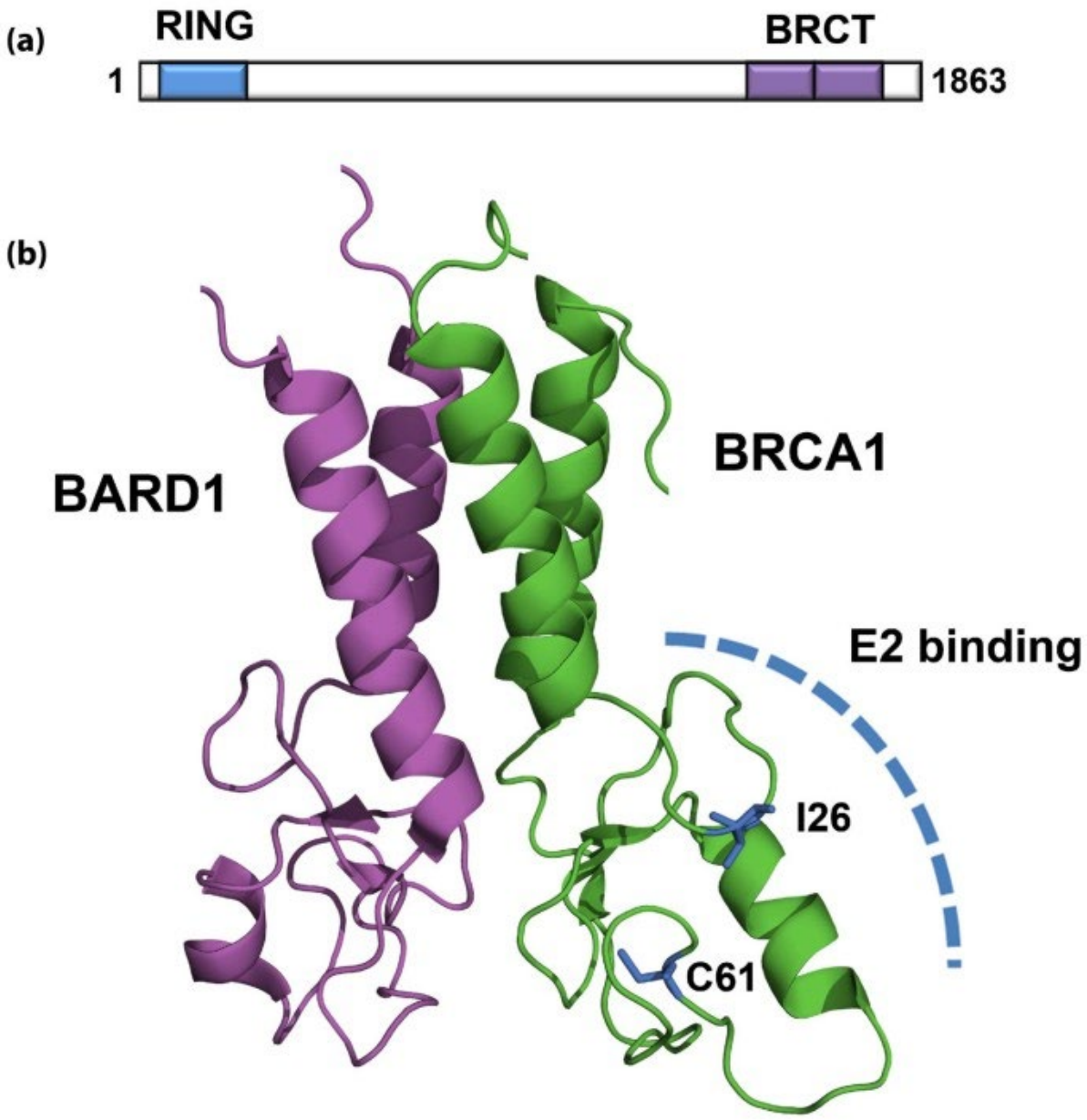
Hereditary cancer. How Breast cancer is caused by inherited BRCA1 gene mutations



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Abstract

Hereditary Breast and Ovarian Cancer (HBOC) typically arises from mutations inherited in the germline. BRCA1 and BRCA2 mutations are predominant contributors to HBOC, accounting for the majority of cases. These mutations are associated with 10% of ovarian cancer cases and 3-5% of breast cancer cases [4].

BRCA1 has crucial domains, including a zinc-binding RING domain, forming an ubiquitin ligase complex (E3) with BARD1. Ubiquitination, activated by ATP, removes damaged proteins and regulating cell functions. The RING finger enables substrate and ubiquitination-conjugating enzyme (E2) interaction, essential for BRCA1's function in tumor suppression. [4,5] The structure of the BRCA1/BARD1 heterodimer has revealed valuable insights into the function of the BRCA1 RING domain. Mutations in cysteine residues coordinating Zn2+ atoms in Site I and Site II can significantly impact function and increase cancer risk. Mutations in these sites alter the RING domain's folding, reduce Zn2+ binding, and decrease ubiquitin ligase activity and BRCA1/BARD1 interactions. These mutations are associated with an elevated risk of breast cancer [4].

BRCA1 proteins are vital for preserving genomic integrity by facilitating accurate DNA repair via homologous recombination. When BRCA1 function is lost, non-tumorigenic cells can transform into cancer stem cells (CSCs). Non-functional BRCA1 and mismatch repair genes contribute significantly to familial cancers. Defects in homologous recombination and mismatch repair pathways provide treatment options. Approximately 50% of hereditary cancer cases involve genes responsible for DNA repair and genome maintenance. In breast cancer, genetic factors like BRCA1 mutations increase the risk, although most genetic drivers remain unidentified. A deeper understanding of these risks can aid in personalized cancer prevention and screening strategies, as HBOC is often associated with BRCA1 and BRCA2 mutations, leading to elevated breast and ovarian cancer risks along with reduced risks of other cancer types [9,10,11]. The goal of this review is to underline how inherited BRCA1 gene mutations cause breast cancer.

Introduction

Cancer is a big cause of globally death, 1 in 6 deaths. Cancer can appear in each organ when cells grow uncontrollably and destructively; as a result, it spreads and attacks other body parts. This process is referred to as metastasis. Widespread metastases is the primary cause of death from cancer [1]. In about 5 to 10 percent of cancers is found to be hereditary. These cancers are often referred to as inherited cancers, what actually is inherited from the parents is the mutated gene that can lead to cancer [2]. The genetic changes that can lead to cancer mainly affect three types of genes; proto-oncogenes, tumor suppressor genes and DNA repair genes. Cell growth and division are controlled by tumor suppressor genes. Uncontrolled cell division can take place in cells when there are certain alterations in the tumor suppressor genes [3].
BRCA1 is a tumor suppressor gene, which can cause HBOC syndrome. HBOC syndrome stands for Hereditary Breast and Ovarian Cancer. HBOC is usually caused by a mutation in the inherited germline. With the majority of the HBOC cases the BRCA1 and BRCA2 mutations are responsible for cancer. 10 percent of ovarian cancer cases and 3-5% of breast cancer cases, are associated with BRCA1 or BRCA2 mutations. Women have a 70-80 % risk of developing breast cancer and a 50% risk of developing ovarian cancer, in the presence of a BRCA1 mutation. There is a 50-60 % risk of developing breast cancer and a 30% risk of developing ovarian cancer, when women are carrying a BRCA2 mutation [4].

The RING domain and the function of ubiquitination

Breast and ovarian cancer were first linked to the tumor suppressor gene BRCA1 in 1994. Over the years there are a number of studies that demonstrate processes regulating tumor development involvement of the role of the BRCA genes [5]. The BRCA1 gene is located on chromosome 17th (17q21.31). It is 126.033 kb, it consists of 24 exons and 1,863 amino acids [6,11]. BRCA1 consists of several domains that are essential for its multiple functions. For the interaction of BRCA1 and BARD1 (BRCA1 Associated RING Domain protein 1) the N-terminal region carries a zinc-binding finger domain RING (Really Interesting New Gene) and the formation of E3 ubiquitin ligase complex. These domains are essential for the interactions. At the C-terminus, two BRCT (BRCA1 C-terminal) domains that bind to phosphopeptides facilitate the interaction between BRCA1 and crucial partner proteins like CtIP (C-terminal binding protein 1 (CtBP1) interacting protein), BRCA1 A Complex Subunit (ABRAXAS), and BRCA1 interacting protein C-terminal helicase 1 (BRIP1/FACJ). The central segment of BRCA1, encoded by exons 11-13, frequently exhibits mutations in breast cancer patients. This region comprises two nuclear localization signals (NLS) and a coiled coil domain essential for interaction with BRCA2 through partner and localizer of BRCA2 (PALB2) [5].

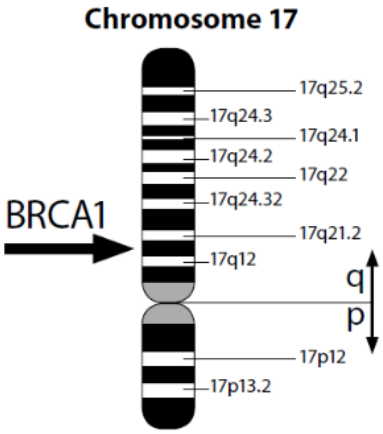

Figure 1: Chromosome 17 with the BRCA1 gene. [12]

The E3 ubiquitin ligase complex is an domain that is involved with the interaction between BRCA1 and BARD1. Ubiquitination removes toxic and damaged proteins and identifies the protein substrate that needs to undergo ubiquitination. It operates in cell death, cell signaling and cell cycle regulation. Ubiquitination-activating enzyme is the start of ubiquitination, it activates ubiquitination with adenosine triphosphate (ATP). A thioester bond is formed between the active site on the cysteine residue on E1 and the C-terminal of the carboxyl group of ubiquitination. With this formation, the ubiquitination is transferred to de ubiquitination-conjugating enzyme (E2). Through a catalytic reaction between ubiquitination ligase (E3) and ubiquitination.

Ubiquitination transfer from E2 to E3. Within the next step of ubiquitination an isopeptide bond between the carboxyl group on the C-terminal of ubiquitination and the lysine ε-amino group of the substrate is formed. This bond is formed through a transfer from the ubiquitination to the lysine residue on the specific substrate, that will bound to E3. Eventually, an ubiquitination chain forms on the substrate. Proteasomal degradation at the 26S proteasome activates when an ubiquitination chain is formed, a signal is sent, the ubiquitination and along with the substrate (Figure 2). The RING finger allows a protein substrate and E2 enzyme to work in conjunction with it, this is known as an adapter-type ligase. [8] The mutations that effect the RING E3 ligase function and BRCA1/BARD1 suggest that the ubiquitin ligase activity of BRCA1 is essential for its tumor suppressor function [4].

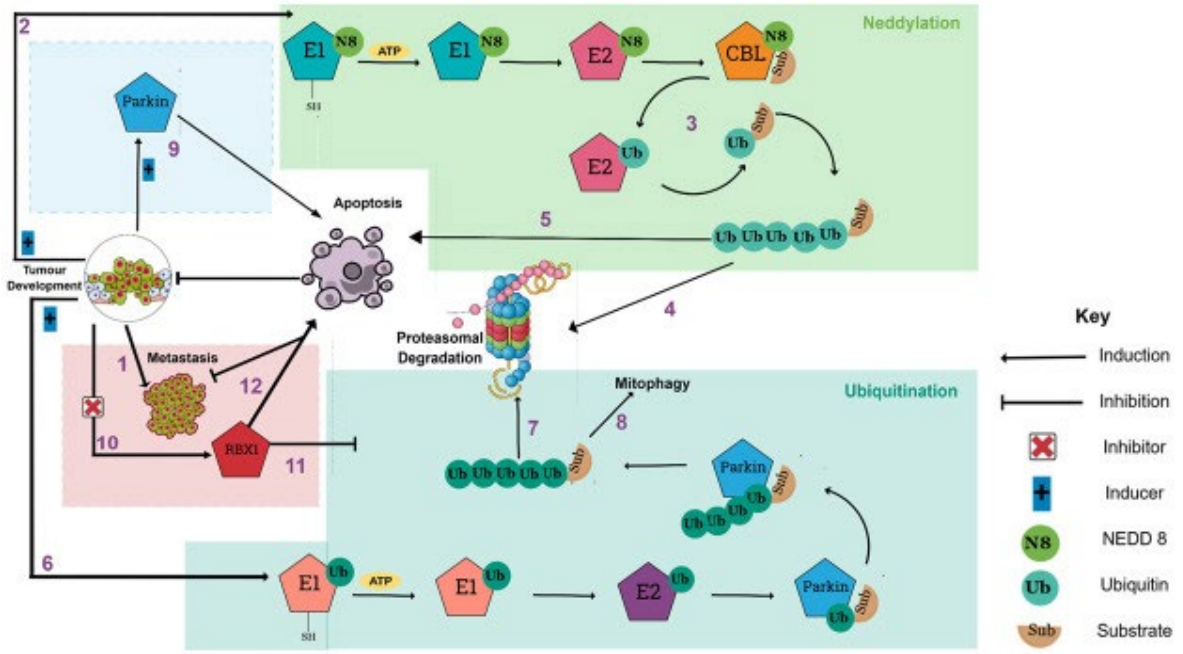

Figure 2: A model representing the various targets of RING finger proteins for use in cancer therapeutics.
(1) Tumor development can lead directly to metastasis. Metastasis can occur due to overexpression of certain RING finger proteins such as RBX1; (2) Inducing of certain RING finger proteins such as CBL will lead to neddylation of the proteins that interact with them; (3) Neddylation causes a conformational change to occur and recruit ubiquitin; (4) And ultimately lead to proteasomal degradation; (5) Or can lead to apoptosis, which leads to the inhibition of tumor development; (6) Inducing of RING finger proteins such as Parkin; (7) This will lead to ubiquitination of the proteins that interact with them and ultimately lead to proteasomal degradation; (8) Or can also lead to mitophagy through a joint autophagic machinery; (9) The induction of Parkin can also lead to apoptosis; (10) Inhibition of RING finger proteins such as RBX1; (11) Will cause the inhibition of E3 activity such as ubiquitination; (12) But does lead to apoptosis and thus inhibiting metastasis. [8]

BRCA1/ BARD1 RING and it's cancer related mutations

The BRCA1 and BARD1 from an archetypal RING-type E3 ligase. A large heterodimer is formed via their N-terminal RING domains. Humans have more than 600 of this large E3 class. Substrate binds simultaneously and an ubiquitination-conjugation (E2) enzyme that holds activated ubiquitination (see Figure 3,4). It is imperative for the structure to include distinctive regions for both substrate and E2 binding. RING domains specifically bind to E2s, potentially leaving the rest of the complex available for substrate binding. Notably, within the human proteome, only BRCA1 and BARD1 possess both an N-terminal RING domain and C-terminal tandem BRCT (BRCA1 C-terminal) domains, both characterized by significant intrinsically disordered content indicative of molecular scaffolds (see Figure 3). Additionally, BARD1 features an ankyrin-repeat domain (Ank) adjacent to its BRCT domain, typically associated with protein-protein interactions [7].

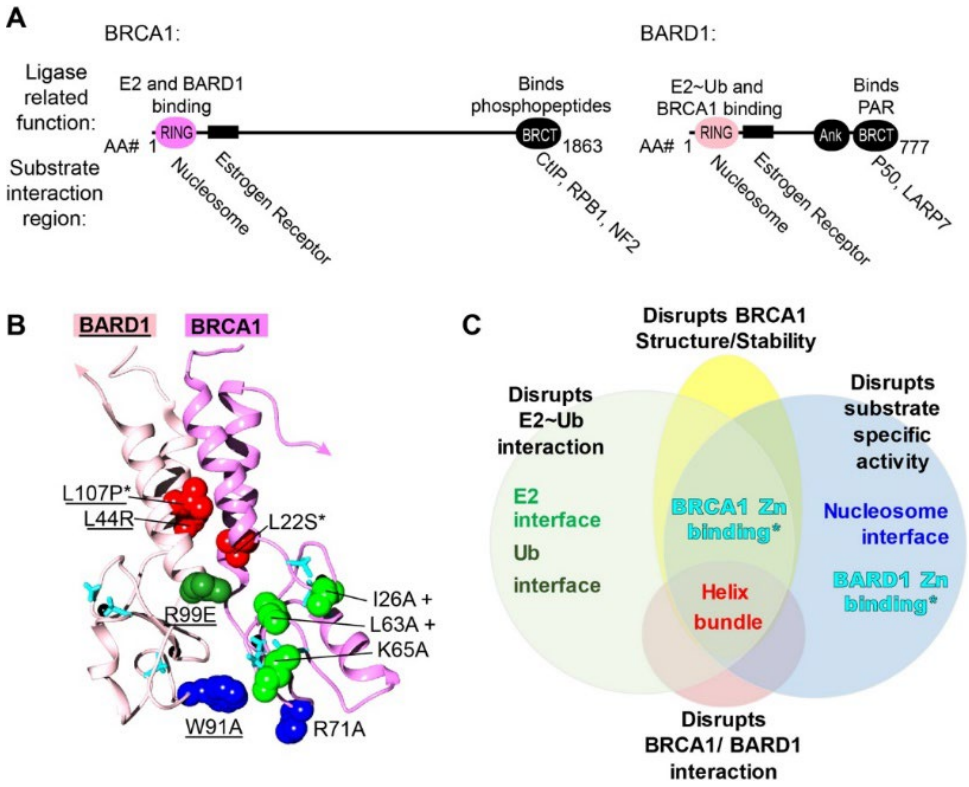

Figure 3: An overview of the features of the BRCA1/BARD1 Ubiquitin ligase.
A) The domain structure and function of BRCA1 and BARD1 domains. Folded domains of BRCA1 and BARD1 are depicted with ovals and their corresponding domain names: really interesting new gene (RING), BRCA1 C-terminal (BRCT), and ankyrin repeats (ARD). Substrate binding domains within the intrinsically disordered regions are represented with rectangles. Domain functions related to E3 ligase activity are listed above domains. Substrates are listed below the region of protein with which they interact. B) The solution structure of RING domains from BRCA1 and BARD1 (PDB 1JM7) are shown in magenta and pink respectively. The sidechains at mutation sites used to study the structure/function relationship are shown in spheres or sticks: BARD1 mutations are underlined, cancer-associated mutations are either labeled with an asterisks or in the case of zinc-coordinating mutation sites depicted in cyan sticks (BRCA1 C24R, C39S/R/Y/W, C44S/Y/F, C47S/Y/F, C61G, C64R/Y/W and BARD1 C53W, C71Y, and C83R). The colors of mutation site sidechains correspond with their functions in Panel C. C) Mutations are categorized in a Venn diagram according to the properties and functions they affect. [7]

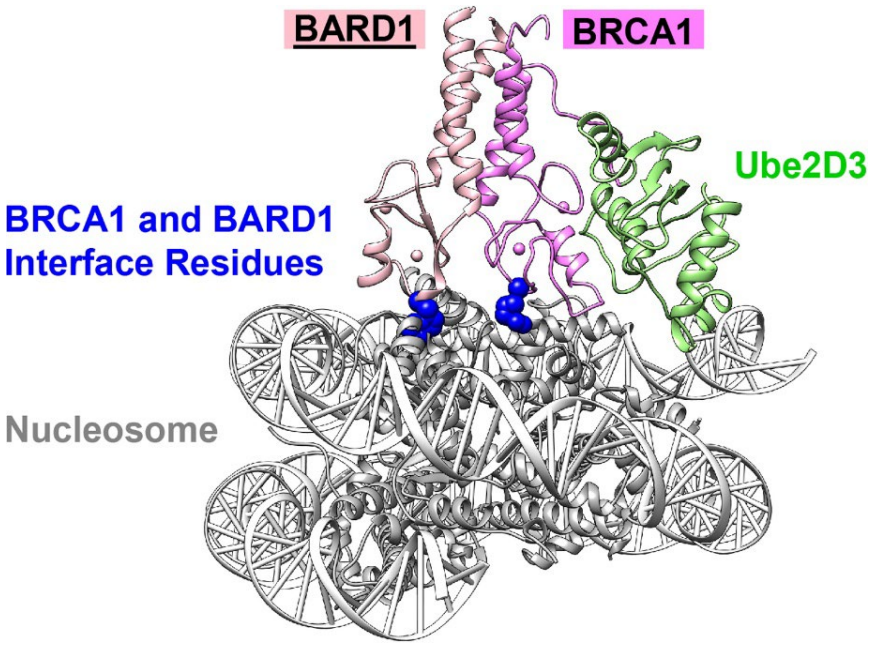

Figure 4: BRCA1 and BARD1 bind the nucleosome substrate and E2 simultaneously.
The cryo-EM structure (PDB 7JZV) reveals that both BRCA1 and BARD1 RING domains (magenta and pink, respectively) contain critical residues (BRCA1 Arg 71 and BARD1 Trp 91, blue) that contact the nucleosome surface (gray). The E2, Ube2D3 (green), binds to a distinct interface on BRCA1 allowing the heterodimer to coordinate substrate and E2 simultaneously. The complex allows for transfer of ubiquitin from the E2 onto the dynamic C-terminal end of H2A (unresolved in the structure). [7]

The structure of the BRCA1/BARD1 heterodimer gained information about the structure-function of the BRCA1 RING domain. The heterodimer of the BRCA1/BARD1 RING is stabilized by the four helix bundle, and it creates a large buried hydrophobic region. The orientation of the BRCA1 RING finger maintain the orientation of the RING finger duo to the flanking alpha-helices. The ligase activity of BRCA1 increases because of the interaction between BRCA1 and BARD1. Therefore the nuclear export sequence, located on the C-terminal helix BRCA1 And BARD1 RING domain is inactivated. Besides the interactions between BRCA1 and BARD1 of the four helix bundle, a few inter-RING interactions may occur as well. The RING finger structure is stabilized with two Zn2+ atoms. This is coordinated by Zn2+ binding loops named Site I and Site II. Site I consist of four cysteine residues and Site II consists of three cysteine and one histidine residue [4].
Mutation of cysteine residues coordinating Zn2+ atoms is clinically significant, leading to altered function and an increased cancer risk. Mutating residues in Site I result in RING domain folding changes. Site II residue mutations cause structural alterations and reduced Zn2+ binding, with subsequent studies revealing decreased ubiquitin ligase activity and BRCA1/BARD1 interactions. These findings suggest that mutations in Site I and Site II impact BRCA1 ubiquitin ligase activity by affecting heterodimerization or BRCA1 interaction. The abundance of RING domain mutations associated with elevated breast cancer risk, coupled with the influence of chemotherapeutic drugs on RING domain activity, underscores the crucial role the RING domain plays in tumor suppression [4].

Inheritance of mutated BRCA1 gene

BRCA1 proteins are crucial for maintaining genomic integrity by ensuring accurate DNA repair through homologous recombination. Non-tumorigenic cells can transform into cancer stem cells (CSCS), and drive the rumor evolution, when BRCA losses its function. Recent research has shown that cancer cells in the same tumor can vary significantly in their ability to initiate tumors. CSCs initiate tumor growth because of the unique capacity for long-term self-renewal and differentiation of various tumor cell types. There is a chance that CSCs acts as a driving force behind cancer evolution. Given their high genomic instability in tumor cell and the extensive self-renewal and clonogenic potential [11].

Significant contributors to familial cancers are non-functional BRCA1 or related mismatch repair (MMR) genes. On the other hand, DNA repair defects homologous recombination (HR) and MMR pathways offer treatment options. About 50 percent of the known hereditary cancer drivers involve genes responsible for DNA repair and genome maintenance. For instance, in breast cancer, women with close relatives affected by the disease face a doubled risk, but only 15%-25% of hereditary breast/ovarian cancer patients carry the BRCA1 mutation. The majority of genetic drivers remain unidentified. Limited knowledge of these genetic risk factors hinders effective cancer screening and prevention strategies. A deeper understanding of these risks can help identify patients who need more aggressive risk reduction measures, while minimizing those without predisposing genes from unnecessary interventions [10].

The most common malignant cancer in a woman is breast cancer. Within these cases 10% of these are hereditary. In almost half of these hereditary cases, cancer predisposition is linked to genetic variants in BRCA1 and BRCA2 genes. The hereditary breast and ovarian cancer (HBOC) syndrome shows an autosomal dominant inheritance pattern with high but not complete penetrance and is associated with BRCA1 and BRCA2. Carriers of germline BRCA1/BRCA2 pathogenic variants have an increased risk of developing breast (BRCA1: 55-72%, BRCA2: 45-69%) and ovarian cancer (BRCA1: 39-44%, BRCA2: 11-17%). Additionally, carriers face a reduced risk of other cancer types such as prostate, pancreatic, and melanoma, with BRCA2 mutation carriers having higher risks for these cancers [9].

Conclusion

In conclusion, hereditary breast and ovarian cancer (HBOC) is primarily attributed to mutations in the BRCA1 gene, which plays a crucial role in preserving genomic integrity and facilitating DNA repair through homologous recombination. Mutations in BRCA1, particularly in the RING domain, can lead to an increased risk of breast and ovarian cancer. These mutations impair the ubiquitin ligase activity of BRCA1, impacting its tumor-suppressing function. The identification of these genetic risk factors has been instrumental in understanding the hereditary nature of some cancers and can guide personalized cancer prevention and screening strategies [4,5,10].

Furthermore, the study of BRCA1 and related mismatch repair genes has revealed their significant role in familial cancers. DNA repair defects in homologous recombination and mismatch repair pathways offer potential treatment options. Despite the importance of these genetic drivers in hereditary cancer cases, the majority of them remain unidentified. This knowledge gap hinders effective cancer screening and prevention strategies, highlighting the need for a deeper understanding of these risks to identify patients who require more aggressive risk reduction measures while sparing those without predisposing genes from unnecessary interventions [10].

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