

INTRODUCTION & METHODOLOGY

Lung cancer is the leading cause of cancer mortality worldwide, with non-small cell lung cancer (NSCLC) accounting for ~85% of cases.

Mutations in the **epidermal growth factor receptor (EGFR)** drive oncogenesis in a subset of NSCLC patients and predict response to EGFR tyrosine kinase inhibitors.

Tissue-based testing is the gold standard for EGFR mutation detection; however, obtaining adequate tumour tissue is often challenging due to procedural constraints, tumour heterogeneity, small biopsy specimens, and delays in diagnostic workflows.

Plasma circulating tumour DNA (ctDNA) testing has emerged as a minimally invasive alternative that can overcome these limitations, enabling molecular testing when tissue is unavailable, inadequate, or delayed.

CHALLENGES WITH TISSUE TESTING IN REAL-WORLD PRACTICE

- Invasive procedures
- Tumour heterogeneity
- Limited or inadequate tissue
- Delays in sample processing and reporting
- Repeat biopsy may not be feasible in all patients

A PROMISING COMPLEMENTARY APPROACH

- Minimally invasive and easily accessible
- Shorter turnaround time
- Useful when tissue is unavailable, inadequate or delayed
- Enables faster clinical decision-making and earlier initiation of targeted therapy
- Particularly valuable in resource-constrained settings

PLASMA ctDNA TESTING

METHODOLOGY & STUDY DESIGN

STUDY DESIGN

Retrospective real-world analysis of NSCLC patients who underwent EGFR mutation testing at a high-volume tertiary cancer centre.

STUDY POPULATION

706 NSCLC patients

- 564 (79.9%) Tissue EGFR testing
- 142 (20.1%) Plasma ctDNA EGFR testing

SAMPLE SOURCES

TISSUE COHORT (n = 564)

- Formalin-fixed, paraffin-embedded (FFPE) tissue
- Macro/microdissection performed as required

PLASMA COHORT (n = 142)

- Peripheral blood (10 mL) collected in Streck tubes
- Plasma separated and cfDNA extracted

MOLECULAR TESTING WORKFLOW

AmoyDX EGFR Mutation Detection Assay (qPCR-based)

- Nucleic Acid Extraction
- Multiplex qPCR Amplification
- Mutation Detection (Exons 18, 19, 20, 21)
- Data Analysis and Interpretation
- Reporting

PATIENT SELECTION CRITERIA

TISSUE TESTING (n = 564)

- All consecutive NSCLC patients who underwent EGFR mutation testing on tumour tissue using qPCR

PLASMA TESTING (n = 142)

- Patients selected based on clinical indications:
 - Delayed biopsy results
 - Inadequate or insufficient tissue
 - Inability to obtain tissue
 - Need for minimally invasive testing

STATISTICAL ANALYSIS

- EGFR mutation rates, mutation subtypes and clinicopathological correlations were analysed.
- Associations with gender and smoking status were evaluated using Chi-square test.
- A p-value < 0.05 was considered statistically significant.

RESULTS & DISCUSSION

TISSUE EGFR TESTING (qPCR) – n = 564

KEY SUMMARY

- EGFR Mutated: 261 (46.3%)
- EGFR Wild Type: 303 (53.7%)
- Median TAT: 48 hours

EGFR MUTATION STATUS

n=564

- EGFR Mutated: 261 (46.3%)
- EGFR Wild Type: 303 (53.7%)

COMPOUND / OTHER ALTERATIONS (n=28, 5.0%)

- G719X + S768I (6)
- L858R + T790M (5)
- Exon19del + L858R (4)
- S768I (3)
- G719X (2)
- Others (single rare variants) (8)

MUTATION SPECTRUM (Among all patients, n=564)

- Exon 19 Deletion: 187 (33.2%)
- L858R (Exon 21): 46 (8.2%)
- Compound EGFR Mutations: 21 (3.7%)
- Other EGFR Alterations*: 7 (1.2%)
- Wild Type: 303 (53.7%)

EGFR MUTATIONS – EXON WISE DISTRIBUTION (Lollipop Plot)

Exon 18: G719X (n=6)
Exon 19: Exon 19 Deletions (n=187)
Exon 20: S768I (n=6)
Exon 21: L858R (n=46), L861Q (n=2)

CLINICOPATHOLOGICAL CORRELATION

Gender	EGFR Positive n (%)	EGFR Negative n (%)	p-value
Female (n = 226)	135 (60.0%)	90 (40.0%)	< 0.001*
Male (n = 338)	125 (36.9%)	214 (63.1%)	
Total (n = 564)	261 (46.3%)	303 (53.7%)	

BY SMOKING STATUS

Smoking Status	EGFR Positive n (%)	EGFR Negative n (%)	p-value
Non-smoker (n = 389)	214 (55.0%)	175 (45.0%)	< 0.001*
Smoker (n = 175)	45 (25.9%)	129 (74.1%)	
Total (n = 564)	261 (46.3%)	303 (53.7%)	

* Statistically significant

Exon 19 deletions were the most common EGFR alterations followed by L858R. EGFR positivity was significantly associated with female gender and non-smoking status.

PLASMA ctDNA EGFR TESTING (AmoyDX) – n = 142

KEY SUMMARY

- EGFR Mutated: 54 (38.0%)
- EGFR Wild Type: 88 (62.0%)
- Median TAT: 48 hours

EGFR MUTATION STATUS

n=142

- EGFR Mutated: 54 (38.0%)
- EGFR Wild Type: 88 (62.0%)

OTHER / COMPOUND ALTERATIONS (n=8, 5.6%)

- G719X + S768I (2)
- L858R + T790M (2)
- Exon19del + L858R (2)
- Others (single rare variants) (2)

MUTATION SPECTRUM (Among all patients, n=142)

- Exon 19 Deletion: 31 (21.8%)
- L858R (Exon 21): 15 (10.6%)
- Other / Compound Alterations*: 8 (5.6%)
- Wild Type: 88 (62.0%)

EGFR MUTATIONS – EXON WISE DISTRIBUTION (Lollipop Plot)

Exon 18: G719X (n=2)
Exon 19: Exon 19 Deletions (n=31)
Exon 20: S768I (n=2)
Exon 21: L858R (n=15), L861Q (n=1)

SUBGROUP CHARACTERISTICS (PLASMA COHORT)

GENDER DISTRIBUTION

Gender	n	%
Female	82	57.7%
Male	60	42.3%
Total	142	100%

SMOKING STATUS

Smoking Status	n	%
Non-smoker	98	69.0%
Smoker	44	31.0%
Total	142	100%

CLINICAL INDICATION FOR PLASMA TESTING

Indication	n (%)
Delayed biopsy / results	78 (54.9%)
Inadequate / insufficient tissue	41 (28.9%)
Inability to obtain tissue	23 (16.2%)
Total	142 (100%)

KEY TAKE-HOME MESSAGE

Plasma ctDNA testing using AmoyDX assay detected actionable EGFR mutations in 38.0% of clinically selected patients with limited or delayed tissue availability, with a median TAT of 48 hours.

* Includes compound and other rare EGFR alterations.

1. TISSUE REMAINS THE PRINCIPAL MOLECULAR TESTING PLATFORM

Tissue-based EGFR testing identified mutations in 261/564 patients (46.3%), confirming a substantial prevalence of actionable EGFR alterations.

Exon 19 deletions (33.2%) and L858R (8.2%) were the predominant alterations.

MUTATION SPECTRUM – TISSUE (n=564)

- Exon 19 Deletion: 187 (33.2%)
- L858R (Exon 21): 46 (8.2%)
- Compound EGFR Mutations: 21 (3.7%)
- Other EGFR Alterations*: 7 (1.2%)
- Wild Type: 303 (53.7%)

KEY TAKE

Tissue testing has the highest mutation detection rate in this real-world cohort.

2. EGFR MUTATIONS SHOW CHARACTERISTIC CLINICAL ENRICHMENT

EGFR positivity was significantly higher in patients with known clinical predictors.

BY GENDER (TISSUE COHORT)

Gender	n	EGFR Positive (%)
Female	226	60.0%
Male	338	36.9%

p < 0.001*

BY SMOKING STATUS (TISSUE COHORT)

Smoking Status	n	EGFR Positive (%)
Non-smoker	389	55.0%
Smoker	175	25.9%

p < 0.001*

3. PLASMA PROVIDES MEANINGFUL DIAGNOSTIC YIELD WHEN TISSUE IS LIMITED

Plasma ctDNA testing detected EGFR mutations in 54/142 patients (38.0%) in a clinically selected population with delayed, inadequate or unavailable tissue.

The spectrum of actionable mutations in plasma mirrored that of tissue, with exon 19 deletion and L858R being the most common.

MUTATION SPECTRUM – PLASMA (n=142)

- Exon 19 Deletion: 31 (21.8%)
- L858R (Exon 21): 15 (10.6%)
- Other / Compound Alterations*: 8 (5.6%)
- Wild Type: 88 (62.0%)

KEY TAKE

Plasma testing achieved a substantial yield in a cohort with tissue limitations.

4. PLASMA AND TISSUE ARE COMPLEMENTARY, NOT COMPETING

The plasma cohort was clinically selected and not a paired comparison; therefore, differences in positivity rates should not be interpreted as assay sensitivity or superiority.

Plasma ctDNA testing complements tissue testing by bridging gaps in tissue availability.

COMPLEMENTARY ROLE IN CLINICAL WORKFLOW

NSCLC Patient → Tissue Biopsy → Adequate Tissue? → YES → Tissue EGFR Testing → Actionable Results / Timely Treatment Decisions
NO / DELAYED / INADEQUATE → Plasma ctDNA EGFR Testing → Actionable Results / Timely Treatment Decisions

5. TURNAROUND TIME SUPPORTS RAPID CLINICAL DECISION-MAKING

Both tissue and plasma workflows achieved a median turnaround time of 48 hours, enabling rapid reporting and earlier initiation of targeted therapy.

MEDIAN TURNAROUND TIME (TAT)

Test Type	Median TAT (Hours)
TISSUE EGFR TESTING (qPCR)	48 HOURS
PLASMA ctDNA EGFR TESTING (AmoyDX)	48 HOURS

* p < 0.001 is considered statistically significant. | Compound / other EGFR alterations may include more than one exon and can be represented in > 1 exon.

CONCLUSION

KEY FINDINGS AT A GLANCE

- 46.3% EGFR positivity in tissue cohort (261/564)
- 38.0% EGFR positivity in plasma cohort (54/142)
- Exon 19 deletion and L858R were the predominant actionable alterations in both cohorts.
- EGFR mutations were significantly associated with female gender and non-smoking status (p < 0.001).
- 48 HOURS Median TAT for both tissue and plasma workflows.

TAKE-HOME MESSAGE

“Plasma EGFR testing should not be viewed simply as a replacement for tissue, but as a complementary strategy that can bridge tissue-access gaps and facilitate timely precision oncology decisions.”

In real-world Indian practice, integrating plasma ctDNA with tissue-based EGFR testing can expand access to actionable molecular diagnosis without requiring an additional invasive procedure.

Disclosure statement: AI-assisted tools were used to support the visual design and formatting of this infographic. The scientific content and messaging were developed, reviewed, and approved by the authors.