Molecular profiling comparison of BRCA1/2-mutated and BRCA1/2 non-mutated triple-negative breast cancer (TNBC)

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Abstract

Background: Triple-negative breast cancer (TNBC) is one type of breast cancer that remains challenging because of its aggressive nature and the lack of effective targeted therapy for it. Molecular profiling has revealed different subtypes, indicating a potential for promising targeted therapy such as androgen blockade and PARP inhibition in some TNBCs. The purpose of this study is to identify differences in BRCA1/2 mutated and non-mutated TNBC to shed light on potential therapeutic options in both subtypes, utilizing a multiplatform approach.

Methods: A cohort of 386 triple-negative breast cancer specimens were tested via a multiplatform profiling service (Caris Life Sciences, Phoenix, AZ) consisting of gene sequencing (next generation sequencing [NGS]), protein expression (immunohistochemistry [IHC]) and gene amplification (fluorescence or chromogenic in situ hybridization [FISH or CISH]). Primary and metastatic specimens were evaluated. Tumor specimens with any BRCA1 and/or BRCA2 mutation (i.e. pathogenic or variant of unknown significance) were categorized as "BRCA1/2-mutated", while all others were considered "BRCA1/2 non-mutated".

Results: In our cohort, 16.3% (63/386) of specimens were BRCA1/2-mutated while 83.7% (323/386) had no BRCA1/2 alteration detected. Amongst the highest rates of protein expression in BRCA1/2-mutated and non-mutated specimens were biomarkers like TOPO1 (63.5% and 63.4%), EGFR (65.2% and 67.4%), and the immune checkpoint biomarker, PD-1 (65.1% and 61.9%), with non-statistically significant differences. Differences noted between BRCA1/2-mutated and non-mutated specimens were detected by IHC in AR (11.1% versus 22.0%, p=0.0585) and PTEN (47.6% versus 59.6%, p=0.0941), with both trending but not achieving statistical significance. The highest overall mutation rate in both BRCA1/2-mutated and non-mutated were TP53 (80.6% and 73.1%, p=0.2659). Differences were also noted between BRCA1/2-mutated and non-mutated specimens by NGS in APC (6.3% versus 1.9%, p = 0.0644) and PIK3CA (11.1% versus 25.8%, p = 0.0137), with PIK3CA being statistically significant.

Conclusion: Multiplatform tumor profiling identified differences in molecular profiles between BRCA1/2 mutated and BRCA1/2 non-mutated TNBC. Our findings raise the possibility for future investigation of potential combination therapeutic targeted therapy. Increased AR overexpression in BRCA1/2 non-mutated specimens is consistent with reports from other institutions. Further studies utilizing tumor profiling to elucidate the biological differences in TNBC subtypes are warranted, to optimally include patients on clinical trials with specific targeted therapy and possibly improve treatment options.

Methods

A cohort of 386 triple-negative breast cancer specimens, verified internally using FDA-approved platforms, were tested via a multiplatform profiling service (Caris Life Sciences, Phoenix, AZ) consisting of gene sequencing (next generation sequencing or NGS), protein expression (immunohistochemistry or IHC) and gene amplification (fluorescence or chromogenic in situ hybridization [FISH or CISH]). Depth of sequencing by NGS was 1500X. Primary and metastatic specimens were included in this cohort.

BRCA1 and BRCA2 was assessed using the Illumina MiSeq NGS, a platform with a sensitivity to detect mutations or variants as low as 20% population of cells. Tumor specimens with a BRCA1 and/or BRCA2 mutation (i.e. pathogenic or variant of unknown significance) were categorized as "BRCA1/2-mutated", while specimens with no mutations detected in either BRCA1 and BRCA2 were categorized as "BRCA1/2 non-mutated". Both tests had to be performed to be included in the overall cohort.

Results

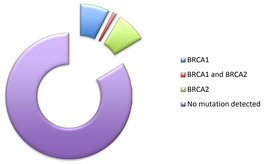
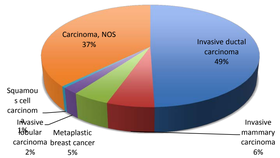


Figure 1 – Demographics. All TNBC specimens (n = 386) analyzed were female, with a median age of 56 years (standard deviation +/- 11.8). In the BRCA1/2-mutated cohort, 44.4% (28/63) were under 50 years old. By contrast, 27.6% (89/323) of patients in the BRCA1/2 non-mutated were under 50.

Overall, 19.5% (63/386) of TNBC profiled contained BRCA1 and/or BRCA2 aberrations. In the BRCA1/2-mutated cohort, 46.0% (29/63) were BRCA1-mutated, 6.3% (4/63) were BRCA1 and BRCA2-mutated, and 47.6% (30/63) were BRCA2-mutated

Figure 2 – Histologies represented in the cohort. The majority of specimens profiled were classified as either invasive ductal carcinomas (IDC) or unspecified breast carcinomas. Of note, one of the two squamous cell carcinoma (SCC) specimens analyzed contained a BRCA1 mutation.



Results (continued)

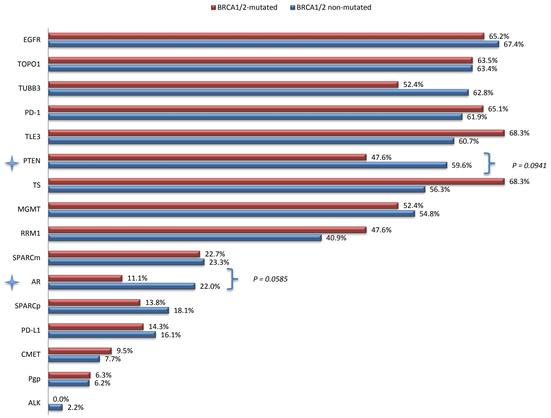


Figure 3 – Immunohistochemistry (IHC) in BRCA1/2-mutated and non-mutated samples. Comparisons were made between various potentially theranostic IHCs evaluating protein overexpression. For some biomarkers, expression rates between BRCA1/2-mutated and non-mutated specimens were similar or not statistically significant, as evidenced by EGFR (65.2% and 67.4%), TOPO1 (63.5% and 63.4%), and PD-1 (65.1% and 61.9%, respectively). However, trends were identified, with androgen receptor (AR) showing a difference of expression between BRCA1/2-mutated and non-mutated specimens of 11.1% and 22.0%, respectively (p = 0.0585). PTEN also showed a trend toward lower expression in BRCA1/2-mutated (47.6% versus 59.6%), indicating dysregulation of the PIK3CA/AKT/mTOR pathway in BRCA1/2-mutated populations.

Biomarker	BRCA1/2-mutated	BRCA1/2 non-mutated
MET	0% (0/62)	0% (0/312)
TOP2A	0% (0/63)	0% (0/321)

Figure 4 – In situ hybridization in BRCA1/2-mutated and non-mutated specimens. In this cohort of breast cancer, no MET or TOP2A amplification was detected in either cohort.

Results (continued)

Biomarker	BRCA1/2-mutated	BRCA1/2-non-mutated
ABL1	0.0%	1.0%
AKT1	3.2%	3.1%
APC	6.3%	1.9%
ATM	0.0%	3.5%
BRAF	0.0%	0.6%
MET	1.6%	1.3%
EGFR	0.0%	0.3%
ERBB2	0.0%	1.3%
FBXW7	3.2%	0.6%
FLT3	0.0%	0.3%
HNF1A	3.5%	0.0%
HRAS	1.8%	1.5%
IDH1	0.0%	0.3%

Biomarker	BRCA1/2-mutated	BRCA1/2-non-mutated
JAK3	1.6%	1.9%
KRAS	1.6%	2.5%
NOTCH1	0.0%	0.3%
NRAS	0.0%	0.3%
PIK3CA	11.1%	25.8%
PTEN	3.2%	5.5%
RB1	3.2%	0.6%
RET	4.9%	1.6%
SMAD4	0.0%	1.2%
STK11	1.6%	2.3%
TP53	80.6%	73.1%
VHL	0.0%	0.3%

Figure 5 – NGS sequencing in BRCA1/2-mutated and non-mutated samples. PIK3CA/AKT/mTOR pathway aberrations were detected in both TNBC cohorts. Of note, PIK3CA mutations were significantly lower in BRCA1/2-mutated specimens compared to non-mutated specimens (11.1% versus 25.8%, p = 0.0137). APC trended higher in the BRCA1/2-mutated versus non-mutated group (6.3% versus 1.9%, p = 0.0644).

Conclusions

- Multi-omic profiling can identify differences in the underlying biology of TNBC, particularly between TNBC with and without BRCA1/2 mutations.
- Higher PIK3CA mutation rates in the non-BRCA mutated TNBC cohort warrants further investigation in clinical trials, particularly in a population with historically worse survival rates in comparison to BRCA1/2-mutated populations.
- Biomarkers like AR deserve further study to assess whether a subgroup of non-BRCA1/2 TNBC may derive benefit from hormonal agents.

References

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