On behalf of Illumina, I respectfully request the NCCN Genetic/Familial High-Risk Assessment: Breast and Ovarian guideline panel to review the enclosed to support the use of next-generation sequencing (NGS)-based multi-gene testing in preference to single-gene testing.

**Specific Requested Changes:**

We propose that multi-gene testing be supported as the preferable way to begin testing when interrogating high- and medium-risk mutations in the Genetic/Familial High-Risk Assessment: Breast and Ovarian NCCN Guidelines v 1.2018.

Similarly, we propose that the discussion statement favoring the performance of single-gene testing over multi-gene testing be removed from the Genetic/Familial High-Risk Assessment: Breast and Ovarian NCCN Guidelines v 1.2018.

**FDA Clearance:**

The recommendation to use an NGS-based technique is not associated with any specific FDA-cleared product/s.

**Rationale:**

The NCCN guidelines currently include recommendations for screening for known high-penetance mutations in several high-risk genes (BRCA1/2, TP53, and PTEN), as well as multiple moderate-risk genes (ATM, BRIP1, CDH1, CHEK2, NBN, PALB2, RAD51C, RAD51D, and STK11). The presence of pathogenic variants in these genes can alter the clinical management of breast and/or ovarian cancer. Abundant clinical data demonstrate that NGS-based multi-gene testing identifies more women at risk of developing breast cancer, including triple-negative breast cancer, compared to BRCA1/2 single-gene testing alone. In a recent study, a 25-gene panel was used to retrospectively analyze the frequency of pathogenic variants in 35,409 women with a single diagnosis of breast cancer. Multi-gene testing identified pathogenic variants in 9.3% of these women, with more than half of these pathogenic variants in genes other than BRCA1 or BRCA2. Thus, compared to BRCA testing, multi-gene testing more than doubled the number of women that could potentially benefit from a change in treatment strategy according to current NCCN guidelines. Comparable results were obtained with a 21-gene panel in a recent study that analyzed breast cancer risk in 65,037 women with breast cancer referred for multigene testing. After excluding BRCA1 and BRCA2 variants, pathogenic variants were detected in 6.2% of these women with breast cancer. Moreover, the efficacy and cost-effectiveness of single-gene BRCA1/2 testing versus multi-gene NGS testing has been modeled in hypothetical cohorts of 40-year and 50-year-old cohorts of women meeting current NCCN criteria for genetic testing for breast cancer or other hereditary syndromes associated with breast cancer. The study estimated that multi-gene testing, followed by risk-reduction management, could cost-effectively improve the life expectancy by 0.007-0.012 life years for women at risk of breast cancer.

Multiple studies have documented higher diagnostic yields for NGS-based multi-gene tests compared with single-tests for BRCA mutations. Prospective and retrospective clinical studies have demonstrated that NGS testing is perfectly concordant with traditional single-gene testing, across more than 750 variants, and can identify clinically significant mutations that are not detectable by single-gene testing. In a recent study, NGS testing detected 28 specific mutations in the SCN1A gene that were missed by Sanger sequencing, due in part to technical limitations of Sanger sequencing. Similarly, evaluation of PIK3CA gene mutations in 186 breast carcinomas by NGS demonstrated that 4.8% of breast tumors had PIK3CA mutations that were not detected by Sanger sequencing.

**Proposed Changes**

**Current Excerpt 1:** Footer “k” of Figure BR/OV-2: Breast and/or Ovarian Cancer Genetic Assessment

"In some cases, multi-gene testing may be a preferable way to begin testing over the single-gene testing process."
Proposed Change:
Delete "In some cases."

New statement:
"Multi-gene testing is a preferable way to begin testing over the single gene testing process."

Current Excerpt 2: Discussion (MS-10): Genetic/Familial High-Risk Assessment: Breast and Ovarian

"Second, in some cases, next-generation sequencing may miss some mutations that would have been detected with traditional single-gene analysis."

Proposed Change
Delete "Second, in some cases, next-generation sequencing may miss some mutations that would have been detected with traditional single-gene analysis."

The following articles are submitted in support of using NGS as a multi-gene testing technique. We would like to acknowledge the contributions of NCCN panel members, who are also co-authors or co-contributors in some of these publications.


Sincerely,
Dr. Amy Mueller MD
Medical Director, Oncology

Amy Mueller MD