Development and Validation of a 34-Gene Inherited Cancer Predisposition Panel Using Next-Generation Sequencing

Background

- Approximately 5% to 10% of all cancers are attributed to inherited genetic variants that are pathogenic or likely pathogenic.[1]
- Assessing predisposition for inherited cancer can be complex: variants in specific genes can be associated with risk of many cancer types, and specific cancer types can be associated with variants in multiple genes.
- Multigene panels can identify hereditary cancer syndromes that were not initially suspected by the clinician.[2,3] Panels available for clinical use incorporate different laboratory procedures and bioinformatics pipelines, so specific panels should be rigorously validated.
- **Objective:** The investigators designed, developed, and validated a targeted next-generation sequencing (NGS) panel of 34 genes associated with hereditary cancer susceptibility. The clinical utility of the assay for variant detection was additionally assessed in specimens submitted for genetic analysis.

Methods

- The NGS panel targeted genes that increase cancer risk ≥2-fold or confer a 5% lifetime risk. Cancer types included breast, ovary, colon, rectum, pancreas, endometrium, prostate, neuroendocrine system, and others.
- For validation, the study included 133 unique deidentified specimens; 33 had known variant status (single nucleotide variants [SNVs], small insertions and deletions [indels], copy-number variants [CNVs]).
  - For CNVs, a CNV-flagging algorithm developed and validated in-house was used, and all flagged CNVs were confirmed by microarray.
- Consecutive clinical specimens were analyzed with the 34-gene NGS panel for SNVs, small indels, and CNVs.

Results

- For SNVs and small indels, sensitivity and specificity were both 100%.
- For CNVs, sensitivity was 100% and specificity 98% when the in-house CNV-flagging algorithm was used; specificity rose to 100% after applying confirmation by microarray.
- Of the first 500 clinical specimens tested in the 34-gene inherited cancer predisposition panel:
  - 53 pathogenic/likely pathogenic variants were detected in 13 genes of 49 individuals (9.8% of 500 patients).
    - Of the 13 genes, 8 are not typically included in testing for Lynch syndrome or *BRCA*-related breast and ovarian cancer syndrome.
    - Among the 41 individuals who met National Comprehensive Cancer Network criteria for only *BRCA1/2* testing,[4] clinically actionable variants were identified in non-*BRCA1/2* genes (eg, *ATM*, *BARD1*, *CDH1*, *CHEK2, PALB2*) for 22 individuals (54%)*.*

Conclusions

- Validation of the 34-gene panel designed, developed, and validated at Quest Diagnostics demonstrated strong test performance characteristics, including high sensitivity and specificity.
- The panel identified pathogenic or likely pathogenic variants in almost 10% of clinical specimens, confirming its clinical utility in assessing predisposition for inherited cancers.

References
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