July 24, 2024

Hologic, Inc.
Howard Liu
Regulatory Affairs Specialist
10210 Genetic Center Drive
San Diego, California 92121

Re: K233352
Trade/Device Name: Aptima HCV Quant Dx Assay
Regulation Number: 21 CFR 866.3170
Regulation Name: Nucleic Acid-Based Hepatitis C Virus Ribonucleic Acid Tests
Regulatory Class: Class II
Product Code: MZP
Dated: June 28, 2024
Received: July 1, 2024

Dear Howard Liu:

We have reviewed your section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (the Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. Although this letter refers to your product as a device, please be aware that some cleared products may instead be combination products. The 510(k) Premarket Notification Database available at https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfpmn/pmn.cfm identifies combination product submissions. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration. Please note: CDRH does not evaluate information related to contract liability warranties. We remind you, however, that device labeling must be truthful and not misleading.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Additional information about changes that may require a new premarket notification are provided in the FDA guidance documents entitled "Deciding When to Submit a 510(k) for a Change to an Existing Device" (https://www.fda.gov/media/99812/download) and "Deciding When to Submit a 510(k) for a Software Change to an Existing Device" (https://www.fda.gov/media/99785/download).

Your device is also subject to, among other requirements, the Quality System (QS) regulation (21 CFR Part 820), which includes, but is not limited to, 21 CFR 820.30, Design controls; 21 CFR 820.90, Nonconforming product; and 21 CFR 820.100, Corrective and preventive action. Please note that regardless of whether a change requires premarket review, the QS regulation requires device manufacturers to review and approve changes to device design and production (21 CFR 820.30 and 21 CFR 820.70) and document changes and approvals in the device master record (21 CFR 820.181).

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Part 801 and Part 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR Part 803) for devices or postmarketing safety reporting (21 CFR Part 4, Subpart B) for combination products (see https://www.fda.gov/combination-products/guidance-regulatory-information/postmarketing-safety-reporting-combination-products); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820) for devices or current good manufacturing practices (21 CFR Part 4, Subpart A) for combination products; and, if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR Parts 1000-1050.

Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to https://www.fda.gov/medical-devices/medical-device-safety/medical-device-reporting-mdr-how-report-medical-device-problems.

For comprehensive regulatory information about medical devices and radiation-emitting products, including information about labeling regulations, please see Device Advice (https://www.fda.gov/medical-devices/device-advice-comprehensive-regulatory-assistance) and CDRH Learn (https://www.fda.gov/training-and-continuing-education/cdrh-learn). Additionally, you may contact the Division of Industry and Consumer Education (DICE) to ask a question about a specific regulatory topic. See the DICE website (https://www.fda.gov/medical-devices/device-advice-comprehensive-regulatory-assistance/contact-us-division-industry-and-consumer-education-dice) for more information or contact DICE by email (DICE@fda.hhs.gov) or phone (1-800-638-2041 or 301-796-7100).

Sincerely,

Maria I. Garcia -S

Maria Garcia, Ph.D.
Assistant Director
Division of Microbiology Devices
OHT7: Office of In Vitro Diagnostics
Office of Product Evaluation and Quality
Center for Devices and Radiological Health

Enclosure

DEPARTMENT OF HEALTH AND HUMAN SERVICES
Food and Drug Administration

Indications for Use

Form Approved: OMB No. 0910-0120
Expiration Date: 07/31/2026
See PRA Statement below.

510(k) Number *(if known)*
K233352

Device Name
Aptima HCV Quant Dx Assay

Indications for Use *(Describe)*
The Aptima HCV Quant Dx assay is a real-time transcription-mediated amplification (TMA) test used for both detection and quantitation of hepatitis C virus (HCV) RNA in fresh and frozen human serum and plasma from HCV-infected individuals.

Plasma may be prepared in ethylenediaminetetraacetic acid (EDTA), anticoagulant citrate dextrose (ACD) solution, and plasma preparation tubes (PPT). Serum may be prepared in serum tubes and serum separator tubes (SST). Specimens are tested using the Panther system for automated specimen processing, amplification, detection, and quantitation. Specimens containing HCV genotypes 1 to 6 are validated for detection and quantitation in the assay.

The Aptima HCV Quant Dx assay is indicated for use as an aid in the diagnosis of active HCV infection in the following populations: individuals with antibody evidence of HCV infection with evidence of liver disease, individuals suspected to be actively infected with HCV antibody evidence, and individuals at risk for HCV infection with antibodies to HCV. Detection of HCV RNA indicates that the virus is replicating and, therefore, is evidence of active infection. Detection of HCV RNA does not discriminate between acute and chronic states of infection.

The Aptima HCV Quant Dx assay is also indicated for use as an aid in the management of HCV infected patients undergoing HCV antiviral drug therapy. The assay can be used to measure HCV RNA levels periodically prior to, during, and after treatment to determine sustained virological response (SVR) or nonsustained virological response (NSVR). Assay performance characteristics have been established for individuals infected with HCV and treated with certain direct-acting antiviral agents (DAA) regimens. No information is available on the assay's predictive value when other therapies are used. The results from the Aptima HCV Quant Dx assay must be interpreted within the context of all relevant clinical and laboratory findings.

The Aptima HCV Quant Dx assay is not approved for use as a screening test for the presence of HCV RNA in blood or blood products.

Type of Use *(Select one or both, as applicable)*

☒ Prescription Use (Part 21 CFR 801 Subpart D) ☐ Over-The-Counter Use (21 CFR 801 Subpart C)

CONTINUE ON A SEPARATE PAGE IF NEEDED.

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HOLOGIC®

510(k) SUMMARY
Aptima HCV Quant Dx Assay

I. SUBMITTER

Hologic, Inc.
10210 Genetic Center Drive
San Diego, CA 92121

Contact Person: Howard Liu
Regulatory Affairs Specialist
howard.liu@hologic.com
Phone: (858) 410-8853

Date Prepared: July 18, 2024

II. DEVICE

Proprietary Name of Device:	Aptima HCV Quant Dx assay
Regulation Description:	Nucleic acid-based hepatitis C virus ribonucleic acid tests
Classification Panel/Medical Specialty:	Microbiology
Regulation Number:	21 CFR 866.3170
Regulatory Class:	Class II
Product Code:	MZP

III. PREDICATE DEVICE

The predicate device is the Aptima HCV Quant Dx assay; P160023, originally approved as a Class III PMA product, reclassified to a Class II 510(k) product on November 26, 2021.

IV. DEVICE DESCRIPTION

Clearance of this pre-market application will 1) enable additional reagent on-board / off-board cycling, allowing operators to load reagents onto the Panther System 8 times instead of 5 times; and 2) update the Assay Definition Module (ADM) to detect, flag and invalidate results impacted by a faulty or flickering LED, using estimated background minimum limits. These changes do not introduce any changes to the original design, method of manufacture, assay procedure, principle of operation, mechanism of action, conditions of use or hardware of the Panther instrument, or to the results interpretation for the cleared assay.

Principles of the Procedure

The principles of the procedure are unchanged.

The Aptima HCV Quant Dx assay is a nucleic acid amplification test that uses real-time TMA technology to detect and quantitate HCV RNA for aiding diagnosis or to establish baseline viral load, as well as to measure on-treatment and post-treatment responses. The assay targets a conserved region of the HCV genome, detecting and quantitating genotypes 1, 2, 3, 4, 5, and 6. The assay is standardized against the 2nd WHO International Standard for Hepatitis C Virus (NIBSC Code 96/798).12

The Aptima HCV Quant Dx assay involves three main steps, which all take place in a single tube on the Panther system: target capture, target amplification by TMA, and detection of the amplification products (amplicon) by the fluorescent labeled probes (torches).

During target capture, viral RNA is isolated from specimens. The specimen is treated with a detergent to solubilize the viral envelope, denature proteins, and release viral genomic RNA. Capture oligonucleotides hybridize to highly conserved regions of HCV RNA, if present, in the test specimen. The hybridized target is then captured onto magnetic microparticles that are separated from the specimen in a magnetic field. Wash steps remove extraneous components from the reaction tube.

Target amplification occurs via TMA, which is a transcription-mediated nucleic acid amplification method that utilizes two enzymes, Moloney murine leukemia virus (MMLV) reverse transcriptase and T7 RNA polymerase. The reverse transcriptase is used to generate a DNA copy (containing a promoter sequence for T7 RNA polymerase) of the target sequence. T7 RNA polymerase produces multiple copies of RNA amplicon from the DNA copy template. The Aptima HCV Quant Dx assay utilizes the TMA method to amplify a portion of the 5' UTR of the HCV genome. Amplification of this region is achieved using specific primers which are designed to amplify HCV genotypes 1, 2, 3, 4, 5, and 6.

Detection is achieved using single-stranded nucleic acid torches that are present during the amplification of the target and that hybridize specifically to the amplicon in real time. Each torch has a fluorophore and a quencher. When the torch is not hybridized to the amplicon, the quencher is in close proximity of the fluorophore and suppresses the fluorescence. When the torch binds to the amplicon, the quencher is moved farther away from the fluorophore and it will emit a signal at a specific wavelength when excited by a light source. As more torches hybridize to amplicon a higher fluorescent signal is generated. The time taken for the fluorescent signal to reach a specified threshold is proportional to the starting HCV concentration. Each reaction has an internal calibrator/internal control (IC) that controls for variations in specimen processing, amplification, and detection. The concentration of a sample is determined by the Panther system software using the HCV and IC signals for each reaction and comparing them to calibration information.

Assay Components

The reagents required to perform the Aptima HCV Quant Dx assay are available in 100-test kits. The kits are packaged in 3 boxes containing 11 reagents which are required for sample processing. A description of the components that are required to perform the Aptima HCV Quant Dx assay are detailed in **Table 1.** In addition, two ancillary kits are required to run the assay (**Table 2**).

Table 1: Reagents Required to Perform the Aptima HCV Quant Dx Assay

Box	Components Description
1	Amplification Reagent
	Enzyme Reagent
	Promoter Reagent
	Amplification Reconstitution Solution
	Enzyme Reconstitution Solution
	Promoter Reconstitution Solution
	Target Capture Reagent
2	Positive Calibrator
3	Negative Control
	Low Positive Control
	High Positive Control

Table 2: Ancillary Kits Required to Perform the Aptima HCV Quant Dx Assay

Panther Run Kit for Real Time Assays (for real time assays only)
Panther System Run Kit (when running non-real time-TMA assays in parallel with real time-TMA assays)

Instrumentation

The Aptima HCV Quant Dx assay has been designed for and validated on the Panther system. The Panther system is an integrated hardware and software system that together with the Aptima HCV Quant Dx assay fully automates all the steps necessary to perform the assay from sample preparation through amplification of nucleic acid, detection, data reduction and amplicon inactivation.

V. INDICATIONS FOR USE

Intended Use

The intended use for the Aptima HCV Quant Dx Assay will remain unchanged:

The Aptima HCV Quant Dx assay is a real-time transcription-mediated amplification (TMA) test used for both detection and quantitation of hepatitis C virus (HCV) RNA in fresh and frozen human serum and plasma from HCV-infected individuals.

Plasma may be prepared in ethylenediaminetetraacetic acid (EDTA), anticoagulant citrate dextrose (ACD) solution, and plasma preparation tubes (PPT). Serum may be prepared in serum tubes and serum separator tubes (SST). Specimens are tested using the Panther® system for automated specimen processing, amplification, detection, and quantitation. Specimens containing HCV genotypes 1 to 6 are validated for detection and quantitation in the assay.

The Aptima HCV Quant Dx assay is indicated for use as an aid in the diagnosis of active HCV infection in the following populations: individuals with antibody evidence of HCV infection with evidence of liver disease, individuals suspected to be actively infected with HCV antibody evidence, and individuals at risk for HCV infection with antibodies to HCV. Detection of HCV RNA indicates that the virus is replicating and, therefore, is evidence of active infection. Detection of HCV RNA does not discriminate between acute and chronic states of infection.

The Aptima HCV Quant Dx assay is also indicated for use as an aid in the management of HCV infected patients undergoing HCV antiviral drug therapy. The assay can be used to measure HCV RNA levels periodically prior to, during, and after treatment to determine sustained virological response (SVR) or nonsustained virological response (NSVR). Assay performance characteristics have been established for individuals infected with HCV and treated with certain direct-acting antiviral agents (DAA) regimens. No information is available on the assay's predictive value when other therapies are used. The results from the Aptima HCV Quant Dx assay must be interpreted within the context of all relevant clinical and laboratory findings.

The Aptima HCV Quant Dx assay is not approved for use as a screening test for the presence of HCV RNA in blood or blood products.

VI. COMPARISON OF TECHNOLOGICAL CHARACTERISTICS WITH THE PREDICATE DEVICE

A comparison of the Aptima HCV Quant Dx assay with the updated 7.2.9 system software to the predicate device, the Aptima HCV Quant Dx assay (P160023, originally approved as a Class III PMA product, reclassified to a Class II 510(k) product on November 26, 2021), is summarized in **Table 3** (similarities) and **Table 4** (differences).

Table 3: Similarities Between Predicate Device and Subject Device

Item	Predicate Device: Aptima HCV Quant Dx assay (P160023)	Subject Device: Aptima HCV Quant Dx assay
Technology Principle of Operation	Target Capture (TC), Transcription-Mediated Amplification (TMA)	Same
Platform	Automated Panther System	Same
Assay Targets	Hepatitis C RNA	Same
Assay Results	Qualitative & quantitative	Same
Function	Detection of RNA from Hepatitis C	Same
Intended Use	The Aptima HCV Quant Dx assay is a real-time transcription-mediated amplification (TMA) test used for both detection and quantitation of hepatitis C virus (HCV) RNA in fresh and frozen human serum and plasma from HCV-infected individuals. Plasma may be prepared in ethylenediaminetetraacetic acid (EDTA), anticoagulant citrate dextrose (ACD) solution, and plasma preparation tubes (PPT). Serum may be prepared in serum tubes and serum separator tubes (SST). Specimens are tested using the Panther® system for automated specimen processing, amplification, detection, and quantitation. Specimens containing HCV genotypes 1 to 6 are validated for detection and quantitation in the assay. The Aptima HCV Quant Dx assay is indicated for use as an aid in the diagnosis of active HCV infection in the following populations: individuals with antibody evidence of HCV infection with evidence of liver disease, individuals suspected to be actively infected with HCV antibody evidence, and individuals at risk for HCV infection with antibodies to	Same

Item	Predicate Device: Aptima HCV Quant Dx assay (P160023)	Subject Device: Aptima HCV Quant Dx assay
	HCV. Detection of HCV RNA indicates that the virus is replicating and, therefore, is evidence of active infection. Detection of HCV RNA does not discriminate between acute and chronic states of infection. The Aptima HCV Quant Dx assay is also indicated for use as an aid in the management of HCV infected patients undergoing HCV antiviral drug therapy. The assay can be used to measure HCV RNA levels periodically prior to, during, and after treatment to determine sustained virological response (SVR) or nonsustained virological response (NSVR). Assay performance characteristics have been established for individuals infected with HCV and treated with certain direct-acting antiviral agents (DAA) regimens. No information is available on the assay's predictive value when other therapies are used. The results from the Aptima HCV Quant Dx assay must be interpreted within the context of all relevant clinical and laboratory findings. The Aptima HCV Quant Dx assay is not approved for use as a screening test for the presence of HCV RNA in blood or blood products.	

Table 4: Differences Between Predicate Device and Subject Device

Item	Predicate Device: Aptima HCV Quant Dx assay (P160023)	Subject Device: Aptima HCV Quant Dx assay
Reagents can be loaded onto the Panther system	5 times	8 times
Results impacted by faulty or flickering LED in Panther instrument	Read as normal, potentially producing incorrect results	Marked as invalid results

VII. PERFORMANCE DATA

Data provided in support of the substantial equivalence determination consist of the following validation activities:

- Intended Use Testing
- Assay Verification
- Software Verification and Validation

Intended Use Testing

Results from the Aptima HCV Quant Assay on Panther and Panther Fusion System Software 7.2.9 were compared to those from the previous software version, 7.2.7 (reported to the FDA in the 2022 PMA annual report for the Aptima HCV Quant Dx Assay, P160023/R004).

The results met established acceptance criteria, confirming that assay performance is equivalent to the prior comparators, and demonstrating that the system is capable of performing the Aptima HCV Quant Dx Assay version 5.3.5.1 on the Panther and Panther Fusion System Software 7.2.9.

Sample Type	Target Conc (log IU/mL)***	Valid N	Expected Result	Allowable Number of Outliers	Standard Deviation*	Result	Met?
QC Control A	Negative	120	Not Detected	1 False positive****	N/A	1 false positive	Met
QC Control B	2.16	40	±0.5 logs from Value Assignment**	1/30	≤ 0.25 log copy*	100% Positivity Ave diff between expected and recovered is -0.05 log IU/mL and largest SD is 0.07 log IU/mL	Met
QC Control D	6.52	40	±0.5 logs from Value Assignment**	0/25	≤ 0.25 log copy	100% Positivity Ave diff between expected and recovered is -0.12 log IU/mL and largest SD is 0.03 log IU/mL	Met
QC Panel S	1.13	80	95% Positivity	N/A	N/A	100% Positivity	Met

* Outliers may be removed from calculation of Standard Deviation
** ±0.50 logs from Target Concentration if Value Assignment is unavailable
*** Using conversion factor of 4.4 HCV copies/IU
**** 95% specificity with 95% confidence interval

Assay Verification (5 to 8 reagent reload change)

Aptima HIV-1 Quant, Aptima HCV Quant Dx and Aptima HBV Quant Assay reagents were subjected to at least 72 cumulative hours on the Panther system with the reagents uncapped. Testing included 5-10 cycles of reagent storage between the Panther system and the assigned reagent storage temperature of 2-8°C. Assay testing was performed on Day 0 and after cycling. Five cycles are the current maximum of cycles allowed for each assay. Testing included at least one reagent lot of each assay stored on-board 1 Panther system at the environmental extremes for reagent evaporation loss (30°C and 20% relative humidity). Performance was evaluated using prepared virus and armored RNA (aRNA) spiked into negative human serum matrix (BI0052).

Aptima HBV and HCV reagents successfully completed 10 on and off on-board cycles with a cumulative 72 hours on board stability.

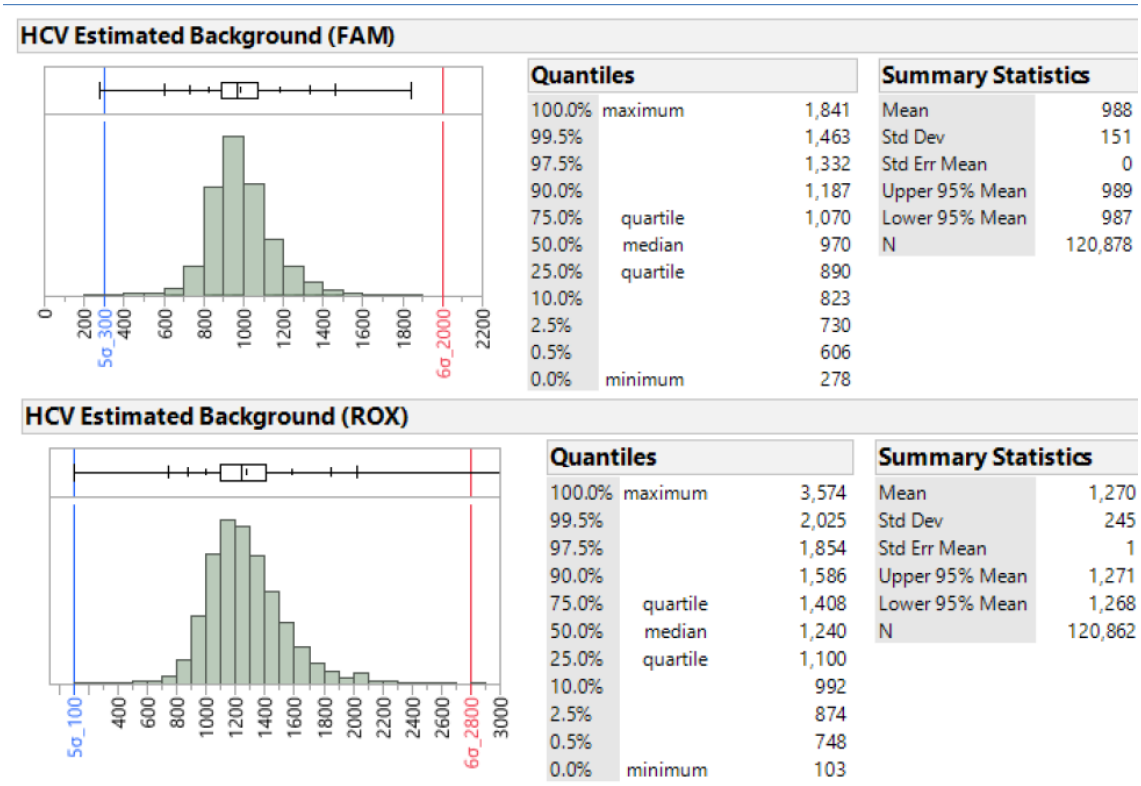
HCV 3.3 log IU/mL					
Kit Cycles	**Total Hours**	**Cycle Avg LogIU/mL**	**Cycle Std Dev**	**Baseline Avg LogIU/mL**	**Diff (Cycle-Baseline)**
5	35	3.22	0.05	3.20	0.02
10	72	3.28	0.06	3.10	0.18

HCV 7.0 log IU/mL					
Kit Cycles	**Total Hours**	**Cycle Avg LogIU/mL**	**Cycle Std Dev**	**Baseline Avg LogIU/mL**	**Diff (Cycle-Baseline)**
5	35	7.45	0.15	7.56	-0.11
10	72	7.30	0.07	7.56	-0.26

Assay Verification (Flickering LED process control change)

Fluorescent curve files and logs from 381 field instruments and 445,910 unique test orders from March 2018 to March 2022 were analyzed. Test orders with outlier estimated background values were excluded by specific root-cause analysis where applicable. Incomplete fluorescent curve files due to a date split were excluded from analysis.

HCV Estimated Background distributions were analyzed by each channel, FAM and ROX as seen below. Data were approximately normal for each channel. Proposed estimated background limits would be 5σ or five standard deviations from the mean, respectively. One test order was flagged by FAM estimated background limit and was also flagged by QNS; this test order was considered a true fault.



This analysis supports the implementation of the proposed Estimated Background minimum limits as seen in Table 1 for HIV, HCV, HBV, and HSV Panther viral assays. New estimated background limits will increase fault detection for these assays.

	FAM	HEX	ROX
HIV	200	1,000	150
HCV	300	N/A	100
HBV	300	200	100
HSV	300	150	100

Software Verification and Validation

SW V&V testing was performed to ensure that the Panther and Panther Fusion System Software, Version 7.2.9 conforms to user needs and intended uses, and that the requirements implemented through software are consistently fulfilled. Software V&V testing of applicable software-specific Product Requirements Document (PRD), Software Requirement Specifications (SRS), and Product Specification Document (PSD) requirements, including regression testing, as applicable, were performed, with the results summarized below. All anomalies generated as part of this testing were reviewed in Defect Review Board (DRB) meetings in accordance with the Software Anomaly Management Work Instruction, 04-03-21-001-WI.

Requirements	Pass	Fail
PRM Requirements	23	0
PRD Requirements	29	0
SRS Requirements	210	0
PSD Requirements	4	0
Regression testing of Anomalies Addressed and Implemented Features and Change Requests	12	0

The reporting and assessment of anomalies was conducted in accordance with the Software Anomaly Management Work Instruction, 04-03-21-001-WI and Product Safety Risk Management Procedure, 04-03-12-SOP. Each software anomaly was assessed for impact to patient and user safety. The assessment determined there are no known software anomalies that exceed the severity level of "Negligible". The following table summarizes the total number of identified anomalies associated with Panther and Panther Fusion System Software, Version 7.2.9.

Severity Level	Definition	Total
Catastrophic	Directly results in patient or user death	0
Critical	Directly results in irreversible injury (permanent impairment or life-threatening injury)	0
Serious	Results in reversible injury or impairment requiring professional medical intervention	0
Minor	Results in temporary injury or impairment not requiring professional medical intervention or blood supply delay or treatment delay	0
Negligible	Results in discomfort/no impact	12

It has been concluded from this testing that the Panther and Panther Fusion System Software provides safe and effective performance with the essential features for the end user to meet the intended use.

SW V&V testing was also performed for the Panther Aptima HCV ADM on Panther System Software Version 7.2.9. As above, system Validation activities included testing of applicable requirements or specifications from the Product Requirements Document (PRD), Product Risk Mitigator (PRM/ FRM), Product Specification Document (PSD) and Software Requirement Specification Document (SRS) as well as testing of modifications associated with hardware, anomaly fixes and/or Software Change Requests (SCR).

There were no anomalies identified during the V&V of the Panther Aptima HCV ADM on Panther System Software Version 7.2.9.

It has been concluded from the overall testing that the Panther Aptima HCV assay on the Panther System Software Version 7.2.9 meets the intended use and safety requirements.

Additional Studies

The FDA issued a hold letter on January 3, 2024 with a request for additional information in two parts as described below, with Hologic responses following:

1. **Intended Use Testing**

- An Intended Use Testing study using a minimum of four (4) clinical specimens including: three (3) specimens/samples with HCV RNA concentration around 3xLLoQ (30 IU/ml) and one (1) HCV RNA negative clinical specimen. Samples may be prepared by diluting a high positive sample in negative clinical matrix. FDA recommends testing 10 replicates of each sample per run, using previous Aptima HCV software version 5.2.5.1 and the updated version 5.3.5.1.
- A revised study report including a clear description of the study and the nature of samples used (e.g., natural or contrived), data analysis, summary of the results and conclusions, and line data in Excel format.

The requested study was carried out, Aptima HCV Assay Software versions 5.2.5.1 and 5.3.5.1 successfully completed and met all acceptance criteria with low level clinical panel. The Aptima HCV Assay Software version 5.2.5.1 and Aptima HCV Assay Software version 5.3.5.1 both resulted in 100% not detected for the negative clinical matrix panel. The Aptima HCV Assay Software version 5.2.5.1 and Aptima HCV Assay Software version 5.3.5.1 both resulted in 100% positivity for the 30 IU/mL (low positive) clinical positive panel. Acceptance criteria for this panel is positivity only and has no accuracy or precision specifications. The requested files are included in this supplement.

2. On-Board Stability (OBS) and Reagent Cycling

To support the additional reagent on-board/off-board cycling, please provide the following:

- An OBS and reagent cycling study including a minimum of one negative sample, a sample around 3xLLoQ: 30 IU/ml, and a moderate positive or high positive sample. Please include a minimum of 5 replicates per sample for each timepoint with more replicates tested at baseline to establish a robust baseline. Please perform the testing using the updated Panther system software version 7.2.9 and Aptima HCV software version 5.3.5.1 or provide a justification for why software changes would not impact the reagent stability results.
- A revised study report, including a detailed study description and the nature of samples used (e.g., natural or contrived), data analysis, summary of the results, line data in Excel format, and conclusions clearly stating the supported OBS and reagent cycling claim. Note, in order to support a claim of 8 times reagent cycling, please evaluate at minimum, the number of cycles previously claimed (5 cycles), the number of cycles currently claimed (8 cycles) and an additional number of cycles that goes beyond the one currently claimed (9 to 10 cycles) using the same kit when possible. This ensures that the reagent stability does not change one time point after the desired claim.

The requested study was carried out, and Aptima reagents successfully completed five, eight, and ten on and off on-board cycles with a cumulative 72 hours on board stability and met all acceptance criteria.

VIII. CONCLUSIONS

The validation study results demonstrate that the Aptima HCV Quant Dx assay on the 7.2.9 Panther system software performs comparably to the predicate device and supports a substantial equivalence decision.