Germline vs. Somatic Genetic Testing: Their Increasing Use and Application

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INTRODUCTION

With the rise of personalized medicine within oncology, genetic testing has become increasingly important. At the time of initial diagnosis, many patients are candidates for upfront evaluation of their germline (inherited) DNA and/or their somatic (tumor) DNA, depending on their cancer type. These results can determine therapy in the adjuvant, maintenance, and recurrent setting. Germline and somatic variants also can have a significant impact on a cancer prognosis, determining additional cancer risks and recommended screening. In addition, this information can help to determine if family members have an elevated risk to develop cancer and if so, management options for early detection or risk reduction.

Prior to the implementation of wide-panel genomic sequencing of tumors, multiple techniques were used to identify oncogenic markers such as immunohistochemistry (IHC) and fluorescence in situ hybridization (FISH) which are still commonly used today. In 2013, Foundation Medicine first published validation of their somatic next-generation sequencing assays which demonstrated a high sensitivity of 95-99% as well as a three times higher identification rate of actionable mutations compared to diagnostic tests.[1] The most common tumor specimens analyzed in this study were lung (18%), breast (14%), and cancers of unknown primary (9%). With the introduction of this technology, there has been a rapid increase in the use of somatic genomic sequencing over the past decade.

This article is dedicated to describing the differences between germline and somatic testing and reviewing their applications. Although both testing methods are used within a variety of cancer types, for the purposes of this article, we will focus primarily on their application within gynecologic oncology.

GERMLINE TESTING

An individual's germline DNA is formed by combining half of the mother's DNA from the egg and half of the father's DNA from the sperm. Pathogenic variants (PVs) are passed from parent to offspring and since that variant is present at the time of fertilization, it gets copied into every cell of the body. Many hereditary cancer syndromes follow autosomal dominant inheritance patterns, which translates to a 50% chance for a parent to pass the PV to their offspring.

Germline genetic testing is typically performed on lymphocyte DNA from blood or a combination of lymphocyte and buccal cells from saliva. There are some cases where blood and saliva cannot be used to perform germline testing. For example, DNA extracted from blood or saliva of a patient that has been diagnosed with a hematological cancer may be tumor DNA and therefore not indicative of a germline variant. A blood or saliva sample from patients who have undergone an allogeneic bone marrow transplant would analyze the DNA of the donor rather than the patient. In these cases, a skin punch biopsy with fibroblast culturing is recommended to obtain DNA.[2]

Germline testing is the standard test offered to patients with a personal and/or family history of cancer suggestive of a hereditary cancer predisposition syndrome. Germline PVs account for approximately 5–10% of all cancers. When an inherited PV is identified, it predicts what types of cancers a patient is at risk to develop. The type of cancer risk changes depending on the affected gene because genes are assigned different functions depending on the body part.

In gynecologic oncology, the National Comprehensive Cancer Network's (NCCN) criteria for germline testing for ovarian cancer are less stringent as compared to endometrial cancer. All patients with epithelial ovarian cancers, regardless of age at diagnosis, are recommended to pursue germline testing, whereas patients with endometrial cancers must be diagnosed under age 50, have a synchronous or metachronous Lynch syndrome-related cancer (colorectal, endometrial, gastric, ovarian, pancreatic, urothelial, brain, biliary duct, and small intestine) or have a family history of endometrial cancer.[2,3] Although every year the NCCN's genetic testing criteria broadens to encompass more patients with endometrial cancer, studies such as Levine et al involve an even wider population recommending germline testing for all endometrial cancers regardless of age at diagnosis and family history.[4]

SOMATIC TESTING

Somatic testing is performed on surgical pathology or a cancer tissue biopsy to elucidate the genomic profile of cancer cells (sequencing hundreds of genes) and assess for PVs that can be targeted for treatment. While germline genetic testing identifies PVs that exist within every cell in the body, somatic testing identifies PVs that exist within cancer cells only. The cancer cells may have distinct genetic mutations, new and different from the patient's germline cells, that are responsible for malignant proliferation. The potential for the discovery of additional genetic mutations within the cancer cells beyond which exists in the patient's germline is why this separate testing is recommended. The main questions being asked are: What is the genetic profile of the tumor and how is it different from the genetic profile of the patient's germline or normal tissue? Does one, or both, have a genetic variant that led to the patient's cancer diagnosis and do we have treatments to target it?

The commencement of The Cancer Genome Atlas (TCGA) project in 2006 by the National Cancer Institute and National Human Genome Research Institute deepened our understanding of the molecular characteristics of cancer.[5,6] They developed a genomic database of over 20,000 primary tumors spanning 33 cancer types. Analysis of this data introduced us to new subclassifications within cancer types, as well as revealed important genomic similarities between cancers of different primary organ types. The TCGA project, in addition to the International Cancer Genome Consortium, laid the foundation for subsequent research on the clinical implications of these genomic alterations and their use as targets for novel therapeutics.[7] For an assortment of cancer types, there are a wide array of targeted treatments and immunotherapy, in addition to numerous genomic-based clinical trials that are available to patients depending on their individual tumor genomic profile.

GENETIC TESTING AND TREATMENT

Precision medicine is a growing field utilizing genomic sequencing to therapeutically target patient-specific genomic alterations. Within gynecologic oncology, there is a growing need for targeted therapy and immunotherapy for both initial cancer treatment as well as maintenance therapy. Given these impactful clinical applications, healthcare providers should be aware of when germline and somatic genomic testing is appropriate for their patients.

In February 2022, the American Society of Clinical Oncology (ASCO) published a clinical opinion statement on the indications for somatic genomic testing in patients with metastatic or advanced solid tumor cancer types.[8] This statement supports performing somatic multigene panel genomic sequencing in patients if there is a known biomarker-linked approved therapy for that cancer. For example, the FDA-approved PARP inhibitor, olaparib, is used within germline or somatic BRCA1/2 mutated patients with ovarian, pancreatic, prostate, or HER2-negative breast cancer. The guidelines also recommend multigene panel genomic sequencing in solid tumors to assess microsatellite instability status and tumor mutational burden for the application of the FDA-approved immunotherapy, pembrolizumab. Somatic genomic sequencing for these purposes should be performed at appropriately certified laboratories.[8]

In the setting of recurrent disease, somatic testing is increasingly important with the expanding targeted therapy and immunotherapy applications. Particularly with the introduction of tissue and tumor site-agnostic treatments, such as pembrolizumab, which was the first FDA-approved tumor-agnostic treatment in 2017, multigene panel genomic sequencing can provide options for alternative therapies, especially in successive lines of treatments.[8] Evaluation of a multigene panel genomic sequencing not only evaluates the application of treatments already FDA-approved but also allows for assessment of eligibility for biomarker-selective clinical trials. Utilizing the National Institutes of Health's clinical trial database (http://clinicaltrials.gov) can potentially offer patients a wider scope of treatment options, if not restricted by location, and should be considered in the clinical decision-making process.

GENETIC TESTING AND HEREDITARY CANCER SURVEILLANCE

Somatic testing alone can reveal genetic variants that are suggestive of a germline PV. This should then prompt the need for subsequent germline testing due to its association with additional cancer risks for the patient and their family members.[2,3] A 2019 study of 2,308 patients diagnosed with a variety of tumor types found that 5% of patients had PVs on somatic testing that triggered referral for germline testing.[9] Of the 41% who completed germline testing, 74% had a germline PV identified.[9] The somatic genes found to harbor a PV prompting follow-up germline testing included, but are not limited to, BRCA1/2, PALB2, BRIP1, MSH2/6, and RAD51C/D. When this occurs, the patient should be referred to a genetic counselor/professional for follow-up counseling and germline testing. For example, a physician may order somatic genomic testing for a patient with recurrent metastatic endometrial cancer (not meeting guidelines for somatic testing in the upfront setting) that identifies a PALB2 pathogenic variant. Assuming she did not previously qualify for germline testing based on age or family history, she is now a candidate for blood or saliva testing to assess for the presence or absence of this somatic PALB2 variant. If the PALB2 variant is also identified within her germline this would increase cancer risks for both the patient and her family members who would benefit from additional medical care that could detect cancers earlier or reduce the risk of developing cancer.

Knowing that a patient has a germline PV provides additional treatment options, but it can also identify when high-risk cancer surveillance is necessary. The NCCN provides recommended medical management guidelines for the majority of established hereditary cancer genes/syndromes.[2,3] These guidelines allow providers to offer patients more intense screening such as breast imaging every 6 months (BRCA1/2) or colonoscopy screening every 1–3 years (Lynch syndrome).[2,3] The guidelines also provide the option of risk-reducing surgeries such as removal of the ovaries (i.e. BRCA1/2, BRIP1, RAD51C/D)[2] which has demonstrated a decrease in morbidity and mortality.[10]

Genetic testing not only benefits the patient but also has a meaningful impact on the family. Discovering that a germline PV exists allows for cascade testing to identify relatives who also carry the family PV and, therefore, have an increased risk of cancer. The majority of known hereditary cancer genes travel in an autosomal dominant inheritance pattern. This means that the patient's first-degree blood relatives have a 50% chance of also inheriting the same PV. Once a relative undergoes counseling and testing and is found to carry the known family PV, the respective recommendations for increased screening, and medical or surgical management can be made. Additionally, in most cases, relatives who test negative for an established family PV (true negatives) do not need risk reducing surgeries or high-risk surveillance which can be a relief to that individual.

Cancers arise when two events cause genetic damage which stops the gene from functioning, known as the Two-Hit hypothesis. The two genetic events could be a combination of two random events (sporadic cancers) or one inherited germline PV and one random event (germline cancers). Paired somatic and germline testing can help to clarify what cancers were caused sporadically. Two PVs identified in tumor tissue (double somatic PVs) with negative germline testing have been shown to cause sporadic cancers.[11] Similar to true negative testing, relatives of patients with sporadic cancers would not have to pursue high-risk screening but may tailor screening based on the family history. For example, individuals with no known hereditary cancer syndrome, but whose first-degree relative was diagnosed with colon cancer, should pursue colonoscopies at age 40 (or 10 years prior to the relative's age of diagnosis) and repeat this exam every 5 years compared to the general population screening recommendation that starts at age 45 and repeats every 10 years.[12]

INFORMED CONSENT

Multiple organizations have written position statements regarding informed consent for genetic testing, which include discussing incidental or secondary findings as well as the accuracy and limitations of genetic testing.[13-15] With both somatic and germline genetic testing, it is common to identify incidental PVs that have no association with the primary reason for testing. For example, informed consent should include making patients aware that the identification of a germline PV, and thus germline validation testing, may be recommended based on their therapeutic somatic test result.

Accuracy and testing limitations are important components that need explanation before patients make a decision about genetic testing. Not all labs offering somatic testing have the option of including germline validation testing. Laboratories having only the capability to perform somatic testing can inform the ordering provider that a PV may be germline; however, additional samples will need to be collected and sent to a secondary laboratory for confirmatory germline analysis.

Both germline and somatic testing can miss identifying PVs. In 2022, Terraf et al found that somatic testing alone failed to detect 10.5% of clinically actionable germline PVs.[16] On the other hand, Hampel et al (2021) identified that 3.5% of germline negative endometrial cancers had double somatic PVs in the mismatch repair genes.[17] For this reason, providers should consider the combination of upfront somatic and germline genetic testing in order to provide accurate and effective patient care.

TREATMENT IMPLICATIONS AND GYNECOLOGIC ONCOLOGY

Ovarian cancer is an example within gynecologic oncology in which both germline and somatic testing are recommended at the time of initial diagnosis. Per the NCCN guidelines, all patients with a personal history of epithelial ovarian cancer should undergo germline genetic testing.[2] Additionally, multigene somatic sequencing is increasingly important for prognosis and treatment of ovarian cancer and should be performed upfront at the time of diagnosis. There is growing evidence of favorable outcomes with targeted therapy in both BRCA1/2 and homologous recombination deficient tumors. For patients diagnosed with ovarian cancer having the inability to repair double-strand DNA breaks, 13–21% harbored a germline BRCA1/2 mutation, and an additional 6–7% had somatic BRCA1/2 variants.[8,18-21] Furthermore, approximately 50% of high-grade serous ovarian carcinomas are homologous recombination deficient.[22-24]

Poly-ADP ribose polymerase (PARP) is an important protein involved in DNA repair pathways, particularly in base excision repair of single strand breaks.[25] PARP inhibitors block these repair pathways, ultimately leading to double strand breaks and targeted cell death of tumors with homologous recombination repair deficiencies, such as BRCA1/2 mutations.[26] This mechanism has been the focus of numerous studies on the various PARP inhibitors within several tumor types.

The PARP inhibitor olaparib is FDA-approved for use as upfront maintenance therapy in patients with advanced

high-grade serous or endometrioid ovarian, fallopian tube or peritoneal cancer and either a germline or somatic BRCA1/2 variant. This approval was based on the 2018 data reporting an incredibly promising progression free survival advantage with a 70% lower risk of disease progression or death.[27] The SOLO1 randomized, double-blind, placebo-controlled, international phase III trial recently published powerful overall survival data on olaparib as upfront maintenance therapy.[28] Patients were randomly assigned olaparib 300 mg BID or placebo for up to 2 years after demonstrating a complete or partial response after platinum-based chemotherapy. At 7 years, 67% of patients receiving olaparib were alive, compared to 46.5% of patients receiving placebo (HR 0.55, 95% CI, 0.40 to 0.76; P<0.0004). Patients receiving olaparib had a median time to first subsequent treatment of 64 months, compared to only 15 months for patients receiving placebo (HR 0.37, 95% CI, 0.28–0.48).[28] The potential to achieve long-term remission is an exciting breakthrough and further highlights the importance of identifying those harboring BRCA1/2 germline and somatic variants early in their treatment course to provide all eligible patients the opportunity towards a cure.

TAKE HOME POINTS

- Germline genetic testing evaluates a patient's inherited DNA while somatic genetic testing evaluates tumor DNA. A patient may qualify for germline and/or somatic genetic testing based on family history or a specific cancer diagnosis.
- Somatic genomic sequencing has increasing clinical applications for cancer prognosis and treatment. Based on results, patients may be candidates for either FDA-approved or experimental targeted treatments and immunotherapies.
- Somatic test results may prompt germline testing. These incidental findings must be included in the informed consent process.
- Genetic counselors/professionals are valuable resources to help determine and facilitate the necessary genetic testing, interpret test results, and work closely with the patient's healthcare team to implement the most effective cancer risk-reducing and preventive plan.

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Conflicts of Interest

The authors report no conflicts of interest.

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