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Evaluation of the analytical performance of six rapid diagnostic tests for the detection of viral hepatitis B and C in Lubumbashi, Democratic Republic of Congo

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ABSTRACT

Rapid diagnostic tests (RDTs) are widely used in Lubumbashi for the diagnosis of viral hepatitis B and C. To date, there are no works that have been carried out in Lubumbashi to independently assess the performance of such tests. This study aimed at assessing the effectiveness of RDTs for the detection of HBsAg and anti-HCV antibodies in order to identify infected blood donors in Lubumbashi. A total of 300 serum samples (100 HBsAg positive samples; 100 anti-HCV positive samples and 100 HBsAg and anti-HCV negative samples) were tested simultaneously using the 6 locally used RDTs and as gold standard the chemiluminescent assays for HBsAg and the RT-TMA for HCV detection.

The six evaluated RDTs demonstrated a sensitivity and a negative predictive value (NPV) of 100 % whereas the specificity and positive predictive value (PPV) varied from 46 % to 98.1 %. SB BioLine HBsAg test performed best in this study with 100 % of sensitivity, 97.1 % of specificity, 100 % of NPV and 96.9 % of PPV. Furthermore, sensitivity, specificity, NPV and PPV for SB BioLine HCV test were as follows: 100 %, 98.1 %, 100 % and 93.9 %. Therefore, SD BioLine tests (HBsAg, HCV) would be selected as the first line RDTs for the detection and the diagnostic of hepatitis B and C. They can prevent blood-borne transmission of HBV and HCV in areas with limited incomes as Lubumbashi.

1. Introduction

Liver diseases are major health concern with world consequences (Parry et al., 2017). Hepatitis B and C are major public health problems worldwide. According to the World Health Organization (WHO), 350 million people are infected with the hepatitis B virus (HBV) and 130–170 million by the hepatitis C virus (HCV) worldwide (Fouelifack et al., 2018). HBV and HCV have been called silent killers because the diseases they cause often progress without apparent symptoms up to complications such as cirrhosis and liver cancer, which cause 720,000 and 470,000 deaths annually, respectively. The regions of Africa and the Western Pacific represent 68 % of people infected with HBV, while the regions of Europe and the eastern Mediterranean are more affected by HCV (Peeling et al., 2017).

Serological tests are generally used as part of the first line diagnostic test strategy for chronic infection with hepatitis B or C viruses, in order to detect potentially infected people and to make them benefit from a possible clinical evaluation and treatment (Parry et al., 2017; Tang et al., 2017; Poiteau et al., 2017, 2018).

Surface Antigen (HBsAg) is a key marker for laboratory detection and diagnosis of HBV infection. The presence of anti-HBs reveals both immunity associated with resolved infection or vaccination. A number of rapid diagnostic tests (RDTs) have been developed for the detection of HBsAg. Most of them meet the analytical sensitivity recommended by the WHO of 0.13 IU/mL, depending on the target population. However, in recent assessments, the analytical sensitivity of each RDT varied considerably, while the specificity appeared satisfactory (Kosack et al., 2014; Poiteau et al., 2017; Mane et al., 2019).

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The diagnosis of hepatitis C is based on the detection of antigen, antibody or viral genome in serum or plasma samples. An enzyme-linked immunosorbent assay (ELISA) or a chemiluminescent immunoassays (CLIA) is extremely sensitive for the detection of anti-HCV antibodies in people at risk (De Paula Scalioni et al., 2014; Fourati et al., 2018).

RDTs have many advantages since their simplicity makes them suitable in the event of a rapidly fatal disease and in remote areas without specialized technicians and infrastructure (D'Acremont et al., 2011). In addition, with respect to blood transfusion, RTDs allow a rapid exclusion of blood donors infected with HBV or HCV. However, in order to correctly interpret them, it is essential to know their analytical and clinical performances.

Several commercial rapid tests for the detection of HBs antigen and anti-HCV antibodies are available on the Congolese market, but their performance characteristics have not been determined independently yet. The assessment of their performance could facilitate decisions regarding the marketing authorization of RDTs in Lubumbashi. RDTs performance indicators are usually given by the manufacturers but some of them do not mention these data in the insert. Nevertheless, we evaluated the analytical performance of all available RDTs since they are commonly used in clinical biology laboratories in Lubumbashi (DRC).

The quality assurance of blood transfusions in Africa remains a challenge due to economic and logistical constraints. RDTs are commonly used in many African blood transfusion centers to screen for transfusion-transmissible infections.

Finally, as part of our research project on viral hepatitis B and C in Lubumbashi, screening of HBV and HCV was mainly done using RDTs. Thereafter, prior to perform the real-time polymerase chain reaction assay (qPCR), the positive samples were confirmed on Liaison XL automatic analyzer and differences in specificity were observed. Therefore, a study was initiated aiming to assess the analytical performance of six rapid diagnostic tests (RDTs) used in Lubumbashi for the screening and the diagnostic of viral infections with HBV and HCV. We used as reference methods, the Liaison XL chemiluminescent assays for HBV and the RT-TMA for HCV. This study would allow us to choose RDTs with best performance as first line HBV and HCV tests in Lubumbashi.

2. Materials & methods

2.1. Framework and period of study

A panel of human sera was selected anonymously from June 2019 to March 2020 in Lubumbashi, DRC. The evaluation of RTDs was performed at the Institut de Recherche Expérimentale et Clinique (IREC), Pôle de Microbiologie, Université Catholique de Louvain.

2.2. Composition of the reference samples panel

Three hundred samples were selected solely on the basis of their status with regard to viral hepatitis B and C as described below. The serum samples were obtained from blood donors who presented themselves to the blood bank of Hôpital Jason Sendwe and Cliniques universitaires de Lubumbashi, 2 major referral hospitals in Lubumbashi, DRC. Blood sera chosen for the evaluation included 100 serum samples from HBV positive individuals, 100 serum samples from HCV positive individuals, 100 serum samples from HBV and HCV negative people. Serum samples were selected using One Step Hepatitis Test Strip (HBsAg, anti-HVC) RDTs. Thereafter, they were frozen and stored at −20 °C.

2.3. Rapid diagnostic test (RDT)

The 6 RDTs evaluated in this study comprised the One Step Hepatitis B surface antigen Test® Strip (Accurate, China), One Step Strip Style HBsAg Rapid Screen Test® (Suzhou Medical Supplies CO., LTD, China), SD Bioline HBsAg WB (Multi SD Standard Diagnostics, INC, Republic of Korea), One Step Hepatitis C Virus Test® Strip (Accurate, China), Hepatitis C Virus Antibody (HCV) Test® Strip (Suzhou Medical Supplies CO., LTD, China), SD BioLine HCV (SD Standard Diagnostics, INC, Republic of Korea).

The RDTs are qualitative tests based on the lateral immunochromatographic technique for the detection of hepatitis B virus surface antigen (HBsAg) and anti-HCV antibodies in human whole blood, serum and plasma. Monoclonal and polyclonal antibodies are used to specifically identify HBsAg whereas the HCV-colloid conjugate antigen reacts with anti-HCV antibodies. They are sensitive tests and the results can be read visually without any equipment (Amini et al., 2017; Barbosa et al., 2017). All tests were performed according to the manufacturer's recommendations.

2.4. The Liaison XL automatic analyzer (DiaSorin, Saluggia, Italy)

The Liaison XL method is a direct, sandwich, two-stage incubation test, based on the chemiluminescent technique. The immunoassay analyzer automatically calculates the HBsAg concentrations expressed in IU/mL and classifies the results. The dosing interval is: 0.030–150 IU/mL HBsAg. Characteristics of the evaluated RDTs and the reference test for the detection of HBsAg are listed in Table 1.

2.5. Real-Time TMA for HCV viral load determination

The hepatitis C viral load was quantified in real time on the Panther® automatic analyzer (Hologic, USA). The Aptima HCV Quant Dx Assay is a nucleic acid amplification assay that uses real-time transcription-mediated amplification (RT-TMA) to detect and quantify HCV RNA. This test targets a conserved region of the HCV genome for the detection and quantification of genotypes 1, 2, 3, 4, 5, and 6.

2.6. Limit of detection (LoD) study

We studied the limit of detection of the RDTs compared to the Liaison XL method by preparing 10-fold dilution of our samples. Three HBV positive sera were selected and a 10-fold serial dilution of serum was performed, 6 dilutions for each sample. The first serum sample with a viral load of 7 $\log_{10}$ UI/mL and HBsAg concentration of >150 IU/mL, the second serum sample with undetectable DNA and HBsAg concentration of >150 IU/mL and the third serum sample with a viral load of 2 $\log_{10}$ UI/mL and HBsAg concentration of 0.06 IU/mL. The viral load these sera was determined by home-made TaqMan PCR, targeting the P gene on the LightCycler 96 (Roche, Germany). The standard has been calibrated against the WHO standard and results were reported in IU/mL. Range of quantification 10^E2 IU/mL to 10^E8 IU/mL. Each dilution was tested for the detection of HBsAg simultaneously by the evaluated RDTs and the liaison XL.

2.7. Analysis of samples

The analysis of the samples took place in two places: (i) In Lubumbashi (Hôpital Jason Sendwe and Cliniques universitaires) where the selection of positive and negative samples in HVB and HVC has been done using as RDTs One Step Hepatitis B Surface Antigen Test Strip and One Step Hepatitis C Virus Test Strip. Samples were then stored at −20 °C. Further, (ii) in Brussels (Laboratoire de Microbiologie, UCLouvain) where samples were first thawed and then tested again with the same RDTs as those used in Lubumbashi at the time of selection. This allowed us to check if the freeze-thaw process could impact viral survival. Finally, all the samples, both negative and positive, were tested in parallel by the four other RDTs and by Liaison XL automatic analyzer for HBV, and RT-TMA for HCV.

The performance indicator parameters that we evaluated were as follows: sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV).

Table 1
Characteristics of the evaluated RDTs and the reference test for the detection of HBsAg.

RDTs	Supports	Manufacturer sensitivity (%)	Manufacturer specificity (%)	Technique	Sample volume (µl)	Test duration (minutes)	Price per test ($)	Firms	Firm's countries
A	Cellulose strips	> 99	97	Immuno-chromatography	10	15	0.3	Accurate	China
B	Cellulose strips	Not known	99	Immuno-chromatography	10	10–20	0.3	Suzhou Medical Supplies CO., LTD	China
C	Cassette	100 (98.3–100)	100 (99.5–100)	Immuno-chromatography	100	20	0.4	SD Standard Diagnostics, INC	Republic of Korea
D	Cellulose strips	> 99	98.6	Immuno-chromatography	10	15	0.3	Accurate	China
E	Cellulose strips	Not known	99	Immuno-chromatography	10	15	0.3	Suzhou Medical Supplies CO., LTD	China
F	Cassette	100 (98.3–100)	100 (98.8–100)	Immuno-chromatography	100	20	0.4	SD Standard Diagnostics, INC	Republic of Korea
G	Automatic analyzer	100 (99,10–100)	99.98(99.89–100)	Chemiluminescence	300	60	3	Diasorin S.p.A	Italy

A = One Step Hepatitis B Surface Antigen Test Strip; B = One Step strip Style HBsAg Rapid Screen Test; C = SD BioLine HBsAg WB (Multi); D = One Step Hepatitis C Virus Test Strip; E= Hepatitis C Virus Antibody (HCV) Test (Strip); F = SD BioLine HCV; G = liaison XL Murex automatic analyzer (HBV).

2.8. Statistical analysis

The results were entered into the Excel database. Statistical analysis was performed on EPI INFO™ version 7.2.4.0. (Epi Info Software, CDC, Atlanta, USA). Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) for each rapid test were assessed and their respective 95 % confidence intervals (95 % CI) were calculated. The concordance between the results of the rapid tests and those of Liaison XL®Quant (HBsAg) and RT- TMA (HCV) was evaluated for each test by Kappa statistics.

2.9. Ethical committee approval

This study obtained the approval of the medical ethical committee of the Université de Lubumbashi, under the approval number: UNILU / CEM / 095/2017.

3. Results

3.1. Detection of HBsAg and HCV using RDTs, Liaison XL and RT-TMA

Using Liaison XL assay as reference test for HBV and RT-TMA as reference test for HCV, out of the 200 tested samples we obtained 94 HBsAg positive samples and 106 HBsAg negative samples. Similarly, with respect to HCV, we obtained 46 positive samples and 154 negative samples.

The sensitivity and the negative predictive value (NPV) were about 100 % for all the six RDTs in both types of infections. In terms of specificity, only the SD BioLine RDT showed specificity of 97,1 % for HBV and 98,1 % for HCV. The SD BioLine presented a positive predictive value (PPV) of 96,9 % for hepatitis B and 93,3 % for hepatitis C. The RDT One Step Hepatitis B surface Antigen Test Strip and One Step strip Style HBsAg Rapid Screen Test presented 3% of false positives for HBV and 27 % of false positives for HCV, while SD BioLine HCV presented only 1,5% of false positives. One step Hepatitis Test Strip (HBsAg, HCV) demonstrated 47 % of (true positives) TP for HBsAg and 23 % for HCV.

One Step Hepatitis B Surface Antigen Test Strip and One Step strip Style HBsAg Rapid Screen Test showed kappa values above 97.6 % and SD BioLine HBsAg Test demonstrated kappa value of 100 %. With respect to the detection of HCV, One Step Hepatitis C Virus Test Strip and Hepatitis C Virus Antibody (HCV) Test (Strip) had kappa values above 49.3 % whereas SD BioLine HCV presented kappa value above 96.7 % (Table 2).

3.2. Limit of detection (LoD) study

We studied the limit of detection of the RDTs compared to the reference method (Liaison XL) by preparing 10-fold dilution of our samples. The results of this study are reported in Table 3.

From this study, it appeared that when the HBsAg level was high (>150 IU/mL), all the evaluated tests kept their sensitivity up to 10 times dilution. However, the reference method (Liaison XL) was more sensitive than the evaluated RDTs since it was positive up to 1/100.000 dilution. On the other hand, when the HBsAg level was low (0.06 IU/mL), the dilution of serum sample (10 times) caused the loss of sensitivity of RDTs as well as that of Liaison XL automatic analyzer. Therefore, the detection limit (LoD) for the tested RDTs and the Liaison XL automatic analyzer could be estimated as being ≥ 0.06 IU/mL of HBsAg (Table 3).

4. Discussion

4.1. Characteristics of the evaluated tests according to the manufacturers

The performance characteristics of different RDTs may be overestimated for commercial reasons by the manufacturers. SD BioLine (HBsAg, HVC) tests, One Step Hepatitis B Surface Antigen Test Strip and One Step strip Style HBsAg Rapid Screen Test presented specificities of ≥ 94 %, the two other tested RDTs including One Step Hepatitis C Virus Test Strip and Hepatitis C Virus Antibody Test (strip) had in our study a specificity of 65 % while the manufacturers assigned them a specificity of 97–99%. Nevertheless, the evaluated RDTs have some advantages in comparison to the Liaison XL automatic analyzer (CLIA) used in this study as reference method, in particular the duration of the test (15−20 min instead of 60 min for CLIA), the required volume sample for analysis (10 µL-100 µL instead of 175 µL-300 µL for CLIA) and the cost of analysis which is ten times more expensive for the Liaison XL automatic analyzer than on RDTs (4€ instead of 0.3−0.4€). The mentioned characteristics of the evaluated RDTs make them useful tools for HBV and HVC diagnosis particularly for emergency situations and in the areas where there are no infrastructure and specialized laboratory technicians. However, despite its high costs, the Liaison XL method, particularly that of the latest generation, is more reliable than the RDTs. The Liaison XL detection is based on the chemiluminescent technique whereas the RDTs are qualitative tests based on the lateral immuno-chromatographic technique.

Of the 6 evaluated RDTs, only the SD BioLine (HBsAg and HCV) had as support a cassette. The other four RDTs had cellulose bands as support. The good analytical performance of SD BioLine RDTs could be linked to their cassette support.

In addition to serum used as biological material in this study, the evaluated RDTs could use also, plasma or whole blood as biological matrix, which is an advantage for low-income countries, since viral hepatitis can be diagnosed everywhere as quickly as possible (Okpokam et al., 2015). Similarly, other researchers have shown that RDTs using serum, plasma, whole capillary blood, can be used in place of standard

Table 2
Sensitivity and specificity of the 6 rapid diagnostic tests compared to the Liaison XL®Quant (HBsAg) and RT-PCR (HCV).

Rapid Test	Total (N)	TP a	FP b	TN d	FN c	% Sensitivity (IC 95 %)	% Specificity (IC 95 %)	% PPV (IC 95 %)	% NPV (IC 95 %)	% Kappa (IC 95 %)
HBsAg marker										
Reference test	200	94	0	106	0					
A	200	94	6	100	0	100 (95,4 – 100)	94,3 (92,7 – 98,3)	94,0 (87,5 – 98,1)	100,0 (98,9 – 100,0)	97,6 (87,6 – 100,0)
B	200	94	6	100	0	100 (95,4 – 100)	94,3 (92,7 – 98,3)	94,0 (87,5 – 98,1)	100,0 (98,9 – 100,0)	97,6 (87,6 – 100,0)
C	200	94	3	103	0	100 (97,7 – 100)	97,1 (96,4 – 100,0)	96,9 (79,3 – 99,3)	100,0(99,6 – 100,0)	100,0 (99,1 – 100,0)
RT-TMA (HCV)										
Reference test	200	46	0	154	0					
D	200	46	54	100	0	100 (91,6 – 100)	64,9 (58,2 – 96,2)	46,0 (39,9 – 67,3)	100,0 (96,1 – 100,0)	49,3 (47,7 – 54,3)
E	200	46	54	100	0	100 (91,6 – 100)	64,9 (58,2 – 96,2)	46,0 (39,9 – 67,3)	100,0 (96,1 – 100,0)	49,3 (47,7 – 54,3)
F	200	46	3	151	0	100 (96,9 – 100)	98,1 (96,7 – 100,0)	93,9 (85,6 – 99,0)	100,0 (98,8 – 100,0)	96,7 (89,1 – 100,0)

A : One Step Hepatitis B Surface Antigen Test Strip, B: One Step strip Style HBsAg Rapid Screen Test, C: SD BioLine HBsAg WB (Multi), D: One Step Hepatitis C Virus Test Strip, E: Hepatitis C Virus Antibody (HCV) Test (Strip), F: SD BioLine HCV, TP = True Positive, FP = False Positive, TN = True Negative, FN = False Negative, PPV = Positive predictive value, NPV = Negative predictive value.

Table 3
Limit of detection of evaluated RDTs and liaison XL for the detection of HBsAg.

RDTs	viral load = 7 Log_{10}, HBsAg > 150 IU/ mL						undetectable viral load, HBsAg <150 IU /mL						viral load = 2 Log_{10}, AgHBs = 0.06 IU /mL					
	10^1	10^2	10^3	10^4	10^5	10^6	10^1	10^2	10^3	10^4	10^5	10^6	10^1	10^2	10^3	10^4	10^5	10^6
Reference test	+	+	+	+	+	–	+	+	+	+	–	–	–	–	–	–	–	–
A	+	–	–	–	–	–	+	–	–	–	–	–	–	–	–	–	–	–
B	+	–	–	–	–	–	+	–	–	–	–	–	–	–	–	–	–	–
C	+	–	–	–	–	–	+	–	–	–	–	–	–	–	–	–	–	–

A: One Step Hepatitis B Surface Antigen Test Strip, B: One Step strip Style HBsAg Rapid Screen Test, C: SD BioLine HBsAg WB (Multi).

enzyme-linked immunosorbent assays to facilitate screening for HBsAg and anti-HCV antibodies, and improve access to care, as suggested by recent recommendations from the international scientific society and WHO (Poiteau et al., 2017, 2018).

4.2. Performance of the rapid diagnostic tests

The analytical performance of the 6 RDTs commonly used in Lubumbashi was assessed by using the Liaison XL automatic analyzer and RT-PCR as reference methods for HBsAg et HCV detection respectively. The performance indicator parameters studied were sensitivity, specificity, PPV and NPV. At the end of this study, out of the six evaluated RDTs, only the SD BioLine (HBsAg, anti-HCV) RDTs demonstrated sensitivity and specificity of 100 % for the detection of HBsAg, sensitivity of 100 % and specificity of 97 % for the detection of anti-HCV antibodies. This result has been previously observed in HBV or HVC coinfected individuals with HIV using as RDTs the Vikia HBsAg® and Wama Immuno-Rapido HBV®, Bioeasy Teste Rapido HCV® and Wama Immuno-Rapido HCV® (Barbosa et al., 2017). Furthermore, in a study carried out in Tanzania, Franzeck et al. (2013) found a sensitivity and a specificity of 96 % and 100 %, respectively for RDT Determine HBsAg serum and whole blood from HIV-1 positive patients, which is in concordance with the data of the present study. De Paula Scalioni et al. (2014) have shown in their investigations that rapid HCV screening tests evaluated in high-risk individuals living in large urban centers, such as injecting drug users, had sensitivities varying from 90.8 to 99.9 %, and specificities from 92.1 to 99.9 %. These results are comparable to those obtained in our study for the RDT from SD BioLine. On the other hand, the remaining 4 RDTs including One Step Hepatitis (B surface Antigen, C Virus Test Strip) and One Step strip Style (HBsAg, C Virus Antibody) presented sensitivity and specificity of 100 % and 94 % respectively for the detection of HBsAg, while for the detection of anti-HCV antibodies, they presented a sensitivity and a specificity of 100 % and 65 % respectively. These results are in agreement with those of Poiteau et al. (2017); Mane et al. (2019) who indicated in their study that most RDTs developed for the detection of HBsAg meet the analytical sensitivity recommended by the WHO of 0.13 IU/mL, depending on the target population. However, our data are not in line with those of Chevaliez et al., 2016 who reported that the analytical sensitivity of each evaluated RDT varied considerably, while the specificity appeared satisfactory.

The SD BioLine (HBsAg, HCV) RDTs meet the WHO criteria recommending that RDTs should have a sensitivity of ≥ 99 % and a specificity of ≥ 95 % (Djimadoum et al., 2018). We therefore suggest their use as the first-line RDT for screening and diagnostic of HBV and HCV in the DRC.

In our study, the indicators of the analytical performance of the evaluated RDTs were different between the SD BioLine (HBsAg, HCV) RDTs and the 4 other ones. Several factors may explain the difference in results between these methods. Indeed, Parry et al. (2017) showed in their study that the causes of false reactivity are diverse and include cross-reactions due to antibody responses to other pathogens, immunizations, and autoantibodies. In such circumstances, the implementation of a 2-tier algorithm with a second serological test in order to confirm the reactivity initially observed can improve the differentiation of the false positive samples.

The agreement ratio between the evaluated RTDs and reference test (Liaison XL) for the detection of HBsAg was excellent because the agreement's kappa coefficient was higher than 81 %. With respect to the detection of HCV, SD bioline HCV showed kappa coefficient of 96.7 % whereas One Step Hepatitis C Virus Test Strip and Hepatitis C Virus Antibody (HCV) Test (Strip) showed kappa coefficient of 49.3 % because of the poor specificities presented by these two RDTs. Discordant results in specificities between the specificities observed in this study and those indicated by the manufacturers might be attributed to the overestimation of the performance indicators for commercial reasons by some manufacturers and also to the quality of reference tests used for the evaluation of the RDTs.

Regarding the limit of detection, even if the evaluated RDTs showed a sensitivity of 100 %, the study showed also that the Liaison XL method is more sensitive than the evaluated RDTs. Indeed, when the HBsAg level is >150 IU /mL, the reactivity of RDT stopped at $1/10^1$ dilution while that of Liaison XL went up to $1/10^4$ dilution. Variation of limit of detection of HBsAg by enzyme immunoassays and RTDs has been already described in different studies of different areas of the world (Barbosa et al., 2017). Scheiblauer et al., 2010 evaluated the sensitivity of 19 HBsAg rapid assays

and the results of this study were as follows: 1 RDT detected 1.5 IU/mL, 2 RDTs demonstrated sensitivities ranging from 1.7 to < 4 IU/mL, 3 RDTs detected 4 IU/mL, and 13 RDTs did not detect any of the standard dilutions as positive. These last tests could only detect undiluted serum samples with HBsAg concentration > 4 UI/mL. In our study, the detection limit for the tested RDTs and the Liaison XL automatic analyzer has been estimated being ≥ 0.06 IU/mL of HBsAg.

The current study had some limitations: first, the sample size was limited and included only blood donors who were apparently healthy individuals. Even if they were infected with HBV or HCV, it should be interesting in the future to carry out such evaluations using a large sample made up of individuals coinfected since according to some studies (Barbosa et al., 2017), coinfection may affect the reliability of RDTs; second, all the evaluated RDTs were of Asian origin, evaluating tests from other areas of the world could give other insights.

5. Conclusion

The aim of our study was to assess the analytical performance of six RDTs used in Lubumbashi (DRC) for the diagnosis of viral hepatitis B and C. Based on the obtained results and taking into account the WHO recommendations in terms of reliability criteria for rapid diagnostic tests, RDT Bioline (HBsAg, anti-HCV) may be eligible as a first line test for the detection and the diagnostic of viral hepatitis B and C in Lubumbashi (DRC). They should therefore very soon be used systematically in the screening of blood donors for HBV and HCV in Lubumbashi since they could prevent blood-borne transmission of HBV and HCV in areas with limited incomes. On the other hand, taking into account the relative lack of specificity of RDTs as further demonstrated in this study, it will be wise for any positive sample to be subject to a confirmation testing that could be carried out by a reliable method such as ELISA, Liaison XL or PCR. In addition, it would also be necessary to investigate the possible causes of the lack of specificity of the evaluated RDTs in order to improve the level of their reliability since they remain the most easily accessible tests locally. The sensitivity of evaluated RDTs was in line with the WHO recommendations. To the best of our knowledge, this is the first study reporting independently the assessment of the performances of RDTs used for the detection of viral hepatitis B and C in Lubumbashi. Our study highlighted the need for analytical and clinical performance evaluation of any new RDT prior to any Congolese marketing authorization.

Author statement

We are pleased to resend the revised version of our manuscript along with responses to reviewer's concerns. The manuscript revision has been revised in accordance with a few well-founded suggestions from the reviewer.

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Declaration of Competing Interest

The authors report no declarations of interest.

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