EGFR Mutation Analysis (Droplet Digital PCR)

EGFR mutations has been identified in 50% of lung adenocarcinoma in East Asian patients. Patients with tumors harboring sensitizing EGFR mutations are responsive to 1st generation tyrosine kinase inhibitors (TKIs) such as erlotinib (Tarceva®). However, most patients receiving EGFR TKIs are susceptible to development of resistant mutations, predominately secondary point mutation in exon 20 of EGFR (T790M).

The ***EGFR* Mutation Analysis** test uses a method called digital droplet PCR (ddPCR) for ultra-sensitive detection of *EGFR* mutations, including **EGFR TKI sensitizing mutations (Exon 19 del, and Exon 21 L858R), as well as resistance mutation (Exon 20 T790M).**

The test is based on accumulated evidence that *EGFR* mutations matching those in tumours are present in bodily fluids (such as blood) in minute quantities, and that ultra-sensitive methods can be used to detect them in many instances.

Ultra-sensitivity for detection of EGFR sensitizing mutations

Analytical performance of ddPCR (EGFR sensitizing mutations)[1]

	Sensitivity	Specificity	Concordance*
Sensitizing mutations (Exon 19 del and L858R)	**95.9%**	**100%**	**98.8%**

*Concordance with tumor EGFR mutation status

ddPCR: Highest sensitivity & accuracy for EGFR T70M detection

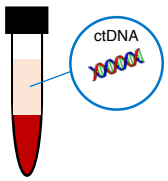
Performance of different plasma (EGFR T790M) assays[2] in AURA study

	Cobas EGFR mutation test	Therascreen EGFR ARMS PCR	ddPCR	BEAMing dPCR
Sensitivity	41%	29%	**71%**	71%
Specificity	100%	100%	**83%**	67%
Concordance with tumor T790M status	57%	48%	**74%**	70%

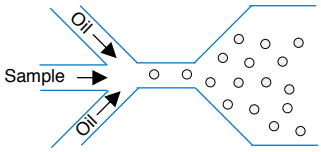
ARMS, amplification refractory mutation system; dPCR, digital polymerase chain reaction; ddPCR, Droplet Digital polymerase chain reaction; EGFR, epidermal growth factor receptor; PCR, polymerase chain reaction.

1. Mellert, Hestia, et al. "Development and Clinical Utility of a Blood-Based Test Service for the Rapid Identification of Actionable Mutations in Non–Small Cell Lung Carcinoma." The Journal of Molecular Diagnostics 19.3 (2017): 404-416.
2. Thress, Kenneth S., et al. "EGFR mutation detection in ctDNA from NSCLC patient plasma: A cross-platform comparison of leading technologies to support the clinical development of AZD9291." *Lung Cancer* 90.3 (2015): 509-515

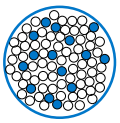
Overview of assay workflow



Extraction of circulating tumor DNA from blood



Sample partitioned into 20,000 individual droplets

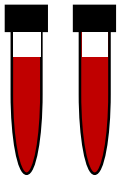


PCR reaction within individual droplets simultaneously

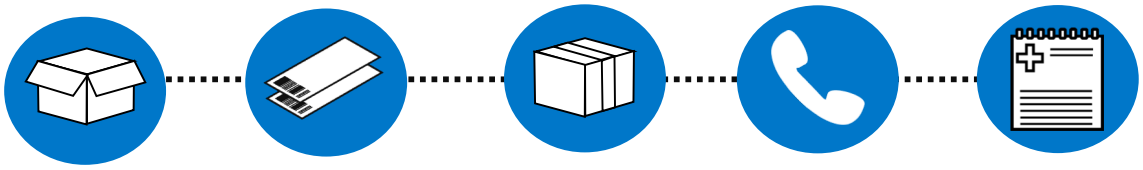
Detection of mutation-positive DNA templates

In brief, the method involves, extraction of circulating tumor DNA(ctDNA) from blood plasma, and partitioning the DNA samples such that individual DNA templates can be amplified and assessed in individual droplets. By preparing tens of thousands of these droplets for each blood sample, rare mutant-positive DNA templates can be detected reliably without the interference of more common DNA templates without mutations.

Specimen requirements and format

Sample Format		Descriptions
Specimen Format (Blood tubes)		2 x 10ml Streck Cell-free DNA BCT (fully filled).

Service workflow



Obtain specimen collection kits from our local representative

Collect specimen in accordance to required sample format

Prepare specimen as per **Shipping instructions.**

Complete sample requisition forms

Contact our local representative to arrange pick-up

We will contact you upon receipt of the specimen and send your test report back within ***5-7*** business days