RESEARCH ARTICLE

Clinical diagnostic performance of light-initiated chemiluminescent assay compared with the Architect chemiluminescence immunoassay for detection of HCV antibody

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Abstract

Background: Hepatitis C virus antibody (anti-HCV) test had been approved as a preliminary screening test for HCV infection. Light-initiated chemiluminescent assay (LiCA) was a homogenous method. We aimed to assess the clinical diagnostic performance of LiCA and compare it with that of chemiluminescence immunoassay (CLIA) which was widely used in clinical laboratories.

Methods: A total of 10 772 patients from the Peking University Third Hospital were enrolled. The serum samples were detected on the ChIVD LiCA500 and Abbott Architect i2000SR platforms. Recombinant immunoblot assay (RIBA) and HCV RNA assay were used for confirmation.

Results: The negative agreement rate between ChIVD LiCA anti-HCV assay and Abbott Architect anti-HCV assay was 99.91%, the positive agreement rate was 37.31%, the total agreement rate was 98.74%, and the kappa coefficient (κ) was 0.519. The sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of ChIVD LiCA anti-HCV assay were 96.39%, 99.95%, 89.58%, and 99.97%, respectively, which were superior to those of Abbott Architect anti-HCV assay (93.98%, 99.25%, 51.90%, and 99.95%, respectively).

Conclusion: ChIVD LiCA anti-HCV assay was a highly sensitive, specific homogenous method with good diagnostic performance, and was applicable for the routine screening of HCV infection in clinical laboratories.

KEYWORDS
Abbott Architect anti-HCV assay, ChIVD LiCA 500 analyzer, HCV antibody, HCV RNA, hepatitis C, recombinant immunoblot assay

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Shuo Yang and Ruifeng Yang These authors contributed equally to this work

1 | INTRODUCTION

The World Health Organization (WHO) estimated that approximately 71 million people were living with chronic hepatitis C virus (HCV) worldwide and 399 000 people died from cirrhosis, hepatocellular carcinoma, and liver function failure caused by HCV infection in 2015.[1] Since 2016, direct-acting antivirals (DAAs) have been strongly recommended by WHO to all patients diagnosed with HCV infection, irrespective of genotype or disease stage.[2] Nowadays, HCV infection can be cured by antiviral treatment; however, many hepatitis C patients are asymptomatic,[3] and therefore, the screening of HCV infection is of great significance. And HCV serology test has been approved as a preliminary screening test for patients who were at high risk or have a history of HCV risk exposure. It is recommended to perform anti-HCV assay for initial detection of serological evidence of past or present infection prior to supplementary HCV RNA for evidence of viraemic infection.[1,4] Rapid and accurate serology methods with high sensitivity and specificity are required for screening to identify patients with HCV infection in clinical practice.

Numerous methods have been developed to detect HCV antibodies, including ELISA, chemiluminescence, and electrochemiluminescence assays.[5-8] Though with high sensitivity and accuracy, these methods exist certain disadvantages. For example, ELISA is time-consuming and laborious, and all the above immunoassays also require several washing steps as well as solid-phase immobilization of antibodies.

LiCA is an unusually robust and sensitive homogeneous double-antigen sandwich immunoassay method based on nanoparticle pairs coated with antigens or antibodies and oxygen channeling, which is capable of rapid, accurate, sensitive quantitative determination of a wide range of analytes.[9] With the outstanding advantages, LiCA has been widely accepted by clinical laboratories for reliable determination of TSH,[10] HBsAg,[11] sIgE against egg white allergens,[12] and tumor markers (CEA, CA15-3, and PSA)[13] etc, with lower detection limits.

However, evidence is sparse regarding the clinical performance of LiCA for detecting HCV antibody. The principle of LiCA is based on two different nanoparticles with the diameter of 200 nm. One nanoparticle contains an HCV-Ag coated chemiluminescer as reagent 1, whereas the other contains a streptavidin-coated photosensitizer which binds to bioavidin labeled HCV-Ag as reagent 2, and either HCV IgM or IgG can be recognized to form Ag-Ab-Ag complex. Under the excitation of laser, ionic oxygen transfers between two particles, producing chemiluminescent emission (Figure 1). The number of photons is converted into the target molecule concentration by single photon counter and mathematical fitting.

In this study, we evaluated the clinical performance of LiCA for detecting HCV antibody and compared the results obtained by ChIVD LiCA 500 system with those obtained by Abbott Architect i2000SR analyzer.

2 | SUBJECTS AND METHODS

2.1 | Subjects

A total of 10 772 patients from the Peking University Third Hospital were enrolled. In the first stage of our prospective study, 10 672 consecutive fresh serum samples without any missing or selectively gathered ones from June 4, to June 27, 2018. In the second stage, 100 serum samples with reactive results of Architect i2000SR were collected from August to September 2018. Our study had been approved by the Ethics Committee of Peking University Third Hospital.

2.2 | Serum anti-HCV and HCV RNA determination

All serum samples were centrifuged at 2575 g for 10 minutes. These fresh samples were determined for HCV antibody on the
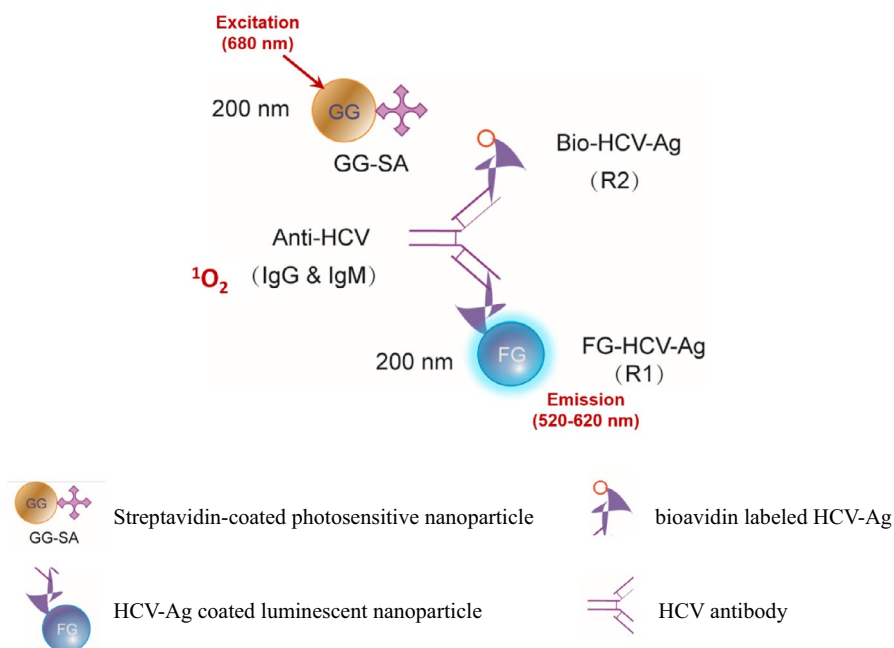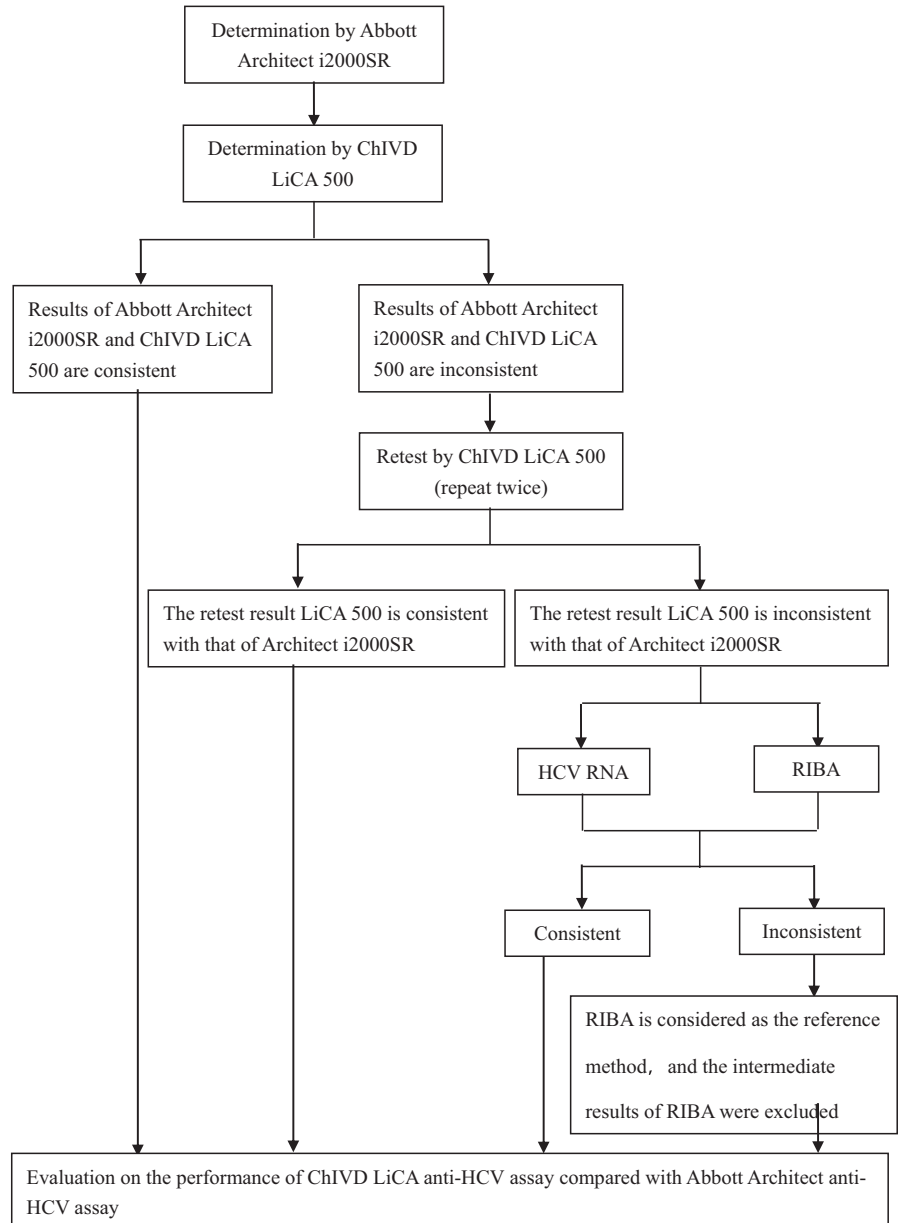

FIGURE 1 Principle of ChIVD LiCA for detecting HCV antibody

FIGURE 2 The flow diagram of serum anti-HCV determination

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                    ┌─────────────────────────┐
                    │   Determination by Abbott│
                    │    Architect i2000SR     │
                    └─────────────────────────┘
                                │
                    ┌─────────────────────────┐
                    │   Determination by ChIVD │
                    │        LiCA 500          │
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Figure: flow diagram with the following boxes:

- Determination by Abbott Architect i2000SR
- Determination by ChIVD LiCA 500
- Results of Abbott Architect i2000SR and ChIVD LiCA 500 are consistent
- Results of Abbott Architect i2000SR and ChIVD LiCA 500 are inconsistent
- Retest by ChIVD LiCA 500 (repeat twice)
- The retest result LiCA 500 is consistent with that of Architect i2000SR
- The retest result LiCA 500 is inconsistent with that of Architect i2000SR
- HCV RNA
- RIBA
- Consistent
- Inconsistent
- RIBA is considered as the reference method, and the intermediate results of RIBA were excluded
- Evaluation on the performance of ChIVD LiCA anti-HCV assay compared with Abbott Architect anti-HCV assay

Architect i2000SR (Abbott Diagnostics) and ChIVD LiCA 500 platforms. Residual samples after detection were stored at −80°C for further confirmation. All results were expressed in S/CO ratio, with S/CO < 1.0 indicating nonreactive result, while S/CO ≥ 1 indicating reactive result. If two results of one sample were inconsistent, the sample was retested by ChIVD LiCA 500 system. In addition, all the samples with inconsistent results between Architect i2000SR and ChIVD LiCA 500 were confirmed by recombinant immunoblot assay (RIBA) and HCV RNA. RIBA is a reference method for detection of HCV antibodies which could be bound to HCV antigens (Core 1, Core 2, Helicase, NS3, NS4, and NS5) immobilized on the test strip (Mikrogen Diagnostics). Results of RIBA were expressed as negative, intermediate, or positive. And HCV RNA copies quantitatively determined by nucleic acid amplification testing (COBAS® AmpliPrep/COBAS® Taqman® HCV Quantitative Test, version 2.0, Roche Molecular Systems, Branchburg, USA). The limit of detection (LoD) for HCV RNA is 15 IU/mL. The cutoff value for HCV RNA is considered as C_t value is below the limit for the assay, while C_t value for HCV is above the limit for the assay or no C_t value is obtained, "HCV RNA negative" is reported, otherwise "HCV RNA positive" is reported. If the RIBA and HCV RNA tests gave discrepant results, we used RIBA as judgement criterion, while the intermediate results of RIBA were excluded (Figure 2). All methods were performed as recommended by the manufacturers.

2.3 | Statistical analysis

All data were analyzed by statistical software SPSS 19.0 (SPSS Inc). The sensitivity, specificity, positive predictive values (PPV), and negative predictive values (NPV) were calculated. The agreement between ChIVD LiCA 500 and Abbott Architect i2000SR assay for the detection of HCV antibody was evaluated based on the positive

agreement rate, the negative agreement rate, the total agreement, and kappa coefficient (κ) calculated using 2 × 2 contingency table. The κ value of 0.41-0.6 indicated moderate agreement between the two anti-HCV assays, the κ value of 0.61-0.8 indicated strong agreement, and the κ value of >0.8 indicated almost perfect agreement.

3 | RESULTS

3.1 | Comparison of ChIVD LiCA 500 and Abbott Architect i2000SR assay for detection of HCV antibody

Of the 10 772 serum samples tested on Abbott Architect i2000SR and CHIVD LiCA 500 platforms, results of 136 samples were inconsistent. Test results showed that there were 201 positive samples in Architec ti2000SR, and there were 85 positive samples in ChIVD LiCA 500 (Table 1). The negative agreement rate was 99.91% (calculated by a/(a + c)), the positive agreement rate was 37.31% (calculated by d/(b + d)), the total agreement rate was 98.74% (calculated by (a + d)/(a + b+c + d)), and the κ value was 0.519, indicating moderate agreement between the two anti-HCV assays. All samples were divided into three groups according to the S/CO ratio of Architect i2000SR, that is <1.0, 1.0-5.0 and >5.0 (Table 2).

3.2 | Analysis of samples with inconsistent results by HCV RNA and RIBA

A total of 136 samples with inconsistent results were retested by ChIVD LiCA 500 twice. All results were consistent with these of the first determination by LiCA 500, suggesting the method based on LiCA had a good reproducibility. Therefore, results of all 136 samples were confirmed by the standard reference methods of HCV RNA and RIBA. Among the 136 samples with inconsistent results between Architect i2000SR and ChIVD LiCA 500, only one sample of HCV RNA was positive, as well as the result of RIBA (Table 3).

3.3 | Diagnostic performance of ChIVD LiCA 500 and Abbott Architect i2000SR anti-HCV assays for the detection of HCV infection

According to results obtained by the standard reference methods of HCV RNA and RIBA, 43 samples with intermediate results were excluded, and 10 729 samples with nonreactive or reactive results

were used to calculate the sensitivity and specificity (Table 4). PPV and NPV for LiCA 500 and Architect i2000SR were assessed for the consecutive 10 672 samples, and 22 intermediate results were excluded, the PPV and NPV of ChIVD LiCA anti-HCV assay were 89.58% and 99.97%, which were superior to those of Abbott Architect anti-HCV assay (51.90% and 99.95%).

4 | DISCUSSION

With the application of DAA, WHO proposed to eliminate HCV by 2030. To eliminate HCV as a public health threat, 90% of patients infected with HCV need to be diagnosed.[1] Therefore, the focus of research has shifted from "how to treat" to "how to screen among different populations." Identification of patients with latent infection is the most valuable way in reducing the disease burden. Because of the atypical symptoms, the diagnosis of hepatitis C mainly depends on clinical laboratory tests, including anti-HCV, HCV antigen,[14] HCV RNA,[15] and genotype.[16]

Serology tests are recommended for screening of HCV infection, which need to be highly sensitive, specific, rapid, and accurate for detection of nearly all affected individuals. As the screening method, chemiluminescence and electrochemiluminescence immunoassays for HCV antibody (the antibodies of HCV core, NS3-4, and NS5 proteins) have been widely used to diagnose HCV infection in clinical laboratories. Previous studies have compared the diagnostic performance of different methods and revealed that the false positive rates of these assays are relatively high.[7,17-19] Also, these methods for anti-HCV detection require several washing steps so that the detection speed is limited.

Recently, LiCA system for anti-HCV detection has been developed. It is a simple, rapid, and high-throughput method, requires no washing steps to effectively avoid washing pollution, can recognize both IgG and IgM of HCV antibodies, and is not interfered by nonspecific IgG.

In the present study, 10 772 patients were enrolled and analyzed, and the negative agreement rate between ChIVD LiCA anti-HCV assay and Abbott architect i2000SR anti-HCV assay was 99.91%; however, the positive agreement rate was only 37.31% due to the high false positive rate of Architect i2000SR assay. Among the 10 772 samples tested by both methods, results of 136 samples were inconsistent. S/CO ratios of 116/136 samples were between 1.0 and 5.0. The large number of false positive samples in practice seriously disturbed the judgement of clinicians. The nonspecific interferents in serum (such as fibrin, heterophil antibody), patients in window period, and individuals with immunodeficiency (such as patients with HIV infection, chemotherapy, and stem cell transplantation) might be the main causes of false positive results. In addition, false positive results often occurred in pregnant women, the elderly, or dialysis patients.

For the samples with inconsistent results, we retested them by ChIVD LiCA 500 twice. All results of retested samples were consistent with the previous ones, suggesting that the LiCA assay had

TABLE 1 Comparison of results in ChIVD LiCA 500 and Abbott architect i2000SR assay for detection of HCV antibody (n = 10 772)

		Architect i2000SR		
		Negative (n)	Positive (n)	Total (n)
LiCA 500	Negative (n)	10 561 (a)	126 (b)	10 687
	Positive (n)	10 (c)	75 (d)	85
	Total (n)	10 571	201	10 772

TABLE 2 Results of HCV antibody divided by S/CO ratio of Architect i2000SR (n = 10 772)

Architect i2000SR		LiCA 500		Consistency	
S/CO ratio	n	Negative (n)	Positive (n)	Agreement ratio (%)	Inconsistent (n)
S/CO < 1	10 571	10 561	10	99.91%	10
1 ≤ S/CO ≤ 5	137	116	21	15.33%	116
S/CO > 5	64	10	54	84.38	10

TABLE 3 Confirmation and verification of samples with inconsistent results between LiCA 500 and Architect i2000SR (n = 136)

Architect S/CO ratio	Inconsistent (n)	Standard reference method (HCV RNA + RIBA)		
		Reactive (n)	Nonreactive (n)	Intermediate (n)
S/CO < 1	10	5	5	0
1 ≤ S/CO ≤ 5	116	2	76	38
S/CO > 5	10	1	4	5

TABLE 4 Diagnostic performance of ChIVD LiCA 500 and Abbott Architect i2000SR for detection of HCV antibody in the whole population (n = 10 729)

	LiCA 500		Architect i2000SR	
	True (n)	False (n)	True (n)	False (n)
Negative (n)	10 641	3	10 566	5
Positive (n)	80	5	78	80
Sensitivity (%)	96.386		93.976	
Specificity (%)	99.953		99.249	

a good reproducibility. Therefore, RIBA and HCV RNA assay were used to confirm the samples with inconsistent results. However, according to the Centers for Disease Control and Prevention (CDC) 2013 Guidelines for Laboratory Testing and Result Reporting on Antibodies to Hepatitis C Virus, it only recommended HCV RNA as a supplemental test for anti-HCV confirmation, while RIBA was no longer recommended because it was time-consuming, labor-intensive, and indeterminate for weakly reactive HCV antibody.[20] However, because of the high specificity, we used RIBA as well as HCV RNA to confirm the 136 samples with inconsistent results in the study. A total of 8 samples were nonreactive, 85 samples were reactive, and 43 samples were intermediate. Most of the samples with the S/CO ratio between 1.0 and 5.0 were nonreactive by the detection of HCV RNA and RIBA (76/116). For the 116-weak reactive samples with the S/CO ratio between 1.0 and 5.0, 38 samples were indeterminate by confirmatory methods. Previous studies had revealed that the diagnostic performance of electrochemiluminescence immunoassays was better than Architectanti-HCV assay and other comparative assays.[17,21] ChIVD LiCA anti-HCV assay showed a high sensitivity of 96.39% and excellent specificity of 99.95%, PPV of 89.58%, and NPV of 99.97%, which were superior to those of the Architect anti-HCV assay. And the diagnostic performance of ChIVD LiCA anti-HCV assay was similar to that of Elecsys reported in other studies.[17]

5 | CONCLUSION

In conclusion, the CHIVD LiCA anti-HCV assay provided a highly sensitive, specific, rapid, and reliable tool for screening HCV infection in clinical laboratories. Nonetheless, weak reactive samples should be confirmed by HCV RNA or followed up for HCV antibody.

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REFERENCES

1. *Guidelines for the Care and Treatment of Persons Diagnosed with Chronic Hepatitis C Virus Infection*. Geneva, Switzerland: World Health Organization; 2018. http://www.ncbi.nlm.nih.gov/books/NBK531733/.
2. *Guidelines for the Screening Care and Treatment of Persons with Chronic Hepatitis C Infection: Updated Version*. Geneva, Switzerland: World Health Organization; 2016.
3. Ward JW, Mermin JH. Simple, effective, but out of reach? Public health implications of HCV drugs. *N Engl J Med*. 2015;31;373(27): 2678–2680.
4. Guidelines Study Group VH. Turkey 2017 clinical practice guidelines on recommendations for screening diagnosing and managing hepatitis C virus. *Turk J Gastroenterol*. 2017;28(Suppl 2):90-93.
5. Khudur Al-Nassary MS, Mahdi BM. Study of hepatitis C virus detection assays. *Ann Med Surg*. 2018;36:47-50.
6. Safi MA. Hepatitis C: an overview of various laboratory assays with their mode of diagnostic cooperation. *Clin Lab*. 2017;63(5):855-865.
7. Hyun J, Ko DH, Kang HJ, Whang DH, Cha YJ, Kim HS. Evaluation of the VIDAS anti-HCV assay for detection of hepatitis C virus infection. *Ann Lab Med*. 2016;36(6):550-554.
8. Tang W, Chen W, Amini A, et al. Diagnostic accuracy of tests to detect hepatitis C antibody: a meta-analysis and review of the literature. *BMC Infect Dis*. 2017;17(Suppl 1):695.
9. Ullman EF, Kirakossian H, Singh S, et al. Luminescent oxygen channeling immunoassay: measurement of particle binding kinetics by chemiluminescence. *Proc Natl Acad Sci USA*. 1994;91(12):5426-5430.

10. Monneret D, Guergour D, Vergnaud S, Laporte F, Faure P, Gauchez AS. Evaluation of LOCI technology-based thyroid blood tests on the dimension vista analyzer. *Clin Biochem*. 2013;46(13–14):1290-1297.

11. Ullman EF, Kirakossian H, Switchenko AC, et al. Luminescent oxygen channeling assay (LOCI): sensitive, broadly applicable homogeneous immunoassay method. *Clin Chem*. 1996;42(9):1518-1526.

12. Bian Y, Liu C, She T, et al. Development of a light-initiated chemiluminescent assay for the quantitation of sIgE against egg white allergens based on component-resolved diagnosis. *Anal Bioanal Chem*. 2018;410(5):1501-1510.

13. Dolscheid-Pommerich RC, Keyver-Paik M, Hecking T, et al. Clinical performance of LOCI™-based tumor marker assays for tumor markers CA 15–3, CA 125, CEA, CA 19–9 and AFP in gynecological cancers. *Tumor Biol*. 2017;39(10):1010428317730246.

14. Duchesne L, Njouom R, Lissock F, et al. HCV Ag quantification as a one-step procedure in diagnosing chronic hepatitis C infection in Cameroon: the ANRS 12336 study. *J Int AIDS Soc*. 2017;20(1):21446.

15. Yao JD, Young S, Heilek GM, et al. Diagnosis and monitoring of HCV infection using the cobas(®) HCV test for use on the cobas(®) 6800/8800 systems. *J Clin Virol*. 2018;102:63-69.

16. Stelzl E, Appel HM, Mehta R, et al. Evaluation of the new cobas® HCV genotyping test based on real-time PCRs of three different HCV genome regions. *Clin Chem Lab Med*. 2017;55(4):517-521.

17. Yue ZH, Xia CS, Wang H. Performance evaluation of the mindray anti-HCV assay for the detection of hepatitis C virus infection. *J Clin Lab Anal*. 2018;32(9):e22600.

18. Kim B, Ahn HJ, Choi MH, Park Y. Retrospective analysis on the diagnostic performances and signal-to-cut-off ratios of the Elecsys Anti-HCV II assay. *J Clin Lab Anal*. 2018;32(1):e22165.

19. Wang L, Chen W, Yu Y. The Performance of the Abbott i2000SR for measuring serum markers of infectious diseases. *J Clin Lab Anal*. 2017;31(1). https://doi.org/10.1002/jcla.22015 Epub 2016 Jun 15.

20. Centers for Disease Control and Prevention (CDC). Testing for HCV infection: an update of guidance for clinicians and laboratorians. *MMWR Morb Mortal Wkly Rep*. 2013;62(18):362-365.

21. Esteban JI, van Helden J, Alborino F, et al. Multicenter evaluation of the Elecsys® anti-HCV II assay for the diagnosis of hepatitis C virus infection. *J Med Virol*. 2013;85(8):1362-1368.