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Spectrum and Incidence of Primary and Therapy-Related Hematologic Malignancies in Individuals with BRCA1 and BRCA2 Pathogenic Variants

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SPECTRUM AND INCIDENCE OF PRIMARY AND THERAPY-RELATED HEMATOLOGIC MALIGNANCIES IN INDIVIDUALS WITH *BRCA1* AND *BRCA2* PATHOGENIC VARIANTS

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Therapy-related myeloid neoplasms (t-MN) are rare and deadly hematologic malignancies that develop following exposure to cytotoxic therapies such as radiation, chemotherapy, and poly (adenosine diphosphate-ribose)-ADP polymerase (PARP) inhibitors. Preliminary evidence suggests that germline *BRCA1* and *BRCA2* pathogenic and likely pathogenic (P/LP) variants may increase susceptibility to t-MNs due to the genes' established role in DNA damage response. There is also evidence that individuals with *BRCA1/2* P/LP variants may be more susceptible to developing primary hematologic malignancies. We reviewed medical records of 706 individuals with *BRCA1/2* P/LP variants to assess hematologic malignancy diagnoses and t-MN development. Our study population was 5.1% male and 94.9% female, 58% had *BRCA1* P/LP variants and 42% had *BRCA2* P/LP variants, and the majority (59.92%) identified as Caucasian. Twenty-one hematologic malignancies were identified (2.97%): non-Hodgkin lymphoma in 9/706 individuals (1.27%), chronic myeloid leukemia and multiple myeloma each in 2/706 individuals (0.28%), respectively, and acute myeloid leukemia, unspecified leukemia, and Hodgkin lymphoma each in 1/706 individuals (0.14%). Therapy-related myeloid neoplasms were seen in 5/706 individuals (0.71%), a significantly higher incidence than 0.13/100,000 (0.0013%) observed in the general population (p-value: 0.000001). The estimated 20-year risk of t-MN development is 2.11% (95%

CI 0.74 – 5.96). This study supports the assertation that germline *BRCA1/2* P/LP variants increase the risk of t-MNs.

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INTRODUCTION

Therapy-related myeloid neoplasms (t-MNs) are a distinct subgroup of myeloid malignancies encompassing acute myeloid leukemia (AML), myelodysplastic syndrome (MDS), and myelodysplastic syndrome/myeloproliferative neoplasms (MDS/MPN) that occur due to exposure to cytotoxic agents such as chemotherapy and/or radiation[1]. Therapy-related myeloid neoplasms are one of the most serious late effects of chemotherapy as they have a very poor prognosis, with ten-year survival rates of less than 20%[2].

The associations between certain therapies and the latency period to development of t-MNs has been well-characterized. Radiation therapy and alkylating agents like carboplatin are associated with a t-MN latency period of 5-7 years[3]. These neoplasms commonly harbor cytogenetic abnormalities involving partial or complete loss of chromosomes 5 and 7[3]. Topoisomerase II inhibitors like doxorubicin (Adriamycin), etoposide, and epirubicin are associated with t-MNs that have a shorter latency period of 2-3 years after cytotoxic exposure[3]. These neoplasms commonly have translocations involving chromosomes 11 and 21[3]. Preliminarily, poly (adenosine diphosphate-ribose)-ADP polymerase (PARP) inhibitors have also been identified as a risk factor for development of t-MN[4-12]. The incidence of t-MN following PARP inhibitor exposure is approximately 0.5-2%[4-11]. The characteristics of these malignancies are not as well-characterized since the first PARP inhibitor approved by the Food and Drug Administration (FDA) was not approved until 2014[13, 14].

Although some risk factors for t-MN development have been identified, the exact risk and etiology are not well understood. Hematologic malignancies and breast cancer are the two most common primary malignancies associated with development of t-MNs[15]. Due to the established contribution of the Fanconi anemia/DNA repair pathway to development of both

breast cancer and hematologic malignancies, it has been speculated that pathogenic variants in *BRCA1* and *BRCA2* (*BRCA1/2*), as well as the other *FANC* genes, may increase an individual's susceptibility to developing t-MNs following cytotoxic therapy[16, 17].

There is evidence that individuals with *BRCA1/2* germline pathogenic/likely pathogenic (P/LP) variants are not at an increased risk to develop increased or severe hematologic toxicity as compared to other women with breast cancer during treatment with chemotherapy[18]. However, the DNA damage response and repair in individuals with *BRCA1/2* P/LP variants in response to cytotoxic therapy has not been compared to DNA damage response in people without *BRCA1/2* P/LP variants[18]. Moreover, there is evidence that they have an increased lifetime risk of developing t-MNs when compared to other breast cancer survivors[19].

Moreover, recent studies have shown the incidence of germline P/LP variants in hereditary breast cancer predisposition genes appears enriched in women who develop t-MNs. These variants may be present in as many as 20% of breast cancer survivors with t-MNs, despite hereditary breast cancer accounting for only 5-10% of breast cancer cases overall[19, 20]. This increased incidence of P/LP variants in hereditary breast cancer genes further supports a possible association between the Fanconi anemia DNA repair pathway and t-MNs[19]. However, several of these hematologic malignancies developed outside of the expected latency period or in patients who did not undergo cytotoxic therapy[19]. This may be an indication that haploinsufficiency of these genes increases the risk of developing hematologic malignancies independent from any prior therapy.

The incidences of non-breast and ovarian cancers diagnosed in individuals with *BRCA1/2* P/LP variants were described by Mersch et al[21]. Their report included the incidence of leukemia and lymphoma. They did not report a significant risk for these cancers in individuals

with *BRCA1/2* P/LP variants, however the study did not include MDS, MPN, or t-MNs, which could have been missed.

To our knowledge there has not been a study with a large enough sample size to achieve statistical power that has looked specifically at the incidence and spectrum of hematologic malignancies in people with *BRCA1/2* P/LP variants. Understanding this relationship is important to establish effective screening, treatment, and prognostication guidelines for individuals with *BRCA1/2* P/LP variants. The aim of our study is to investigate the incidence and characteristics of therapy-related and *de novo* hematologic malignancies in individuals with BRCA1/2 individuals.

METHODS

Study Population and Data Collection

Our study population was taken from the University of Texas MD Anderson Cancer Center Clinical Cancer Genetics Program Database. All individuals were seen at our institution either for diagnosis and treatment of a cancer or for a second opinion. Individuals who were diagnosed with a *BRCA1/2* P/LP variant between 1997 and 2015 and had at least one cancer diagnosis were eligible for inclusion. Individuals with a second P/LP variant in another cancer-predisposing gene and/or those who had not been diagnosed with cancer were excluded from analysis.

Data was collected by a single researcher (RR) by retrospective chart review. Due to the large population size, individuals who met study inclusion criteria were randomized using a random number generator to decrease potential bias based on time since diagnosis. Demographic information including sex, age, race/ethnicity, gene (*BRCA1/2*) and variant,

family history of hematologic malignancies, vital status, and cancer risk factors were collected from patient medical records. Cancer-specific information including cancer diagnoses, age at diagnosis, blood count parameters, and treatment regimens were collected from patient medical records. Date of first and last encounter at our institution were collected to calculate follow up and latency periods.

All data was collected in Qualtrics and de-identified at the point of collection. This study was approved by the Institutional Review Boards at The University of Texas MD Anderson Cancer Center and The University of Texas Health Science Center.

Statistical Analysis

Malignancy-specific data were recorded for each primary cancer. Multiple primaries of the same cancer type were counted as separate diagnoses while recurrences and/or metastases were included with the primary cancer as a single diagnosis. Cancer therapies were defined as cytotoxic if they are known to cause DNA damage. For this study, cytotoxic chemotherapy, radiation, and PARP inhibitors were considered cytotoxic. For individuals who received multiple lines of therapy or multiple rounds of the same therapy, the youngest age at exposure was used for analysis of follow-up and calculating the t-MN latency period.

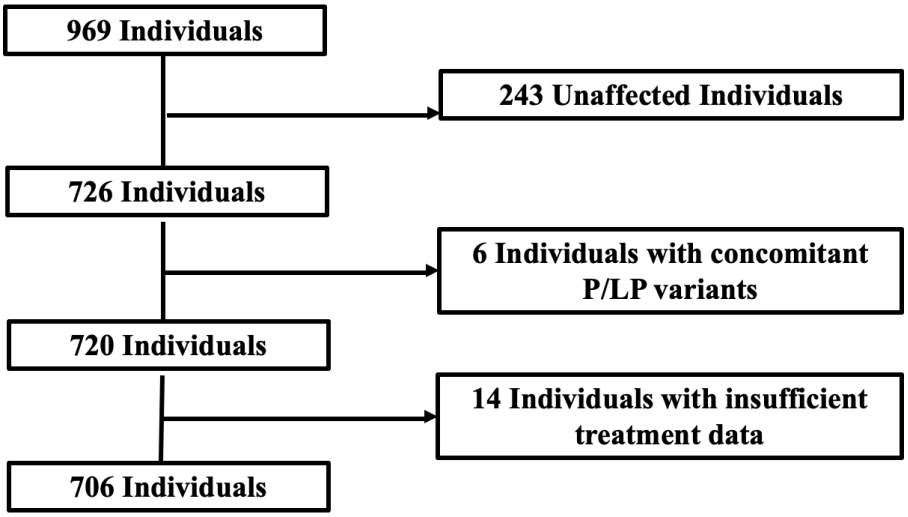
Statistical analysis was performed using Stata (v.13, College Station, TX) [22]. Statistical significance was set at a p-value of <0.05. Fisher's exact test of independence was used to compare demographic factors between the cohort who developed hematologic malignancies and the cohort who did not. It was also used to compare cytotoxic exposure, cancer incidence, and family history of hematologic malignancy between the individuals who developed a t-MN and those who did not. The incidence of t-MN is reported as a percentage with an Agresti-Coull

95% confidence interval. A one-sample proportion test was used to compare incidence of t-MN in our study to the incidence in the general population. A survival-time analysis was used to calculate follow-up time. Follow-up time in person-years was reported as medians and interquartile ranges (IQR). Survival models were utilized to assess hazard functions (reported with 95% confidence intervals, 95CI) at 1, 2, 5, 10, 15 and 20 years post-cytotoxic exposure. Cox proportional hazards model was used to assess differences between males and females and reported as hazard ratios (HR) with 95CI.

RESULTS

We identified 1,979 individuals with a *BRCA1/2* P/LP variant seen at our institution between 1997 and 2015. Of these, 969 were randomly selected and reviewed. Two hundred and forty-one individuals were excluded from analysis because they had not been diagnosed with cancer at the time of data collection and analysis. Six patients were excluded due to concomitant germline pathogenic variants including two individuals who also had Lynch syndrome, three individuals with *CDKN2A* pathogenic variants, and one individual with a *RUNX1* pathogenic variant. Fourteen individuals were excluded from analysis due to insufficient information available about their treatment. The final cohort was 706 individuals (n=706).

Figure 1: Establishing the Study Cohort



Our study population was 5.1% male and 94.9% female, 58% had *BRCA1* P/LP variants and 42% had *BRCA2* P/LP variants, and the majority (59.92%) identified as Caucasian. Additional demographic information is provided in Table 1. To evaluate the representativeness of our *BRCA1/2* cohort, we reviewed the incidence of classic *BRCA*-related cancers. The vast majority (70.11%, 495/706) of individuals were diagnosed with at least one breast cancer, 12.6% (89/706) had a second breast primary, and 2.55% (18/706) were diagnosed with pancreatic cancer. Moreover, 31.3% (210/670) of women were diagnosed with ovarian cancer and 30.56% (11/36) of men were diagnosed with prostate cancer.

Table 1: Demographic Characteristics (n=706)

Characteristic	Total n (%)	Individuals with Hematologic Malignancy, n (%)	Individuals without Hematologic Malignancy, n (%)	P-Value
Sex				**0.086**
Male	36 (5.1)	3 (14.29)	33 (4.82)	
Female	670 (94.9)	18 (85.71)	652 (95.18)	
***BRCA* P/LP Variant**				**1.00**
BRCA1	412 (58.36)	12 (57.14)	400 (58.39)	
BRCA2	294 (41.64)	9 (42.86)	285 (41.61)	
Ethnicity				**0.003***
Ashkenazi Jewish	50 (7.08)	5 (23.81)	45 (6.57)	
Caucasian	423 (59.92)	7 (33.33)	416 (60.73)	
African/African American	59 (8.36)	0 (0.00)	59 (8.61)	
Hispanic	102 (14.45)	3 (14.29)	99 (14.45)	
Asian/Pacific Islander	33 (4.67)	2 (9.52)	31 (4.53)	
Multiracial	36 (5.10)	4 (19.05)	32 (4.67)	
Native American	1 (0.14)	0 (0.00%)	1 (0.15)	
Unknown	2 (0.28)	0 (0.00%)	2 (0.29)	
Family History of Hematologic Malignancies				**0.827**
Yes	116 (16.43)	4 (19.05)	112 (16.35)	
No	557 (78.90)	16 (76.19)	541 (78.98)	
Unknown	33 (4.67)	1 (4.76)	32 (4.67)	

In total, 21 hematologic malignancies were identified in 21 individuals. No individuals analyzed developed more than one hematologic malignancy. There were no significant differences in alcohol use, tobacco use, chemical exposure, *BRCA1* or *BRCA2* variant, sex, or presence/absence of a family history of hematologic malignancies between individuals who developed a hematologic malignancy and those who did not (Table 1). There was a significant difference in ethnicity between the two groups. There were significantly more Ashkenazi Jewish and multiracial individuals in the cohort who developed hematologic malignancies when compared to Caucasian individuals than in the cohort who did not (p=.003).

Table 2: Incidence of Hematologic Malignancies

Hematologic Malignancy (n=21)	Individuals Diagnosed, n	Overall Incidence (%), n=706
Non-Hodgkin Lymphoma	9	1.27
Hodgkin Lymphoma	1	0.14
Chronic Myeloid Leukemia	2	0.28
Multiple Myeloma	2	0.28
Acute Myeloid Leukemia	1	0.14
Therapy-Related Myeloid Neoplasm	5	0.71
Unspecified Leukemia	1	0.14

Of the 21 hematologic malignancies identified, the majority were non-Hodgkin lymphoma (NHL). Eight hematologic malignancies were the primary cancer diagnosed in each respective individual: four with NHL, two with chronic myeloid leukemia (CML), and one with acute myeloid leukemia (AML) and Hodgkin lymphoma (HL), respectively.

Table 3: Individuals with Therapy-Related Myeloid Neoplasms (t-MN)

Patient	Primary Cancer(s)	Chemotherapy Agents	Additional Cytotoxic Therapies	Latency Period	t-MN	Cytogenetic Findings	Somatic Mutations
1	Ovarian Cancer	Carboplatin ⊹ Paclitaxel Doxorubicin ǂ	PARP Inhibitor	5 years	MDS/AML	None	None
2	Breast Cancer	Flourouracil Epirubicin ǂ Cyclophosphamide ⊹ Unknown Agents	Radiation	2 years	AML	47,XX,t(9;11)(p22;q23), +der(9)t(9;11)	None*
3	Pancreatic Neuroendocrine Tumor	Carboplatin ⊹ Cisplatin ⊹ Etoposide ǂ Erlotinib Temozolomide ⊹ Everolimus Irinotecan	None	5 years	MDS	46, X, -Y, +8; 45, X, -Y	None*
4	Breast Cancer (2) Ovarian Cancer	Carboplatin ⊹ Paclitaxel Doxorubicin ǂ Flououracil Cyclophosphamide ⊹	PARP Inhibitor Radiation	18 years	AML	46,XX,t(9;11)(p22;q23)	*TP53, GATA2(2), KRAS*
5	Ovarian Cancer	Carboplatin ⊹ Paclitaxel Doxorubicin ǂ Bevacizumab	None	15 years	MDS	45, XX, -7/idem, del(11)(q23)	None

⊹ alkylating agents; ǂ topoisomerase II inhibitors
MDS = Myelodysplastic Syndrome
AML = Acute Myeloid Leukemia
*Patient did not receive full molecular work-up following t-MN diagnosis

The observed study incidence of t-MN was 0.71% (95I: 0.28%-1.87%) which is significantly higher than the general population incidence of 0.0013% reported from 2001-2014 (p-value=0.001)[23]. There were no statistically significant differences between individuals who developed t-MNs and those who did not by number of cytotoxic therapies, type of cytotoxic therapy (radiation vs. chemotherapy vs. PARP inhibitors), number of primary cancers, or family history of hematologic malignancies.

The hazard functions for developing t-MN at various time points are listed in Table 4. The overall hazard function for the whole cohort was 2.11% (95CI: 0.74 – 5.96). A Cox proportional hazards regression model to assess the difference in hazard between males and females resulted in an approximately 10-fold higher, non-age adjusted, hazard of t-MN in males compared to females (HR:9.92, 95CI: 1.09 – 90.73).

Table 4: Risk for Developing Therapy-related Myeloid Neoplasms (t-MN) Over Time

Time Since Cytotoxic Exposure (Years)	Individuals at Risk, n	Individuals Diagnosed with t-MN, n	Hazard function	95CI*
1	624	0	0	
2	586	1	0.0017	0.0002 - 0.0121
5	469	2	0.0060	0.0019 - 0.0185
10	254	0	0.0060	0.0019 - 0.0185
15	153	1	0.0125	0.0039 - 0.0392
20	92	1	0.0211	0.0074 - 0.0596

* 95CI = 95% confidence interval

Overall, 642/706 (90.93%) individuals received cytotoxic therapy, putting them at risk to develop t-MN. These 642 individuals were included in the survival analysis and contributed a total of 6,739 person-years of information. The median follow-up time for individuals in our study was 8.12 person-years (IQR: 4.62 – 14.04), with the longest follow-up being 40.63 years.

DISCUSSION

The aim of this study was to examine the incidence and characteristics of hematologic malignancies, particularly t-MN, in a large cohort of individuals with *BRCA1/2* P/LP variants. Our cohort was representative of the increased cancer risks in *BRCA1/2*-positive individuals in that the majority analyzed were diagnosed with breast cancer and expected proportions had ovarian, prostate, and pancreatic cancer. The demographics of our cohort are also representative of *BRCA1/2*-positive individuals diagnosed with cancer: the majority are female, and nearly even between *BRCA1/2* variants.

Interestingly, the proportion of Ashkenazi Jewish individuals, as compared to individuals of European descent, was significantly higher in the cohort who developed hematologic malignancies than in the cohort that did not. This could suggest an increased risk for Ashkenazi Jewish individuals who have *BRCA1/2* P/LP variants to develop hematologic malignancies. Given that the number of individuals who developed hematologic malignancies is relatively small, the significance of the Ashkenazi Jewish individuals may be due to the increased prevalence of *BRCA1/2* mutations in the Ashkenazi Jewish population as founder mutations. However, it is possible that there is a potential biological significance of these Ashkenazi Jewish founder mutations which have a particular propensity for hematologic malignancies and further studies are needed to elucidate this relationship.

Nearly all hematologic malignancies identified in this study cohort were lymphoid, primarily NHL. NHL is the most common hematologic malignancy in the general population, so it is not surprising that this is reflected in our cohort of individuals with *BRCA1/2* P/LP

variants[12, 24]. However, this relatively high incidence of lymphoid malignancies suggests a possible association between *BRCA1/2* and general risk of lymphoid malignancy.

The cytogenetic characteristics of the t-MNs identified in our study were similar to those reported in the literature. Translocations between chromosomes 9 and 11 are common in patients who have been exposed to topoisomerase II inhibitors and are seen in approximately 11% of t-MN overall[3, 25, 26]. Forty percent (2/5) of t-MN resulting from topoisomerase II inhibitor therapy showed t(9;11). Similarly, monosomy 7 and 7q deletions are enriched in t-MN patients as compared to patients with *de novo* leukemia and are most common in individuals who have been exposed to radiation and alkylating agents[3, 26]. The individual with monosomy 7 developed a t-MN following exposure to an alkylating agent which is in accordance with patterns observed in the literature[3]. In contrast, trisomy 8 without a complex karyotype, as seen in patient 3, is less common in t-MN than in *de novo* leukemia[26]. It was noted that the loss of chromosome Y observed in patient 3 may have been clonal, but more likely due to age-related non-disjunction. Although many of the patients in our study had cytogenetic characteristics that are commonly reported in t-MNs, it was unusual that none of them had complex cytogenetics. Complex karyotypes are common in t-MN, and the lack of complex karyotypes identified in our study could indicate a role of BRCA1/2 P/LP variants as a contributing factor to t-MN development[26].

The molecular characteristics of t-MNs in our study were also similar to those reported in the literature. Up to 40% of t-MNs in the general population have a somatic *TP53* variant[25, 27]. TP53 mutations in t-MNs are also highly-associated with treatment regimen and may be seen in as many as 55% of t-MNs following treatment with platinum-based chemotherapy[28]. These mutations commonly precede t-MN development along with the acquisition of additional

cytogenetic or molecular changes. Our study is limited in that two of the five t-MNs identified did not receive a full molecular workup of their myeloid malignancy, and thus were not tested for *TP53* variants. All three of the t-MNs analyzed for *TP53* variants did receive platinum-based chemotherapy. Of the t-MNs analyzed for *TP53* variants, 33% (1/3) were identified to have somatic *TP53* variants. It is possible that the underlying germline *BRCA1/2* P/LP variants are sufficient to promote acquisition of somatic and molecular aberrations leading to t-MN in individuals without somatic *TP53* or other variants. This could also explain why we identified increased rates of t-MN as compared to the general population.

In the general population, the most common primary cancers prior to t-MN development are breast cancer and hematologic malignancies, particularly non-Hodgkin lymphoma, multiple myeloma, and Hodgkin lymphoma[15]. The incidence of t-MNs that develop from non-Hodgkin lymphoma is also increasing over time, and the incidence following treatment for breast cancer and Hodgkin lymphoma has remained constant; however, the incidence of t-MNs following treatment for ovarian cancer is decreasing over time[25]. Therefore, it is unusual that most of the t-MNs in this cohort were identified following ovarian cancer treatment, while only two developed after breast cancer treatment, and none following treatment for a hematologic malignancy. This may be, in part, due to a difference in treatment between individuals with *BRCA1/2*-associated ovarian cancer versus non-*BRCA* ovarian cancers, namely the use of PARP inhibitors. Our institution was a site for PARP inhibitor clinical trials before Olaparib's approval by the FDA in 2014[14]. Two of the individuals who developed t-MN following ovarian cancer treatment were treated with PARP inhibitors through a clinical trial. It is difficult to determine if PARP inhibitors themselves are responsible for an increased risk of t-MN or if the correlation exists because nearly all PARP inhibitor clinical studies have been performed in *BRCA1/2*

positive individuals, which alone appears to confer an increased risk of t-MN. Moreover, risk for t-MN following PARP inhibitor treatment seems to be highest in individuals who have not received other cytotoxic therapies, and evidence is conflicting if additional risk is conferred within the setting of several prior cytotoxic therapies[12, 29]. However, PARP inhibitors were not approved as a first line therapy until 2020, so the contribution of PARP inhibitors to t-MN risk independent from other cytotoxic therapies requires further investigation[30].

The reported risk of t-MN following treatment for HL is approximately 0.7-1.7% depending on the treatment regimen, and HL is a primary cancer associated with a high risk of t-MN, comparable to risk following treatment for breast cancer [25, 26, 31]. In our cohort, the overall risk of t-MN is estimated to be 2.11% by 20 years. In contrast, the reported risk of t-MN following treatment for non-Hodgkin lymphoma is approximately 0.7-1.7% depending on the treatment regimen[31] and NHL is associated with the highest known risk of t-MN development[15, 23]. The risk calculated in our study is higher than this, further demonstrating that the risk of t-MN following cytotoxic exposure is increased in individuals with germline *BRCA1/2* P/LP variants; however, it is difficult to compare these risks when looking at such specialized populations. Our study also identified a nearly 10-fold increased risk of t-MN development in males as compared to females. Further analysis is necessary to understand if this difference in risk is due to confounding factors, a relatively small sample size, or if it is a true relationship.

The observed study incidence of t-MN was 0.71%. This is significantly higher than the general population incidence of 0.0013% reported from 2001-2014 using SEER data (p-value=0.000001)[23]. Our analysis is limited as our study population included only individuals with *BRCA1/2* mutations who developed cancer; however, despite this, the incidence of t-MN in our study is significantly higher than the reported population incidence.

Moreover, the incidence of t-MN reported in our study is likely a very conservative estimate, therefore a significant underestimate, due to the limitations of a retrospective chart review. For example, the individual who was described to have "unspecified leukemia" (Table 2) was treated for breast cancer with doxorubicin two years prior to her leukemia diagnosis, which would be within the expected latency for a t-MN following topoisomerase II inhibitor exposure. It is likely that she is merely a representative example of other individuals who similarly were treated at our institution for a primary cancer, yet went on to develop a t-MN that was diagnosed and/or treated elsewhere.

Future studies should examine the relationship between t-MNs and germline *BRCA1/2* P/LP variants more robustly using a prospective study design. A prospective design would allow for longer-term follow-up, even if individuals underwent diagnosis and treatment at different institutions. It would also allow for a better understanding the t-MN risk associated with PARP inhibitors as, with time, more individuals treated with them will outlive the latency period. Our study has several limitations. It was performed at a single site which poses challenges for generalizing this data to the general population and limits our demographic diversity. This also limited our ability to capture individuals who may have developed t-MN and been treated at other institutions. Moreover, we analyzed only individuals that were identified to have a *BRCA1/2* P/LP variant in 2015 or earlier to analyze individuals who had outlived most of the latency period necessary for t-MN development. However, in doing so, we limited both our overall study population size and the number of individuals who had been treated with PARP inhibitors.

Overall, our study adds to the growing evidence of an increased incidence of t-MN in individuals with germline *BRCA1/2* likely P/LP variants after treatment with cytotoxic therapies

and PARP inhibitors. Although t-MN is still a rare complication, its incidence is expected to continue to increase over time as cancer survivorship increases, and appears to be a uniquely increased risk among men and women with *BRCA1/2* pathogenic variants. Due to the dismal five- and 10-year survival rates for t-MN, understanding a patient's risk is critical when considering treatment recommendations and when counseling patients about treatment options. Thus, along with this study, further studies are needed to better characterize this risk and elucidate its relationship to specific therapies as cancer treatment continues to evolve.

REFERENCES

1. Vardiman JW, Thiele J, Arber DA, Brunning RD, Borowitz MJ, Porwit A, Harris NL, Le Beau MM, Hellstrom-Lindberg E, Tefferi A, Bloomfield CD. The 2008 revision of the World Health Organization (WHO) classification of myeloid neoplasms and acute leukemia: rationale and important changes. *Blood*. Jul 30 2009;114(5):937-51. doi:10.1182/blood-2009-03-209262

2. Hulegårdh E, Nilsson C, Lazarevic V, Garelius H, Antunovic P, Rangert Derolf Å, Möllgård L, Uggla B, Wennström L, Wahlin A, Höglund M, Juliusson G, Stockelberg D, Lehmann S. Characterization and prognostic features of secondary acute myeloid leukemia in a population-based setting: a report from the Swedish Acute Leukemia Registry. *Am J Hematol*. Mar 2015;90(3):208-14. doi:10.1002/ajh.23908

3. Godley LA, Larson RA. Therapy-related myeloid leukemia. *Semin Oncol*. Aug 2008;35(4):418-29. doi:10.1053/j.seminoncol.2008.04.012

4. Coleman RL, Fleming GF, Brady MF, Swisher EM, Steffensen KD, Friedlander M, Okamoto A, Moore KN, Efrat Ben-Baruch N, Werner TL, Cloven NG, Oaknin A, DiSilvestro PA, Morgan MA, Nam JH, Leath CA, Nicum S, Hagemann AR, Littell RD, Cella D, Baron-Hay S, Garcia-Donas J, Mizuno M, Bell-McGuinn K, Sullivan DM, Bach BA, Bhattacharya S, Ratajczak CK, Ansell PJ, Dinh MH, Aghajanian C, Bookman MA. Veliparib with First-Line Chemotherapy and as Maintenance Therapy in Ovarian Cancer. *N Engl J Med*. 12 2019;381(25):2403-2415. doi:10.1056/NEJMoa1909707

5. Coleman RL, Oza AM, Lorusso D, Aghajanian C, Oaknin A, Dean A, Colombo N, Weberpals JI, Clamp A, Scambia G, Leary A, Holloway RW, Gancedo MA, Fong PC, Goh JC, O'Malley DM, Armstrong DK, Garcia-Donas J, Swisher EM, Floquet A, Konecny GE, McNeish

IA, Scott CL, Cameron T, Maloney L, Isaacson J, Goble S, Grace C, Harding TC, Raponi M, Sun J, Lin KK, Giordano H, Ledermann JA, investigators A. Rucaparib maintenance treatment for recurrent ovarian carcinoma after response to platinum therapy (ARIEL3): a randomised, double-blind, placebo-controlled, phase 3 trial. *Lancet*. Oct 2017;390(10106):1949-1961. doi:10.1016/S0140-6736(17)32440-6

6. González-Martín A, Pothuri B, Vergote I, DePont Christensen R, Graybill W, Mirza MR, McCormick C, Lorusso D, Hoskins P, Freyer G, Baumann K, Jardon K, Redondo A, Moore RG, Vulsteke C, O'Cearbhaill RE, Lund B, Backes F, Barretina-Ginesta P, Haggerty AF, Rubio-Pérez MJ, Shahin MS, Mangili G, Bradley WH, Bruchim I, Sun K, Malinowska IA, Li Y, Gupta D, Monk BJ, Investigators PE-OG-. Niraparib in Patients with Newly Diagnosed Advanced Ovarian Cancer. *N Engl J Med*. 12 2019;381(25):2391-2402. doi:10.1056/NEJMoa1910962

7. Mirza MR, Monk BJ, Herrstedt J, Oza AM, Mahner S, Redondo A, Fabbro M, Ledermann JA, Lorusso D, Vergote I, Ben-Baruch NE, Marth C, Mądry R, Christensen RD, Berek JS, Dørum A, Tinker AV, du Bois A, González-Martín A, Follana P, Benigno B, Rosenberg P, Gilbert L, Rimel BJ, Buscema J, Balser JP, Agarwal S, Matulonis UA, Investigators E-ON. Niraparib Maintenance Therapy in Platinum-Sensitive, Recurrent Ovarian Cancer. *N Engl J Med*. 12 2016;375(22):2154-2164. doi:10.1056/NEJMoa1611310

8. Ray-Coquard I, Pautier P, Pignata S, Pérol D, González-Martín A, Berger R, Fujiwara K, Vergote I, Colombo N, Mäenpää J, Selle F, Sehouli J, Lorusso D, Guerra Alía EM, Reinthaller A, Nagao S, Lefeuvre-Plesse C, Canzler U, Scambia G, Lortholary A, Marmé F, Combe P, de Gregorio N, Rodrigues M, Buderath P, Dubot C, Burges A, You B, Pujade-

Lauraine E, Harter P, Investigators P-. Olaparib plus Bevacizumab as First-Line Maintenance in Ovarian Cancer. *N Engl J Med*. 12 2019;381(25):2416-2428. doi:10.1056/NEJMoa1911361

9. Ledermann JA, Harter P, Gourley C, Friedlander M, Vergote I, Rustin G, Scott C, Meier W, Shapira-Frommer R, Safra T, Matei D, Fielding A, Spencer S, Rowe P, Lowe E, Hodgson D, Sovak MA, Matulonis U. Overall survival in patients with platinum-sensitive recurrent serous ovarian cancer receiving olaparib maintenance monotherapy: an updated analysis from a randomised, placebo-controlled, double-blind, phase 2 trial. *Lancet Oncol*. Nov 2016;17(11):1579-1589. doi:10.1016/S1470-2045(16)30376-X

10. Pujade-Lauraine E, Ledermann JA, Selle F, Gebski V, Penson RT, Oza AM, Korach J, Huzarski T, Poveda A, Pignata S, Friedlander M, Colombo N, Harter P, Fujiwara K, Ray-Coquard I, Banerjee S, Liu J, Lowe ES, Bloomfield R, Pautier P, investigators SE-O. Olaparib tablets as maintenance therapy in patients with platinum-sensitive, relapsed ovarian cancer and a BRCA1/2 mutation (SOLO2/ENGOT-Ov21): a double-blind, randomised, placebo-controlled, phase 3 trial. *Lancet Oncol*. 09 2017;18(9):1274-1284. doi:10.1016/S1470-2045(17)30469-2

11. Penson RT, Valencia RV, Cibula D, Colombo N, Leath CA, Bidziński M, Kim JW, Nam JH, Madry R, Hernández C, Mora PAR, Ryu SY, Milenkova T, Lowe ES, Barker L, Scambia G. Olaparib Versus Nonplatinum Chemotherapy in Patients With Platinum-Sensitive Relapsed Ovarian Cancer and a Germline BRCA1/2 Mutation (SOLO3): A Randomized Phase III Trial. *J Clin Oncol*. 04 2020;38(11):1164-1174. doi:10.1200/JCO.19.02745

12. Nitecki R, Melamed A, Gockley AA, Floyd J, Krause KJ, Coleman RL, Matulonis UA, Giordano SH, Lu KH, Rauh-Hain JA. Incidence of myelodysplastic syndrome and acute myeloid leukemia in patients receiving poly-ADP ribose polymerase inhibitors for the treatment

of solid tumors: A meta-analysis of randomized trials. *Gynecol Oncol*. Mar 2021;doi:10.1016/j.ygyno.2021.03.011

13. Konecny GE, Kristeleit RS. PARP inhibitors for BRCA1/2-mutated and sporadic ovarian cancer: current practice and future directions. *Br J Cancer*. Nov 2016;115(10):1157-1173. doi:10.1038/bjc.2016.311

14. AstraZeneca. Lynparza (olaparib) [package insert]. U.S. Food and Drug Administration website. https://www.accessdata.fda.gov/drugsatfda_docs/label/2020/208558s014lbl.pdf. Revised May 2020. Accessed March 31, 2021.

15. McNerney ME, Godley LA, Le Beau MM. Therapy-related myeloid neoplasms: when genetics and environment collide. *Nat Rev Cancer*. 08 2017;17(9):513-527. doi:10.1038/nrc.2017.60

16. D'Andrea AD. BRCA1: a missing link in the Fanconi anemia/BRCA pathway. *Cancer Discov*. Apr 2013;3(4):376-8. doi:10.1158/2159-8290.CD-13-0044

17. Bagal B, Kumar R, Gaur T, Talreja V, Bonda A, Patkar N, Shetty D, Kowtal P, Subramanian PG, Gupta S, Sarin R, Hasan SK. Characterization of therapy-related acute leukemia in hereditary breast-ovarian carcinoma patients: role of BRCA1 mutation and topoisomerase II-directed therapy. *Med Oncol*. Apr 2020;37(5):48. doi:10.1007/s12032-020-01371-z

18. West AH, Knollman H, Dugan J, Hedeker D, Handorf EA, Nielsen SM, Bealin LC, Goldblatt LG, Willems H, Daly MB, Afghahi A, Olopade OI, Hulick PJ, Shagisultanova E, Huo D, Obeid E, Churpek JE. Hematologic toxicity in BRCA1 and BRCA2 mutation carriers during chemotherapy: A retrospective matched cohort study. *Cancer Med*. 09 2019;8(12):5609-5618. doi:10.1002/cam4.2471

19. Churpek JE, Marquez R, Neistadt B, Claussen K, Lee MK, Churpek MM, Huo D, Weiner H, Bannerjee M, Godley LA, Le Beau MM, Pritchard CC, Walsh T, King MC, Olopade OI, Larson RA. Inherited mutations in cancer susceptibility genes are common among survivors of breast cancer who develop therapy-related leukemia. *Cancer*. Jan 2016;122(2):304-11. doi:10.1002/cncr.29615

20. Tran G, Helm M, Litton J. Current Approach to Breast Cancer Risk Reduction for Women with Hereditary Predispositions to Breast Cancer. *Current Breast Cancer Reports*. 2016;8:165-174. doi:https://doi.org/10.1007/s12609-016-0220-9

21. Mersch J JM, Park M, Nebgen D, Peterson SK, Singletary C, Arun BK and Litton JK. Cancers associated with BRCA1 and BRCA2 mutations other than breast and ovarian. *Cancer*. Jul 2015;121(14):2474-5. doi:10.1002/cncr.29357

22. Stata Statistical Software: Release 13. StataCorp. 2013.

23. Morton LM, Dores GM, Tucker MA, Kim CJ, Onel K, Gilbert ES, Fraumeni JF, Curtis RE. Evolving risk of therapy-related acute myeloid leukemia following cancer chemotherapy among adults in the United States, 1975-2008. *Blood*. Apr 2013;121(15):2996-3004. doi:10.1182/blood-2012-08-448068

24. Smith A, Howell D, Patmore R, Jack A, Roman E. Incidence of haematological malignancy by sub-type: a report from the Haematological Malignancy Research Network. *Br J Cancer*. Nov 2011;105(11):1684-92. doi:10.1038/bjc.2011.450

25. Heuser M. Therapy-related myeloid neoplasms: does knowing the origin help to guide treatment? *Hematology Am Soc Hematol Educ Program*. Dec 2016;2016(1):24-32. doi:10.1182/asheducation-2016.1.24

26. Kayser S, Döhner K, Krauter J, Köhne CH, Horst HA, Held G, von Lilienfeld-Toal M, Wilhelm S, Kündgen A, Götze K, Rummel M, Nachbaur D, Schlegelberger B, Göhring G, Späth D, Morlok C, Zucknick M, Ganser A, Döhner H, Schlenk RF, AMLSG G-A. The impact of therapy-related acute myeloid leukemia (AML) on outcome in 2853 adult patients with newly diagnosed AML. *Blood*. Feb 2011;117(7):2137-45. doi:10.1182/blood-2010-08-301713

27. Bolton KL, Ptashkin RN, Gao T, Braunstein L, Devlin SM, Kelly D, Patel M, Berthon A, Syed A, Yabe M, Coombs CC, Caltabellotta NM, Walsh M, Offit K, Stadler Z, Mandelker D, Schulman J, Patel A, Philip J, Bernard E, Gundem G, Ossa JEA, Levine M, Martinez JSM, Farnoud N, Glodzik D, Li S, Robson ME, Lee C, Pharoah PDP, Stopsack KH, Spitzer B, Mantha S, Fagin J, Boucai L, Gibson CJ, Ebert BL, Young AL, Druley T, Takahashi K, Gillis N, Ball M, Padron E, Hyman DM, Baselga J, Norton L, Gardos S, Klimek VM, Scher H, Bajorin D, Paraiso E, Benayed R, Arcila ME, Ladanyi M, Solit DB, Berger MF, Tallman M, Garcia-Closas M, Chatterjee N, Diaz LA, Levine RL, Morton LM, Zehir A, Papaemmanuil E. Cancer therapy shapes the fitness landscape of clonal hematopoiesis. *Nat Genet*. 11 2020;52(11):1219-1226. doi:10.1038/s41588-020-00710-0

28. Leonard DG, Travis LB, Addya K, Dores GM, Holowaty EJ, Bergfeldt K, Malkin D, Kohler BA, Lynch CF, Wiklund T, Stovall M, Hall P, Pukkala E, Slater DJ, Felix CA. p53 mutations in leukemia and myelodysplastic syndrome after ovarian cancer. *Clin Cancer Res*. May 2002;8(5):973-85.

29. Morice PM, Leary A, Dolladille C, Chrétien B, Poulain L, González-Martín A, Moore K, O'Reilly EM, Ray-Coquard I, Alexandre J. Myelodysplastic syndrome and acute myeloid leukaemia in patients treated with PARP inhibitors: a safety meta-analysis of randomised

controlled trials and a retrospective study of the WHO pharmacovigilance database. *Lancet Haematol*. Feb 2021;8(2):e122-e134. doi:10.1016/S2352-3026(20)30360-4

30. GlaxoSmithKline. Zejula (niraparib) [package insert]. U.S. Food and Drug Administration website. https://www.accessdata.fda.gov/drugsatfda_docs/label/2020/208447s015s017lbledt.pdf. Revised April 2020. Accessed April 19, 2021.

31. Eichenauer DA, Thielen I, Haverkamp H, Franklin J, Behringer K, Halbsguth T, Klimm B, Diehl V, Sasse S, Rothe A, Fuchs M, Böll B, von Tresckow B, Borchmann P, Engert A. Therapy-related acute myeloid leukemia and myelodysplastic syndromes in patients with Hodgkin lymphoma: a report from the German Hodgkin Study Group. *Blood*. Mar 2014;123(11):1658-64. doi:10.1182/blood-2013-07-512657

32. Takahashi K, Wang F, Kantarjian H, Doss D, Khanna K, Thompson E, Zhao L, Patel K, Neelapu S, Gumbs C, Bueso-Ramos C, DiNardo CD, Colla S, Ravandi F, Zhang J, Huang X, Wu X, Samaniego F, Garcia-Manero G, Futreal PA. Preleukaemic clonal haemopoiesis and risk of therapy-related myeloid neoplasms: a case-control study. *Lancet Oncol*. 01 2017;18(1):100-111. doi:10.1016/S1470-2045(16)30626-X

33. Guru Murthy GS, Hamadani M, Dhakal B, Hari P, Atallah E. Incidence and survival of therapy related myeloid neoplasm in United States. *Leuk Res*. 08 2018;71:95-99. doi:10.1016/j.leukres.2018.07.013

34. Li J, Smith A, Crouch S, Oliver S, Roman E. Estimating the prevalence of hematological malignancies and precursor conditions using data from Haematological Malignancy Research Network (HMRN). *Cancer Causes Control*. 08 2016;27(8):1019-26. doi:10.1007/s10552-016-0780-z

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