iC SARS-CoV-2 Test and iC (COVID-19) Validation

The Tempus iC SARS-CoV-2 and iC (COVID-19) tests detect the presence of SARS-CoV-2 RNA (the causative viral agent of the COVID-19 clinical syndrome) in upper respiratory tract specimens from people at risk for COVID-19. These assays use reverse-transcription coupled with a multiplex polymerase chain reaction (RT-PCR) targeting three SARS-CoV-2 genes: N (nucleocapsid gene), S (spike gene), and ORF1ab (open reading frame 1ab gene).

The assays include positive and negative plate level controls as well as a sample level internal control. For anterior nares (AN or nasal) samples collected at home by the patient, the iC SARS-CoV-2 Test also targets RNase P. Together, these verify the efficacy of sample collection, RNA extraction, and PCR amplification.

The laboratory workflow has been highly automated and consists of RNA extraction using the PerkinElmer Chemagic Viral DNA/RNA 300 H96 Kit on a Chemagic 360 instrument for the iC SARS-CoV-2 Test or using the Beckman Coulter RNAdvance Viral Nucleic Acid Kit on a Beckman Coulter i7 instrument for the iC (COVID-19) test (for HCP collected samples only). RT-PCR is done using the Applied Biosystems EUA TaqPath COVID-19 Combo Kit, the Applied Biosystems TaqMan SARS-CoV-2 RNase P Assay Kit (when testing home collection samples), and the Applied Biosystems QuantStudio 7 Flex Real-Time PCR system.

Validation of the Tempus iC SARS-CoV-2 and iC (COVID-19) tests was performed on upper respiratory swab specimens in our CLIA-certified, high-complexity, CAP-accredited laboratory. Accuracy and limit of detection (LOD) were determined using known or contrived positive and negative samples and dilution series. Following the Chemagic 360 extraction workflow, accuracy was determined by testing 61 known positive and negative samples collected in VTM for concordance. Positive samples showed 100% concordance and negative samples showed 96.8% concordance. Accuracy in Mawi medium was determined by testing 60 known positive and negative samples. Positive and negative samples showed 100% concordance with expected results. Accuracy in saline was determined by testing 30 contrived positive clinical replicates at high, medium and low concentrations and 30 negative replicates. Positive and negative samples showed 100% concordance with expected results. Following the Beckman Coulter i7 extraction workflow (used with HCP collected samples only), accuracy was determined by testing 60 known positive and negative samples for concordance. Positive samples showed 100% concordance and negative samples showed 100% concordance. Lastly, the test can detect 250 viral copies per mL in VTM or Mawi medium and 500 viral copies per mL in saline. Additional performance information can be found in Table 1 below.

Validation of the Tempus iC SARS-CoV-2 and iC (COVID-19) tests established a robust assay for the detection of SARS-CoV-2 RNA with high sensitivity and specificity in upper respiratory swab samples. The detection of SARS-CoV-2 RNA does not rule out co-infection with other viruses or bacteria, and negative results do not preclude an infection by SARS-CoV-2. The patient's healthcare provider should assess results along with the patient's medical information and clinical observations in order to make treatment or management decisions.

The EUA authorized test:

- has not been FDA cleared or approved;
- has been authorized by FDA under an EUA for use by the authorized laboratories;
- has been authorized only for the detection of nucleic acid from SARS-CoV-2, not for any other viruses or pathogens; and
- is only authorized for the duration of the declaration that circumstances exist justifying the authorization of emergency use of in vitro diagnostics for detection and/or diagnosis of COVID-19 under Section 564(b)(1) of the Federal Food, Drug and Cosmetic Act, 21 U.S.C. § 360bbb-3(b)(1), unless the authorization is terminated or revoked sooner.

TABLE 1: Performance specifications in upper respiratory samples

Validation assay	iC SARS-CoV-2 Test *Chemagic 360*	iC (COVID-19) *Beckman i7*
Limit of detection	250 viral copies per mL in VTM or Mawi Medium; 500 viral copies per mL in saline	250 viral copies per mL in Mawi Medium
Sensitivity	VTM: 99.9% Mawi: 99.9% Saline: 99.9%	Mawi: 99.9%
Specificity	VTM: 96.8% Mawi: 99.9% Saline: 99.9%	Mawi: 99.9%