



## Multicenter clinical evaluation of alinity m HCV assay performance

Stéphane Chevaliez, Francesco Onelia, Monia Pacenti, Emily Goldstein, Juan-Carlos Galán, Laura Martínez-García, Alba Vilas, Allison Glass, Leana Maree, Maria Krügel, et al.

### ► To cite this version:

Stéphane Chevaliez, Francesco Onelia, Monia Pacenti, Emily Goldstein, Juan-Carlos Galán, et al.. Multicenter clinical evaluation of alinity m HCV assay performance. Journal of Clinical Virology, 2020, 129, pp.104531 -. <10.1016/j.jcv.2020.104531>. <hal-03491380>

**HAL Id: hal-03491380**

**<https://hal.science/hal-03491380v1>**

Submitted on 18 Jul 2022

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons CC BY-NC 4.0 - Attribution - Non-commercial use - International License

**Manuscript title:** Multicenter Clinical Evaluation of Alinity m HCV Assay Performance

**Authors:** Stéphane Chevaliez<sup>a,b,\*†</sup>, Francesco Onelia<sup>c†</sup>, Monia Pacenti<sup>d</sup>, Emily Goldstein<sup>e</sup>, Juan-Carlos Galán<sup>f</sup>, Laura Martínez-García<sup>f</sup>, Alba Vilas<sup>g</sup>, Allison Glass<sup>h</sup>, Leana Maree<sup>h</sup>, Maria Krügel<sup>h</sup>, Robert Ehret<sup>i</sup>, Heribert Knechten<sup>j</sup>, Patrick Braun<sup>j</sup>, Gudrun Naeth<sup>j</sup>, Sara Bonanzinga<sup>k</sup>, Kathy Jackson<sup>k</sup>, Klara Abravaya<sup>l</sup>, Jens Dhein<sup>m</sup>, Shihai Huang<sup>l</sup>, Ajith M. Joseph<sup>l</sup>, Danijela Lucic<sup>l</sup>, Natalia Marlowe<sup>l</sup>, Michael J. Palm<sup>l</sup>, Karin Pfeifer<sup>m</sup>, Dan Toolsie<sup>l</sup>, Birgit Reinhardt<sup>m</sup>, Martin Obermeier<sup>i</sup>, Rory Gunson<sup>e</sup>

**Author affiliations:**

<sup>a</sup> National Reference Center for Viral Hepatitis B, C, and D, Department of Virology, Hôpital Henri Mondor, Université Paris-Est, Créteil, France

<sup>b</sup> INSERM U955, Créteil, France

<sup>c</sup> Università di Padova, Padua, Italy

<sup>d</sup> Azienda Ospedaliera di Padova, Padua, Italy

<sup>e</sup> West of Scotland Specialist Virology Centre, Glasgow, UK

<sup>f</sup> Servicio de Microbiología. Hospital Universitario Ramón y Cajal and Instituto Ramón y Cajal de Investigación Sanitaria (IRYCIS) and CIBER en Epidemiología y Salud Pública (CIBERESP), Madrid, Spain

<sup>g</sup> Laboratori de Referència de Catalunya, El Prat de Llobregat, Spain

<sup>h</sup> Lancet Laboratories, Johannesburg, South Africa

<sup>i</sup> Medizinisches Infektiologiezentrum Berlin, Germany

<sup>j</sup> Laboratory Dr. Knechten, Medical Center for HIV and Hepatitis, Aachen, Germany

<sup>k</sup> Victorian Infectious Diseases Reference Laboratory, Royal Melbourne Hospital, at the  
Peter Doherty Institute for Infection and Immunity, Victoria, 3000, Australia

<sup>l</sup> Abbott Molecular, Des Plaines, IL, USA

<sup>m</sup> Abbott GmbH, Wiesbaden, Germany

**\* Corresponding author:** Stéphane Chevaliez, Department of Virology, Hôpital Henri  
Mondor, 51 avenue du Maréchal de Lattre de Tassigny, 94010 Créteil, France. Tel: +33-  
1-4981-2828; Fax: +33-1-4981-2839. E-mail: [stephane.chevaliez@aphp.fr](mailto:stephane.chevaliez@aphp.fr)

<sup>†</sup>These authors contributed equally to this work.

**Abstract word count:** 250 words

**Article word count:** 2494 words

## 1   **Introduction**

2   The World Health Organization (WHO) estimates that 71 million people worldwide are  
3   chronically-infected with hepatitis C virus (HCV), and nearly 600,000 individuals die  
4   each year from complications of HCV, primarily cirrhosis and hepatocellular carcinoma  
5   (HCC) [1]. Hepatitis C virus is the leading virus-related cause of HCC in North America,  
6   Europe, Japan, parts of central Asia, and northern Africa and the Middle East [2].  
7   Despite the development of highly active, all-oral, well-tolerated, short-duration drugs  
8   with high cure rates (>95%), chronic HCV infection remains a global public health  
9   concern. The endpoint of therapy is sustained virologic response (SVR), defined as  
10   undetectable HCV RNA in serum or plasma 12 or 24 weeks after the treatment  
11   cessation. SVR is associated with reduced liver inflammation, fibrosis regression, and  
12   increased quality of life and overall survival.

13         For detection and quantification of HCV RNA in clinical practice, sensitive and  
14   accurate nucleic acid technologies (NATs) are recommended by international clinical  
15   practice guidelines [3, 4]. HCV RNA assays are now widely used in clinical virology  
16   laboratories for both diagnosis and treatment monitoring, and are partly or fully  
17   automated. With a broad range of linear quantification [up to  $10^8$  international units per  
18   milliliter (IU/mL)], a limit of detection (LOD) in the order of 15 IU/mL or less, and an  
19   identical lower limit of quantification (LLOQ), NATs for HCV RNA are well suited to meet  
20   clinical needs [5].

21         The Alinity™ m HCV assay (Abbott Molecular, Des Plaines, IL, USA) is a  
22   polymerase chain reaction (PCR) dual-probe assay that makes use of the fully  
23   automated Alinity m system, which eliminates the need for batch processing of samples  
24   and automates all aspects of nucleic acid testing from sample loading to result

reporting. With the Alinity m system, the first 12 HCV assay results are obtained in less than 2 hours, with 12 additional results reported every 16 minutes thereafter. Here, we examined the performance of the new Alinity m HCV assay compared to three different commercially available HCV assays for accurate HCV RNA detection and quantification in a very large series of patients chronically infected with different HCV genotypes.

## **Materials and Methods**

### *Clinical specimens and study sites*

Serum (n=406) and plasma (n=1401) samples from patients with chronic HCV infection were collected at nine health care sites in Europe, South Africa, and Australia (Azienda Ospedaliera di Padova, Padua, Italy; Hôpital Universitaire Henri Mondor, Créteil, France; Hospital Universitario Ramón y Cajal, Madrid, Spain; Laboratori de Referència de Catalunya, El Prat de Llobregat, Spain; Lancet Laboratories, Johannesburg, South Africa; Medizinisches Infektiologiezentrum Berlin, Germany; Laboratory Dr. Knechten, Aachen, Germany; Victorian Infectious Diseases Reference Laboratory, Melbourne, Australia; West of Scotland Specialist Virology Centre, Glasgow, UK).

Genotype information based on phylogenetic analysis of a portion of the NS5B gene using a deep sequencing-based assay (Sentosa® SQ HCV Genotyping Assay, Vela Diagnostics, Singapore) or an in-house Sanger method as previously described [6] was available for 375 clinical specimens: 214 genotype 1, 35 genotype 2, 59 genotype 3, 40 genotype 4, 2 genotype 5, and 10 genotype 6. The remaining 15 clinical samples were from patients infected with a 2k/1b (n=7) or 1b/3a (n=3) recombinant strain or a mixed 1a plus 3a infection (n=5).

The study was performed in accordance with the principles of Good Clinical Practice and conducted in adherence with the Declaration of Helsinki. Only residual samples for routine HCV testing were used. All samples were anonymized by assigning an identification number without any patient information to each sample prior to the start of the study.

#### *HCV RNA detection and quantification*

Clinical specimens were tested with the Alinity m HCV assay and the commercially available HCV assay used for routine testing at each study site. Comparator assays were based on either real-time PCR [RealTime HCV (Abbott Molecular) or cobas® HCV Test (Roche Molecular Systems, Pleasanton, CA)] or transcription-mediated amplification [TMA; Aptima® HCV Quant Dx assay (Hologic Inc., San Diego, CA)].

Alinity m HCV assay: In a fully automated process, HCV RNA was isolated on the Alinity m System from 600 µL of plasma or serum according to the manufacturer's instructions. The fluorescence curves were analyzed by the Alinity m system software, versions 1.2, 1.3, or 1.3.1. The dynamic range of quantification of the Alinity m HCV assay is 12 to  $1 \times 10^8$  IU/mL (1.1 to 8.0 Log IU/mL) with an LOD of 5.11 IU/mL in plasma and serum.

RealTime HCV assay: HCV RNA was processed from 500 µL of plasma or serum using the *m2000sp*™ and the *m2000rt* analyzers for automated sample extraction and real-time PCR amplification and detection, according to the manufacturer's instructions. The data were analyzed with Abbott RealTime *m2000rt* software, version 8.1. The dynamic range of quantification of this assay is 12 to  $1 \times 10^8$

IU/mL (1.1 to 8.0 Log IU/mL), with an LOD of 10.5 IU/mL in plasma and 7.2 IU/mL in serum.

cobas HCV assay: HCV RNA was extracted, purified and amplified from 500  $\mu$ L of plasma or serum on the cobas 6800 System according to the manufacturer's instructions (Roche Molecular Systems). The dynamic range of quantification is 15 to  $1 \times 10^8$  IU/mL (1.2 to 8.0 Log IU/mL), with an LOD of 8.5 IU/mL in plasma and 9.6 IU/mL in serum.

Aptima HCV Quant Dx assay: HCV RNA was isolated from 500  $\mu$ L of plasma using the Panther® analyzer system according to the manufacturer's instructions. The dynamic range of quantification of Aptima HCV Quant Dx is 10 to  $1 \times 10^8$  IU/mL (1.0 to 8.0 Log IU/mL), with an LOD of 4.3 IU/mL in plasma.

The conserved 5' untranslated region (5' UTR) is the target region of the primers and probes in all four assays, respectively.

#### *Analytical Assessment of Alinity m HCV – Precision, Detectability and Reproducibility*

Assay precision and detectability were evaluated across 4 study sites by using a non-commercial panel, consisting of different concentrations of HCV RNA and generated by dilution of HCV-positive clinical samples in normal human plasma (Exact Diagnostics, Fort Worth, TX, USA). Each panel contained a predetermined amount of HCV RNA adjusted based on the RealTime™ HCV assay (Abbott Molecular). The nine panel members contained target concentrations of  $1 \times 10^2$  IU/mL (2.00 Log IU/mL), 25 IU/mL (1.40 Log IU/mL) and 10 IU/mL (1.00 Log IU/mL) for each HCV RNA genotype 1a, 1b, and 3a, respectively. Mean, standard deviation (SD) and percent coefficient of

deviation (% CV) were evaluated at concentrations >10 IU/mL and detectability was evaluated at 10 IU/mL.

Reproducibility was assessed by evaluating assay controls from multiple reagent lots used across all testing sites. Overall mean, SD and CV of each assay's quality controls (QC) were evaluated.

### *Statistical analysis*

Descriptive statistics are reported as means  $\pm$  SD. Relationships between quantitative variables were analyzed by Deming regression. Differences in quantification between the assays were evaluated by Bland Altman analysis. All statistical analyses were performed using PC SAS version 9.3 (SAS, Cary, NC, USA).

## **Results**

### *Linear quantification by Alinity m HCV*

The linearity of Alinity m HCV assay was assessed by testing serial dilutions of a serum specimen collected from a patient infected with genotype 2, which contained  $8.30 \times 10^6$  IU/mL (6.90 Log IU/mL) HCV RNA. To prepare 8 diluted concentrations, with the lowest one near the LLOQ of the assay, a serial dilution with a dilution factor of 4.2 was performed. Each dilution was tested in triplicate in the same run. Means and ranges were compared with the theoretical HCV RNA levels. The expected difference in quantitation between the concentration levels was 0.62 Log IU/mL. The observed differences between subsequent dilutions were comparable and compatible with the expected values ( $0.81 \pm 0.12$ ;  $0.62 \pm 0.11$ ;  $0.72 \pm 0.06$ ;  $0.57 \pm 0.00$ ;  $0.85 \pm 0.05$ ;  $0.53 \pm$



0.17;  $0.64 \pm 0.08$  Log IU/mL). Figure 1 shows the measured values obtained with the serial dilutions compared to the expected values (correlation coefficient,  $r=1.00$ , linear regression equation,  $y = 0.9858x + 0.0016$ ).

#### *Precision, detectability and reproducibility of Alinity m HCV*

Precision was evaluated by testing a panel consisting of dilutions of clinical samples with 100 IU/mL (2.00 Log IU/mL) and 25 IU/mL (1.40 Log IU/mL) HCV RNA of genotype 1a, 1b, or 3a. As shown in Table 1, the coefficients of variation varied from 6.6% to 13.5% for Alinity m. At a concentration near the LLOD, the Alinity m HCV assay exhibited a high level of detectability (>98%). Reproducibility was characterized by a coefficient of variation of 4.3% for the low positive control (LPC) and of 1.7% for the high positive control (HPC) across the three different HCV control lots used during the study (Table 1).

#### *Clinical performance of Alinity m HCV assay*

In total, 1807 samples from HCV-seropositive individuals were retrospectively tested with Alinity m HCV assay at the nine participating study sites. Results were compared to previous routine test results obtained by using one of the three comparator assays: RealTime HCV assay (n=1505), cobas HCV assay (n=186), and Aptima HCV Quant Dx assay (n=116).

Of the 1505 samples tested by the Alinity m HCV and Abbott RealTime HCV assays, 883 fell within the dynamic range of both assays (Table 2). Figure 2 and Table 3 show the relationships between HCV RNA levels quantified with both assays. A strong correlation between the two assays was found ( $r = 0.98$ ; Deming regression equation,  $y$

= 1.00 x + 0.16). A weak bias across the quantitative range of the two assays was observed using Bland Altman analysis ( $0.13 \pm 0.283$  Log IU/mL; Table 3), indicating no major difference in quantification according to the HCV RNA level. In 95.4% (842/883) samples, the difference between the assays was within  $\pm 1.96$  times the SD of the mean bias. Of the remaining samples, 1.7% (15/883) were measured higher and 2.9% (26/883) measured lower with the Alinity m HCV assay.

Of the 186 samples tested by the Alinity m HCV and cobas HCV assays, 163 fell within the dynamic range of both assays (Table 4). A strong correlation between the two assays was found ( $r = 0.96$ ; Deming regression equation,  $y = 0.90 x + 0.48$ ; Figure 3, Table 3). Bland Altman analysis resulted in a negligible bias across the quantitative range of the two assays ( $-0.01 \pm 0.454$  Log IU/mL; Table 3). No major difference in quantification was observed according to the HCV RNA level. In 94.1% (175/186) samples, the difference between the assays was within  $\pm 1.96$  times the SD of the mean bias. Of the remaining samples, 4.8% (9/186) were higher and 1.1% (2/186) lower with the Alinity m HCV assay.

Of the 116 samples tested by the Alinity m HCV and Aptima HCV Quant Dx assays, 18 fell within the dynamic range of both assays (Table 5). A strong correlation between the two assays was found ( $r = 0.99$ ; Deming regression equation,  $y = 1.02 x + 0.02$ ; Figure 4, Table 3). A weak bias of HCV RNA level was observed by Bland Altman analysis across the quantitative range of the two assays ( $0.14 \pm 0.232$  Log IU/mL; Table 3). No major difference of quantification was observed according to the HCV RNA level as all samples showed a difference between the assays within  $\pm 1.96$  times the SD of the mean bias.

### *Influence of HCV genotype on HCV RNA quantification*

The influence of the HCV genotype on HCV RNA quantification was assessed by comparing results of the Alinity m HCV and RealTime HCV assays of a subset of serum and plasma samples from 375 patients infected with HCV genotypes 1 to 6. A strong relationship was found between HCV RNA levels measured in the same specimens with the Alinity m HCV assay and RealTime HCV assay, regardless of the HCV genotype (data not shown).

### **Discussion**

In this international multicenter study evaluating a large number of clinical specimens, we showed that the new Alinity m HCV assay accurately quantifies HCV RNA levels. Results indicated that there is no impact of the HCV genotype on assay performance based on previous studies [7]. The performance of the Alinity m HCV assay was comparable to several HCV assays that are currently widely used in clinical practice, including the RealTime HCV assay and the cobas HCV assay.

An excellent correlation was found between the Alinity m HCV assay and comparator HCV assays, with correlation coefficients  $\geq 0.96$ . We observed only a low difference in HCV RNA levels measured with the Alinity m HCV assay, with an overall bias ranging from -0.01 to 0.14 log IU/mL. Overall, more than 95% of the quantifiable patient results with the Alinity m HCV assay were less than  $\pm 1.96$  SD of the mean bias different from the respective comparator assays (94%-100%). The observed low differences in performance will most likely have no implications for clinical practice.

A larger number of specimens was observed with detectable HCV RNA < LOQ by either RealTime HCV or Aptima HCV Quant Dx compared to undetectable HCV RNA

190 results with Alinity m HCV (Tables 2 and 5). One hypothesis will be additional free/thaw  
191 cycles of specimens prior to testing with Alinity m HCV compared to testing with  
192 RealTime HCV. A higher detection rate obtained with Aptima HCV compared to Alinity  
193 m HCV could be due to the lower LOQ of 10 IU/mL of the Aptima HCV assay compared  
194 to 12 IU/mL of the Alinity m HCV assay.

195         This study has several limitations. The small proportion of clinical specimens  
196 containing genotypes 5 and 6 tested reflects the HCV genotype distribution in Europe.  
197 Thus, although our results strongly suggest that the performance of the Alinity m HCV  
198 assay will be high for all genotypes, further studies are warranted. The utility of the  
199 Alinity m HCV assay for longitudinal HCV RNA monitoring in patients receiving direct-  
200 acting antiviral (DAA) therapy is also not known. Unfortunately, no clinical data were  
201 available and the proportion of clinical samples collected from patients receiving  
202 antiviral treatment was unknown. However, the large number of clinical specimens  
203 (16.9%) with detectable but nonquantifiable HCV RNA by any test were probably  
204 collected in patients receiving antiviral therapy (week 4 or end of therapy). Finally,  
205 although we tested more than 100 samples with the Aptima HCV Quant Dx assay, the  
206 number of samples quantified with both the Alinity m and Aptima HCV assays was  
207 relatively small (18 samples).

208         In conclusion, with this first field evaluation of the Alinity m HCV assay, we  
209 showed that the newly developed real-time PCR-based Alinity m HCV assay is  
210 sensitive, reproducible, and accurately quantifies HCV RNA levels in serum and plasma  
211 samples from patients with chronic HCV infection. Quantification is linear across the full  
212 dynamic range, covering values observed in untreated and DAA-treated patients. In  
213 addition, the Alinity m platform is fully automated with a consolidated, continuous, and

random-access workflow. With its processing capacity of 300 samples per 8-hour shift and the capability to immediately process urgent (STAT) samples, it enables same-day reporting of HCV test results. Rapid diagnostic testing can shorten the time between diagnosis and treatment, which may improve patient care and outcomes. The Alinity m HCV assay can thus confidently be used to detect and quantify HCV RNA in clinical practice.

## **Acknowledgements**

The authors would like to thank the statistics team at Abbott Molecular for assistance with data analysis. The authors declare that the results have been reported in an unbiased and fair manner.

## **References**

- [1] World Health Organization (WHO). Combating Hepatitis B and C to Reach Elimination by 2030. May 2016.
- [2] Yang JD, Hainaut P, Gores GJ, Amadou A, Plymoth A, Roberts LR. A global view of hepatocellular carcinoma: trends, risk, prevention and management. *Nat Rev Gastroenterol Hepatol*. 2019;16(10):589-604.
- [3] Pawlotsky J-M, Negro F, Aghemo A, Berenguer M, Dalgard O, Dusheiko G, et al. EASL Recommendations on Treatment of Hepatitis C 2018. *J Hepatol*. 2018;69:461-511.

235 [4] Ghany MG, Marks KM, Morgan TR, Wyles DL, Aronsohn AI, Bhattacharya D, et al.  
236 Hepatitis C Guidance 2019 Update: AASLD-IDSA Recommendations for Testing,  
237 Managing, and Treating Hepatitis C Virus Infection. Hepatology. 2019.

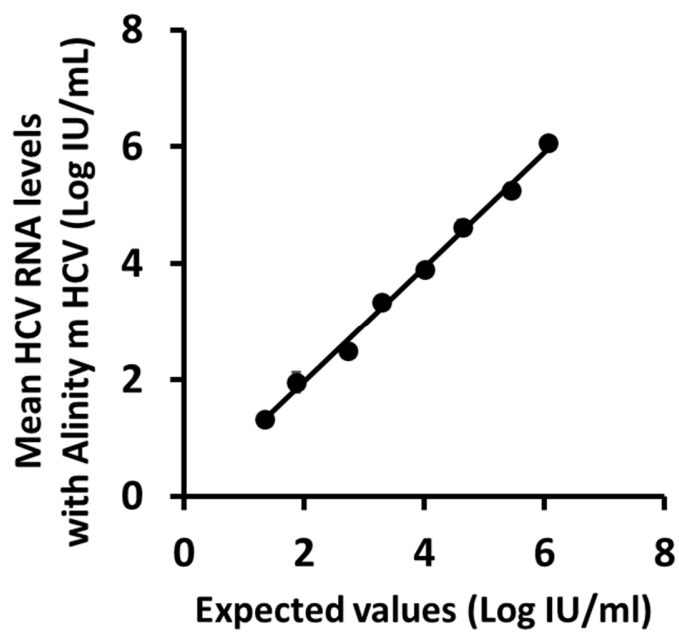
238 [5] Chevaliez S. Strategies for the improvement of HCV testing and diagnosis. Expert  
239 Rev Anti Infect Ther. 2019;17:341-7.

240 [6] Rodriguez C, Soulier A, Demontant V, Poiteau L, Mercier-Darty M, Bouvier-Alias M,  
241 et al. A novel standardized deep sequencing-based assay for hepatitis C virus genotype  
242 determination. Sci Rep. 2018;8:4180.

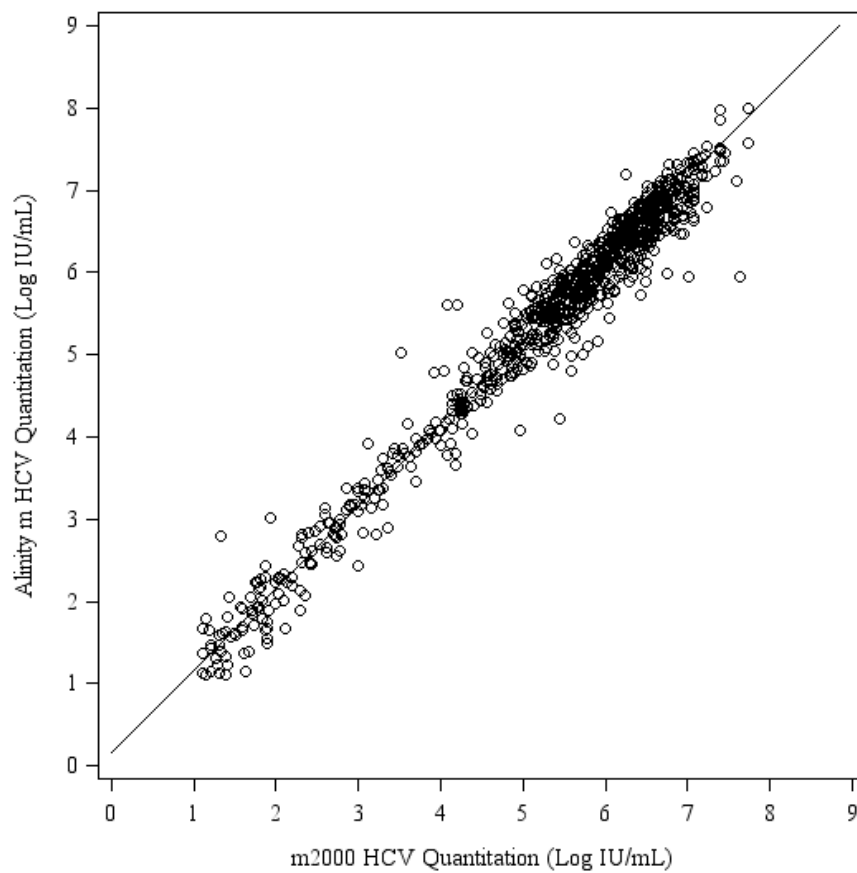
243 [7] Vermehren J, Kau A, Gartner BC, Gobel R, Zeuzem S, Sarrazin C. Differences  
244 between two real-time PCR-based hepatitis C virus (HCV) assays (RealTime HCV and  
245 Cobas AmpliPrep/Cobas TaqMan) and one signal amplification assay (Versant HCV  
246 RNA 3.0) for HCV RNA detection and quantification. J Clin Microbiol. 2008;46:3880-91.

## Figures

**Figure 1. Alinity m HCV assay linearity.** HCV RNA quantification by the Alinity m HCV assay of a dilution series of a clinical serum specimen containing  $8.30 \times 10^6$  IU/mL ( $6.90 \text{ Log IU/mL}$ ) HCV RNA.

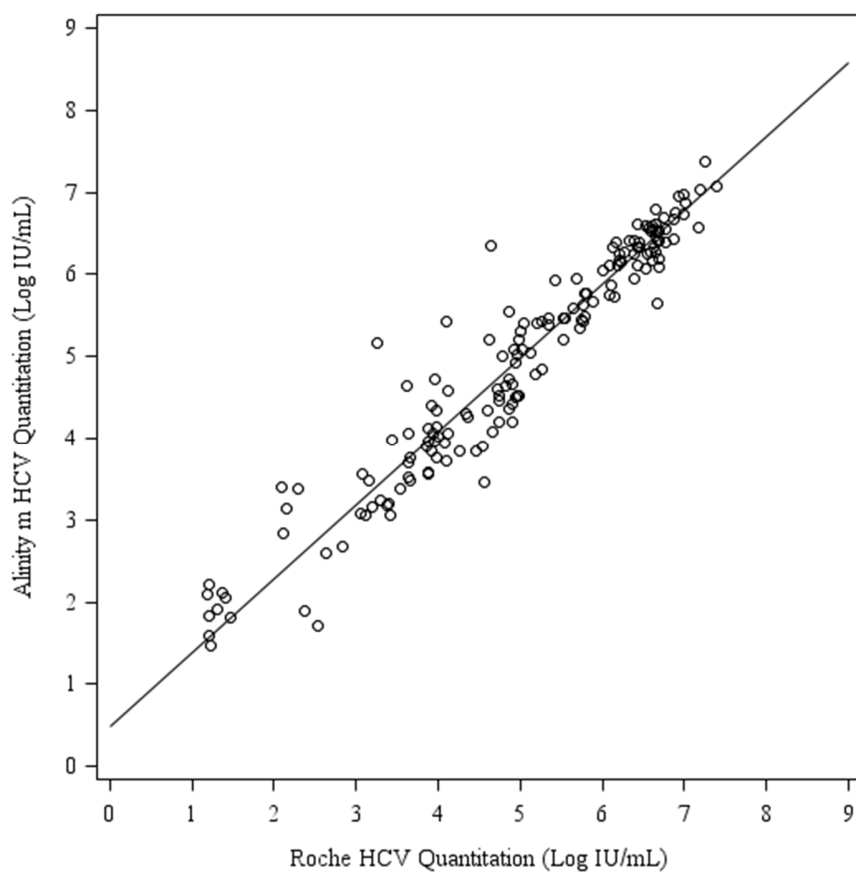


**Figure 2. Alinity m HCV assay and RealTime HCV assay performance.** Deming regression of HCV RNA levels showing correlation between the Alinity m HCV assay and RealTime HCV assay.



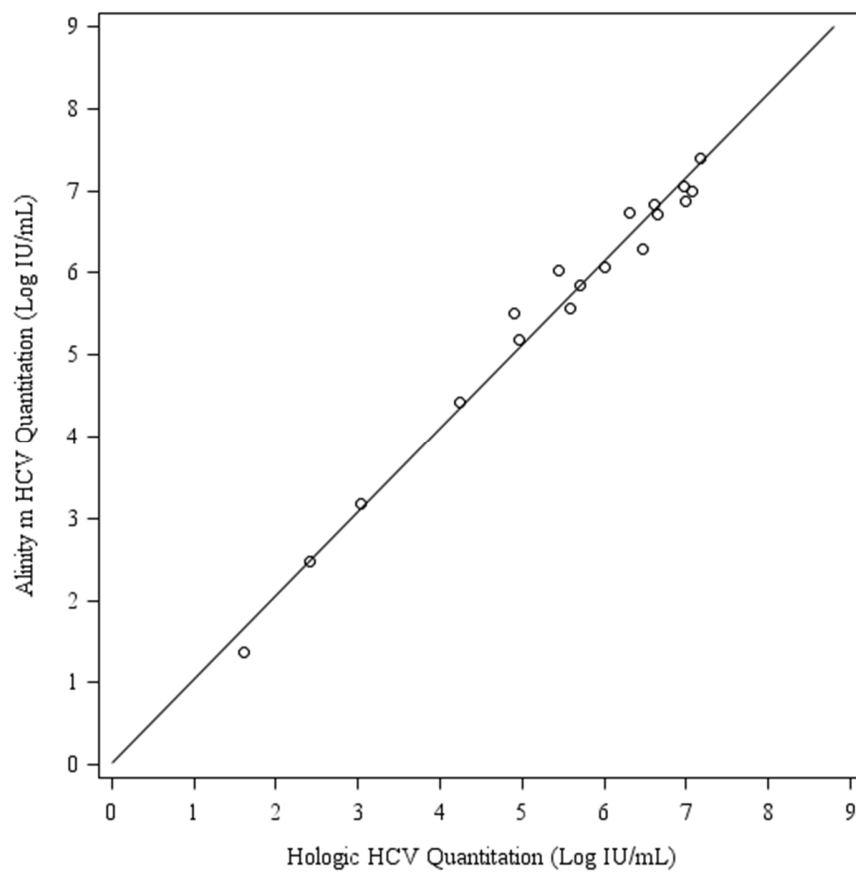


**Figure 3. Alinity m HCV assay and cobas HCV assay performance.** Deming regression of HCV RNA levels showing correlation between the Alinity m HCV assay and cobas HCV assay.



**Figure 4. Alinity m HCV assay and Aptima HCV Quant Dx assay performance.**

Deming regression of HCV RNA levels showing correlation between the Alinity m HCV assay and Aptima HCV Quant Dx assay.



## Tables

**Table 1.** Precision, Detection Rates, and Reproducibility of the Alinity m HCV Assay

	Panel Member (Genotype) / Control	Target HCV RNA* (Log IU/mL)	No. of Replicates	Mean of Measured HCV RNA (Log IU/mL)	SD (Log IU/mL)	CV (%)	No. of Replicates Detected	Detection Rate (%)
<b>Precision</b>	GT 1a-1	2.00	58	2.05	0.17	8.1	-	-
	GT 1a-2	1.40	54	1.38	0.16	11.9	-	-
	GT 1b-1	2.00	56	2.23	0.15	6.6	-	-
	GT 1b-2	1.40	59	1.54	0.21	13.5	-	-
	GT 3a-1	2.00	56	2.24	0.17	7.5	-	-
	GT 3a-2	1.40	56	1.65	0.16	9.8	-	-
<b>Detection Rate</b>	GT 1a-3	1.00	58	-	-	-	57	98.3
	GT 1b-3	1.00	57	-	-	-	57	100.0
	GT 3a-3	1.00	60	-	-	-	60	100.0
<b>Reproducibility</b>	LPC	2.71-2.81	215	2.70	0.12	4.3	-	-
	HPC	6.02-6.23	215	6.06	0.11	1.7	-	-

SD: standard deviation; CV: coefficient of variation; GT: genotype; LPC: low positive control; HPC: high positive control

\*For LPC and HPC, the range reflects the different target concentrations across the control lots used during the study.

**Table 2.** Comparison of HCV RNA Levels Using the Alinity m HCV and RealTime HCV Assays

		RealTime HCV (n)		
		Not detected	<12 IU/mL	≥12 IU/mL
Alinity m HCV (n)	Not detected	342	160	0
	<12 IU/mL	20	64	2 <sup>a</sup>
	≥12 IU/mL	8 <sup>b</sup>	24 <sup>c</sup>	883*

\* One additional sample was above the upper limit of quantification (8.00 Log IU/mL) for both methods. Another additional sample was >8.00 Log IU/mL with Alinity m but quantified with RealTime HCV.

<sup>a</sup> RealTime results ranged from 1.28 -1.40 Log IU/mL.

<sup>b</sup> Alinity m results ranged from 1.09 - 1.93 Log IU/mL.

<sup>c</sup> Alinity m results ranged from 1.09-1.80 Log IU/mL.

**Table 3.** Performance Comparison of Alinity m HCV and Comparator HCV Assays

Comparator Assay	Quantified Samples (n)	Correlation Coefficient (r)	Mean of Bias (Alinity m - Comparator; Log IU/mL)	SD of Bias (Log IU/mL)
RealTime HCV	883	0.983	0.13	0,283
cobas HCV	163	0.959	-0.01	0.454
Aptima HCV Quant Dx	18	0.991	0.14	0.232

**Table 4.** Comparison of HCV RNA Levels Using the Alinity m HCV and cobas HCV Assays

		cobas HCV (n)		
		Not detected	<15 IU/mL	≥15 IU/mL
Alinity m HCV (n)	Not detected	17	3	0
	<12 IU/mL	1	1	0
	≥12 IU/mL	0	1 <sup>a</sup>	163

<sup>a</sup> Alinity m sample quantitated at 1.72 Log IU/mL.

**Table 5.** Comparison of HCV RNA Levels Using the Alinity m HCV and Aptima HCV Quant Dx Assays

		Aptima HCV Quant Dx (n)		
		Not detected	<10 IU/mL	≥10 IU/mL
Alinity m HCV (n)	Not detected	44	50	0
	<12 IU/mL	0	4	0
	≥12 IU/mL	0	0	18