Review Article

# DSS1 in clinical laboratory medicine and oncology from RNA metabolism and R-loop homeostasis to BRCAness and PARP inhibitor response

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**ABSTRACT**

Cancer genome testing routinely reports driver alterations, mutational signatures, and DNA repair states, yet key clinical phenotypes—tumor behavior, relapse risk, and chemoresistance—are not fully explained by genotype alone. Deleted in split-hand/split-foot 1 (DSS1) is a small, acidic, intrinsically disordered protein conserved across eukaryotes and shared by several large multiprotein assemblies, making it a plausible functional modifier of these phenotypes. This review summarizes DSS1 biology from the perspective of clinical laboratory medicine, focusing on how DSS1 links RNA metabolism, R-loop homeostasis, and homologous recombination repair (HRR) and how this linkage may inform biomarker development. Mechanistically, DSS1 behaves as a stabilizing "molecular glue" in protein complexes and serves as a core component of RNA export/quality-control modules, including TREX-2 and the TREX-2.1/REX complex, which regulate co-transcriptional messenger ribonucleoprotein remodeling, nuclear export, and RNA surveillance—processes closely tied to R-loop (DNA:RNA hybrid) control and transcription-coupled DNA damage. DSS1 is also an obligate cofactor of BRCA2: DSS1 binding stabilizes BRCA2, improves its solubility, and limits off-target interactions, supporting repair-competent BRCA2 in the nucleus. We then review clinicopathological evidence that DSS1 dysregulation correlates with cancer behavior, including shorter relapse-free survival and a chemoresistant phenotype. Finally, we propose that DSS1-low, *BRCA*-proficient tumors may acquire BRCAness, providing a rationale for PARP inhibitor responses beyond current BRCA-focused companion diagnostics, and we outline how standardized DSS1 measurement (mRNA and/or protein), integrated with BRCA status and other HRR-related signatures, could refine molecular testing and therapeutic stratification in routine practice.

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**Key Words**
BRCAness, DSS1, PARP inhibitor, R-loop, transcription-coupled DNA damage

## I. Introduction

Cancer genome testing has transformed oncology from descriptive histology to molecular pathology by reading out driver alterations, mutational signatures, and repair-pathway states. These readouts reflect multiple mutagenic and repair processes and illustrate not only oncogenic mechanisms but also actionable alterations that predict chemosensitivity[1)2)]. To interpret them further, we need to map each alteration onto its

underlying oncogenic pathway. Some alternative patterns come from outside exposures (e.g., UV light or tobacco), while others are caused by enzymatic mutagenesis (e.g. activation-induced cytidine deaminase (AID) / apolipoprotein B mRNA editing catalytic polypeptide-like (APOBEC) family), mismatch-repair failure, or replication stress[3)4)]. In some contexts, transcription itself may also add DNA damage both controllably and pathologically, the latter of which is called transcription-coupled DNA damage[5)6)].

Deleted in split-hand/split-foot 1 (DSS1) was originally described through genetic mapping studies of split-hand/split-foot malformation (SHFM), where it appeared within a critical chromosomal region on 7q21.3–q22.17). However, the developmental abnormalities in SHFM are now more commonly attributed to dysregulation of genes near DSS1, such as DLX5 and DLX6, than to a DSS1-specific mechanism[8)]. Subsequent biochemical and genetic analyses revealed that DSS1 is a highly conserved, small (~70 amino acids in yeast; ~70–80 in mammals), acidic, and intrinsically disordered protein with surprisingly broad functional diversity[7)9)–12)].

Orthologs of DSS1, termed Sem1 in budding yeast, are conserved across eukaryotes[10)13)]. DSS1/Sem1 is now recognized as a component of multiple large protein complexes, including the 19S lid of the 26S proteasome, a stable subunit of the TRanscription–EXport-2 (TREX-2) and TREX-2.1/Repressor of EXport (REX) complexes, and an essential cofactor of BRCA2 in homologous recombination (HR)[14)–22)]. Therefore, DSS1 contributes to several fundamental processes: protein degradation, mRNA biogenesis and export, transcription-coupled genome maintenance, DNA repair, and replication stress responses. Accordingly, genetic and functional alterations in DSS1 or its partner complexes can affect genomic instability, oncogenesis, chemoresistance, and tumor development[23)–29)].

Here, we review DSS1 from the perspective of clinical laboratory medicine. We first outline its molecular and structural properties, as well as its role as an obligate cofactor of BRCA2 in HR and R-loop control. We then summarize clinicopathological and experimental evidence linking DSS1 dysregulation to cancer behavior. Finally, we show the emergence of BRCAness and poly (adenosine diphosphate-ribose) polymerase inhibitor (PARPi) sensitivity in DSS1-low, *BRCA*-proficient tumors, and discuss how DSS1 could be developed as a biomarker and therapeutic modifier in cancer genome testing.

## II. Structural and functional roles of DSS1

### *Molecular and structural features of DSS1*

The human DSS1 gene is located on chromosome 7q21 and encodes a small acidic protein of ~70 amino acids[7)13)]. Orthologous genes (Sem1 in *Saccharomyces cerevisiae, dss1*$^{+}$ in *Schizosaccharomyces pombe*, etc.) show strong sequence conservation of acidic and hydrophobic motifs[11)13)30)]. DSS1 lacks extensive secondary structure under physiological conditions, being classified as an intrinsically disordered protein[31)32)]. Nuclear magnetic resonance spectroscopy and other biophysical analyses indicate that DSS1 can transiently form helical segments and multiple short linear motifs, which become stabilized upon binding to structured partners[33)34)].

This intrinsic disorder gives DSS1 two key properties: 1) binding plasticity, allowing DSS1 to interact with numerous partners through distinct interfaces, and 2) molecular "glue" behavior, whereby DSS1 often binds at the interface between two or more structured subunits and stabilizes their assembly. Small, acidic, intrinsically disordered proteins such as DSS1 function as molecular glues in the PCI-containing complexes, inserting between PCI domain proteins to stabilize and regulate large multiprotein assemblies[11)12)17)]. Comprehensive interactome analyses, particularly in *S. pombe*, have revealed that DSS1 associates with a broad range of complexes, including the 19S lid of the 26S proteasome, the TREX-2 and TREX-2.1/REX complexes, the Thp3–Csn12–Sem1 complex, and mitotic septin assemblies[14)-22)35)]. This diversity suggests that DSS1 functions more as a general scaffolding cofactor for PCI-containing complexes than as a single-pathway factor. In addition, DSS1 forms an obligate complex with the non-PCI tumor suppressor BRCA2[13)].

### *DSS1 in the TREX-2 complex, mRNA nuclear export, and R-loop formation*

The TREX-2 complex is a conserved assembly that couples transcription to mRNA export at the nuclear pore complex (NPC). In mammals, TREX-2 is built on a germinal center–associated nuclear protein (GANP) scaffold, with DSS1, PCI domain–containing 2 (PCID2), enhancer of yellow 2 (ENY2), and centrin 2/3 (CETN2/3) as additional subunits (**Figure 1**)[18)19)22)]. TREX-2 interacts with the NPC and transcriptional regulators (e.g., mediator), thereby facilitating the delivery of nascent transcripts to nuclear pores[36)-38)]. In yeast, Sem1-deficient cells show defective mRNA export, impaired transcription elongation, and accumulation of

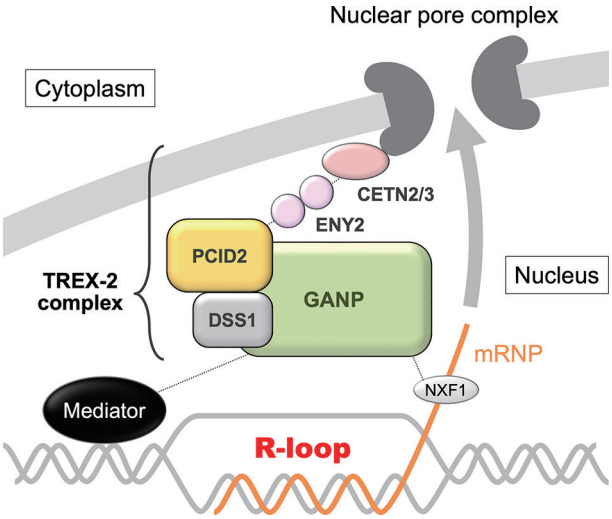


**Figure 1** Schematic representation of mRNA nuclear export through the nuclear pore complex. In mammals, the TRanscription–EXport-2 (TREX-2) complex comprises germinal center–associated nuclear protein (GANP), deleted in split-hand/split-foot 1 (DSS1), PCI domain–containing 2 (PCID2), enhancer of yellow 2 (ENY2), and centrin 2/3 (CETN2/3). TREX-2 associates with messenger ribonucleoprotein (mRNP) particles, in part via the mRNA export receptor nuclear RNA export factor 1 (NXF1), and facilitates the targeting of transcripts to nuclear pores for export to the cytoplasm. Impaired TREX-2 function can lead to nuclear retention of mRNA and is proposed to increase the opportunity for nascent RNA to re-anneal to its template DNA, forming DNA:RNA hybrids (R-loops). Excessive or persistent R-loops can promote transcription-associated DNA damage, including replication stress and DNA double-strand breaks, and thereby contribute to mutagenesis and genome instability. Adapted from Pathol Int 2024; 74(3): 103–18 with permission and minor modification[19].

DNA damage; this phenotype is similar to that of cells lacking other TREX-2 components or the principal mRNA export receptor Mex67/nuclear RNA export factor 1 (NXF1)[26)39)-42)]. In mammals, DSS1/Sem1 is similarly included in the TREX-2 complex, although its dynamics may vary among cell types or conditions.

TREX-2 is now recognized as an important factor in R-loop homeostasis. R-loops are three-stranded structures that form when the nascent RNA transcript slips back and re-anneals to its DNA template, creating a DNA:RNA hybrid and leaving the non-template DNA strand displaced as a single-stranded DNA (ssDNA). R-loops can have useful functions under physiological conditions. They form at certain promoters and transcription terminator regions, where they help control transcription initiation and termination and influence local chromatin structure[43)-47)]. In B cells, R-loops at immunoglobulin switch regions are deliberately formed as intermediates of class-switch recombination, because the exposed ssDNA is targeted by AID to trigger programmed DNA breakage and joining[48)-51)]. In other systems, including mitochondrial DNA, R-loop-like structures contribute to replication initiation by providing both an RNA primer and a structural platform for DNA polymerases[52)-55)]. In these contexts, R-loops are formed at specific loci and removed once their role is fulfilled, maintaining what is often referred to as R-loop homeostasis.

However, when R-loops become too abundant, too stable, or mislocalized, they turn from useful intermediates into threats to genome integrity. Persistent R-loops act as roadblocks for replication forks, leading to fork stalling, reversal, or collapse, and thereby causing replication stress and DNA double-strand breaks (DSBs)[56)-59)]. The displaced ssDNA is chemically vulnerable and prone to damage and erroneous repair; therefore, excessive R-loops drive mutagenesis and are recognized as a major source of transcription-coupled DNA damage[60)-62)]. Factors that play a role in RNA processing and export, helicases that unwind DNA:RNA hybrids, and topoisomerases that relieve torsional stress help prevent harmful R-loop accumulation; when these systems fail, R-loops build up and contribute to oncogenesis[63)-65)].

The TREX-2 complex is positioned at the nuclear pore, where it helps move nascent RNA transcripts away from their template DNA by anchoring actively transcribed genes near the pore basket and handing the transcripts off to the export receptor NXF1[38)66)-68)]. By reducing the dwell time of free RNA near its template DNA, the TREX-2 complex is thought to lower the chance that RNA will re-anneal and form R-loops. Collectively,

the TREX-2 complex may promote co-transcriptional messenger ribonucleoprotein (mRNP) remodeling and efficient export. Although the exact contribution of DSS1 within the TREX-2 complex to R-loop suppression remains incompletely defined, DSS1 deficiency may impair TREX-2 function and thus indirectly promote R-loop accumulation and transcription-coupled DNA damage.

### *DSS1 in the TREX-2.1/REX complex*

DSS1 also plays a key role in the human TREX-2.1 complex, which was recently described[21]. The TREX-2.1 complex is a trimer composed of LENG8, DSS1, and PCID2: LENG8 and PCID2 are PCI domain proteins, and DSS1 binds their PCI-type helical scaffolds in the same way as in GANP–PCID2 of the TREX-2 complex. Structural studies show that the TREX-2.1 complex binds the DEAD-box helicase DDX39B (UAP56), a core component of another mRNA nuclear export machinery, the THO/TREX complex.

Functionally, the TREX-2.1 complex behaves as an mRNP remodeling factor that controls DDX39B activity. Biochemical studies have shown that the TREX-2.1 complex binds directly to DDX39B, stimulates its ATPase activity, and can displace it from RNA[21]. More recently, the same LENG8–DSS1–PCID2 module has been identified as the core of the REX complex, which acts in nuclear RNA quality control. In that pathway, LENG8 is recruited to incompletely processed or intron-retaining transcripts and targets them for degradation instead of allowing them to be exported from the nucleus[21,69]. Together, these findings indicate that DSS1 is an essential factor in RNA metabolism, including post-splicing control, mRNA nuclear export, and nuclear RNA surveillance. However, whether DSS1-driven abnormalities in RNA processing directly contribute to oncogenesis remains unclear.

### *DSS1 as an obligatory cofactor of BRCA2 in homologous recombination*

When a DNA DSB occurs, the sister chromatid serves as a template for the homologous recombination repair (HRR) pathway. BRCA2 is an important HRR factor that loads RAD51 recombinase onto ssDNA at DSBs and stalled replication forks[70-72]. DSS1 forms a stable complex with the BRCA2 DNA-binding domain (DBD) and is considered an obligate partner of BRCA2[13,73]. Biochemical and structural studies show that DSS1 binds to multiple surfaces of the BRCA2 DBD, including the helical domain and the oligonucleotide/oligosaccharide-binding (OB) folds[25,73]. This interaction stabilizes BRCA2, improves its solubility, and promotes efficient RAD51 filament formation on ssDNA. DSS1 depletion or *BRCA2* mutations that weaken DSS1 binding reduce the steady-state level of BRCA2 in the nucleus, shift it into less-soluble, partially aggregated states, and decrease nuclear HRR activity[23,74-76].

In addition, DSS1 prevents binding of the BRCA2 DBD to double-stranded DNA (dsDNA). In the absence of DSS1, BRCA2 exhibits increased binding to dsDNA, which could misdirect RAD51 to inappropriate regions[25]. Moreover, loss of DSS1 induces aberrant BRCA2 oligomeric states that are less competent for proper RAD51 loading[75,77]. Thus, the BRCA2–DSS1 partnership is essential for maintaining stable, correctly assembled, repair-competent BRCA2 molecules in the nucleus (**Figure 2**). This tuning mechanism provides a useful conceptual framework: DSS1 acts as an allosteric

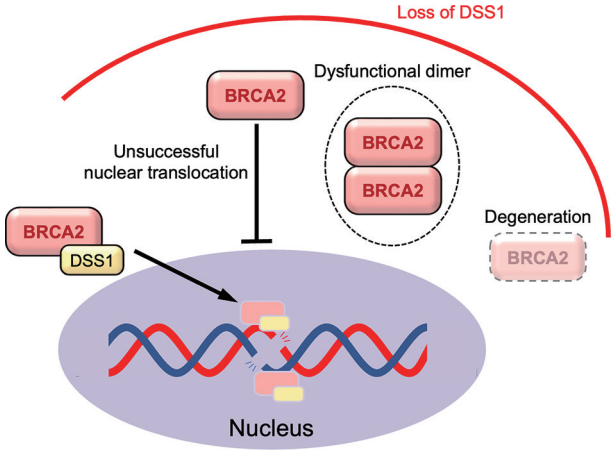


**Figure 2** Schematic representation of BRCA2 dysfunction in the absence of DSS1. Loss of DSS1 destabilizes BRCA2, leading to altered nuclear distribution, formation of aberrant and nonfunctional BRCA2 dimers/oligomers, and BRCA2 degradation mediated by chaperones.

modulator that optimizes BRCA2's substrate specificity in HRR and replication fork protection while reducing off-target interactions that could be deleterious.

BRCA2 also participates in R-loop processing. Several observations suggest that BRCA2 helps prevent harmful R-loop accumulation. BRCA2 interacts with RNA polymerase II and factors that control promoter-proximal pausing, and thereby helps avoid prolonged stalling of the transcription machinery, a situation that favors R-loop formation[78)79)]. In addition, BRCA2 works together with R-loop–processing factors such as the RNA helicase DDX5 and the ribonuclease RNase H2 to dissolve DNA:RNA hybrids and repair R-loop–associated lesions[79)80)]. Moreover, interactions between BRCA2 and subunits of the TREX-2 complex help suppress R-loop–induced genomic instability[81)].

Thus, it is plausible that DSS1, by jointly acting in the TREX-2 complex and BRCA2, coordinates mRNA metabolism with HR-mediated processing.

### III. DSS1 dysregulation in breast cancer

Our work and that of others have examined DSS1 expression in human cancers[27)-29)]. Because DSS1 participates in mRNA metabolism, R-loop homeostasis, and HRR machinery, its dysfunction is thought to be closely associated with oncogenesis and chemosensitivity in various cancers.

Of all cancer types, the association between DSS1 expression and breast cancer has been examined most thoroughly. We have reported that, in invasive breast carcinomas, high DSS1 expression was associated with significantly shorter relapse-free survival although the DSS1 level did not correlate strongly with classical clinicopathological factors such as tumor size, nodal status, histological grade, hormone receptor status, or Ki-67 index[27)]. This indicates that DSS1 is an independent marker that is not simply a surrogate for proliferation or advanced stage. Interestingly, functional assays using breast cancer cell lines complemented the clinical data[27)]. DSS1-overexpressing MCF7 cells (estrogen receptor α (ERα)-positive, p53 wild type) showed decreased chemosensitivity to DNA-damaging agents such as camptothecin and etoposide. Cell-cycle analysis and apoptosis assays showed that these cells had a smaller sub-G1 fraction after drug exposure than control cells, indicating increased chemoresistance. Conversely, siRNA-mediated DSS1 knockdown suppressed proliferation and induced apoptosis in both MCF7 and drug-resistant MDA-MB-231 (triple-negative, p53-deficient) cells. When combined with camptothecin or etoposide, DSS1 knockdown further enhanced drug-induced cell death, particularly in MDA-MB-231 cells.

Another study of ours extended these findings by analyzing DSS1 together with PCID2 and BRCA2 in breast carcinomas. DSS1 expression was consistently higher in tumor tissues than in normal breast tissue, whereas PCID2 expression was similar between normal and malignant tissues (**Figure 3**)[28)]. When patients were divided into high- and low-expression groups for DSS1, BRCA2, and PCID2, only DSS1 showed a significant association with poor relapse-free survival. This confirmed DSS1 as the key clinical predictor among these three factors. In vitro analysis, propidium iodide staining, single-cell gel electrophoresis (comet assays), and clonogenic survival assays to assess chemosensitivity revealed that DSS1 depletion in breast cancer cell lines increased sensitivity to several DNA-damaging agents, whereas DSS1 overexpression conferred chemoresistance[28)].

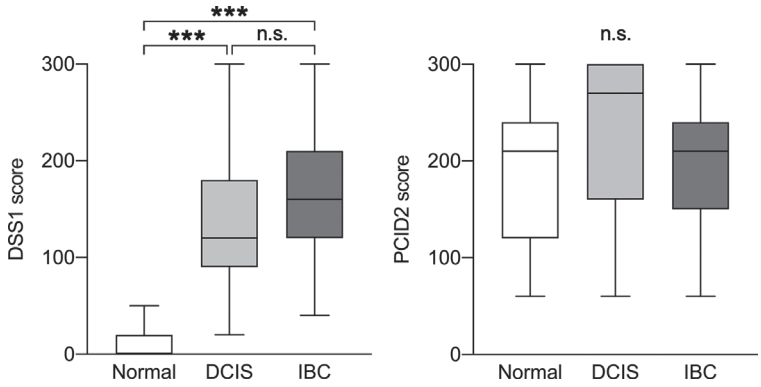


**Figure 3** DSS1 (left) and PCID2 (right) expression scores in normal breast tissue, ductal carcinoma in situ (DCIS), and invasive breast carcinoma (IBC). Expression scores (0–300) were calculated by multiplying staining intensity (0–3; weak to strong) by the percentage of positive cells (0%–100%) in immunohistochemistry. Box plots show the median (center line) and the 25th and 75th percentiles (box); whiskers indicate the range. ***$P < 0.001$; n.s., not significant. Adapted from Lab Invest 2021; 101 (8): 1048–59 with permission and minor modification[28)].

Importantly, BRCA2 expression itself did not significantly affect chemosensitivity under the same conditions, even though DSS1 depletion reduced BRCA2 protein levels. This indicates that the change in drug response after DSS1 knockdown is not simply a consequence of reduced BRCA2. Furthermore, PCID2 behaved similarly to DSS1 at the functional level.

Taken together, DSS1 is a clinically relevant modifier of breast cancer behavior that sits squarely within the scope of clinical laboratory medicine. Functional data revealed that high DSS1 is not just a passive marker but a surrogate for a tumor's ability to tolerate genotoxic stress, whereas low DSS1 defines a potentially druggable vulnerability. For clinical laboratory medicine, this means that DSS1 expression is a candidate prognostic and predictive biomarker that could, after further validation, be incorporated alongside ER, progesterone receptor (PR), human epidermal growth factor receptor 2 (HER2), Ki-67, and p53 to refine risk stratification and anticipate response to standard chemotherapy.

## IV. DSS1 dysregulation in renal cancer

The work of others has indicated that DSS1 is upregulated in metastatic clear cell renal cell carcinoma (ccRCC) and promotes both primary tumor growth and metastasis[29]. High DSS1 expression correlates with a gene signature of proliferation and with poorer outcomes in renal cancer datasets. In addition, large-scale CRISPR screening has identified DSS1 among genes that are essential for the survival of many ccRCC cell lines, suggesting that ccRCC cells depend on DSS1 for fitness.

In metastatic ccRCC cell lines and mouse models, DSS1 promotes tumor growth and lung metastasis[29]. DSS1 interacts directly with LC3 and, via the E3 ligase TRIM25, enhances K63-linked polyubiquitination of LC3B, which leads to suppression of autophagic flux. DSS1 knockdown increases autophagic vacuoles and LC3 puncta, while DSS1 overexpression has the opposite effect, indicating that DSS1 functions as a negative regulator of autophagy in ccRCC. Functionally, when autophagy is restored (by DSS1 silencing or pharmacologic manipulation), ccRCC cells show reduced proliferation, migration, invasion, and metastatic potential. Furthermore, this autophagy block feeds directly into epithelial–mesenchymal transition (EMT) control. DSS1-mediated inhibition of autophagy stabilizes the EMT transcription factor TWIST1, which is otherwise degraded through autophagy.

Overall, DSS1's repertoire may extend beyond genomic maintenance, linking it to autophagy regulation, EMT, and metastatic signaling pathways.

## V. BRCAness and PARPi chemosensitivity in DSS1-low cancers

Cancer genome profiling is now indispensable in modern clinical laboratory medicine. In breast, ovarian, and prostate cancers, both hereditary and sporadic

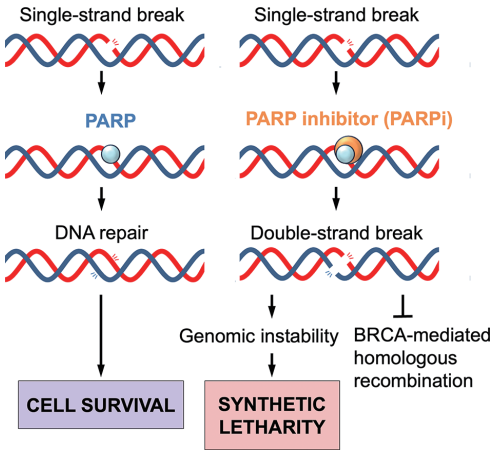


**Figure 4** Schematic representation of DNA repair by poly(adenosine diphosphate-ribose) polymerase (PARP) and the basis of PARP inhibitor (PARPi)–induced synthetic lethality. PARP recognizes SSBs and promotes their repair, supporting cell survival. PARPi inhibit PARP activity at SSBs, causing unrepaired lesions to be converted into DNA double-strand breaks (DSBs) during replication. When homologous recombination repair (HRR), including BRCA1/2-dependent pathways, is intact, such DSBs can be repaired; when HRR is defective, DSB repair fails and cell death ensues. Thus, the combination of HRR deficiency and PARP inhibition is expected to selectively kill HR-deficient cells, a phenomenon termed "synthetic lethality." Notably, clinical responses to PARPi have also been observed in subsets of ovarian and prostate cancers without detectable BRCA mutations by current companion diagnostics, suggesting that additional mechanisms beyond canonical BRCA-dependent synthetic lethality may contribute to PARPi sensitivity.

*BRCA* mutations lead to impaired HRR for DNA damage, contributing to oncogenesis and malignant progression. For such tumors, PARPi have long been expected to exhibit high therapeutic efficacy based on "synthetic lethality." PARPi block the repair of DNA single-strand breaks (SSBs); as a result, in cancer cells that already have defective HRR due to *BRCA* mutations, unrepaired SSBs are converted into DSBs during replication and become essentially irreparable, which selectively kills *BRCA*-mutated cancer cells while largely sparing normal cells (**Figure 4**).

However, recent clinical trials in ovarian and prostate cancers have shown that PARPi can be effective even in patients who test negative on current companion diagnostics for *BRCA* mutations[82)83)]. There is a subset of cancer cells that, despite being *BRCA*-proficient, display biological features similar to *BRCA*-mutated cells—a condition referred to as "BRCAness." Nevertheless, the detailed molecular mechanisms underlying this phenotype, as well as reliable pathological diagnostic methods, have yet to be established.

We therefore hypothesize that, because DSS1 is required for optimal BRCA2 function, DSS1 depletion might disrupt the BRCA2–DSS1 interface and thereby induce BRCAness even in *BRCA2*-proficient cells. We found that, among ovarian cancer subtypes, DSS1 expression was significantly lower in serous carcinoma than in other histological types such as endometrioid, clear cell, and mucinous carcinomas (**Figure 5**). This pattern is consistent with the clinical trial data showing that PARPi are effective in patients with serous carcinoma, irrespective of the results of current companion diagnostics for *BRCA* mutations. In prostate specimens, our small series further suggested that DSS1 expression was generally higher in high-grade prostatic intraepithelial neoplasia (PIN) and appeared to gradually decrease as the Gleason pattern increased from 3 to 5 in acinar adenocarcinomas (**Figure 6**). Functional analyses also indicated that DSS1 depletion reduced HRR activity and enhanced PARPi sensitivity (unpublished data).

Taken together, these observations support a model in which reduced DSS1 expression induces a BRCA-like HRR deficiency and thereby contributes to a BRCAness phenotype that enhances PARPi sensitivity. From the perspective of clinical laboratory medicine, DSS1 expression is therefore a promising biomarker candidate to identify BRCAness in *BRCA*-proficient tumors and to refine the indication for PARPi therapy beyond current BRCA-focused companion diagnostics. In the future, standardized DSS1 assays and integrated interpretation with BRCA status and other HRR-related signatures may help clarify the pathobiology of BRCAness and improve the design of molecular testing panels in routine practice.

## VI. Conclusion

From the viewpoint of clinical laboratory medicine, DSS1 is important because it links several key pathways that we already try to assess in daily practice: mRNA metabolism, R-loop homeostasis, HRR activity, replication stress, and even autophagy and EMT in some tumors. Current cancer genome tests mainly tell us what is mutated (for example, BRCA1/2 status or HRR gene

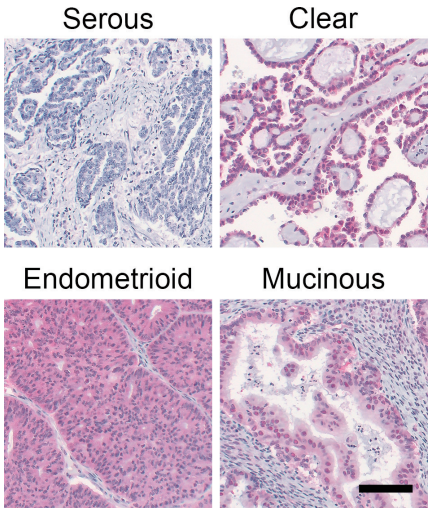


**Figure 5** Representative immunohistochemistry for DSS1 in ovarian carcinoma subtypes (unpublished data). DSS1 expression is maintained in clear cell, endometrioid, and mucinous carcinomas, whereas it is reduced in serous carcinoma. Reduced DSS1 expression may destabilize BRCA2 and potentially contribute to a "BRCAness" phenotype. These observations are consistent with clinical trial data showing PARP inhibitor activity in serous ovarian carcinoma beyond current BRCA-focused companion diagnostics.

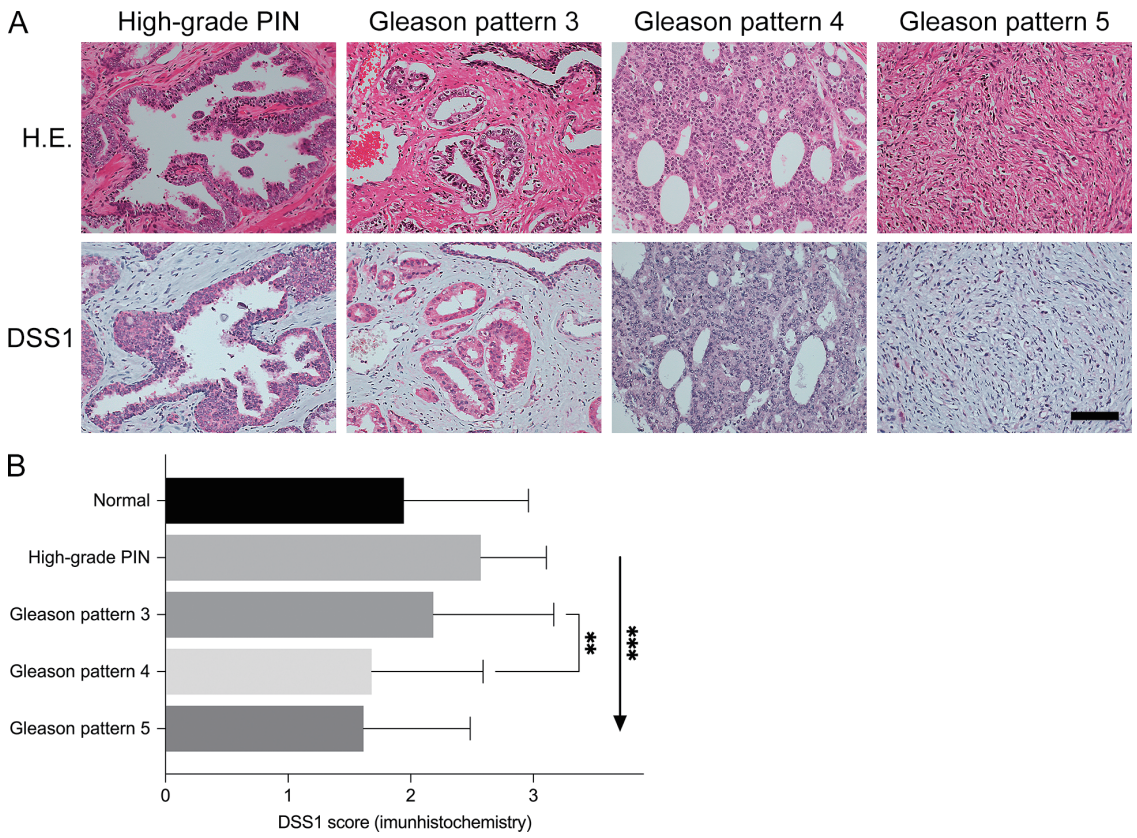


**Figure 6** DSS1 expression in prostate cancer (unpublished data). (A) Representative immunohistochemistry for DSS1 across prostatic lesions and Gleason patterns. (B) DSS1 staining is relatively high in high-grade prostatic intraepithelial neoplasia (PIN) and decreases in acinar adenocarcinoma with increasing Gleason pattern (3–5), as assessed by the Cochran–Armitage trend test (***$P < 0.001$). In addition, the Kruskal-Wallis test followed by Dunn's multiple comparison test showed that DSS1 expression in Gleason pattern 4 carcinoma is significantly lower than that in Gleason pattern 3 carcinoma (**$P = 0.003$). These findings suggest that DSS1 expression tends to decline with histologic progression, and that BRCAness may occur in higher-grade prostate cancer.

panels), but they often cannot explain why some *BRCA*-proficient tumors behave as if they were HR-deficient, or why tumors with similar genotypes show very different responses to chemotherapy and PARPi. Because DSS1 is an essential cofactor of BRCA2 and a shared component of TREX-2 and TREX-2.1/REX complexes, its expression and function may provide a more "functional" readout of these pathways.

The next step for clinical laboratories is to turn this concept into practical tools. DSS1 could be measured at the RNA level in NGS panels and at the protein level by immunohistochemistry, and its status could then be interpreted together with *BRCA* mutations, HR deficiency scores, and other markers of transcription- and replication-related DNA damage. Prospective studies that correlate DSS1 status with prognosis and treatment response, especially to PARPi and DNA-damaging regimens, will be crucial. If such data accumulate and assays are standardized, DSS1 has the potential to become a routine biomarker in cancer genome testing—helping us not only to classify tumors, but also to predict which patients truly have BRCAness, which tumors are likely to be chemoresistant, and where new therapeutic vulnerabilities may lie.

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## Disclosure of Conflicts of Interest

The authors have no conflicts of interest to declare.

**References**


1) Alexandrov LB, Nik-Zainal S, Wedge DC, et al. Signatures of mutational processes in human cancer. Nature. 2013; 500(7463): 415-21.
2) Helleday T, Eshtad S, Nik-Zainal S. Mechanisms underlying mutational signatures in human cancers. Nat Rev Genet. 2014; 15(9): 585-98.
3) Burns MB, Temiz NA, Harris RS. Evidence for APOBEC3B mutagenesis in multiple human cancers. Nat Genet. 2013; 45(9): 977-83.
4) Roberts SA, Gordenin DA. Hypermutation in human cancer genomes: footprints and mechanisms. Nat Rev Cancer. 2014; 14(12): 786-800.
5) Santos-Pereira JM, Aguilera A. R loops: new modulators of genome dynamics and function. Nat Rev Genet. 2015; 16(10): 583-97.
6) Gaillard H, Aguilera A. Transcription as a threat to genome integrity. Annu Rev Biochem. 2016; 85: 291-317.
7) Crackower MA, Scherer SW, Rommens JM, et al. Characterization of the split hand/split foot malformation locus SHFM1 at 7q21.3-q22.1 and analysis of a candidate gene for its expression during limb development. Hum Mol Genet. 1996; 5(5): 571-9.
8) Birnbaum RY, Everman DB, Murphy KK, et al. Functional characterization of tissue-specific enhancers in the DLX5/6 locus. Hum Mol Genet. 2012; 21(22): 4930-8.
9) Sowińska-Seidler A, Socha M, Jamsheer A. Split-hand/foot malformation - molecular cause and implications in genetic counseling. J Appl Genet. 2014; 55(1): 105-15.
10) Jäntti J, Lahdenranta J, Olkkonen VM, et al. SEM1, a homologue of the split hand/split foot malformation candidate gene Dss1, regulates exocytosis and pseudohyphal differentiation in yeast. Proc Natl Acad Sci U S A. 1999; 96(3): 909-14.
11) Tomko RJ Jr, Hochstrasser M. The intrinsically disordered Sem1 protein functions as a molecular tether during proteasome lid biogenesis. Mol Cell. 2014; 53: 433-43.
12) Kragelund BB, Schenstrøm SM, Rebula CA, et al. DSS1/Sem1, a multifunctional and intrinsically disordered protein. Trends Biochem Sci 2016; 41(5): 446-59.
13) Marston NJ, Richards WJ, Hughes D, et al. Interaction between the product of the breast cancer susceptibility gene BRCA2 and DSS1, a protein functionally conserved from yeast to mammals. Mol Cell Biol. 1999; 19(7): 4633-42.
14) Sone T, Saeki Y, Toh-e A, et al. Sem1p is a novel subunit of the 26 S proteasome from Saccharomyces cerevisiae. J Biol Chem. 2004; 279(27): 28807-16.
15) Paraskevopoulos K, Kriegenburg F, Tatham MH, et al. Dss1 is a 26S proteasome ubiquitin receptor. Mol Cell. 2014; 56(3): 453-61.
16) Jani D, Lutz S, Hurt E, et al. Functional and structural characterization of the mammalian TREX-2 complex that links transcription with nuclear messenger RNA export. Nucleic Acids Res. 2012; 40(10): 4562-73.
17) Ellisdon AM, Dimitrova L, Hurt E, et al. Structural basis for the assembly and nucleic acid binding of the TREX-2 transcription-export complex. Nat Struct Mol Biol. 2012; 19(3): 328-36.
18) Sakai Y, Phimsen S, Okada S, et al. The critical role of germinal center-associated nuclear protein in cell biology, immunohematology, and hematolymphoid oncogenesis. Exp Hematol. 2020; 90: 30-8.
19) Sakai Y, Kuwahara K. Carcinogenesis caused by transcription-coupled DNA damage through GANP and other components of the TREX-2 complex. Pathol Int. 2024; 74(3): 103-18.
20) Crespo R, Ne E, Reinders J, et al. PCID2 dysregulates transcription and viral RNA processing to promote HIV-1 latency. iScience. 2024; 27(3): 109152.
21) Clarke BP, Gao S, Mei M, et al. Structural mechanism of DDX39B regulation by human TREX-2 and a related complex in mRNP remodeling. Nat Commun. 2025; 16(1): 5471.
22) Rezano A, Gondo N, Sakai Y, et al. Tumorigenesis caused by aberrant expression of GANP, a central component in the mammalian TREX-2 complex-lessons from transcription-coupled DNA damages. Int J Mol Sci. 2024; 25(24): 13612.
23) Li J, Zou C, Bai Y, et al. DSS1 is required for the stability of BRCA2. Oncogene. 2006; 25(8): 1186-94.
24) Zhao W, Vaithiyalingam S, San Filippo J, et al. Promotion of BRCA2-Dependent Homologous Recombination by DSS1 via RPA Targeting and DNA Mimicry. Mol Cell. 2015; 59(2): 176-87.
25) Huang Y, Li W, Foo T, et al. DSS1 restrains BRCA2's engagement with dsDNA for homologous recombination, replication fork protection, and R-loop homeostasis. Nat Commun. 2024; 15(1): 7081.
26) Faza MB, Kemmler S, Jimeno S, et al. Sem1 is a functional component of the nuclear pore complex-associated messenger RNA export machinery. J Cell Biol 2009; 184(6): 833-46.
27) Rezano A, Kuwahara K, Yamamoto-Ibusuki M, et al. Breast cancers with high DSS1 expression that potentially maintains BRCA2 stability have poor prognosis in the relapse-free survival. BMC Cancer. 2013; 13: 562.
28) Gondo N, Sakai Y, Zhang Z, et al. Increased chemosensitivity via BRCA2-independent DNA damage in DSS1- and PCID2-depleted breast carcinomas. Lab Invest. 2021; 101(8): 1048-59.
29) Chen X, Liu Q, Wu J, et al. DSS1 inhibits autophagy to acti-

vate epithelial-mesenchymal transition in a pro-metastatic niche of renal cell carcinoma. Nat Commun. 2025; 16(1): 6769.

30) Funakoshi M, Li X, Velichutina I, et al. Sem1, the yeast ortholog of a human BRCA2-binding protein, is a component of the proteasome regulatory particle that enhances proteasome stability. J Cell Sci. 2004; 117(Pt 26): 6447-54.

31) Schenstrøm SM, Rebula CA, Tatham MH, et al. Expanded Interactome of the Intrinsically Disordered Protein Dss1. Cell Rep. 2018; 25(4): 862-70.

32) Ruidiaz SF, Dreier JE, Hartmann-Petersen R, et al. The disordered PCI-binding human proteins CSNAP and DSS1 have diverged in structure and function. Protein Sci. 2021; 30(10): 2069-82.

33) Jacobsen NL, Bloch M, Millard PS, et al. Phosphorylation of Schizosaccharomyces pombe Dss1 mediates direct binding to the ubiquitin-ligase Dma1 in vitro. Protein Sci. 2023; 32(9): e4733.

34) Stefanovie B, Hengel SR, Mlcouskova J, et al. DSS1 interacts with and stimulates RAD52 to promote the repair of DSBs. Nucleic Acids Res. 2020; 48(2): 694-708.

35) Kuang Z, Ke J, Hong J, et al. Structural assembly of the nucleic-acid-binding Thp3-Csn12-Sem1 complex functioning in mRNA splicing. Nucleic Acids Res 2022; 50(15): 8882-97.

36) Fischer T, Strässer K, Rácz A, et al. The mRNA export machinery requires the novel Sac3p-Thp1p complex to dock at the nucleoplasmic entrance of the nuclear pores. EMBO J. 2002; 21(21): 5843-52.

37) Umlauf D, Bonnet J, Waharte F, et al. The human TREX-2 complex is stably associated with the nuclear pore basket. J Cell Sci. 2013; 126(Pt 12): 2656-67.

38) Schneider M, Hellerschmied D, Schubert T, et al. The nuclear pore-associated TREX-2 complex employs mediator to regulate gene expression. Cell. 2015; 162(5): 1016-28.

39) González-Aguilera C, Tous C, Gómez-González B, et al. The THP1-SAC3-SUS1-CDC31 complex works in transcription elongation-mRNA export preventing RNA-mediated genome instability. Mol Biol Cell. 2008; 19(10): 4310-8.

40) Strässer K, Bassler J, Hurt E. Binding of the Mex67p/Mtr2p heterodimer to FXFG, GLFG, and FG repeat nucleoporins is essential for nuclear mRNA export. J Cell Biol. 2000; 150(4): 695-706.

41) Herold A, Suyama M, Rodrigues JP, et al. TAP (NXF1) belongs to a multigene family of putative RNA export factors with a conserved modular architecture. Mol Cell Biol. 2000; 20(23): 8996-9008.

42) Braun IC, Herold A, Rode M, et al. Nuclear export of mRNA by TAP/NXF1 requires two nucleoporin-binding sites but not p15. Mol Cell Biol. 2002; 22(15): 5405-18.

43) Ginno PA, Lott PL, Christensen HC, et al. R-loop formation is a distinctive characteristic of unmethylated human CpG island promoters. Mol Cell. 2012; 45(6): 814-25.

44) Chen PB, Chen HV, Acharya D, et al. R loops regulate promoter-proximal chromatin architecture and cellular differentiation. Nat Struct Mol Biol. 2015; 22(12): 999-1007.

45) Sanz LA, Hartono SR, Lim YW, et al. Prevalent, dynamic, and conserved R-loop structures associate with specific epigenomic signatures in mammals. Mol Cell. 2016; 63(1): 167-78.

46) Skourti-Stathaki K, Kamieniarz-Gdula K, Proudfoot NJ. R-loops induce repressive chromatin marks over mammalian gene terminators. Nature. 2014; 516(7531): 436-9.

47) Skourti-Stathaki K, Proudfoot NJ, Gromak N. Human senataxin resolves RNA/DNA hybrids formed at transcriptional pause sites to promote Xrn2-dependent termination. Mol Cell. 2011; 42(6): 794-805.

48) Yu K, Chedin F, Hsieh CL, et al. R-loops at immunoglobulin class switch regions in the chromosomes of stimulated B cells. Nat Immunol. 2003; 4(5): 442-51.

49) Ramiro AR, Stavropoulos P, Jankovic M, et al. Transcription enhances AID-mediated cytidine deamination by exposing single-stranded DNA on the nontemplate strand. Nat Immunol. 2003; 4(5): 452-6.

50) Chaudhuri J, Tian M, Khuong C, et al. Transcription-targeted DNA deamination by the AID antibody diversification enzyme. Nature 2003; 422(6933): 726-30.

51) Muramatsu M, Kinoshita K, Fagarasan S, et al. Class switch recombination and hypermutation require activation-induced cytidine deaminase (AID), a potential RNA editing enzyme. Cell. 2000; 102(5): 553-63.

52) Xu B, Clayton DA. A persistent RNA-DNA hybrid is formed during transcription at a phylogenetically conserved mitochondrial DNA sequence. Mol Cell Biol 1995; 15(1): 580-9.

53) Xu B, Clayton DA. RNA-DNA hybrid formation at the human mitochondrial heavy-strand origin ceases at replication start sites: an implication for RNA-DNA hybrids serving as primers. EMBO J. 1996; 15(12): 3135-43.

54) Lee DY, Clayton DA. Properties of a primer RNA-DNA hybrid at the mouse mitochondrial DNA leading-strand origin of replication. J Biol Chem 1996; 271(39): 24262-9.

55) Lee DY, Clayton DA. Initiation of mitochondrial DNA replication by transcription and R-loop processing. J Biol Chem. 1998; 273(46): 30614-21.

56) Hamperl S, Bocek MJ, Saldivar JC, et al. Transcription-replication conflict orientation modulates R-loop levels and activates distinct DNA damage responses. Cell. 2017; 170(4): 774-86.e19.

57) Stoy H, Zwicky K, Kuster D, et al. Direct visualization of transcription-replication conflicts reveals post-replicative DNA:RNA hybrids. Nat Struct Mol Biol 2023; 30(3): 348-

59.
58) Promonet A, Padioleau I, Liu Y, et al. Topoisomerase 1 prevents replication stress at R-loop-enriched transcription termination sites. Nat Commun 2020; 11 (1) : 3940.
59) Hamperl S, Cimprich KA. The contribution of co-transcriptional RNA:DNA hybrid structures to DNA damage and genome instability. DNA Repair (Amst) . 2014; 19: 84-94.
60) Crossley MP, Bocek M, Cimprich KA. R-loops as cellular regulators and genomic threats. Mol Cell. 2019; 73 (3) : 398-411.
61) García-Muse T, Aguilera A. R loops: from physiological to pathological roles. Cell. 2019; 179 (3) : 604-18.
62) Allison DF, Wang GG. R-loops: formation, function, and relevance to cell stress. Cell Stress. 2019; 3 (2) : 38-46.
63) Sollier J, Cimprich KA. Breaking bad: R-loops and genome integrity. Trends Cell Biol. 2015; 25 (9) : 514-22.
64) Sollier J, Stork CT, García-Rubio ML, et al. Transcription-coupled nucleotide excision repair factors promote R-loop-induced genome instability. Mol Cell. 2014; 56 (6) : 777-85.
65) Li F, Zafar A, Luo L, et al. R-loops in genome instability and cancer. Cancers (Basel) . 2023; 15 (20) : 4986.
66) Dieppois G, Iglesias N, Stutz F. Cotranscriptional recruitment to the mRNA export receptor Mex67p contributes to nuclear pore anchoring of activated genes. Mol Cell Biol. 2006; 26 (21) : 7858-70.
67) Wickramasinghe VO, McMurtrie PI, Mills AD, et al. mRNA export from mammalian cell nuclei is dependent on GANP. Curr Biol. 2010; 20 (1) : 25-31.
68) Wickramasinghe VO, Andrews R, Ellis P, et al. Selective nuclear export of specific classes of mRNA from mammalian nuclei is promoted by GANP. Nucleic Acids Res. 2014; 42 (8) : 5059-71.
69) Tian L, Liu L, Yoon Y, et al. LENG8 mediates RNA nuclear retention and degradation in eukaryotes. Mol Cell. 2026; 86 (8) : 1478-94.e8.
70) Jensen RB, Carreira A, Kowalczykowski SC. Purified human BRCA2 stimulates RAD51-mediated recombination. Nature. 2010; 467 (7316) : 678-83.
71) Carreira A, Hilario J, Amitani I, et al. The BRC repeats of BRCA2 modulate the DNA-binding selectivity of RAD51. Cell. 2009; 136 (6) : 1032-43.
72) Schlacher K, Christ N, Siaud N, et al. Double-strand break repair-independent role for BRCA2 in blocking stalled replication fork degradation by MRE11. Cell. 2011; 145 (4) : 529-42.
73) Yang H, Jeffrey PD, Miller J, et al. BRCA2 function in DNA binding and recombination from a BRCA2-DSS1-ssDNA structure. Science. 2002; 297 (5588) : 1837-48.
74) Kristensen CN, Bystol KM, Li B, et al. Depletion of DSS1 protein disables homologous recombinational repair in human cells. Mutat Res. 2010; 694 (1-2) : 60-4.
75) Lee M, Shorthouse D, Mahen R, et al. Cancer-causing BRCA2 missense mutations disrupt an intracellular protein assembly mechanism to disable genome maintenance. Nucleic Acids Res. 2021; 49 (10) : 5588-604.
76) Gudmundsdottir K, Lord CJ, Witt E, et al. DSS1 is required for RAD51 focus formation and genomic stability in mammalian cells. EMBO Rep. 2004; 5 (10) : 989-93.
77) Le HP, Ma X, Vaquero J, et al. DSS1 and ssDNA regulate oligomerization of BRCA2. Nucleic Acids Res. 2020; 48 (14) : 7818-33.
78) Shivji MKK, Renaudin X, Williams ÇH, et al. BRCA2 regulates transcription elongation by RNA polymerase II to prevent R-loop accumulation. Cell Rep. 2018; 22 (4) : 1031-9.
79) Sessa G, Gómez-González B, Silva S, et al. BRCA2 promotes DNA-RNA hybrid resolution by DDX5 helicase at DNA breaks to facilitate their repair. EMBO J. 2021; 40 (7) : e106018.
80) D'Alessandro G, Whelan DR, Howard SM, et al. BRCA2 controls DNA:RNA hybrid level at DSBs by mediating RNase H2 recruitment. Nat Commun. 2018; 9 (1) : 5376.
81) Bhatia V, Barroso SI, García-Rubio ML, et al. BRCA2 prevents R-loop accumulation and associates with TREX-2 mRNA export factor PCID2. Nature. 2014; 511 (7509) : 362-5.
82) González-Martín A, Pothuri B, Vergote I, et al. Niraparib in Patients with Newly Diagnosed Advanced Ovarian Cancer. N Engl J Med. 2019; 381 (25) : 2391-402.
83) Agarwal N, Azad AA, Carles J, et al. Talazoparib plus enzalutamide in men with metastatic castration-resistant prostate cancer: final overall survival results from the randomised, placebo-controlled, phase 3 TALAPRO-2 trial. Lancet. 2025; 406 (10502) : 447-60.