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Clinical, Biological, Immunological and Therapeutic Profile of Patients Co-Infected with HIV-HBV and/or HCV in Kinshasa, in the Democratic Republic of the Congo: Multicenter Cross-Sectional Study

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Abstract

Background and Objective: HIV infection is often associated with HBV and HCV infection, together leading to high morbidity and mortality in developing countries. The objective of this study is to describe the clinical, biological, immunological and therapeutic profile of patients co-infected with HIV-HBV and/or HCV. **Methods:** A cross-sectional and descriptive study including 180 people living with HIV (PLWHIV) in the city of Kinshasa province was conducted. Socio-demographic, clinical, biological and serological characteristics were analyzed. **Results:** The frequency of HIV-HBV/HCV co-infection was 23.9%. The distribution of age and sex of patients did not differ significantly according to co-infection status. The notion of pedicure and manicure was significantly more observed in patients free from viral hepatitis (51.1% *versus* 32.6%, p = 0.034). The median duration of knowledge of the HIV status which was longer in the co-infected (4 years *versus* 2 years, p = 0.022). A

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lower median level of GPT was observed in co-infected compared to other patients (14 IU/L *versus* 20 IU/L, p = 0.041). Serum albumin (3.1 g/L versus 3.3 g/L, p = 0.034) and prothrombin (58.3% *versus* 65.6%, p = 0.045) were lower in HIV co-infected-VHB and/or VHC. The median INR was higher in co-infected than in other patients (1.6 *versus* 1.4; p = 0.009). Patients without therapy Antiretroviral (TARV) medication were more numerous in co-infected (20.9% versus 8.0%, p = 0.025). **Conclusions:** The profile of PLWHIV was dominated by the presence of pedicures and manicures with high transaminases and without anti-viral treatment.

Keywords

HIV-HCV-HBV Co-Infection, Clinical Profile, Biology, TARVs, Kinshasa

1. Introduction

The hepatotropic and lymphotropic virus, HCV and HBV induce a specific cytotoxic T immune response which, combined with the production of interferon γ, allows in some patients to eliminate the virus with a frequency of around 11% to 32% in HIV-HCV/HBV co-infected patients [1]. However, the chances of spontaneous recovery from acute hepatitis C or B in people living with HIV (PLWHIV) are lower than without HIV infection, ranging from 11% to 32% [2]-[8]. The transition to the chronic phase of viral C and B infection is the result of the modulating effect exerted by the virus itself on the balance established after infection between costimulatory and pro/anti-apoptotic molecules, between Treg cells and T-helpers but also the establishment of escape mechanisms that allow the virus to remain in the body indefinitely, mainly in hepatocytes [9]. However, there is evidence of HCV or HBV replication in other cell types including lymphocytes. Chronic viral replication of HCV and HBV occurs against a background of permanent immune activation, higher in HIV-HCV/HBV co-infected patients [10], whose long-term effects are only just beginning to be identified and quantified. Several studies have thus demonstrated a higher vascular risk in HCV/HBV mono-infected patients (stroke, obliterating arterial disease of the lower limbs, coronary artery disease), as well as renal failure (by nephroangiosclerosis and extra-membranous glomerulonephritis), diabetes mellitus and cancers, especially extrahepatic, the first of them identified being non-Hodgkin's malignant lymphoma [11] [12] [13] [14]. In the Democratic Republic of the Congo, we do not have sufficient data to support the role of HCV/HBV co-infection in patients infected with HIV. The objective of this study is to describe the clinical, biological, immunological and therapeutic profile of patients co-infected with HIV-HBV and/or HCV.

2. Methods

It was a cross-sectional and descriptive study. It took place in the city of Kinsha-

sa, the political capital of the Democratic Republic of the Congo. In this city, HIV care is organized in all general hospitals, in certain general and university hospitals, in certain private medical centers and in the medical training of certain NGOs. Depending on the availability and completeness of the information sought in our study, we considered two NGOs located in urban-rural areas, namely: the Pediatric Foundation of Kimbondo and the NGO "Community Actions AIDS/Better Future for Orphans of Congo (ACS/AMO-Congo) "which are among the oldest centers in Kinshasa for HIV care. It was during the period from November 10, 2013 to January 10, 2014. The study had consecutively included elderly people of at least 18 years known to have HIV who consulted during the study period at the two selected sites and who freely consented to participate in the study. Inclusion criteria were to be PLWHIV at least 18 years of age consenting to participate in the study and followed in one of the two treatment centers selected for the study, to have a medical file including the parameters sought. To carry out the present study, were used as materials data collection sheet, informed consent sheet, strips for rapid qualitative tests HBsAg and AcVHC of the brand "ACCURATE of Indian manufacture", quantitative tests for the determination of the markers of the hepatitis B (ELISA) brand "DIALAB Austrian manufacture", test for determination of HCV antibodies (ELISA) brand "DIALAB Austrian manufacture", tests for evaluation of hepatic synthetic function.

The actual data collection involved 3 stages: 1) administering a questionnaire to patients to collect socio-demographic information, medical history and risk behaviors for viral hepatitis; 2) the blood test for the determination of markers of the hepatic functions studied and of HBV and HCV; 3) analysis of the medical file of each patient selected in search of clinical, immunological and therapeutic information relating to HIV infection. Socio-demographic parameters included age, sex, occupation, marital status, level of education and religion. Regarding the medical history and risky behavior, we looked for the concept of previous blood transfusion, vaccination against HBV, the number of sexual partners, the type of sexual intercourse, the concept of scarification, circumcision, excision, piercing, drug addiction, knowledge of one's HBV and HCV serological status and the notion of surgical intervention in the past. The analysis of the medical file of each patient allowed us to gather information on the year of the diagnosis of HIV, the clinical stage of the HIV infection, the current ARV treatment regimen and, if applicable, the highest rate, recent CD4; only CD4 counts not older than 3 months before the survey were taken into account. On the blood test, the HBV biological markers sought were HBs Ag, anti-HBs Ab, anti-HBc Ab, HBe Ag and anti-HBe Ab. For HCV, total anti-HCV Abs were assayed. Transaminases (SGOT and SGPT) were the desired markers of hepatic cytolysis. To explore cholestasis, we assayed for γGT, total and direct bilirubin as well as alkaline phosphatases. To assess hepatic synthetic function, prothrombin, serum albumin, and INR were assayed. In terms of the collection process, any patient se-

lected for the study received, after consultation by the center's medical team, the survey questionnaire. After completing the questionnaire, the patient was directed to the laboratory with a token bearing an identification code. Once arrived at the laboratory, a 5 cc venous blood sample was taken in two tubes, a dry tube for hematological and biochemical analyzes and another tube with citrate for serological analyzes. The samples were centrifuged using a German brand A-RD-42-26 device at 1500 revolutions/min/5min, decanted and then stored in the refrigerator at a temperature between 2˚C to 8˚C at the site laboratories. studies. Qualitative tests for HBsAg and for total HCV antibodies were performed on site; the rest of the samples were sent to the Lomo Médical laboratory where hematological, biochemical and serological analyzes were carried out.

A spectrophotometer of the brand "Spectrum" and a "Bain Marie of the HUMAN brand" and ELISA reader "HIMARETADR-FINGLE of the HUMAN brand" were used to carry out the hematological and biochemical analyzes according to the manufacturer's standards. Kinetic methods for GOT, GPT, PAL, γGT; enzymatic for serum albumin, total and direct bilirubin, prothrombin and INR; and immunoenzymatic type ELISA for quantitative tests for viral hepatitis B and C were used. The results were transcribed on an ad hoc form with the corresponding codes.

Definition of concepts

Anti-HBs Ab were found to be positive when in the Elisa test, the anti-HBs Ab titre was >12 IU/L [15];

HIV-HBV co-infection was selected on the basis of the positivity of HBsAg in HIV + patients [15];

HIV-HCV coinfection was selected on the basis of the positivity of anti-HCV Ab in HIV + patients [15].

Statistical analyzes

The processing and analysis of the data collected was carried out using SPSS version 21 software. During the analysis, the age of the patients was categorized into tertiles (<40 years, 40 - 49 years and ≥50 years). CD4 counts were dichotomized at the threshold of their median value (3 years and 303 cells/mm^3, respectively). The descriptive statistics applied include the mean ± standard deviation for continuous quantitative variables with symmetric distribution, the median with extreme values for those with asymmetric distribution, and relative (%) and/or absolute (n) frequencies for qualitative variables. For comparison of means and medians, Student's t test and Wilcoxon/Mann-Whitney nonparametric test were applied. For the analysis of the contingency tables, we used Pearson's chi-square test or Fisher's exact test or linear trend chi-square, as appropriate. For the tests used, the statistical significance level retained was p value < 0.05.

Ethical considerations

The protocol was submitted to the ethics committee of the School of Public Health of the University of Kinshasa and was agreed at number ESP/CE/012/14. Thus, when recruiting patients, anonymity and confidentiality were guaranteed.

3. Results

Clinical profile of HIV-HBV and/or HCV co-infected of the 180 PLWHIV included in the study, 43 had an HIV-HBV-HCV coinfection, a frequency of 23.9% coinfection.

Table 1 presents the clinical profile of all patients and the HIV-HBV and/or HCV co-infection status.

The age and sex distribution of the patients did not differ significantly according to co-infection status. The notion of pedicure and manicure was significantly more

Table 1. Clinical characteristics of patients according to HIV-HBV and/or HCV co-infection status.

Variable	Over all (n = 180)	HIV (n = 137)	HIV+/HBV+/ HCV (n = 43)	p value
Age (year)	44.2 ± 11.0	44.6 ± 10.6	42.8 ± 12.2	0.369
<40%	33.3	29.9	44.2	0.132
40% - 49%	34.4	38.0	23.3	
≥50%	32.2	32.1	32.6	
Gender, %				0.416
Female	76.7	78.1	72.1	
Male	23.3	21.9	27.9	
Transfusion, %	32.8	33.6	30.2	0.684
Pedicure and manicure, %	46.7	51.1	32.6	0.034
Jaundice in the past, %	5.6	5.1	7.0	0.704
Concept of anti-HBV vaccination, %	1.1	1.5	0.0	-
Scarification, piercing, excision, %	40.0	39.4	41.9	0.775
Substance addiction, %	6.7	5.1	11.6	0.161
Number of sexual partners, %				0.125
1	61.1	64.2	51.2	
>1	38.9	35.8	48.8	
Condom use, %	20.0	21.2	16.3	0.484
Surgery in the past, %	45.0	43.8	48.8	0.562
Knowledge of status	1.1	0.7	2.3	0.424
Duration of HIV infection (years)	3 (1 - 20)	4 (1 - 15)	2 (1 - 20)	0.022
Clinical stage of HIV, %				0.194
I	10.0	8.0	16.3	
II	30.6	28.5	37.2	
III	46.1	49.6	34.9	
IV	13.3	13.9	11.6	

observed in patients free from viral hepatitis (51.1% versus 32.6% in co-infected, p = 0.034). The same is true of the median duration of knowledge of the HIV status, which was significantly longer in this latter group (4 years versus 2 years in the co-infected, p = 0.022). The other characteristics noted, in this case, a history of transfusion (32.8%), jaundice (5.6%), anti-HBV vaccination (1.1%), scarification, piercing and excision (40%), drug addiction (6.7%) or surgery (45%), condom use (20%), number of sexual partners, or clinical stage of HIV infection did not differ significantly according to co-infection status.

Biological profile of HIV-HBV and/or HCV co-infected

The data are indicated in Table 2.

Biologically (Table 1), the median levels of SGOT (23 IU/L), gamma-GT (28 IU/L), alkaline phosphatase (160 IU/L), total bilirubin (0.4 g/L) and direct bilirubin (0.1 g/L) did not differ significantly depending on coinfection status. In contrast, a significantly lower median level of SGPT was observed in HIV-HBV and/or HCV coinfected compared to other patients (14 IU/L *versus* 20 IU/L; p = 0.041). The same is true of the mean serum albumin levels (3.1 g/L *versus* 3.3 g/L; p = 0.034) and prothrombin (58.3% *versus* 65.6%; p = 0.045) which were significantly lower in the group of HIV-HBV and/or HCV co-infected. The median INR was significantly higher in co-infected than in other patients (1.6 *versus* 1.4; P = 0.009).

Therapeutic profile of HIV-HBV and/or HCV co-infected

The results are presented in Table 3.

The ARV therapeutic profile of the patients differed significantly depending on the co-infection status. In particular, more patients without ARV medication were observed in the group of HIV-HBV and/or HCV co-infected (20.9% versus 8.0%; p = 0.025) (Table 3).

The median CD4 lymphocyte count did not differ significantly depending on

Table 2. Laboratory characteristics of patients according to HIV-HBV and/or HCV co-infection status.

Variable	Over all (n = 180)	HIV (n = 137)	HIV+/HBV+/ HCV (n = 43)	p
SGOT (UI/L)	23 (1 - 748)	25 (1 - 138)	18 (2 - 748)	0.055
SGPT (UI/L)	19 (4 - 150)	20 (4 - 150)	14 (4 - 144)	0.041
Gamma-GT (UI/L)	28 (4 - 232)	30 (4 - 232)	24 (4 - 129)	0.623
Alkaline phosphatases (IU/L)	160 (18 - 689)	157 (34 - 689)	182 (18 - 565)	0.116
Total bilirubin (g/L)	0.4 (0.1 - 3.9)	0.4 (0.1 - 1.2)	0.5 (0.1 - 3.9)	0.313
Direct Bilirubin (g/L)	0.1 (0.0 - 1.0)	0.1 (0.0 - 0.8)	0.1 (0.0 - 1.0)	0.822
Serum albumin (g/L)	3.2 ± 0.6	3.3 ± 0.6	3.1 ± 0.6	0.034
Prothrombin (%)	63.8 ± 21.0	65.6 ± 21.0	58.3 ± 20.3	0.045
INR	1.4 (1.0 - 5.9)	1.4 (1.0 - 5.9)	1.6 (1.0 - 5.9)	0.009

Data are expressed as mean ± standard deviation or median (range).

Table 3. Therapeutic characteristics of patients according to HIV-HBV and/or HCV co-infection status.

Variable	Over all (n = 180)	HIV (n = 137)	HIV+/HBV+/HCV (n = 43)	p
ARV treatment, %				0.025
No treatment	11.1	8.0	20.9	
AZT + 3TC + NVP	73.9	73.7	74.4	
TDF + 3TC + EFV	11.1	13.9	2.3	
ABC + DDI + [ALUVIA ou LPV/r]	3.9	4.4	2.3	

the HIV-HBV/C co-infection status (347 cells/mm^3 in the co-infected patients versus 303 cells/mm^3 in the non-co-infected; p = 0.606).

4. Discussion

The purpose of this study was to describe the clinical, biological and therapeutic profile of HIV-HBV-HCV co-infection in Kinshasa. With regard to HIV-HBV/HCV co-infection, the age groups < 40 years, 40 - 49 years and ≥50 years represented a frequency of 44.2%, 23.3%, 32.6%, respectively. The difference in frequency between age groups was not statistically significant (p = 0.132). These results are contradictory to other epidemiological studies reported in the literature [16] [17] [18]. The discrepancy of our results compared to the aforementioned studies is justified by the fact that the data of the literature read so far and which mention the age do not speak about the importance of the infection with HBV and HCV compared to each age category. The difference in work methodology took us away, but also the population sizes of the aforementioned studies were almost 2 to 20 times larger than ours [16] [17] [18].

In addition, the female sex is more predominant (72.1%) than the male sex (27.9%). This result is consistent with the fact that in the general population female PLHIV are more common. The practice of manicures and pedicures has emerged as the main risk factor for the occurrence of HIV-hepatitis B and/or C co-infection alongside the duration of the HIV infection. The laboratory profile of co-infected patients was dominated by the high level of GPT 14 (4 - 144), an increase to 3x> normal. These results are close to those of Idoko in Nigerian [19]. A low level of serum albumin, prothrombin and INR was observed with respectively 3.1 ± 0.6 (p = 0.034), 58.3 ± 20.3 (p = 0.045), 1.6 (1.0 - 5.9) (p = 0.009).

Regarding the treatment of hepatitis C and B in PLWHIV, this study confirms the strong dynamics of treatment since the availability of direct viral agents with an efficacy and a tolerance comparable to those observed in people with mono-infected with HIV, including in patients with HIV-HCV/HBV [20]. This dynamic is also observed routinely in France [21]. However, there is a significant percentage of PLWHIV co-infected with HCV and HBV still to be treated, as shown by a survey conducted in June 2016 (*i.e.* almost 2 years after the possibil-

ity of treating all co-infected PLWHIV), at within the ICONE cohort comprising 10,087 PLHIV followed in the centers of the Bourgogne-Franche Comté, Grand Est and Hauts de France regions [21].

The limitations of our study are the fact that it was not multicenter therefore not representative in our country, the cost of biological tests did not allow the study to be extended to other sites. The methodology used in our study does not identify the factors associated with HIV-HBV-HCV infection. We did not test whether the infection was new or old in relation to anti HBc Ab, or assay the viral DNA to separate cases of cure from that of infection.

Apart from these limitations, this work constitutes the first research carried out in this field in Kinshasa. The participation rate which is particularly interesting since the study was carried out in a developing country where the inhabitants are not used to participating in epidemiological studies and research. The biological analyzes were carried out in one of the reference laboratories in our country.

5. Conclusions

The results of our study on the frequency and the clinical-biological profile of HIV and hepatitis B and C virus co-infection in an HIV + population in Kinshasa have shown that:

- Hepatitis B and C are frequent in this population;
- The notion of pedicure and manicure as well as the duration of HIV infection emerged as the clinical profile most associated with HIV/hepatitis B and/or C virus co-infection;
- Acute and occult hepatitis B was the most common typological profile;
- A low SGPT transaminase level, a low serum albumin and prothrombin level as well as a high INR level were the laboratory profile most associated with HIV/viral hepatitis B co-infection and/or C.

Author's Contributions

All authors contributed to data analysis, drafting or revising the article, have agreed on the journal to which the article will be submitted, gave final approval of the version to be published, and agree to be accountable for all aspects of the work.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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APPENDIX: Data Collection Sheet on the Prevalence of Co-Infections by Hepatitis B and C Viruses and Evaluation of Hepatic Function in an HIV+ Population in Kinshasa

1. Identity

Name……
Post name:……
Age:……
Sex:……
Race:……
Civil status:……
Profession:……
Address:……
Province of origin:……
Level of studies completed:……
Religion:……

2. Background

Blood transfusion……..	Yes	No
Blood group…….....	Yes	No
Pedicure and Manicure ……	Yes	No
Jaundice ……	Yes	No
Previous vaccination against		
Hepatitis B……	Yes	No
Scarifications……	Yes	No
Circumcision……	Yes	No
Piercing……	Yes	No
Circumcision……	Yes	No
Drug addiction……	Yes	No
Number of sexual partners One Several	Yes	No
Safe sex	Yes	No
HBV or HCV known before HIV diagnosis	Yes	No

Year of HIV diagnosis……..……..……..……..……..……..
Current antiretroviral treatment……
Antiretroviral regimens……

Surgical intervention in the past	Yes	No

Current stage of HIV infection……

3. Paraclinical

at. Medical Biology

Qualitative HBs antigen……	Positive	Negative
Qualitative HCV antibody……	Positive	Negative

HBs antigen (ELISA):……..……..…….	Positive	Negative
HBs Antibody (ELISA)……..…..…	Positive	Negative
HBc Antibody (ELISA)……..……..…	Positive	Negative
HBe antigen (ELISA)……..……..…	Positive	Negative
HBe (ELISA) Antibody ……..……..…..	Positive	Negative
Total VH Antibodies (ELISA) …..……..	Positive	Negative
ALAT:…….	Normal	High Specify the value:….....
AST:……	Normal	High Specify value:…….....…
Gamma GT ……..	Normal	High Specify value……
Serum albumin…	Normal	Low Specify value……..……
Prothrombin ……	Normal	Low Specify value……..…
Alkaline phosphatase	Normal	Low Specify value……..…
Total bilirubin	Normal	High Specify value……
Direct bilirubin	Normal	High Specify the value……

CD4 (specify value):……

Done in Kinshasa, on……../……../201……..….