

Seroprevalence of Hepatitis B & C and HIV among sickle cell patients in Kinshasa City (Democratic Republic of the Congo): A prospective study

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ABSTRACT

Introduction

Sickle cell disease (SCD) is a genetic disorder characterized by abnormal hemoglobin production, leading to red blood cell deformity and various complications, including anemia and organ damage. Transfusion therapy is a common treatment for SCD complications, but it poses risks, including the transmission of infectious diseases such as hepatitis B, hepatitis C, and HIV. The prevalence of these infections among transfused SCD patients in Kinshasa, the Democratic Republic of the Congo, is not well-documented. This study aimed to determine the seroprevalence of hepatitis B & C and HIV among sickle cell patients who received transfusions in Kinshasa.

Purpose

The general objective of this study was to determine the seroprevalence of hepatitis B & C and HIV among sickle cell patients transfused in Kinshasa. This was a prospective and analytical cross-sectional study carried out during the year 2023 among sickle cell patients treated at the SS Anemia Mixed Medicine Center (CMMASS).

Methods

A total of 100 sickle cell patients were included in the study, with 50 patients who had received transfusions and 50 who had not. Blood samples were collected and analyzed using two methods: the immunochromatographic method with rapid tests and the electrochemiluminescence immunological test (ECLIA). Fisher's exact test was applied with an alpha risk of 0.05 to compare the seroprevalence rates between transfused and non-transfused patients.

Results

Among the transfused sickle cell patients, the seroprevalences of hepatitis B, hepatitis C, and HIV were 6%, 4%, and 3%, respectively. The co-infection rates for HBV-HIV and HCV-HIV were 1% and 2%, respectively. However, there was no significant difference in seroprevalence rates between transfused and non-transfused patients ($p > 0.05$).

Conclusions

These results partially confirm our hypothesis that the seroprevalence of hepatitis B & C and HIV among transfused sickle cell patients would be higher than that of the general population. Further studies with larger sample sizes are needed to confirm these findings and to assess the impact of transfusion therapy on the risk of acquiring these infections in SCD patients.

INTRODUCTION

Sickle cell disease is an inherited hemoglobin disorder. According to the World Health Organization (WHO), nearly 5% of the global population carries a gene responsible for a hemoglobin abnormality (Masengo et al., 2021). Most individuals affected by this disease live in Sub-Saharan Africa, with prevalence rates ranging from 10 to 40%. In the Democratic Republic of the Congo, which is a biodiversity-based country (Kambale et al., 2016), the population uses plants to treat this disease (Ngbolua et al., 2014; Ngbolua et al., 2019). It poses a major public health problem in developing countries (Thiam et al., 2017).

According to the WHO, 300,000 children are born worldwide every year with a major hemoglobin abnormality, the most common of which is sickle cell disease. Most of these births take place in three countries: Nigeria, the Democratic Republic of the Congo (DRC), and India (WHO, 2023). Without appropriate care, 50 to 75% of children born with this anomaly die before the age of 5. Given this health challenge, sickle cell disease has been declared a priority public health disease by the World Health Organization with an estimated prevalence of 2% among newborns (WHO, 2023). It is estimated that 50,000 children are born with sickle cell disease every year and that half of them die before the age of 5. However, early, and appropriate treatment reduces symptoms and prevents serious attacks (Mukinayi et al., 2021). Blood transfusion is a major treatment for sickle cell disease. By providing HbA, it helps prevent and treat the acute and chronic complications of this pathology. However, it is not without risk for the sickle cell patient. In addition to iron overload, the main risk of transfusion in sickle cell disease is anti-erythrocyte alloimmunization, resulting in a conflict between transfused red blood cells and the patient's anti-erythrocyte antibodies, leading to delayed post-transfusion hemolysis. Sickle cell patients, who undergo polytransfusion, have a high rate of alloimmunization compared with the general population (Habibi, 2018a). If transfusion rules are not observed at the level of blood collection, storage, and use; the patient or recipient may run a greater risk than the expected benefit. Risks can be classified into different categories: immuno-hematological risks, infectious risks, and risks of hydro-electrolytic overload. Transmission of hepatitis B, hepatitis C, and human immunodeficiency virus (HIV) is a major risk

associated with blood transfusion (Habibi, 2018b). On the one hand, hepatitis due to the hepatitis B virus theoretically represents a major danger since asymptomatic carriers represent 5 to 20% of the population in black Africa. Worldwide, an estimated 2 billion people have been in contact with this virus at some point in their lives, and 257 million of them remain chronically infected. The prevalence of hepatitis B is highest in sub-Saharan Africa and East Asia, where between 5% and 10% of the adult population has chronic hepatitis B (Kengne et al., 2021). On the other hand, hepatitis due to the hepatitis C virus is one of the viral hepatitis with post-transfusion transmission. Indeed, worldwide, 180 million people are infected with this virus and its seroprevalence in the general population is 0.5 to 1%, but in multi-transfused cases, it varies from 4 to 20% in some studies, and even up to 60% in others (Biswas et al., 2018). The existence of a serological or seroconversion window favors transfusion of contaminated blood that is undetectable by the various standard tests. The number of people living with HIV worldwide in 2020, according to UNAIDS, was 37.7 million (UNAIDS, 2020). One mode of HIV transmission is blood transfusion. Factors contributing to the transmission of infectious diseases through transfusion in Africa include high transfusion rates among certain patient groups, particularly pregnant women, and anemic children; and a high prevalence of human immunodeficiency virus (HIV) in blood donor populations. These factors hurt transfusion safety, increasing the likelihood of transmission of various infectious markers in sickle cell patients (Kabamba & Kabyala, 2013). Seroprevalences of transfusion-transmissible viruses have, for the most part, been studied in various populations, including donors, recipients, sex workers, homosexuals, and others (Aurelie Mayomo Fohoue). However, a few studies exist on the seroprevalence of transfusion-transmitted viruses in the Democratic Republic of Congo (Sack et al., 2013). These date back some ten years. To the best of our knowledge, none of them has been carried out recently on sickle-cell patients transfused in Kinshasa. Taking the above into account, this study sought to answer the question: What would be the seroprevalence of hepatitis B, hepatitis C, and HIV among transfused sickle cell patients in the City of Kinshasa in 2023?

METHODS

Study site and period

This study was carried out in the Democratic Republic of Congo, in the provincial city of Kinshasa. Samples were collected at the Centre de Médecine Mixte et d'Anémie SS (CMMASS). Analyses were performed at the Centre National de Transfusion Sanguine for the analysis of these samples during the period from February 02 to November 02, 2023.

Study population

The study population consisted of transfused sickle cell patients and non-transfused sickle cell patients. Sickle cell patients were selected by random sampling.

Biological material

The biological material used was serum obtained from whole blood collected from sickle cell patients managed at the Centre de Médecine Mixte et d'Anémie SS (CMMASS).

Laboratory equipment and reagents

10 to 100 µl micropipette; Gloves; Hemolysis tube; Syringe; Yellow tip holder; Blue tip holder; Marker for sample numbering; Cobas 8000 (in-house) connected to computer; Analyzer; Data manager; HIV duo (for qualitative determination of HIV p24 antigen and HIV antibodies); HBsAg duo (for quantitative determination of hepatitis B surface antigen); HCV duo (for dual qualitative detection of hepatitis C virus (HCV) core antigen and antibodies to HCV); HIV-1/2 determination kit; HBsAg determination kit; Bioline HCV SD kit; Bioline HCV SD test diluent;

Selection criteria

The following criteria were retained for inclusion in the study: confirmed sickle-cell anemia, informed consent (parent of sickle-cell anemic child); at least one transfusion (for the first group); and transfusion-naïve sickle-cell anemia (for the second group).

Exclusion criteria

Subjects who did not meet the above selection criteria were excluded from the study.

Type of Study

This was a descriptive, analytical, cross-sectional study to determine the seroprevalence of hepatitis B, hepatitis C, and HIV among transfused sickle cell patients in the city of Kinshasa in 2023.

Method of analysis

Sample analysis began with the identification and selection of sickle-cell patients who had consented to take part in the study, and from whom blood samples were taken to test for serological markers of hepatitis B, hepatitis C, and HIV viruses. The first stage involved rapid (immuno-chromatographic) tests (HIV, HBs, and SD Bioline HCV). The second step was electrochemiluminescence immunoassay (ECLIA) on the Cobas 8000 from la Maison Roche to its Cobas e 801 modules.

Ethical considerations

Samples were collected anonymously from sickle cell patients after they had given their consent to be included in the study.

Data processing

Data for this study were entered into the Excel 2013 database and processed using SPSS version 21.0 software. Frequencies and the average age of the population were calculated. The Chi-square test was applied, with Fisher's exact test in the case of small numbers. The significance threshold was 0.05 for comparison of results between the two sickle cell groups.

RESULTS

Table 1 gives the distribution of sickle cell patients by sex

Table 1:
Distribution of sickle cell patients by sex

Sickle cell patients		Gender		Total
		Male	Female	
Transfused	Workforce	20	30	50
	%	20,0	30,0	50,0
Non-transfused	Workforce	27	23	50
	%	27,0	23,0	50,0
Total	Workforce	47	53	100
	%	47,0	53,0	100,0

Table 1 shows that of the 50 transfused sickle-cell patients, 30 (30%) were female and 20 (20%) males. Secondly, this **Table** shows that of the 50 non-transfused sickle-cell patients, 23 (23%) were female and 27 (27%) males. Overall, the female sex was the most represented with 53 sickle cell patients or 53%, followed by the male sex with 47 sickle cell patients or 47%.

Table 2 gives the distribution of sickle cell patients by age group

Table 2: Distribution of sickle cell patients by age group

Age groups (years)		Sickle cell patients		Total
		Transfused	Non-transfused	
1 – 5	Workforce	1	39	40
	%	1,0	39,0	40,0
6 – 11	Workforce	12	11	23
	%	12,0	11,0	23,0
12 – 17	Workforce	30	0	30
	%	30,0	0,0	30,0
18 – 23	Workforce	7	0	7
	%	7,0	0,0	7,0
Total	Workforce	50	50	50
	%	50,0	50,0	50,0

Table 2 shows that the age group most represented among transfused sickle-cell patients was 12 to 17 years, with 30 subjects (30%). On the other hand, the **Table** shows that the most represented age group among non-transfused sickle-cell patients was 1 to 5 years, with 39 subjects (39%). The mean age of the sickle-cell patients was 8±6 years.

Table 3 gives the distribution of transfused sickle cell patients according to the number of transfusions

Table 3:
Distribution of transfused sickle cell patients according to the number of transfusions

Number of transfusions	Workforce	Percentage
1	6	12,0
2	14	28,0
3	8	16,0
4	10	20,0
5	6	12,0
6	6	12,0
Total	50	100,0

The results in **Table 3** provide information on the number of transfusions received by sickle cell patients. This number varied from 1 to 6 times. The two most represented classes were those of 2 transfusions with 14 sickle cell patients, i.e., 28%, followed by 4 transfusions with 10 sickle cell patients, i.e., 20%.

Table 4 gives the seroprevalence of hepatitis B in sickle-cell patients

Table 4:
Seroprevalence of hepatitis B in sickle-cell patients

Sickle-cell patients		Hepatitis B		Total
		Positive	Negative	
Transfused	Workforce	6	44	50
	%	6,0	44,0	50,0
Non-transfused	Workforce	3	47	50
	%	3,0	47,0	50,0
Total	Workforce	9	91	100
	%	9,0	91,0	100,0

Table 4 shows that the seroprevalence of hepatitis B in transfused sickle-cell patients was 6%. Among non-transfused sickle-cell patients, the figure was 3%. The difference was therefore not significant (Fisher's exact test: 1.088; p = 0.290).

Table 5 gives the seroprevalence of hepatitis C in sickle-cell patients

Table 5:
Seroprevalence of hepatitis C in sickle-cell patients

Sickle-cell patients		Hepatitis C		Total
		Positive	Negative	
Transfused	Workforce	4	46	50
	%	4,0	46,0	50,0
Non-transfused	Workforce	2	48	50
	%	2,0	48,0	50,0
Total	Workforce	6	94	100
	%	6,0	94,0	100,0

Table 5 shows that the seroprevalence of hepatitis C in transfused sickle cell patients was 4%. Among non-transfused sickle-cell patients, the figure was 2%. The difference was therefore not significant (Fisher's exact test: 0.702; p = 0.402).

Table 6 gives the HIV seroprevalence in sickle-cell patients

Table 6:
HIV seroprevalence in sickle-cell patients

Sickle-cell patients		HIV		Total
		Positive	Negative	
Transfused	Workforce	3	47	50
	%	3,0	47,0	50,0
Non-transfused	Workforce	1	49	50
	%	1,0	49,0	50,0
Total	Workforce	4	96	100
	%	4,0	96,0	100,0

Table 6 shows that HIV seroprevalence in transfused sickle-cell patients was 3%. Among non-transfused sickle-cell patients, the figure was 1%. The difference was therefore not significant (Fisher's exact test: 1.031; p = 0.310).

Table 7 gives the seroprevalence of HBV, HCV, HIV and HBV & HIV and HCV & HIV co-infection as a function of number of transfusions

Table 7:
Seroprevalence of HBV, HCV, HIV and HBV & HIV and HCV & HIV co-infection as a function of number of transfusions

No of transfusions	Workforce	Results	HBV	HCV	VIH	HBV&HIV	HCV&HIV
1	6	Positive	0	0	0	0	0
		Negative	6	6	6	6	6
2	14	Positive	0	1	0	0	0
		Negative	14	13	14	14	14
3	8	Positive	0	0	1	0	0
		Negative	8	8	7	8	8
4	10	Positive	1	1	0	0	0
		Negative	9	9	10	10	10
5	6	Positive	4	1	1	0	1
		Negative	2	5	5	6	5
6	6	Positive	1	1	1	1	1
		Negative	5	5	5	5	5
Total	50	Positive	6	4	3	1	2
		Negative	44	46	47	49	48

Table 7 first shows that the seroprevalence of hepatitis B was 6%, 4 % of whom had been transfused 5 times. Secondly, the seroprevalence of hepatitis C was 4%, 1% of whom had been transfused 1 time, 1% of whom had been transfused 4 times, 1% of whom had been transfused 5 times, and 1% of whom had been transfused 6 times. Thirdly, the seroprevalence of HIV was 3%, with 1% having been transfused 3 times, 1% having been transfused 5 times and 1% having been transfused 6 times. Fourthly, HBV & HIV co-infection had a seroprevalence of 1% having been transfused 6 times. Finally, HCV & HIV co-infection had a seroprevalence of 2%, of which 1% had been transfused 5 times and a further 1% had been transfused 6 times.

Table 8:
Comparison of seroprevalence of hepatitis B&C and HIV in transfused and non-transfused sickle-cell patients

Serological markers	Results	Transfused sickle cell patients (n=50)	Non-transfused sickle cell patients (=50)	Fisher exact test	p
HBV	Positive	6	3	1,088	0,290 (DNS)
	Negative	44	47		
HCV	Positive	4	2	0,702	0,402 (DNS)
	Negative	46	48		
HIV	Positive	3	1	1,031	0,310 (DNS)
	Negative	47	49		

Table 8 first shows that the seroprevalence of hepatitis B in transfused sickle-cell patients was 6 %. In non-transfused sickle-cell patients, it was 3%. Secondly, this **Table** shows that the seroprevalence of hepatitis C was 4% in transfused

sickle-cell patients. Among non-transfused sickle-cell patients, the figure was 2%. Finally, HIV seroprevalence in transfused sickle-cell patients was 3%. In non-transfused sickle-cell patients, it was 1%. There was therefore no significant difference.

DISCUSSION

This study aimed to determine the seroprevalence of hepatitis B & C and HIV in sickle cell transfusion patients in the city of Kinshasa in 2023. To achieve this, 100 samples were taken during 2023 at the Centre de Médecine Mixte d'Anémie SS, including 50 from transfused sickle cell patients and 50 from non-transfused sickle cell patients.

The results obtained were classified under two headings: sociodemographic data and results relating to the seroprevalence of hepatitis B & C and HIV. Regarding socio-demographic data, gender was the first variable studied. **Table 1** shows that in both groups of sickle-cell patients, the female sex was the most represented (53 %), followed by the male sex (47%). This situation could be explained by the simple fact that the selection of sickle cell patients was random and that female subjects predominated by chance. Sickle cell disease is not sex-linked; it is a genetic disease with autosomal recessive inheritance (Gaye et al., 2022). However, many authors have reported female predominance in studies among sickle cell patients. In the Shongo et al. study (Shongo et al., 2014), females accounted for just over half the cases (53.7%). This predominance was also found in the study by Nacoulma, who in his series found a sex ratio of 1.5 (Nacoulma et al., 2006). On the other hand, other authors, including Diagne et al. (2000) and Mabila et al. (2005) report a slight male predominance. As for the age of the sickle-cell patients studied, **Table 2** of this study shows that the age group most represented among transfused sickle-cell patients was 12 to 17 years, with 30 subjects (30%). On the other hand, this **Table** shows that the most represented age group among non-transfused sickle-cell patients was 1 to 5 years, with 39 subjects (39%). The mean age of sickle cell patients was 8 ± 6 years. The age range most represented among transfused sickle cell patients could be explained by the fact that most sickle cell patients have their first attack between the ages of 6 and 12, which is the primary reason for consultation (Kple-Faget, 2014). These results do not contradict those of the study conducted by

Shongo et al. (Shongo et al., 2014). The 1 to 5 age group predominated among non-transfused sickle-cell patients, simply because they had not yet received medical care, which would end in transfusions, as soon as the attacks began in most cases. Regarding the results concerning the seroprevalence of hepatitis B & C and HIV in transfused sickle cell patients, the study first looked at the number of transfusions per sickle cell patient. The data in **Table 3** provide information on this number. This number varied from 1 to 6 times. The two most represented classes were those of 2 transfusions with 14 sickle cell patients, i.e., 28%, followed by 4 transfusions with 10 sickle cell patients, i.e., 20%. These results are not far removed from those of the study conducted by Gody et al. on the occurrence of HIV and HBV in a cohort of sickle cell children transfused at the Bangui pediatric complex, which shows that the number of transfusions in sickle cell patients varied from 1 to 6 times (Gody, 2014).

Concerning the seroprevalence of hepatitis B in transfused sickle cell patients, **Table 4** shows that the seroprevalence of hepatitis B in transfused sickle cell patients was 6%. This prevalence is close to that found in a study conducted by Sack et al. on the carriage of HBs antigen and anti-HCV antibodies in homozygous sickle cell patients at Yaoundé Central Hospital (Sack et al., 2013). This prevalence increases with the number of transfusions received, especially in patients who have received more than 10 blood transfusions. This would be due to the deficit linked to transfusion safety, which in principle must use WHO standards that reduce the residual risk of blood transfusion (Sack et al., 2013). Whereas in non-transfused sickle cell patients, it was 3%. According to a WHO report, over 91 million Africans are living with hepatitis B or C, and over 8% of the total population of 19 countries is infected with the hepatitis B virus (WHO, 2022a). Regarding the seroprevalence of hepatitis C in transfused sickle cell patients, **Table 5** shows that seroprevalence was 4%. This seroprevalence differs by far from that found in the same study conducted by Sack et al, who found a seroprevalence of 16.6% (Sack et al., 2013). This difference could be explained by the fact that this prevalence was obtained only in sickle cell patients who had been polytransfused (more than 10 transfusions) and therefore more at risk of being infected.

However, it is also reflected in the fact that the prevalence of hepatitis C in the general population is over 1%, whereas in non-transfused sickle-cell patients (controls), it was 2% (WHO, 2022b).

Concerning HIV seroprevalence in transfused sickle cell patients, **Table 7** shows that HIV seroprevalence in transfused sickle cell patients was 3%. This seroprevalence does not deviate from that found in the study conducted by Gody et al, which showed 6% of HIV cases (Gody, 2014). On the other hand, our results differ from those of another study carried out in the Democratic Republic of the Congo (Tshilolo et al., 2007), which showed an 11.3% HIV infection rate among transfused sickle-cell children. The low level of HIV seroprevalence in our study is because the Centre National de Transfusion Sanguine (CNT) currently has automated systems using enzyme-linked immunosorbent assays capable of detecting HIV infection two weeks after infection. This means that all non-conforming blood units can be discarded after biological qualification of the blood, to reduce the risk of HIV transmission through blood transfusion. Among non-transfused sickle-cell patients, the prevalence was 1%. This prevalence is lower than that of the general population, which stands at 1.2% according to the national strategic plan for the response to HIV (PNMLS, 2023b). As far as co-infection is concerned, a 1% HBV-HIV co-infection rate and an HCV-HIV co-infection rate were noted among sickle cell transfusion recipients. This situation can be explained by the high frequency of transfusions received. Indeed, cases of co-infection were found in sickle cell patients who had been transfused 5 or more times. These results concur with those of Gody et al. (Gody, 2014). Furthermore, Fisher's exact test did not show a significant difference between the results of transfused sickle cell patients and controls ($p > 0.05$) as shown in **Table 8**. The use of two methods, namely immunochromatographic methods with rapid tests and immunological methods using electrochemiluminescence, enabled us to obtain results containing less bias. However, this study only involved a small number of sickle-cell patients, given the difficulty of finding controls, i.e., non-transfused sickle-cell patients. We would have liked to use molecular methods for diagnosis. This means that the material to be treated in this field has not yet been exhausted, i.e., comparing the serological status of sickle-cell children with that of their parents. In the future, further

research may be carried out to improve the management of sickle-cell patients.

CONCLUSION AND RECOMMENDATIONS

The study aimed to determine the seroprevalence of hepatitis B & C and HIV in sickle cell transfusion patients in Kinshasa in 2023, involving 100 samples from Centre de Médecine Mixte d'Anémie SS, with 50 from transfusion recipients and 50 from non-transfusion recipients. Immunochromatographic and electrochemiluminescence methods were used to determine the seroprevalence. Results showed hepatitis B seroprevalence at 6%, hepatitis C at 4%, and HIV at 3% in transfusion recipients, with HBV-HIV co-infection at 1% and HCV-HIV co-infection at 2%, not significantly different from controls ($p > 0.05$). Modern techniques in blood qualification and preventive measures should reduce virus transmission risk through transfusions.

Recommendations include systematic vaccination for hepatitis B and C, ensuring high-quality blood availability, thorough blood unit compliance checks, and comprehensive transfusion process monitoring for recipient safety. Future research should include studies on larger and more diverse populations across different regions to better understand the seroprevalence and its variations. Implementing longitudinal studies to track the progression and outcomes of co-infections in sickle cell patients over time could provide deeper insights into their impact on health and treatment responses. It would also be pertinent to investigate the effectiveness of various intervention strategies, such as vaccination programs for Hepatitis B and targeted antiviral therapies, to reduce the incidence and improve the management of co-infections. Examining the barriers to healthcare access and treatment adherence among sickle cell patients can inform policies and programs aimed at improving healthcare delivery and patient outcomes. Additionally, exploring the role of genetic predispositions and environmental factors in the susceptibility to these infections in sickle cell patients could lead to more personalized and effective treatment approaches. Addressing these areas, future research can contribute to better management, prevention, and ultimately, the reduction of co-infections in sickle cell patients in Kinshasa and beyond.

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Ethical Approval: None obtained because CMMASS has no IRB, but the process for establishing one is underway.

Conflicts of Interest: None declared.

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