Tempus xT Validation

The Tempus xT next generation sequencing assay is designed to detect actionable oncologic targets by sequencing tumor samples with matched normal saliva or blood samples, when available. The xT assay[1] covers 648 genes spanning ~3.6 Mb of genomic space. From DNA sequencing, somatic and incidentally detected germline single nucleotide variants (SNVs), insertions and deletions (indels), copy number variants (CNVs), and translocations in 22 genes are detected, along with the TERT promoter region, and 239 sites to determine microsatellite instability status. Tumor mutational burden (TMB) and microsatellite instability (MSI) status are reported. HLA Class I genotyping information is provided on xT tumor normal and tumor only reports for clinical trial matching purposes only, and not for transplantation purposes. Test results are intended to provide tumor molecular information that can be used by clinicians to help inform clinical management when patients are seeking further cancer treatment.

CAP/CLIA validation of the Tempus xT panel in its Chicago, Illinois and Durham, North Carolina laboratories focused on actionable oncologic variants. The assay requires specimens with a tumor content of 20% post macrodissection (minimum 30% for MSI status). For solid tumors, an FFPE tumor sample is sequenced along with a matched normal blood or saliva sample (when available). For circulating hematologic malignancies, a blood or bone marrow sample is sequenced. Clinical sequencing is performed to 500x depth of coverage for tumor specimens and 150x for normal specimens. Performance specifications are listed in Tables 1 and 2 below. These results establish high sensitivity and specificity for the Tempus xT assay.

The xT assay is used across a diverse set of clinical settings including leading academic centers, NCI designated cancer centers, hospital networks, and community hospitals.

TABLE 1: PERFORMANCE SPECIFICATIONS CHICAGO LAB*

Variant Type	Limit of Detection	Analytical Sensitivity	Negative Percentage Agreement
SNVs	5.0% VAF	98.2%	>99.0%
Indels	5.0% VAF	91.1%	>99.0%
Copy Number Alterations	Gain—30.0% tumor purity Loss—40.0% tumor purity	91.4%	>99.0%
Microsatellite Instability	30.0% tumor purity	90.5%	98.4%
Rearrangements	30.0% tumor purity	90.9%	>99.0%

TABLE 2: PERFORMANCE SPECIFICATIONS DURHAM LAB*

Variant Type	Limit of Detection	Analytical Sensitivity	Negative Percentage Agreement
SNVs	5.0% VAF	97.6%	>99.0%
Indels	5.0% VAF	91.4%	>99.0%
Copy Number Alterations	Gain—33.2% tumor purity Loss—38.5% tumor purity	92.7%	>99.0%
Microsatellite Instability	30.0% tumor purity	96.2%	99.6%
Rearrangements	30.0% tumor purity	96.3%	>99.0%

1 Version 4 of the Tempus xT assay. ** Solid tumor performance.*