is responsible for the repair of DNA double-strand breaks. HR repair is a meticulous and accurate mechanism employed to mend damaged DNA. This process relies on an intact sister chromatid or a homologous chromosome, which serves as a template for the repair of DNA breaks. *BRCA1* serves as a scaffold protein that coordinates and regulates DNA repair-related proteins. It interacts with a variety of proteins implicated in HR repair, such as *BRCA2*, RAD51, and PALB2. *BRCA1* aids in the recruitment and stabilization of the RAD51 protein, which is essential for the strand invasion phase of HR repair. *BRCA2* interacts directly with RAD51 and facilitates its transfer onto single-stranded DNA at the DNA break site. This formation of RAD51 filaments facilitates the search for and invasion of the homologous DNA template, resulting in the repair of the double-stranded break. Mutations in *BRCA1* or *BRCA2* that impede their function can impair HR repair and heighten susceptibility to DNA damage accumulation. These mutations can result in genomic instability, as cells become more susceptible to accumulating DNA alterations such as mutations, deletions, and translocations. This can contribute to the eventual development of cancer [5,6].

The availability of genetic mutation data pertaining to the susceptibility and prognostic implications of cancer can offer guidance for therapeutic interventions in cases of BC. At present, ongoing clinical trials are being undertaken to assess the potential of poly (ADP-ribose) polymerase (PARP) inhibitors in the management of BC, specifically in relation to their effectiveness in managing BC with *BRCA* mutations. Hence, the administration of systemic therapy for BC in individuals with *BRCA* mutations can be tailored depending on the unique state of the *BRCA* mutation, rather than relying just on the manifestation of disease symptoms [7].

A number of distinct molecular subtypes that differ considerably in prognosis and therapeutic targets present in cancer cells have been identified by gene expression studies. Microarray technology is a powerful instrument used in molecular biology and genomics to investigate gene expression, DNA sequencing, genotyping, and other genomic applications. It enables researchers to analyze the expression or presence of thousands to millions of genes or DNA sequences simultaneously in a single experiment [8]. Several prior studies have discussed the identification of differentially expressed genes (DEGs) in BC when compared to healthy individuals [9–11]. However, there is a lack of research that specifically examines the analysis of DEGs in BC patients with *BRCA1/2* mutations in comparison to those without such mutations. There is a limited study as well that analyzes the DEGs of PARP inhibitor treatments in BC with *BRCA1/2* mutations. Therefore, it seems important to understand more about those conditions that can be more developed as biomarkers as well.

In this study, BC gene expression profiles from three separate studies (two types of datasets) using microarrays were analyzed. The microarray expression profile datasets GSE25835, GSE40115, and GSE55399 were obtained from the Gene Expression Omnibus (GEO). The DEGs were retrieved based on parameter adjustment p-value of 0.05, log2FC > 1 (upregulated DEGs) and log2FC < −1 (downregulated DEGs). By using this approach, we aimed to identify the biomarkers from DEGs of *BRCA1/2* mutation carriers and *BRCA1/2* mutation carriers that received PARP inhibitor treatment. We hypothesized that there are some DEGs in patients with *BRCA1/2* mutations and that the use of PARP inhibitor treatments will change the biological functions (BFs) as well.

MATERIAL AND METHODS

Identification of differentially expressed genes

The GSE25835 [12], GSE40115 [13], and GSE55399 [14] datasets were retrieved from the GEO database. Dataset GSE25835 uses the Affymetrix HT Human Genome U133A Array Platform (GPL3921), dataset GSE40115 uses Agilent-029949 Custom Sureprint G3 Human GE 8x60K Microarray (GPL15931), and dataset GSE25835 uses the Affymetrix HT Human Genome U133A Array (GPL3921) platform. In this study, we used two different categories of datasets. The first datasets were GSE25835 and GSE40115, which compared the DEGs of BC with *BRCA1/2* mutations and wild-type. Whereas in the GSE55399 dataset, the DEGs were compared between PARP inhibitor treatment and no PARP inhibitor treatment.

To analyze gene expression data, GEO2R software was used to calculate adjusted p-values and FDR by using T test and Benjamini and Hochberg (false discovery rate) [15]. To obtain a significant DEG from the dataset, we standardized the primary adjustment at a p-value of 0.05; the up-regulated DEG is considered if log2FC > 1 and the down-regulated DEG if log2FC < −1. The DEGs derived from the two data sets were searched for overlapping sections using Venny 2.1.0 (https://bioinfogp.cnb.csic.es/tools/venny/) and considered for further analysis [11].

Protein–protein interaction (PPI) networks

The search tool for the retrieval of interacting genes/proteins (STRING) database provides information on PPIs and functional associations. It seeks to facilitate the investigation of protein networks and their relationships with diverse biological processes [16]. By using Cytoscape software [17] (version 3.8.2), a confidence value of 0.9 was employed to achieve a

robust interaction. In addition, the molecular complex detection (MCODE) algorithm was employed to acquire overlapping clusters from the resultant interaction networks. Multiple group determination parameters are employed, including a degree cutoff of 2, a *k*-score of 2, a maximum depth of 100, and a node score cutoff of 0.2 [8,18].

Gene ontology (GO) term and signaling pathway enrichment analysis

The DEGs that had been analyzed were enriched for their gene ontologies using Enrichr (http://maayanlab.cloud/Enrichr/). The ClueGO [19] (version 2.5.7), module of the Cytoscape software was used to investigate the gene annotation interrelationship analysis. The statistical analysis for ClueGO enrichment analysis involved the utilization of a two-tailed hypergeometric test with a significance level of p less than 0.05. In addition, the Benjamini–Hochberg correction, as well as the kappa score of 0.4 were applied to determine significant enrichment. The pathway analysis was conducted utilizing the REACTOME database, accessible at http://www.reactome.org [20].

Survival analysis

Gene expression profiling interactive analysis (GEPIA) [21] was utilized to do a statistical analysis of gene expression data to determine the correlation between gene expression levels and both time and % overall survival (OS). This analysis involved the calculation of Kaplan–Meier curves and performed on the BC dataset and using the Cox proportional hazards model. In addition, the hazard ratio was determined, along with its corresponding 95% confidence intervals. The analysis was conducted to predict the effect of DEG on the survival of BC patients. A significant difference being defined by the p-value score is less than 0.05 [22].

RESULT AND DISCUSSION

BC is the most commonly diagnosed cancer and the primary cause of cancer-related deaths worldwide among females. The prevalence of germline mutations in the *BRCA1* or *BRCA2* genes among BC patients in Asia is estimated to range from 2% to 3% [1]. Both *BRCA1* and *BRCA2* have significant involvement in the process of HR repair, hence contributing to the repair of DNA damage [5]. A dysfunction of *BRCA1/2* results in heightened chromosomal instability, potentially contributing to the development of tumors. Significantly, the absence of *BRCA1/2* also leads to increased cellular susceptibility to interstrand DNA cross-linking agents, including cisplatin, as well as PARP inhibitors, an auspicious new category of anticancer agents [23,24].

Cisplatin and its derivative, carboplatin, which are platinum compounds, exhibit efficacy as chemotherapeutic agents. Nonetheless, relapse does transpire in the majority of female patients who have *BRCA1/2*-mutated cancer which then develop resistance to platinum. Several mechanisms have been identified in scientific research as potential contributors to cisplatin resistance, such as increased glutathione expression and altered drug transport via altered copper transporter expression.

Due to the high sensitivity of *BRCA1/2*-mutant to PARP inhibitors, numerous ongoing clinical trials are evaluating the efficacy of PARP inhibitors in BC patients with *BRCA1/2* mutation. Synthetic lethality is the consequence of inhibiting PARP in HR-deficient cells, such as *BRCA1/2*-mutant, and encouraging clinical trial outcomes in *BRCA*-associated carcinomas have been documented [25]. Selection of systemic therapy for BC can be done by looking at *BRCA* mutation status not only on disease characteristics [7].

Bioinformatics plays an important role in the study of cancer at the molecular level related to prognostics, diagnostics, and therapeutics. The comprehensive bioinformatics analysis and biological database are innovative techniques to bring future perspectives to the field of oncology [26]. In this regard, bioinformatic analysis by using computational gene profiling strategies for BC has been advocated. This study employs a methodology that utilizes two distinct types of datasets, a practice that remains infrequently employed in other research endeavors. Some previous studies applied only one type of dataset, as mentioned in Deng *et al.* [27], Fei *et al.* [28], and Zhou *et al.* [29].The objective is to obtain more extensive findings by identifying DEGs in individuals with *BRCA1/2* mutations and understanding how these genes impact the effectiveness of PARP-inhibitor therapy.

Differentially expressed genes

In this work, we analyzed the gene expression of two types of microarray datasets, namely, GSE25835, GSE40115, and GSE55399, obtained from the GEO database (Table 1). The data set was analyzed using GEO2R to obtain DEG by setting the cutoff criteria, namely the p-value adjustment < 0.05; log2FC > 1 (upregulated DEGs) and log2FC < −1 (downregulated DEGs). From the GSE25835 and GSE40115A datasets, we found DEGs in BC patients with *BRCA1/2* mutation carriers compared to the wild-type carriers. Meanwhile, from the GSE55399 dataset, we found DEGs in BC cell lines treated with PARP inhibitors compared to those without PARP inhibitors.

In the GSE25835, GSE40115, and GSE55399 datasets, a total of 4,503 DEGs were identified (2,312 upregulated and 2,191 downregulated), 900 (495 upregulated and 405 downregulated), and 620 (441 upregulated and 179 downregulated). As shown in Figure 1D–E (Tables S1 and S2), 113 DEGs (60 upregulation and 53 downregulation) were discovered to overlap in the GSE25835

Table 1. Sample information in GSE25835, GSE40115, GSE55399, and GSE173839 datasets.

GEO ID	Samples
GSE25835	Breast tumor tissue from *BRCA1* mutant carriers
	Breast tumor tissue from *BRCA1* wild-type carriers
GSE40115	Breast tumor tissue from *BRCA2* mutant carriers
	Breast tumor tissue from *BRCA2* wild-type carriers
GSE55399	TNBC cell line with *BRCA1* mutation treated using ABT888 (Veliparib)
	TNBC cell line with *BRCA1* mutation without ABT888 (Veliparib) treatment

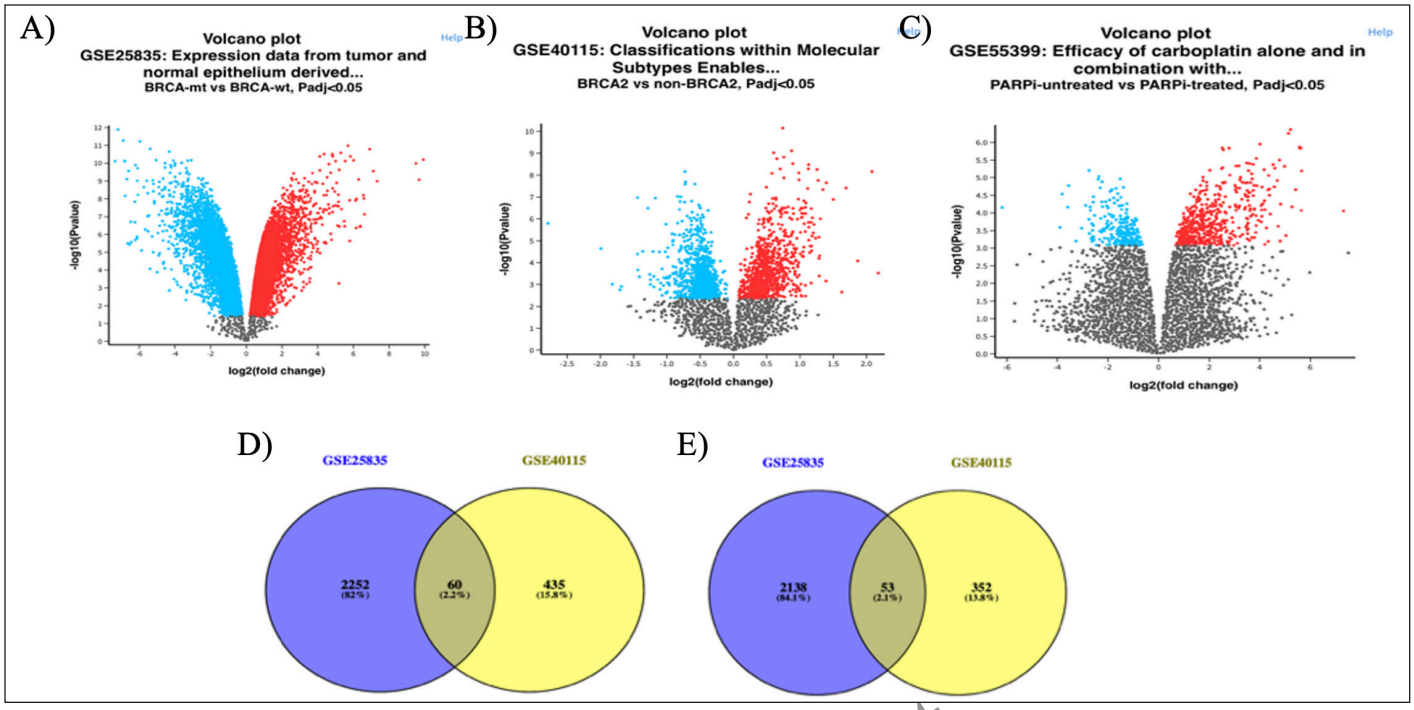


Figure 1. Volcano plot of all the DEGs of A) GSE25835, B) GSE40115, C) GSE55399; Venn diagram of GSE25835 and GSE40115, D) upregulated overlapping DEGs, and E) downregulated overlapping DEGs.

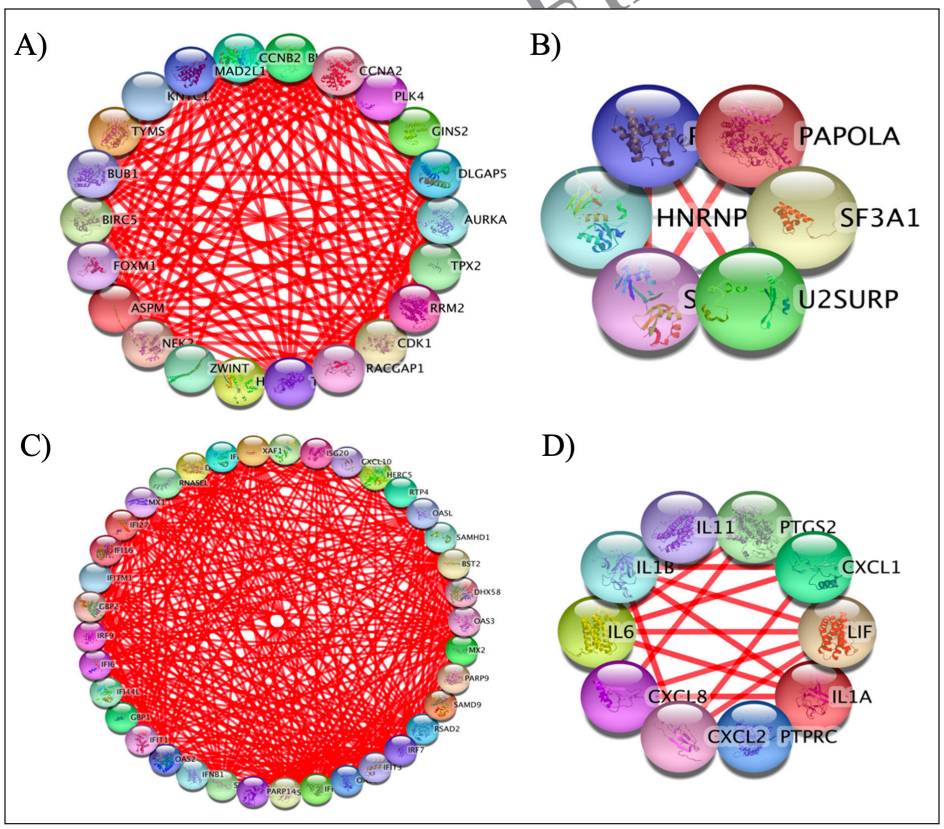


Figure 2. PPI network of overlapping GSE25835 and GSE40115 DEGs: A) upregulated DEGs and B) downregulated DEGs; PPI network of GSE55399 DEGs: C) upregulated DEGs and D) downregulated DEGs.

and GSE40115 datasets. Figure 1A–C depict the volcano graphs for DEGs of GSE25835, GSE40115, and GSE55399, respectively. The DEGs that overlapped between the GSE25835 and GSE40115 datasets were analyzed further.

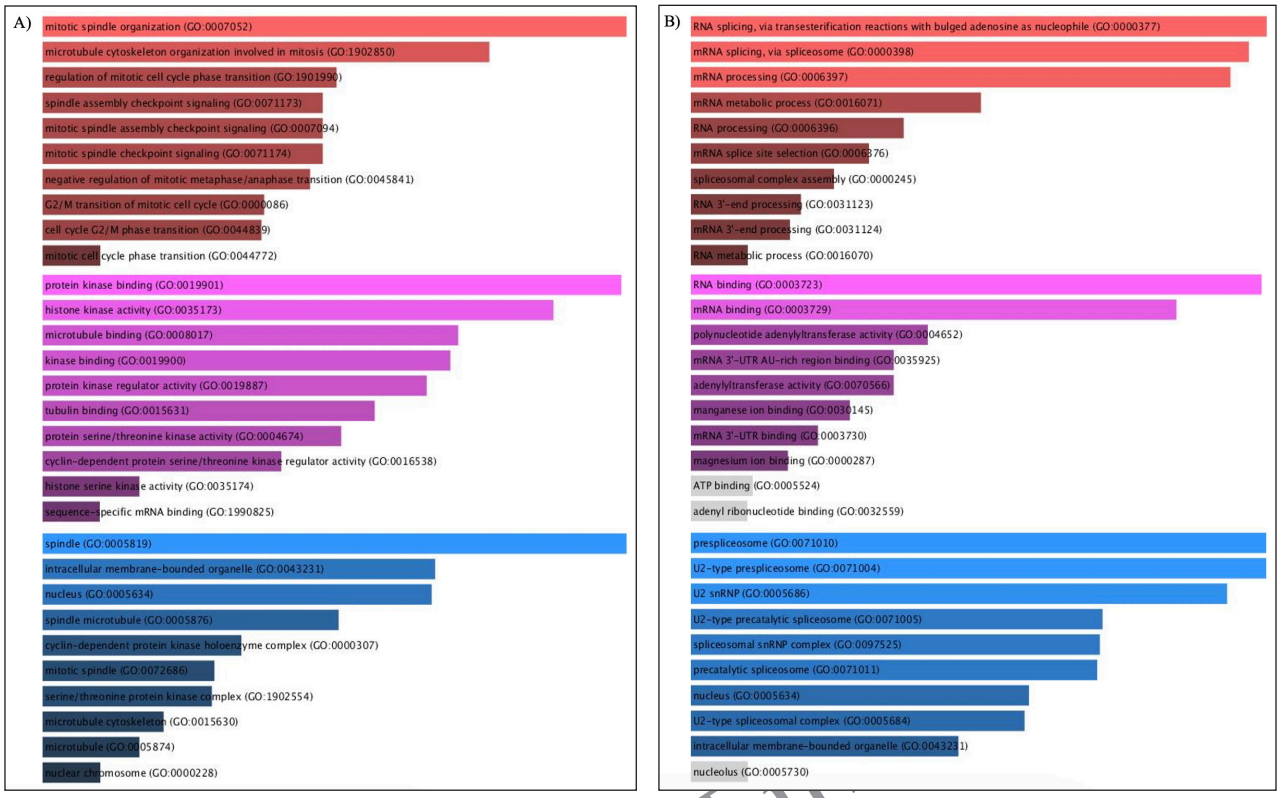


Figure 3. A) GO enrichment analysis of overlapping upregulated GSE25835 and GSE40115 DEGs, B) GO enrichment analysis of overlapping downregulated GSE25835 and GSE40115 DEGs. The top 10 annotations based on their *p*-values are shown for biological processes (red), MFs (magenta), and CCs (blue).

Figure 4. A) GO enrichment analysis of the upregulated GSE55399 DEGs, B) GO enrichment analysis of the downregulated GSE55399 DEGs. The top 10 annotations based on their *p*-values are shown for biological processes (red), MFs (magenta), and CCs (blue).

Prediction of hub genes through PPI network

Physical and functional associations between DEG proteins were evaluated using STRING. The confidence threshold for the minimum required interaction score was set at 0.90. Groups resulting from STRING analysis were visualized using Cytoscape. The nodes represent the number of proteins, while the edges represent their interactions. The MCODE plugin in Cytoscape was utilized to visually depict clusters of closely interconnected network interactions (Fig. 2). This analysis identified hub genes in 22 and 6 nodes for upregulated and downregulated overlapping GSE25835 and GSE40115 DEGs, respectively. These were 37 and 10 nodes for upregulated and downregulated GSE55399 DEGs, respectively. This hub genes were utilized for additional analysis. The DEGs were listed in Tables S1 and S2.

GO enrichment analysis

Based on the GO analysis of significant DEGs, the top 10 annotations were selected for biological process ontology (BP), molecular function (MF), and cellular component (CC). The DEGs that overlapped from GSE25835 and GSE40115, as depicted in Figure 3A, were significantly enriched in the mitotic spindle organization and mitotic cell cycle. Protein kinase binding, histone kinase activity, and cyclin-dependent regulator activity were observed to be upregulated in MF. For CC, significant enrichment was observed in the spindle, cyclin-dependent protein kinase holoenzyme complex, and serine/threonine protein kinase complex. On the other hand, in Figure 3B, RNA splicing was observed to be downregulated in BP. For CC, prespliceosome and spliceosome downregulation was observed. The

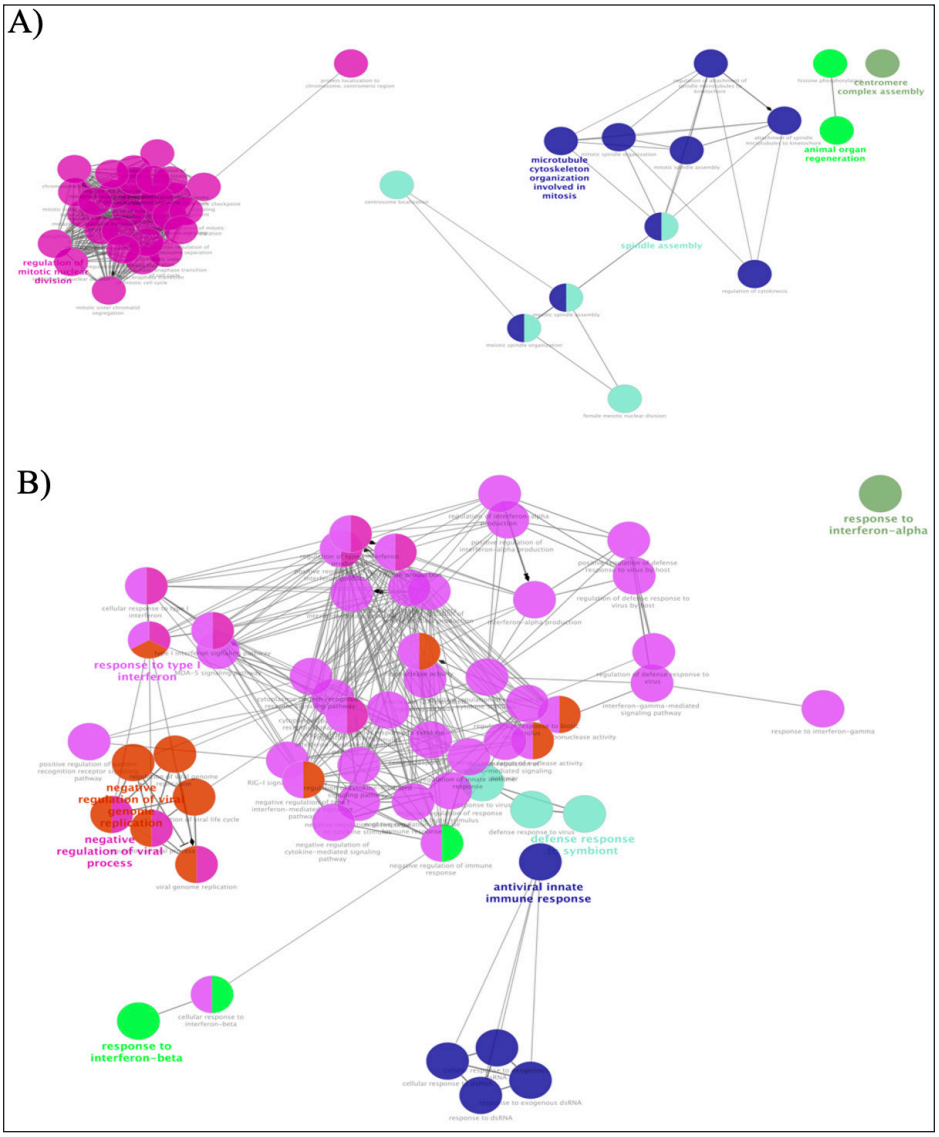
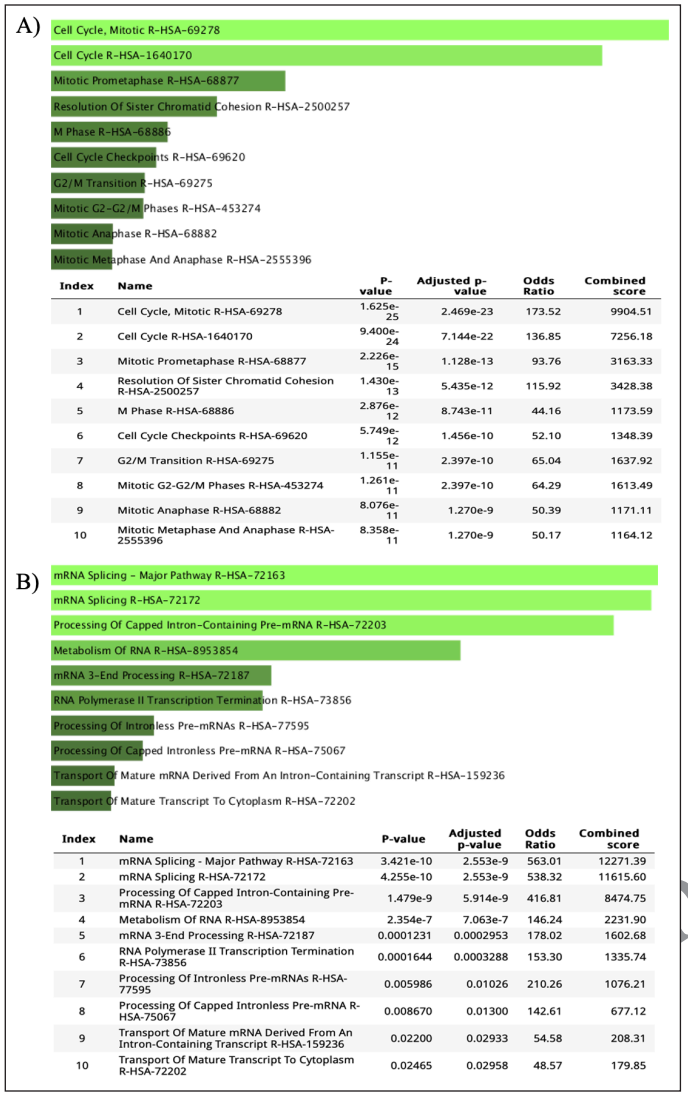


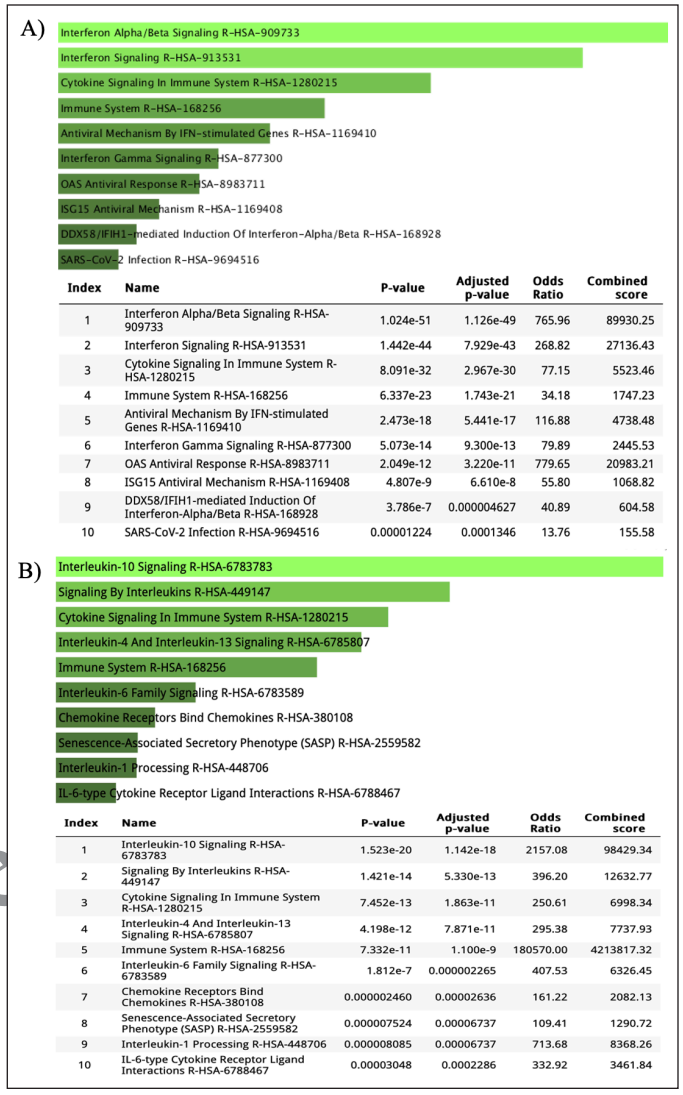
Figure 5. The GO enrichments were visualized using the ClueGO plugin from Cytoscape. A) biological processes (BP) interactions of overlapping upregulated GSE25835 and GSE40115 DEGs; B) biological processes (BP) interactions of upregulated GSE55399 DEGs. The bold fonts indicate the most important functional GO terms.



Index	Name	P-value	Adjusted p-value	Odds Ratio	Combined score
1	Cell Cycle, Mitotic R-HSA-69278	1.625e-25	2.469e-23	173.52	9904.51
2	Cell Cycle R-HSA-1640170	9.400e-24	7.144e-22	136.85	7256.18
3	Mitotic Prometaphase R-HSA-68877	2.226e-15	1.128e-13	93.76	3163.33
4	Resolution Of Sister Chromatid Cohesion R-HSA-2500257	1.430e-13	5.435e-12	115.92	3428.38
5	M Phase R-HSA-68886	2.876e-12	8.743e-11	44.16	1173.59
6	Cell Cycle Checkpoints R-HSA-69620	5.749e-12	1.456e-10	52.10	1348.39
7	G2/M Transition R-HSA-69275	1.155e-11	2.397e-10	65.04	1637.92
8	Mitotic G2-G2/M Phases R-HSA-453274	1.261e-11	2.397e-10	64.29	1613.49
9	Mitotic Anaphase R-HSA-68882	8.076e-11	1.270e-9	50.39	1171.11
10	Mitotic Metaphase And Anaphase R-HSA-2555396	8.358e-11	1.270e-9	50.17	1164.12

Index	Name	P-value	Adjusted p-value	Odds Ratio	Combined score
1	mRNA Splicing - Major Pathway R-HSA-72163	3.421e-10	2.553e-9	563.01	12271.39
2	mRNA Splicing R-HSA-72172	4.255e-10	2.553e-9	538.32	11615.60
3	Processing Of Capped Intron-Containing Pre-mRNA R-HSA-72203	1.479e-9	5.914e-9	416.81	8474.75
4	Metabolism Of RNA R-HSA-8953854	2.354e-7	7.063e-7	146.24	2231.90
5	mRNA 3-End Processing R-HSA-72187	0.0001231	0.0002953	178.02	1602.68
6	RNA Polymerase II Transcription Termination R-HSA-73856	0.0001644	0.0003288	153.30	1335.74
7	Processing Of Intronless Pre-mRNAs R-HSA-77595	0.005986	0.01026	210.26	1076.21
8	Processing Of Capped Intronless Pre-mRNA R-HSA-75067	0.008670	0.01300	142.61	677.12
9	Transport Of Mature mRNA Derived From An Intron-Containing Transcript R-HSA-159236	0.02200	0.02933	54.58	208.31
10	Transport Of Mature Transcript To Cytoplasm R-HSA-72202	0.02465	0.02958	48.57	179.85

Figure 6. Pathway enrichment analysis for overlapping GSE25835 and GSE40115 DEGs: A) upregulated DEGs and B) downregulated DEGs by using REACTOME.



Index	Name	P-value	Adjusted p-value	Odds Ratio	Combined score
1	Interferon Alpha/Beta Signaling R-HSA-909733	1.024e-51	1.126e-49	765.96	89930.25
2	Interferon Signaling R-HSA-913531	1.442e-44	7.929e-43	268.82	27136.43
3	Cytokine Signaling In Immune System R-HSA-1280215	8.091e-32	2.967e-30	77.15	5523.46
4	Immune System R-HSA-168256	6.337e-23	1.743e-21	34.18	1747.23
5	Antiviral Mechanism By IFN-stimulated Genes R-HSA-1169410	2.473e-18	5.441e-17	116.88	4738.48
6	Interferon Gamma Signaling R-HSA-877300	5.073e-14	9.300e-13	79.89	2445.53
7	OAS Antiviral Response R-HSA-8983711	2.049e-12	3.220e-11	779.65	20983.21
8	ISG15 Antiviral Mechanism R-HSA-1169408	4.807e-9	6.610e-8	55.80	1068.82
9	DDX58/IFIH1-mediated Induction Of Interferon-Alpha/Beta R-HSA-168928	3.786e-7	0.000004627	40.89	604.58
10	SARS-CoV-2 Infection R-HSA-9694516	0.00001224	0.0001346	13.76	155.58

Index	Name	P-value	Adjusted p-value	Odds Ratio	Combined score
1	Interleukin-10 Signaling R-HSA-6783783	1.523e-20	1.142e-18	2157.08	98429.34
2	Signaling By Interleukins R-HSA-449147	1.421e-14	5.330e-13	396.20	12632.77
3	Cytokine Signaling In Immune System R-HSA-1280215	7.452e-13	1.863e-11	250.61	6998.34
4	Interleukin-4 And Interleukin-13 Signaling R-HSA-6785807	4.198e-12	7.871e-11	295.38	7737.93
5	Immune System R-HSA-168256	7.332e-11	1.100e-9	180570.00	4213817.32
6	Interleukin-6 Family Signaling R-HSA-6783589	1.812e-7	0.000002265	407.53	6326.45
7	Chemokine Receptors Bind Chemokines R-HSA-380108	0.000002460	0.00002636	161.22	2082.13
8	Senescence-Associated Secretory Phenotype (SASP) R-HSA-2559582	0.000007524	0.00006737	109.41	1290.72
9	Interleukin-1 Processing R-HSA-448706	0.000008085	0.00006737	713.68	8368.26
10	IL-6-type Cytokine Receptor Ligand Interactions R-HSA-6788467	0.00003048	0.0002286	332.92	3461.84

Figure 7. Pathway enrichment analysis for GSE55399 DEGs: A) upregulated DEGs and B) downregulated DEGs by using REACTOME.

downregulation of RNA binding and adenyltransferase activity were observed in MF.

The GO analysis of upregulated GSE55399 DEGs revealed significant interferon signaling enrichment (Fig. 4A). RNA binding, adenyltransferase activity, and nuclease activity were observed to be upregulated in MF. For CC, a notable mitochondrial membrane enrichment was observed. Significant enrichment of the cytokine-mediated signaling pathway was observed for the GSE55399 DEGs for BP (Fig. 4B). We observed a decrease in cytokine signaling and growth factor activity in MF. For CC, a noteworthy enrichment in blebs and the nuclear membrane's outermost layer was observed.

ClueGO enrichment analysis

The Cytoscape ClueGO plugin allows the performance of functional enrichment analysis on genes that have been identified as differentially expressed. ClueGo enables the visualization of gene annotation clusters within PPI networks. Figure 5 depicts the GO for MFs and BF. The statistical method employed for the ClueGO enrichment analysis was the utilization of a two-tailed hypergeometric test with a significance threshold set at 0.05. In addition, the Benjamini–Hochberg correction was applied, and a kappa score of 0.4 was used as the key criterion. The enrichment results obtained from ClueGO suggest that mutations in *BRCA1/2* genes have the potential to alter the regulation of mitosis. In addition, the administration of PARP inhibitors to individuals with *BRCA1/2* mutations is expected to enhance the interferon response and reduce cytokine signaling. To validate our bioinformatics findings, it is necessary to conduct functional validations.

Enrichment of pathways

REACTOME, one of the largest metabolic pathway information databases, was used to analyze the DEG pathway. As a result of mutations in the *BRCA1/2* gene (dataset GSE25835 and GSE40115), DEGs that are upregulated are involved in the

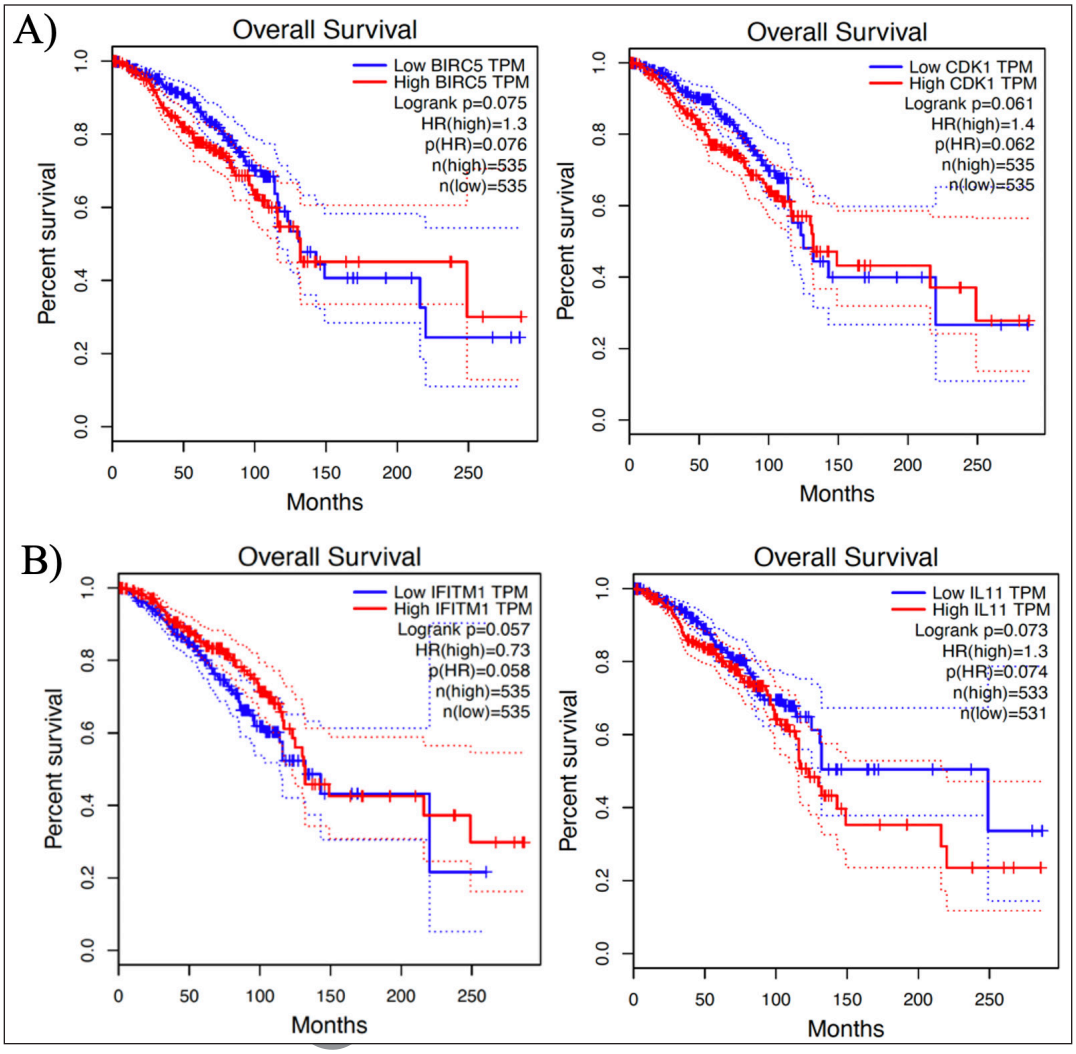


Figure 8. Kaplan–Meir plots that showed OS analysis: A) poor OS due to the upregulated expression BIRC5 and CDK1 and B) poor OS due to the downregulated expression of IFITM1 and upregulated IL11 expression.

cell cycle pathway, while DEGs that are downregulated are involved in the mRNA splicing pathway (Fig. 6). On the other hand, in PARP inhibitory therapy (dataset GSE55399), DEGs that were upregulated were involved in interferon signaling, whereas DEGs that were downregulated were involved in cytokine signaling (Fig. 7).

Survival analysis

The GEPIA results display an analysis of the hub genes' OS. Figure 8 depicts the identification criteria: HR score > 1 (upregulated gene); HR < 1 (downregulated gene) and log-rank $p < 0.05$. This result suggested that the upregulated expression of BIRC5 (logrank $p = 0.075$; HR = 1.3) and CDK1 (logrank $p = 0.061$; HR = 1.4) were associated with an impaired OS, as were the downregulated expression of interferon-induced transmembrane 1 (IFITM1) (logrank $p = 0.057$; HR = 0.73) and the upregulated expression of Interleukin 11 (IL11) (logrank $p = 0.073$; HR = 1.3). Unfortunately, those results were not statistically significant because the log-rank p-score was above 0.05. However, the increased expression of BIRC5 and CDK1 in *BRCA1/2* mutation carriers may be associated with a low OS. In contrast, treatment with a PARP inhibitor may improve OS by upregulating IFITM1 and downregulating IL11 expression.

This result is consistent with previous research indicating that BC patients with an upregulated BIRC5 gene, which codes for Survivin, have a lower OS rate [30,31]. Survivin is one of the apoptosis inhibitors implicated in cell division regulation and apoptosis inhibition [32]. *In vitro* research revealed that endogenously defective or mutant *BRCA1* (MDA-MB-436) cells express a higher level of survivin than do normal cells [33]. Other research revealed that *BRCA1* mutant mouse mammary tumors have low levels of SIRT1 and high levels of Survivin [34].

In mammalian cells, the cyclin-dependent kinases (CDKs), which include CDK2, CDK4, and CDK6, and also CDK1, are crucial regulators of cell cycle progression [35]. Several malignancies, including BC, have been found to exhibit CDK dysregulation that results in increased cell proliferation [36]. Our findings are consistent with previous studies, indicating that BC patients with high CDK1 expression had a worse 5-year recurrence-free survival compared to those with low CDK1 expression [37]. Therefore, the upregulation of

BIRC5 and CDK1 expression in *BRCA1/2* mutant carriers who may have a low OS rate is a promising marker.

The upregulation of IFITM1 gene expression was observed in individuals with *BRCA1/2* mutations who underwent treatment with PARP inhibitors. IFITM1 is a member of the immune-related IFITM protein family that is expressed on the cell surface [37]. The lack of *BRCA2* leads to the activation of cell-intrinsic immunological signaling. In addition, the administration of PARP inhibitors elicits the interferon response in cells and tumors that are defective in *BRCA2*, which aligns with the findings of our works [38]. IFITMs have been the subject of intense research due to their role in viral inhibition, and a considerable body of evidence has established a correlation between IFITMs and cancer growth. The increased expression of IFITM1 has an impact on the expression of HLA-B, potentially affecting the presentation and detection of cancer-causing antigens on the surface of cells by cytotoxic T cells. Consequently, this may hinder the complete elimination of tumor cells [37].

In our study, PARP inhibitor interventions for *BRCA1/2* mutation carriers resulted in decreased IL11 expression. Analysis of OS revealed that downregulation of IL11 expression improved survival [39]. Based on our research, it has been observed in various studies that decreased expression of IL-11 is associated with enhanced survival rates among BC patients. IL-11 belongs to the IL-6 cytokine family, characterized by the utilization of the GP130 signal transducing receptor subunit. The cytokine GP130 facilitates the enhancement of the "cancer-specific capabilities" of the malignant epithelium within the tumor's inflammatory microenvironment, while concurrently suppressing the immunological reaction of innate and adaptive immune cells toward the tumor. IL-11, aside from its documented role as a hemopoietic growth factor, is increasingly becoming recognized for its involvement in the biology of epithelial cancer [40]. Several studies showed as well that PARP inhibitors may improve the treatment of genomically unstable cancers by activation/modulation of the immune system and attenuating the inflammatory response [41–43]. Therefore, the upregulation of IFITM1 and the downregulation of IL11 can serve as a prospective evaluation marker for PARP inhibitor treatment in *BRCA1/2* mutant carriers.

Nevertheless, our investigation has encountered some limitations. The DEGs discovered were derived from an online database. To validate our research, additional tests conducted using patient samples are required. Furthermore, we utilized a dataset sourced from a non-Asian population as a result of the scarcity of datasets available in the database. Further investigation is required to specifically examine the subsequent processes triggered by *BRCA1/2* mutations in BC and the application of PARP inhibitor treatment.

CONCLUSION

We identified hub genes of cell cycle response (CDK1 and BIRC5) that are strongly linked to *BRCA1/2* mutation. PARP inhibitor treatments in *BRCA1/2* mutant carriers are strongly related to the upregulation of IFITM1 (interferon signaling) and the downregulation of IL11 (cytokine signaling). Therefore, PARP inhibitors may improve the treatment by activation/modulation the immune system and attenuating the inflammatory response. However, the dataset used to analyze the DEGs of PARP inhibitor treatments in *BRCA1/2* mutant carriers still used BC cell lines, forthcoming research may be able to use clinical patients as the subjects. Moreover, functional studies are further needed to validate this finding.

AUTHORS' CONTRIBUTIONS

RIP: Formal analysis, write the manuscript; SIW, SSP, FF: Conceptualize and supervise the research.

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CONFLICTS OF INTEREST

The authors declare that they have no competing interests.

ETHICAL APPROVALS

This study does not involve experiments on animals or human subjects.

DATA AVAILABILITY

Datasets were retrieved from the GEO (Gene Expression Omnibus) database (https://www.ncbi.nlm.nih.gov/geo/): GSE25835, GSE40115, and GSE55399.

PUBLISHER'S NOTE

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SUPPLEMENTARY MATERIAL

Supplementary data can be downloaded from the journal's website link: [https://japsonline.com/admin/php/uploadss/4310_pdf.pdf]