Integrated NGS-Based Tissue and Plasma Profiling to Advance Precision Oncology in Breast Cancer

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Introduction

The integration of broad genomic testing has become increasingly vital for personalized treatment planning in patients with breast cancer. By enabling the simultaneous evaluation of multiple genes and pathways, comprehensive genomic profiling provides a deeper understanding of tumor biology, identifies clinically actionable alterations, and guides the selection of targeted therapies with greater precision. This expanded molecular insight supports more informed therapeutic decisions, optimizes patient stratification, and enhances the ability to anticipate treatment response and resistance, ultimately contributing to improved clinical outcomes.

In this study, we evaluated the analytical performance and clinical relevance of a certified for in vitro diagnostics (CE-IVD), 1021-gene comprehensive genomic profiling panel in detecting actionable tumor biomarkers across a diverse cohort of breast cancer cases. The study aimed to validate the robustness of the assay in both tissue and liquid biopsy formats, and to explore its clinical utility in real-world patients with advanced breast cancer.

Methods and Materials

Tumor and blood samples from 353 patients in total were analyzed. The samples included 197 FFPE tumor and 156 plasma cfDNA. Concurrent molecular profiling of tissue and plasma was conducted in 23 cases to evaluate the efficacy of liquid biopsy although tissue samples may have originated from prior procedures. DNA extracted from leukocytes was used as a control to prevent the detection of false-positive results due to clonal hematopoiesis mutations.

Targeted-capture sequencing was performed using the Oncology Multi-Gene Variant Assay (GenePlus) which is a qualitative in vitro diagnostic test for detection of variants in 1021 tumor-related genes, and gene fusions in 38 genes. Sequencing was carried out on the MGI DNBSEQ-G400 platform. NGS data were analyzed via a dedicated bioinformatics pipeline on the Gene Box platform (GenePlus), enabling detection of all major genomic alterations, including gene fusions, Copny Number Variations (CNVs), as well as assessment of tumor mutational burden (TMB) and microsatellite instability (MSI).

Results

Regarding the analysis of 197 FFPE samples, at least one on-label actionable biomarker was identified in 54.8% of cases. Among these samples, PIK3CA mutations were the most frequently detected in 27.4% of the cases. ESR1 mutations were present in 16.2% Mutations in PTEN and AKT1 genes were also observed in 7.6% and 3% of cases, respectively. Additionally, 20% of patients harbored a suspected germline variant in BRCA1/2, PTEN and CDH1 genes.

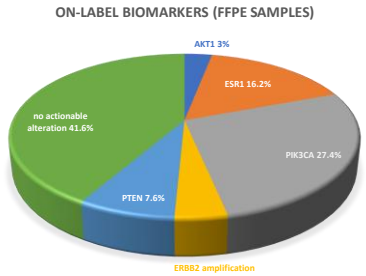

Figure 1. On-label actionable biomarker identified in 54.8% of cases of the FFPE samples

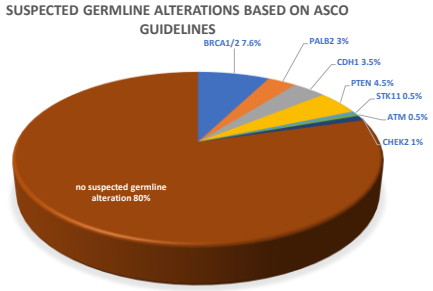

Figure 2. Suspected germline alterations identified in FFPE samples

In the analysis of 156 plasma samples, 46% were positive for an on label-biomarker. Among these, PIK3CA alterations were identified in 17.9% and ESR1 mutations were present in 17.3% of the cases. At lower frequency, variants were also observed in BRCA1/2 (3.8%), PTEN (3.2%), AKT1(1.3%) genes. ERBB2 (1.3%) amplification and an NTRK fusion (0.6%) were also detected. Approximately 20% of plasma samples harbored verified germline variants, most frequently in BRCA1/2 and CHEK2 genes.

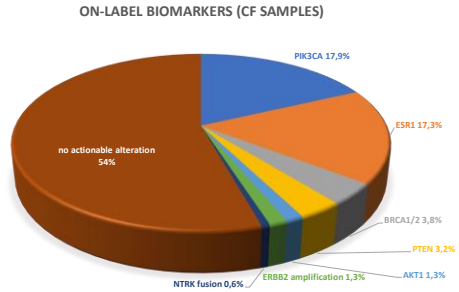

Figure 3. On-label actionable biomarker identified in 54.8% of cases of the Cf samples

In a subset of 23 metastatic Breast Cancer patients, concurrent analysis of tissue and liquid biopsy demonstrated a 70% overall concordance rate on on-label biomarker analysis.

- In two cases, FFPE analysis identified additional on-label mutations not detected in plasma, whereas in one case plasma analysis revealed a second ESR1 mutation not detected in the FFPE sample.
- In two patients, plasma analysis was negative, while tissue profiling identified alterations in ESR1 and PIK3CA genes.
- Importantly, in two cases, liquid biopsy surpassed tissue analysis by detecting an ESR1 variant and an NTRK fusion.

Discussion

This study demonstrates the clinical utility of comprehensive multigene NGS profiling in both tissue and plasma samples from patients with breast cancer, revealing a high rate of actionable variants. The parallel analysis of matched tissue and plasma samples showed a high concordance. In some cases, though, analysis of tissue provided additional information than liquid biopsy and vice versa. These differences emphasize that the two methodologies are complementary rather than interchangeable. Each approach contributes distinct and clinically relevant genomic information that would otherwise remain undetected if relying solely on FFPE tissue analysis or exclusively on liquid biopsy.

Conclusions

In conclusion, integrated NGS analysis of tissue and liquid biopsies provides a powerful platform for precision oncology in breast cancer, supporting therapeutic decisions, resistance monitoring, and hereditary cancer risk assessment. The synergy between tissue- and plasma-based testing enables more accurate therapeutic selection, facilitates earlier detection of emerging resistance mechanisms, and contributes to improved monitoring of tumor evolution throughout the course of treatment. Furthermore, the inclusion of both sample types supports comprehensive assessment of hereditary cancer predisposition, ensuring that clinically actionable germline findings are not overlooked.

Overall, leveraging both tissue and liquid biopsies maximizes the clinical impact of NGS profiling, providing a more complete and actionable view of tumor biology—particularly in patients with advanced or heterogeneous disease, where reliance on a single sample type may fail to capture the full genomic landscape.

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