xE Validation Summary

Tempus xE (version 2) is a whole exome next-generation sequencing assay that analyzes the entire coding region (exome) of the patient's genome. Clinical sequencing is performed to 300x depth of coverage in enhanced regions (648 genes) and 250x depth of coverage in non-enhanced regions for tumor specimens. Normal specimens are sequenced at 150x depth of coverage.

It encompasses 19,433 genes covering a ~34 Mb target region of the human genome and is optimized for formalin fixed paraffin embedded (FFPE) tissue samples. The FFPE tumor tissue is matched to a normal blood or saliva sample to ensure fidelity of somatic variant calling. The xE assay identifies actionable oncogenic variants. From DNA sequencing, somatic and incidentally detected germline single nucleotide variants (SNVs), insertions and deletions (indels), copy number losses (CNLs), and copy number gains (CNGs) are reported.

CAP/CLIA validation of the Tempus xE (version 2) assay focused on actionable oncologic variants. The assay requires specimens with tumor content of at least 40% post macrodissection. Performance specifications are listed in Table 1 below as compared to internally developed orthogonal tests. These results establish high sensitivity and specificity for the Tempus xE (version 2) assay.

xE PERFORMANCE SPECIFICATIONS—(SNVs and INDELs based on xE (version 2) to orthogonal exome panel), Copy Number Gains and Copy Number Losses based on xE (version 2) vs xT (version 4))

Variant Class	Limit of Dectection	Sensitivity	LOD
Enhanced Single Nucleotide Variants	05.0% VAF	97.9%	99.9%
Non-Enhanced Single Nucleotide Variants	10.0% VAF	93.6%	99.9%
Enhanced Insertions and Deletions	05.0% VAF	92.6%	99.9%
Non-Enhanced Insertions and Deletions	10.0% VAF	90.5%	99.9%
Copy Number Gains	30.0% Tumor Purity	85.7%	—
Copy Number Losses	50.0% Tumor Purity	97.5%	—