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# Prevalence and determinants of Hepatitis C Virus Infection and Genotypes in Chronic Hemodialysis Patients in Kinshasa

## *Prévalence et déterminants de l'infection par le virus de l'hépatite C et génotypes chez les hémodialysés chroniques, à Kinshasa*

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**Résumé**

*Contexte et Objectifs*. Le nombre des patients hémodialysés en Afrique subsaharienne est en constante augmentation ; justifiant de ce fait une meilleure prise en charge de ces patients. La présente étude a déterminé la prévalence de l'infection par le virus de l'hépatite C en précisant les génotypes ainsi que les facteurs y associés dans ce groupe de patients. *Méthodes*. 127 patients hémodialysés chroniques ont subi des tests sérologiques à la recherche des anticorps anti-VHC dans plusieurs centres de Kinshasa de février à juin 2018. Le génotype viral a été realisé par la RT-PCR. *Résultats*. La fréquence des anticorps anti-VHC a varié de 0 à 52,9 % dans ce groupe. Les génotypes les plus fréquents ont été les 4 (18/22) et les 2 (2/22) ; étant simultanément rétrouvé chez un patient, et indéterminé chez un autre sujet. Avoir reçu au moins 4 transfusions [7,21 (1,09-10,61; p=0,040)], ne pas être sous EPO [5,81(1,47-12,96); p=0.012)], être en hémodialyse depuis au moins 14 mois [3,63 (1,60-5,05); p=0.035)] et être dialysé dans un centre surchargé [2,06 (0,83-5,86); p=0,073)] étaient associés à un risque plus élevé d'infection par le VHC. *Conclusion*. Ses principaux déterminants sont : le nombre des transfusions sanguines et la durée d'HD ; d'où la nécessité de réduire les transfusions sanguines chez les sujets dialysés par l'administration d'EPO, étant donné le coût prohibitif du traitement contre le VHC dans notre contexte.

**Mots-clés** : Virus de l'hépatite C, Génotypes, Hémodialyse, Facteurs de risque, RD Congo
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**Summary**

*Context and objective*. The steady increase in the number of chronic hemodialysis patients in sub-Saharan Africa (SSA) calls for improved management of those patients. The present study aimed to determine the frequency of hepatitis C virus (HCV) infection, the prevalent genotypes, and the risk factors associated with HCV in hemodialysis patients in Kinshasa (DR Congo). *Methods*. A cross-sectional study was conducted from February to June 2018 in all hemodialysis centers in Kinshasa. Blood samples were collected from 127 chronic hemodialysis patients and tested for the presence of antibodies against HCV. The HCV genotype was identified by real-time polymerase chain reaction (RT- PCR). *Results*. Twenty-two (17.3 %) patients were positive for anti-HCV antibodies, ranging from 0 % to 52.9 % in different centers. Genotype 4 was detected in 18/22 (81.8 %), followed by genotype 2 in 2/22 (9.1%), and both genotypes 2 and 4 in one patient (4.5%). One patient had an undetermined genotype (4.5 %). Having received at least 4 transfusions [7,21 (1,09-10,61); p=0.040)], not being under EPO treatment [5,81(1,47-12,96); p=0.012)], being on hemodialysis for at least 14 months [3,63(1,60-5,05); p=0.035)]and being dialyzed in an overloaded center [2,06(0,83-5,86); p=0.073)] were associated with a greater risk of HCV infection. *Conclusion*. This high HCV prevalence (17.3 %) represents a substantial health burden in HD patients from Kinshasa, DR Congo. It is largely driven by the number of blood transfusions, the duration time in hemodialysis. Observations from the present study underscore the need of reducing the number of blood transfusions in people on dialysis through the administration of erythropoietin, given the unaffordable cost of HCV therapy for most individuals in DR Congo.

**Keywords**: hepatitis C virus, genotype, Hemodialysis, Prevalence, Risk factors, DR Congo.

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## Introduction

The infection risk in hemodialysis (HD) patients is considerable, explained by multiple factors including impaired immunity, comorbidities such as diabetes, and the need for frequent hospitalizations and surgical interventions. HD itself further involves frequent and prolonged exposure to blood by means of vascular access, and the proximity of other patients during dialysis sessions. Thus hepatitis C virus (HCV) infection is rather common in HD patients. Identifying acute HCV infections is often difficult, as most patients tend to be asymptomatic. However, 75-85% of subjects with HCV infection will develop chronic hepatitis, and 10-20% will develop liver cirrhosis over 20 years (1-2). Therefore, it is important that dialysis centers have adequate processes for preventing HCV infection, including training of staff and regular audits.

The Dialysis Outcomes and Practice Patterns Study (DOPPS) reported recently an overall HCV prevalence of 9.9 % among adult hemodialysis patients in high- and middle-income countries (3). Actually, the prevalence varied greatly from one country to another (ranging from 4.1% in Belgium to 20.1% across the Gulf Cooperation Council Countries), and from one dialysis center to another (3). Compared to HCV-negative patients, HCV-positive patients enrolled in DOPPS had a higher risk of all-cause and liver-related mortality and all-cause hospitalization (4). Thus the KDIGO 2018 guideline recommends that each hemodialysis center carefully follows every HCV-infected dialysis patient, measures viral load, assesses the extent of hepatic fibrosis, and establishes optimal treatment strategies (5). Furthermore, with the recent Direct-acting Antiviral Agent (DAA), elimination of HCV from the general population and from dialysis centers may be possible, thereby reducing the considerable morbidity and mortality associated with this disease; but for economic reasons, DAAs are rarely administered in sub-Saharan Africa (SSA) (6). In the absence of a vaccine, routine screening and adherence to standard infection control practices will remain the key strategies for preventing HCV transmission in hemodialysis centers (5).

Data on HCV infection in hemodialysis patients in SSA are scarce. There have been a few single-center studies and none assessed risk factors for hepatitis C virus infection (7-9).

Recent HD studies from SSA generally report higher prevalence of HCV compared to western countries. The present multicentric study aimed to determine the prevalence of HCV infection and genotypes in all Kinshasa (Democratic Republic of Congo) hemodialysis centers and identify the risk factors associated with HCV infection.

## Methods

This cross sectional study was performed in all hemodialysis centers of Kinshasa, the capital city of DR Congo. The parameters of interest related to patients were: age, sex, marital status, blood group, medical history (viral status at the initiation of hemodialysis including Hepatitis B Virus = HBV, Human Immunodeficiency Virus = HIV and HCV), number of transfusions since HD start, nosocomial exposure (Dental care, Piercing or Tattooing, Surgical intervention or endoscopic treatment) IV Drug addiction, and known liver disease.

Concerning parameters related to hemodialysis, we recorded time on hemodialysis in months, cause of chronic kidney disease (CKD) and the type of vascular access (AVF, KT, Prosthesis). Other parameters of interest related to hemodialysis centers were: number of treatment rooms in the center, number of machines in the center, number of staff (doctors, nurses and other caregivers), and number of patients on HD. All centers used high permeability dialyzers and respected the recommended duration of disinfection between two dialysis sessions.

Blood sample (10 ml) was obtained from each patient before the hemodialysis session, and centrifuged immediately. Serum was stored in aliquots at −80°C. All samples were subsequently subjected to liver function tests: alanine aminotransferase (U/l), aspartate

aminotransferase (U/l), gamma-glutamyl transpeptidase (U/l), and bilirubin concentration (mg/dl) in the laboratory of LOMO MEDICAL, Kinshasa. Anti-HCV antibodies were detected by a third-generation ELISA (Huma Reader Single®, France). The third-generation assay of ELISA detects antibodies to three HCV antigens (c22-3, c200, and NS5). In collaboration with the Indian laboratory of HJ Hospital in Kinshasa, HCV-RNA testing was performed using reverse transcription-polymerase chain reaction (RT-PCR) (Cobas 4800®, Roche, France) with a detection limit of 100 copies/ml.

*The operational definitions used were as follows*

* HCV infection: the presence of anti-HCV antibodies by the 3rd generation ELISA and confirmation of the HCV viral infection by RT-PCR;

Chronic hemodialysis: any patient with CKD stage 5 on hemodialysis for more than 3 months;

* HCV seroconversion: any patient included in the study with a previous negative HCV antibody but who during the study had positive serology for HCV by the 3rd generation ELISA method.

Depending on the prices of HD sessions, the HD centers were grouped into category A (which applies low prices = 1/3) and category B (which applies standard prices = 200 $ US).

*Statistical analyses*

Statistical analyses were performed using SPSS 21.0 for Windows (SPSS Inc., Chicago, IL, USA). Comparisons between groups were performed using Student's t-test, Fisher's exact test and the Chi-square test, as appropriate. Logistic regression analysis was used to identify the risk factors associated with HCV infection. A p-value < 0.05 was considered statistically significant.

Ethical considerations

The study protocol was approved by the ethics committee of the Public Health School of the University of Kinshasa (ESP/CE/032/2018). The rules of anonymity were respected both for the names of the patients and the medical structures (hemodialysis centers). All patients screened positive for HCV infection were referred to gastroenterologists for evaluation and treatment.

## Results

A total of 185 patients from 6 hemodialysis centers were considered to participate in this cross-sectional study between February and June 2018. Only 127 patients meeting inclusion criteria and giving informed consent were investigated. The remaining 58 patients could not be tested for the following reasons: acute kidney injury (AKI) (n=6), chronic hemodialysis treatment for less than 3 months (n=18), refusal to take samples (n=2), travel (n=6), or loss to follow-up during the investigation (n=26). Of the 127 patients tested for HCV antibodies, 22 were positive (17.3 %). All positive samples were confirmed by RT-PCR. We had 2 cases of seroconversion (1.9 %) compared to previous tests performed three to six months earlier. Viral load varied between 1,567 and 3,310,000 copies/ml (median: 14,400 copies / ml). Figure 1 shows that genotype 4 was the most common.

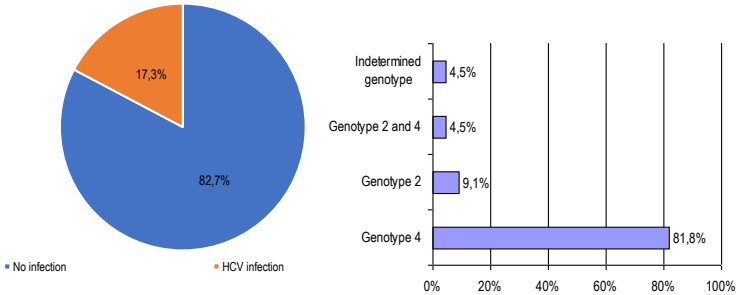


**Figure 1**. Prevalence of HCV infection (A) and HCV genotypes (B) in chronic HD patients in Kinshasa

Table 1 shows that the frequency of infection varied between centers, ranging from 0 to 52.9 %.

**Table 1. Frequency of HCV infection by hemodialysis center**

| Hemodialysis center | Number of rooms | Number of Nurses/doctors | Total number of patients | Patients included | HCV+Prevalence |
|---|---|---|---|---|---|
| 1 A | 2 | 12 | 51 | 17 | 9 (52.9) |
| 2 B | 2 | 8 | 17 | 16 | 5 (31.2) |
| 3 B | 4 | 14 | 54 | 43 | 5 (11.6) |
| 4 B | 2 | 17 | 30 | 24 | 2 (8.3) |
| 5 B | 2 | 10 | 27 | 22 | 1 (4.5) |
| 6 B | 2 | 10 | 5 | 5 | 0 |
| Overall | 14 | - | 184 | 127 | 22 (17.3) |

A: center applying 1/3 of standard dialysis session prices
B: center applying standard dialysis session prices

The HCV infected patients had a time on hemodialysis treatment, were less frequently under EPO and had received more blood transfusions since the start of hemodialysis (table 2).

**Table 2. Patient characteristics according to HCV infection status**

| Variables | Whole group n=127 | $HCV^+$ n=22 | $HCV^-$ n=105 | p |
|---|---|---|---|---|
| Age | 54.7 ± 13.9 | 57.3 ±15.2 | 54.3± 13.7 | 0.36 |
| Sexe | | | | |
| - Men | 82 (64.6) | 15 (68.2) | 67 (63.8) | 0.70 |
| - Women | 39 (30.7) | 7 (31.8) | 38 (36.2) | |
| Time on hemodialysis | 22.4 ± 8.6 | 27.4 ± 8.7 | 21.3 ± 6.2 | < 0.001 |
| Cause of CKD | | | | |
| - Diabetic nephropathy | 47 (37.0) | 7 (31.8) | 40 (38.1) | 0.63 |
| - Nephrosclerosis | 47 (37.0) | 9 (40.9) | 38 (36.2) | |
| - Glomerulonephritis | 25 (19.7) | 6 (27.3) | 19 (18.1) | |
| Vascular access | | | | |
| - Fistula | 24 (18.9) | 1 (4.6) | 23 (21.9) | 0.06 |
| - Catheter | 103 (81.1) | 21 (95.5) | 82 (78.1) | |
| Number of transfusions | 3.9 ± 2.0 | 4.6 ± 1.0 | 3.6 ± 2.2 | 0.04 |
| HBV serology positive | 3 (2;4) | 0 | 3 (2.9) | 0.42 |
| HIV serology positive | 4 (3.1) | 1 (4.5) | 3 (2.9) | 0.70 |
| EPO | 83 (65.4) | 7 (31.8) | 76 (72.4) | < 0.001 |

Results are expressed as means ± standard deviation (SD) or absolute frequency (%). CKD: chronic kidney disease HBV: hepatitis B virus HIV: human immunodeficiency virus EPO: erythropoietin

Multivariate analysis (table 3) showed that having received 4 Transfusions multiplied the risk of being infected by 7.21 (1.09-10.61). The risk was multiplied by 9.59 (1.67-14.94) beyond 8 transfusions. Not receiving EPO treatment increased the risk by 5.81 (1.47-12.96). After 14 months undergoing hemodialysis treatment, the risk of being infected by HCV was multiplied by 3.63 (1.60-5.05). Compared to the 5 other centers, the patients followed in center 1 had a risk multiplied by 2.06 (0.83-5.86).

**Table 3. Risk factors of HCV infection in chronic hemodialysis patients**

| | Univariate analysis | | Multivariate analysis | |
|---|---|---|---|---|
| Factors | p | OR (CI 95%) | P | OR (CI 95%) |
| Catheter vs fistula | 0.039 | 5.89 (1.72-46.16) | 0.363 | 2.83 (0.30-6.49) |
| Number of transfusions | | | | |
| <4 | ref. | | | 1 |
| 4-8 | 0.037 | 5.04 (1.11-22.96) | 0.040 | 7.21 (1.09-10.61) |
| >8 | 0.001 | 11.20 (2.56-49.05) | 0.011 | 9.59 (1.67-14.94) |
| No EPO vs EPO | 0.001 | 5.62 (2.08-15.17) | 0.012 | 5.81 (1.47-12.96) |
| Duration on HD | | | | |
| ≥14 vs <14 months | 0.008 | 4.28 (1.50- 6.22) | 0.035 | 3.63 (1.60-5.05) |
| Center A vs centers B | <0.001 | 8.39 (2.75-25.59) | 0.073 | 2.06 (0.83-5.86) |

Ref: reference; A: center applying 1/3 of standard prices; B: center applying standard dialysis prices

## Discussion

This study shows a high frequency of HCV infection in patients on chronic hemodialysis in Kinshasa, with disparities between centers. Genotype 4 was the most common. The risk factors associated with HCV infection were the number of blood transfusions, the absence of EPO treatment, the time on HD as well as the characteristics of the centers.

Studies in Europe indicate a decrease in incidence of HCV infection in parallel with prevalence in hemodialysis centers (3) over the last 10 years, while others countries, especially in Eastern Europe, maintain a high incidence (10). The frequency of HCV infection we observed is higher than found in hemodialysis centers in Western Europe and North America (3- 4, 10). Reducing the number of blood transfusions in people on dialysis through administration of erythropoietin, aseptic measures including the use of single use disposables such as syringes and use of the right disinfectants, are other factors that have contributed to lowering the frequency of HCV infection in developed countries (5), as did the testing of blood donors for HCV from 1992 with increasingly sensitive tests (11). The recent treatments of HCV, particularly with DAA, may well increase the gap between countries because availability of DAA is not guaranteed in several countries including those in SSA (12). However, the average prevalence in this study is similar to those reported in other dialysis centers in SSA (7-9).

It is also known that in each country, the frequency of HCV infection in hemodialysis varies from center to center (3). This finding was confirmed in our study where we observed a disparity between the centers, with the frequency of HCV infection varying between 0 and 52.9 %. Despite the fact that the number of participants was low (only 17 patients out of 51) in the center practicing lower prices per HD session and the serological status at the initiation of HD was not well established, the high frequency of HCV infection in this center could be explained by the high treatment rates with an inadequate nursing medical staff-patient's ratio probably responsible for nosocomial transmission. Because in a country like the DRC where dialysis is not subsidized and where social security and mutual funds are poorly developed, the consumables used in dialysis inevitably participate in the calculation of the prices of hemodialysis sessions (13). The motivation of nursing/medical staff and their number in relation to patients treated also have an impact on the quality of care. But these aspects were not analyzed in our study. Only 2 patients were seroconverted to HCV. This is nevertheless important knowing that, in accordance with the recommendations of the Congolese Society of Nephrology (SOCONEPH), the previous control went back to 6 months previously, unless the patient had to move to another dialysis center. In this case, serology checks for HCV, HIV, and HBV are carried out systematically. The rate of seroconversion found (1.9 %) is almost identical to that reported in the literature (14-16). The

general measures that are recommended to prevent seroconversion are: limitation of handling and safe disposal of sharp or stinging objects and contaminated waste, hand washing, use of single-use surgical gloves, changed after each patient, training of personnel, the use of machines which allow thermal and chemical disinfection, disinfection of the external surfaces of the machine and HD environment (5).

Knowledge of the HCV genotype is important for the choice of antiviral treatment (12). As in studies conducted in the general population in SSA (17), and even in hemodialysis patients treated in Angola (18), genotype 4 was predominant in our study.

Several factors explain the HCV infection in hemodialysis, but the most frequently reported in the literature are blood transfusions and the duration of treatment in hemodialysis. Other factors were highlighted by the multicenter study (DOPPS) such as male gender, diabetes mellitus, black race, hepatitis B and alcohol (3). In our study, HCV infection was associated with the number of blood transfusions, the duration of hemodialysis, not taking EPO, and the type of hemodialysis center. Strengthening the national health system, improving the country's economic situation, can help increase the use of EPO in hemodialysis; which will reduce the number of transfusions. If a blood transfusion is necessary, a more demanding qualification of the blood must be ensured in the blood banks. The risk associated with the type of hemodialysis center shows that while improving access to treatment thanks to lower prices, the prevention and hygiene measures described above must be observed. Indeed, a link with suboptimal hygienic precautions seems likely. Taking into account the very high frequency of HCV infection in this type of center, i.e. roughly one in 2 patients, we can wonder whether it is not advisable to dialyze these patients in isolated rooms from other uninfected patients? Admittedly this would equal to accepting suboptimal hygienic precautions and should be a last option resort. The hemodialysis setting has unique features that facilitate the transmission of HCV, such as a high risk of blood contamination of surfaces, objects, and devices, and a large number of patients treated simultaneously in a shared space. In SSA countries, risk factors associated with HCV infection may be due to unsafe medical practices or other factors such as familial transmission, mother's HCV status, or illiteracy. HCV prevention and control programs should include health education, increased community awareness towards the disease, controlling infection distribution in healthcare centers, proper sterilization of medical and dental instruments, and ensuring a safe supply of blood and blood products (19).

## Conclusion

This high HCV prevalence (17.3 %) represents a substantial health burden in HD patients from Kinshasa, DR Congo. It is largely driven by the number of blood transfusions, the duration time in hemodialysis. Our observations call the need of reducing the number of blood transfusions in people on dialysis through administration of erythropoietin, given the unaffordable cost of HCV therapy for most individuals in the DR Congo.

### Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be considered as a potential conflict of interest.

### Author's contributions

MJR and MMN conceived the idea, designed and supervised the study, had full access to all data and took responsibility for the integrity of the data. MMN collected the clinical and laboratory data. MJR analyzed data and performed statistical analysis. All co-authors reviewed and approved the final version.

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