Contemporary vs. Traditional DNA-Based Mutation Testing to Detect Hereditary Breast and Ovarian Cancer Syndrome (HBOC) in Women: A Meta Narrative Review

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BACKGROUND

- Breast cancer one of the most common cancers in women
- Inherited genetic mutations in BReast CAncer genes 1 & 2 (BRCA 1/2) cause cancer if wild-type allele is inactive

→ Hereditary Breast and Ovarian Cancer Syndrome

- HBOC caused by an autosomal dominant mutation genetically passed down
  - Predisposes individuals to an increased risk of developing early onset breast and ovarian cancers

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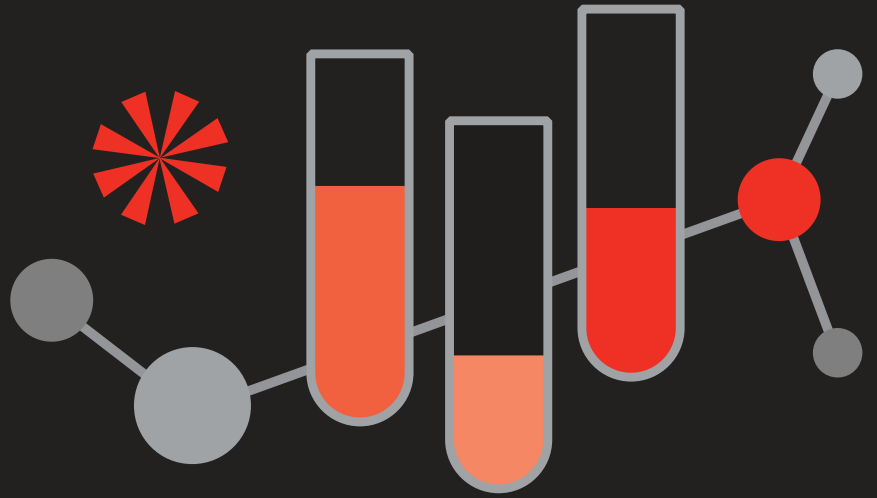
Tumor Group	Gene/Biomarker	Alterations	Indications	Result Interpretation/Significance	Assay Techniques
Invasive Ductal/Lobular Carcinoma	*BRCA1/2***	Inactivating variants	Germline testing, therapeutic	Hereditary breast cancer, predicts response to PARP-inhibitors	CMA, MLPA, NGS, PCR, Sanger
	*CDH1***	Inactivating variants	Germline testing, diagnostic	Inactivated in lobular carcinomas (*in-situ* and invasive), germline pathogenic variants associated with susceptibility to ILC and hereditary diffuse gastric cancer	CMA, IHC (E-cadherin), MLPA, NGS, PCR, Sanger
	ERBB2	Amplification, activating variants	Therapeutic	Molecular/intrinsic subtype classification, predicts response to anti-HER2 therapies	FISH, IHC, NGS
	ER/PR	Increased expression	Prognostic, therapeutic	Molecular/intrinsic subtype classification, predicts response to endocrine therapy	IHC
	IDH2	p.Arg172 hotspot variants	Diagnostic, possibly therapeutic in the future	Associated with tall cell carcinoma with reversed polarity	NGS, PCR, Sanger
	PIK3CA	Activating variants	Therapeutic	Predicts response to alpelisib; enriched in luminal A tumors	NGS, PCR, Sanger
	*PTEN***	Inactivating variants	Germline testing	Cowden syndrome	CMA, MLPA, NGS, PCR, Sanger
	*STK11***	Inactivating variants	Germline testing	Peutz-Jeghers syndrome	CMA, MLPA, NGS, PCR, Sanger
	*TP53***	Inactivating variants	Germline testing	Li-Fraumeni syndrome, enriched in basal-like tumors	CMA, MLPA, NGS, PCR, Sanger
Salivary Gland-Type Neoplasms	*CRTC1::MAML2* *CRTC3:MAML2*	Fusion	Diagnostic	Associated with mucoepidermoid carcinoma	FISH, NGS, RT-PCR
	ETV6::NTRK3	Fusion	Diagnostic, therapeutic	Associated with secretory carcinoma, predicts response to larotrectinib and entrectinib	FISH, IHC (pan-TRK), NGS, RT-PCR
	HMGA2 or *PLAG1*	Fusion	Diagnostic	Associated with pleomorphic adenoma	FISH, IHC (HMGA2, PLAG1), NGS, RT-PCR
	HRAS	p.Gly12, p.Gly13, p.Gln61 hotspot variants	Diagnostic, possibly therapeutic in the future	Associated with adenomyoepithelioma	IHC, NGS, PCR, Sanger
	MYB::NFIB and *MYBL1::NFIB*	Fusion	Diagnostic	Associated with adenoid cystic carcinoma	FISH, IHC/ISH (MYB), NGS, RT-PCR

SIGNIFICANCE

- Contemporary DNA testing using multigene panels has the potential to identify harmful mutations not only in BRCA genes but in other potentially harmful variants that predispose to HBOC.
- It is important for healthcare professionals to recognize susceptible patients early on.
- Contemporary technologies in multigene paneling are improving healthcare by streamlining diagnostic procedures.
- Next-Generation Sequencing is an efficient alternative to the traditional method of Sanger Sequencing.
- These new genetic testing methods may have a positive impact on those that are high risk for HBOC, allowing patients greater options and time to implement preventative measures if needed.



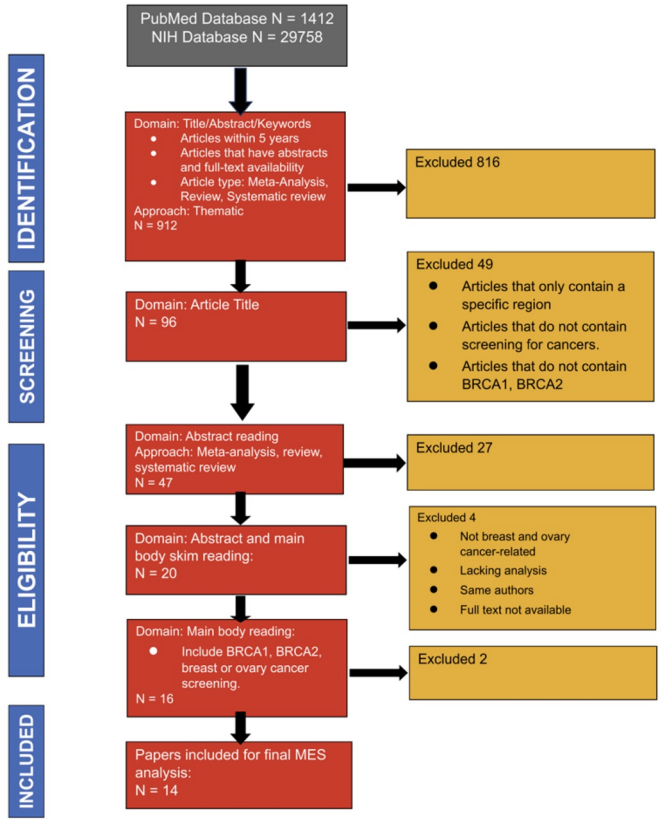
RESEARCH QUESTION

Does the use of multi-gene panel testing increase the probability for detection of HBOC-associated mutations compared to testing only *BRCA1* and *BRCA2*?

HYPOTHESIS

Women who undergo multi-gene panel testing will have a higher detection rate for cancer-predisposing mutations compared to women who receive testing focused only on BRCA1 and BRCA2.

METHODOLOGY



Inclusion Criteria

- Relevance: documents should directly address genetic testing methods for HBOC in women
- Recent publications: preference should be given to recent publications (5-year range).
- Diverse methodologies: experimental, observational, systematic, and meta-synthesis studies allow for a comprehensive examination from various perspectives.

Key Words: Hereditary breast and ovarian cancer syndrome (HBOC), BRCA 1, BRCA2, multi-gene panel testing, Next-generation sequencing, Sanger sequencing

Exclusion Criteria:

- Studies that do not contribute relevant narratives or perspectives to the research question.
- Studies that focus on other types of cancers with no mention of breast cancer, ovarian cancer, or HBOC.
- Literature that lacks credibility, such as unreliable sources or publications with methodological flaws.
- Literature is not from a reputable and reliable source.
- Duplicate publications or multiple reports of the same study.
- Studies published outside the defined 5-year time frame
- Literature written in languages that are not accessible for translation or interpretation.

SAMPLE PROCESSING CRITERIA

Criteria	Sanger sequencing	Next-Generation Sequencing (NGS)
Accuracy	99.9%	99 – 99.9%
Sample Limit	Up to 20 samples	More than 20 samples
Speed (<20 samples)	Fast	Slow
Speed (>20 samples)	Slow	Fast
Sensitivity	15 – 20%	1%
Sample coverage	1 read/sample (300 – 850 bp)	Billions of Reads/Samples (up to 16Tb)

Sanger Sequencing

- Pros:
  - Fast
  - Accurate
  - Cost-effective for < 20 samples
- Cons:
  - Maximum of only 20 samples
  - 1 read/sample

Next-Generation Sequencing (NGS)

- Pros:
  - Fast turnaround time
  - Accurate
  - Can test more than 20 samples
  - High sensitivity allows for detection of low-frequency variants
- Cons:
  - Expensive
  - Time-consuming for 1-20 targets

COSTS OF SEQUENCING

Sequencing Method	Platform	Target size (bp)	Cost (per sample, USD)
Next-Generation Sequencing	WGS	~ $3x10^9$	$1000 - $3000
	WES	~ $6x10^7$	$500 - $2000
	TS	~ $1x10^5$ – $1x10^7$	$300 - $1000
Sanger Sequencing		~ $3x10^2$ – $1x10^3$	$30

Comparison the costs between genetics sequencing methods: NGS (Next Generation Sequencing), WGS (Whole Genome Sequencing), WES (Whole Exome Sequencing), TS (Targeted Sequencing) and Sanger sequencing.

STRENGTHS AND LIMITATIONS

STRENGTHS

- Recent studies (within 5-year timeframe: 2019-2024)
- Appropriate statistical methods were used
- Able to combine large and complex research

LIMITATIONS

- Meta-narrative based on a limited number of sources and information
- Secondary research
- Overused data
- Increased risk of bias

FUTURE DIRECTIONS

- The future of HBOC diagnosis and treatment lies in the field of Genetic Counseling.
- By implementing the use of expansive tests such as multi-gene or NGS, the specific genes that are causing HBOC in patients would be detected early on.
- Doctors and genetic counselors will be able to accurately predict:
  - The effectiveness of treatments
  - The course and risks of metastasis
  - Track inheritable qualities that could be present in an offspring
- These advancements could potentially lessen the severity of HBOC over time.

CONCLUSION

- Our findings **support** the hypothesis because detection rates for HBOC tend to increase when multigene panels are used compared to single gene testing; “significant differences were observed regarding variant pathogenicity ($p<0.0001$) … the multigene panel was able to significantly resolve more high-risk patients” because of the vast panel of genes tested (Molina-Zaya, et al., 2022).
- Study findings support the recommendation for more widespread use of multigene panel genetic tests.
- A change to mainly multigene panel tests is likely to be seen in the future. The high cost of new genetic methods can be justified by the better detection outcomes in diagnosis of HBOC-causing germline mutations.

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