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CASE REPORT

Correlation Between BRCA Mutation And Lymphatic Node Involvement In Advanced Stage Ovarian Cancer

Ade Putra[1,*], Syamel Muhammad[2], Puja Agung Antonius[2]

1. *Departement of Obstetrics and Gynecology, Medical Faculty of Andalas University, Padang, West Sumatera, Indonesia*
2. *Gynecologic Oncology Division of Departement of Obstetrics and Gynecology, Medical Faculty of Andalas University, Padang, West Sumatera, Indonesia*

Corresponding Author: *Ade Putra, adep935@gmail.com*

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Abstract

Background *Gene mutations affecting the Breast Cancer-1 (BRCA1) and Breast Cancer-2 (BRCA2) genes are linked to ovarian and breast cancer which connected with a risk of ovarian cancer of 50% and 20%, respectively. High-grade serous ovarian cancer (HGSC) frequently has lymph node involvement. According to lymphatic drainage routes, the most prevalent site of nodal metastases is in the aortic area. A case of Cancer of Unknown Primary (CUP) with abdominal lymph node enlargement and high carbohydrate antigen 125 levels discovered a germline Breast Cancer 1 (BRCA1) p.L63* variant by a family history inquiry and BRCA analysis, indicating hereditary breast and ovarian cancer syndrome. Therefore we wanted to assess whether BRCA gene status correlates with lymph node involvement in advanced stage ovarian cancer.*

Methods *This was an analytic-observational study with cross sectional study design. The samples in this study were patients diagnosed with advanced stage ovarian cancer who were treated at M Djamil Padang General Hospital, Ibnu Sina Hospital Padang, and Ahmad Mochtar Hospital Bukittinggi by using data from BRCA somatic test results and anatomical pathology examination results. Data were being tested statistically with Chi-square/ Fischer test.*

Result *this study showed 1 patient (3,1%) with positive pathogenic mutation detected in BRCA1 gen with lymphatic node involvement, and 8 (25%) patients without lymphatic node involvement. This study also showed 3 patients (9,4%) with negative pathogenic mutation detected in BRCA1 gen with lymphatic node involvement, and 20 (62,5%) patients without lymphatic node involvement. There is no significant correlation between BRCA mutation and lymphatic node involvement in advanced stage ovarian cancer (p=1,0).*

Conclusion *BRCA mutation has no significant correlation with lymphatic node involvement in advanced stage ovarian cancer.*

Keywords: *BRCA mutation, advanced stage ovarian cancer, lymphatic node.*

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INTRODUCTION

In females, ovarian cancer is the fifth most common cause of malignancy-associated mortality.[1] With an incidence of 7.47%, it is one of the most common gynecologic malignancies in Indonesia. The incidence rises with age, reaching its highest in the 50–54 age range. With a 5-year survival rate of fewer than 30%, stage IIIC had the highest occurrence and the majority of patients (>75%) presented with stage III to IVB.[2–4] The most common subtype of mucinous carcinoma (25.5%) was found in the histology, which showed 88.3% epithelial ovarian cancer.[5]

There isn't currently a recognized etiology that could account for the entire illness. A genetic component is one of the most significant risk factors for ovarian cancer. Ovarian cancer cases with a genetic susceptibility account for 10-15% of cases. Gene mutations affecting the Breast Cancer-1 (BRCA1) and Breast Cancer-2 (BRCA2) genes are linked to ovarian and breast cancer. The genes with the most impact on the incidence of ovarian cancer to date are BRCA1 and BRCA2, which were initially identified in 1994 and 1995. BRCA2 is found on chromosome 13q, and BRCA1 is an oncosuppressor gene on chromosome 17q21. These genes can be deleted or added, which results in an early codon termination and a shorter protein. The genes also participate in the chromatin remodeling process, hence their mutation results in uncontrolled cell proliferation. BRCA1 and BRCA2 mutations are connected with a risk of ovarian cancer of 50% and 20%, respectively.[2,6]

For the past two decades, the majority of hereditary ovarian carcinomas have been attributed to mutations in the BRCA genes. Currently, as a result of next generation sequencing, at least 16 more genes associated with hereditary ovarian cancer have been found, leading to increasing evidence of similar uncommon syndromes associated with gynecologic tumors.[7,8] KIAA0101, RAD51AP1, FAM83D, CEP55, PRC1, CKS2, CDCA5, NUSAP1, ECT2, and TRIP13 were identified as top ten hub genes; SIN3A, VDR, TCF7L2, NFYA, and FOXM1 were identified as predominant transcription factors (TFs) in advanced High-Grade Primary Tumors (HGPTs); and CEP55, PRC1, CKS2, CDCA5, and NUSAP1 were identified as potential biomarkers from hub gene clustering. Further investigation identified hsa-miR-215-5p, hsa-miR-193b-3p, and hsa-miR-192-5p as important miRNAs that target HGPT genes. Overall, our findings highlighted HGPT-associated genes, TFs, miRNAs, and pathways as potential biomarkers, presenting new paths for OC diagnostic and therapeutic approaches.[9]

High-grade serous ovarian cancer (HGSC) frequently has lymph node involvement. According to lymphatic drainage routes, the most prevalent site of nodal metastases is in the aortic area. However, pelvic lymph nodes are also affected, although inguinal metastases are less likely.[10] Extracapsular spread of lymph node metastases is a typical characteristic of lymph node-positive ovarian cancer. Extracapsular (ECG) cancer growth was linked to increased grade, lymph node ratio (LNR), and shorter overall survival. This study suggested that the presence of an ECG could be taken as a sign of tumor aggressiveness.[11]

A case of Cancer of Unknown Primary (CUP) with abdominal lymph node enlargement and high carbohydrate antigen 125 levels discovered a germline Breast Cancer 1 (BRCA1) p.L63* variant by a family history inquiry and BRCA analysis, indicating hereditary breast and ovarian cancer

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syndrome.[12] The BRCA mutational status appears not to be linked with clinical characteristics and post-salvage lymphadenectomy progression-free survival (PSL-PFS), unlike other sites of illness in reccurent ovarian cancer (ROC) patients.[13] We proposed that assessing BRCA gene status could be useful in the decision-making approach to surgery or chemotherapy in lymph node involvement in advanced-stage ovarian cancer.

METHODS

This was an analytic-observational study conducted in a cross-sectional design. The samples in this study were patients diagnosed with advanced stage ovarian cancer who were treated at M Djamil Padang General Hospital, Ibnu Sina Hospital Padang, and Ahmad Mochtar Hospital Bukittinggi. Microscopic review of the cases for confirming the diagnosis and tumor grading were assessed according to World Health Organization (WHO) Classification of Female Genital Tumors, Fifth Edition, 2020.[14]

The Formalin-Fixed Paraffin-Embedded (FFPE) tissue was sent to MedGenome Labs, India and KALGen INNOLAB, Indonesia which have been certified by College of American Pathologist (CAP). Data from BRCA somatic test results and anatomical pathology examination results were used, and the study ran from March 2021 to September 2023. Follow up time was up to eighteen months. Purposive sampling was used to choose research subjects from patients with advanced stage ovarian cancer receiving treatment at M Djamil Padang General Hospital, Ibnu Sina Hospital Padang, and Ahmad Mochtar Hospital Bukittinggi. The study involved 32 patients. Data were analyzed using SPSS ver. 27 for Machintos. Qualitative data were described as number and percentage. Chi-square (Fisher's exact) test was used to examine the relation between qualitative variables as appropriate. A p-value less than or equal to 0.05 was considered statistically significant. All tests were two tailed.

RESULT

Based on the research results, there were 32 patients consisting of 3 patients from Ibnu Sina Hospital Padang, 1 patient from Achmad Mochtar Hospital Bukittinggi and 28 patients from M. Djamil Hospital Padang. This study showed most of the patients age is 61–70 age (34.4%), followed with 31,3% patients 41–50 y.o, and 51–60 y.o., respectively. FIGO stage IIIB was dominant (37.5%) and stage IIB was 28.1%. Of the 32 patients, there were 9 patients (28%) with positive results for pathogenic mutation detected in the BRCA1 gene and 4 patients (13%) with lymphatic node involvement.

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Table 1. Distribution of the variable

VARIABEL	F (%)
Age (years)	
<40	1 (3.1)
41 – 50	10 (31.3)
51 – 60	10 (31.3)
61 – 70	11 (34.4)
FIGO stage	
IA	2 (6.3)
IC	1 (3.1)
IIA	1 (3.1)
IIB	9 (28.1)
IIIA	5 (15.6)
IIIB	12 (37.5)
IIIC	1 (3.1)
IVA	1 (3.1)
BRCA1/2 mutation	
Positive	9 (28,1)
BRCA 1 mutation	5 (55,6)
BRCA 2 mutation	3 (33,3)
Both BRCA1/2 mutation	1 (11,1)
Negative	23 (71,9)
Lymphatic Node Involvement	
Positif	4 (12,5)
Negatif	28 (87,5)

This study showed 1 patient (3,1%) with positive pathogenic mutation detected in BRCA1 gen with lymphatic node involvement, and 8 (25%) patients without lymphatic node involvement. This study also showed 3 patients (9,4%) with negative pathogenic mutation detected in BRCA1 gen with lymphatic node involvement, and 20 (62,5%) patients without lymphatic node involvement. There is no significant correlation between BRCA mutation and lymphatic node involvement in advanced stage ovarian cancer (p>0,05) (Table 2).

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Table 2. The relations between BRCA mutation and Lymphatic Node Involvement.

Variabel	Lymphatic Node Involvement, f (%)		Total	Nilai p
	Positif	Negatif		
BRCA Somatic Test Result				
Positif	1 (0)	8 (100)	9 (100)	**1,000**
Negatif	3 (77,7)	20 (22,2)	23 (100)	
Total	4 (12,5)	28 (87,5)	32 (100)	

DISCUSSION

In this study, the frequency of BRCA mutations in patients with advanced ovarian cancer is examined, and the relationship between BRCA status and lymphatic node involvement is correlated. Different studies have produced conflicting findings about the level of BRCA expression in ovarian cancer that has progressed. The average age was found to be between 61 and 70 years old, with FIGO stage IIIB. Using data from the Swedish Quality Registry for Gynecological Cancer (SQRGC), a population-based cohort study revealed that 497 women with epithelial ovarian or Fallopian tube cancer FIGO stage III or IV had an average age of 66 (30-94) years, with 66% of them falling into the 30-70 age range. Moreover, it said that 77% were categorized as FIGO stage III.[15] The median age of BRCA1 mutation was 49 (32–75) years old, while the median age of BRCA2 mutation was 54 (28–74) years old, according to Singer et al. (2019).[16] According to a study by Amin et al. (2023), 31.2% of patients with ovarian serous carcinoma were diagnosed with FIGO stage IIIC, and their average age was 56 ± 9.9 years.17 A cohort of 109 patients who were evaluable also found that 51.4% of patients had FIGO stage III and that the median age of patients at diagnosis was 59 years.[17]

Additionally, almost one-third of the patients (9/32; 28.1%) showed positive BRCA expression, half of the positive patients (5/9; 55.6%) had a BRCA1 mutation, and roughly one-third of the positive patients (3/9; 33.3%) had a BRCA2 mutation. With an overall mutation rate of 42% (184 cases) among 443 women with a personal history of ovarian cancer (OC), the latest study revealed a greater incidence of germline BRCA mutations. Specifically, 32% (143 cases) of the patients had BRCA1 mutations, and 9% (41 cases) had BRCA2 mutations.[16] Amin et al. (2023) found that 61.2% of the patients they examined had BRCA1 expression.[18]

High-grade serous tubal/ovarian cancer (HGSC) had a 40% (28/70) BRCA1/2 malfunction rate, according to Kalachand et al. This included 14.3% BRCA1 methylation (n=10), 7.1% BRCA1 mutation (n=5), and 18.6% BRCA2 mutation (n=13).[17] BRCA1/2 immunohistochemistry (IHC) was assessed by Manchana et al. as a screening method for germline BRCA1/2 in epithelial ovarian cancer (EOC). 36 individuals (34.3%) had decreased expression of BRCA1 and/or BRCA2. For the identification of germline BRCA1 mutations, loss of BRCA1 expression yielded 66.7% sensitivity, 84.3% specificity, 28.6% positive predictive value (PPV), and 96.4% negative predictive value (NPV). In contrast, the identification of germline BRCA2 mutations by reduction of BRCA2 expression had

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50% sensitivity, 78.8% specificity, 12.5% PPV, and 96.3% NPV.[19]

Just four patients had lymph node involvement, and 25% of them had BRCA mutations. Age, FIGO stage, and lymphatic node involvement in advanced ovarian cancer did not significantly correlate with BRCA mutation, according to our research (p<0,05).

In line with our study, Yamada et al reported a case of cancer of unknown primary (CUP) with adenocarcinoma histology and a germline BRCA1 p.L63* variant. It initially resembled a favorable prognosis type similar to ovarian cancer, but metastases were observed in cervical lymph nodes, indicating a somewhat atypical CUP compared to the typical ovarian cancer-like CUP. This study identified a germline Breast Cancer 1 (BRCA1) p.L63* variant through a family history inquiry and BRCA analysis, indicating hereditary breast and ovarian cancer syndrome. [12]

Bachmann et al clarified the impact of node involvement (affected to resected nodes) in optimally cytoreduced (residual tumour ≤1cm) stage IIIC/IV ovarian cancer. On average, 21.3 pelvic and para-aortic nodes were removed per patient (range 1-60 nodes). Minor nodal involvement (node ratio >0-≤0.5: (59%) was most often detected. Increasing node ratio leads to significant decreased overall survival (p<0.001). Significant best overall survival was associated with minor node involvement (node ratio >0 to ≤0.5). Complete cytoreduction correlated with node affection shows significant best prognostic impact in minor node affection compared to incomplete resection (R>0-≤1cm) independent to nodal status (OS p<0.001).[20]

Henning et al presented a case of a lady in her 60s where increased metabolic activity of the cardiophrenic lymph node was picked up in the investigation of an adenocarcinoma of the lung. Post-thoracoscopic resection histopathological analysis of this lymph node showing an epithelial structure with positive immunohistochemical markers PAX8, WT1, ER, and p16 with a p53 wild type-pattern were the sole presenting features of a high grade serous ovarian carcinoma, that was otherwise undetectable by radiological or hematological screening. Only histopathological analysis after modified radical hysterectomy in gynae-oncological fashion were able to identify a 4 mm lesion within the left fallopian tube.[21]

Cardiophenic lymph nodes drain lymph from the diaphragm, the liver, pleura, pericardium, and anterior abdominal wall into the parasternal lymph nodes and the anterior mediastinal chain.Due to their distribution and contribution to lymphatic drainage of the peritoneal cavity, it is pathophysiologically sensible to assume that invasion of the peritoneal cavity is the main cause of infiltration of the cardiophrenic lymph nodes.While there are reports of isolated distal lymph node recurrences of high grade serous carcinoma and cases where rare thoracic disease masquerades as ovarian malignancy, to our knowledge it has not been reported before that a cardiophrenic lymph node can be the sole presenting feature of such a cancer.[22–24] The literature reflects that involvement of the cardiophrenic lymph nodes are associated with prior involvement of the peritoneum and should therefore be considered a late stage disease process. Furthermore, cardiophrenic involvement on pre-treatment imaging is an indicator of stage IV cancer behavior and consequently associated with poorer outcome of patients.[25,26]

A multicenter, retrospective department database review was investigate the prognostic

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value of lymph node ratio (LNR) in patients with stage III ovarian high-grade serous carcinoma (HGSC) at 6 gynecologic oncology centers in Turkey. A total of 229 node-positive women with stage III ovarian HGSC, an increased LNR was associated with a decrease in 5-year OS from 65.1% for LNR1, 42.5% for LNR2, and 25.6% for LNR3, respectively ($p<0.001$). In multivariate analysis, women with LNR≥0.50 were 2.7 times more likely to die of their tumors.[27]

Karadağ et al retrospectively investigated the number of lymph nodes that need to be removed to detect lymph node positivity and the effect of this number on prognosis. A total of 155 patients underwent lymphadenectomy and the median number of removed lymph nodes was 24. Lymph node positivity was detected in 72 (46.4%) of these patients, while the median number of positive lymph nodes was 4 in the lymph node-positive group. A statistically significant positive correlation was found between the number of lymph nodes removed and the median overall survival (OS) ($r = 0.546$, $p<0.001$). At the same time, when the number of 24 lymph nodes, which is the median number of lymph nodes removed and the value found to detect lymph node positivity in the receiver operating characteristic (ROC) curve, is taken as cut off; mean OS was found to be statistically significantly higher in the group with adequate lymph node dissection compared to the group with insufficient lymph node dissection (46.46±35.22 vs 22.33±21.43; $p < 0.001$, respectively). It was shown that more than 24 lymph nodes are required for adequate lymph node dissection in the patients included in the study, and thus it can contribute positively to the prognosis. With the support of more comprehensive and prospective studies conducted on this subject to this study, clearer data will emerge about the number of lymph nodes that should be removed in an ideal surgery.[28]

CONCLUSION

Ovarian cancer is considered the most fatal gynecological malignancy in the global health. High grade serous carcinoma can have ovarian, tubal or peritoneal origin, with the latter being a rare presentation. Due to poor screening and presentation with non-specific symptoms, ovarian cancer is usually diagnosed in its advanced stage (III–IV) in most patients when the tumor has spread throughout and beyond the abdominal cavity. As advanced ovarian cancer should not be undertreated, therefore risking the patients' progression of disease, lymph node involvement—even in the case of an isolated site—should be considered stage III/IVb. This should always occur in consultation of a multidisciplinary team (MDT) to carefully consider the different results of imaging, histopathological reports, and patient characteristics.

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