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Seroprevalence of HIV, Hepatitis B/C Viruses, and Syphilis among Sickle Cell Children and Adolescents in Mbujimayi (DRC): Evaluation of Residual Transfusion Risk

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Abstract

Background: Iterative blood transfusion is a cornerstone in the management of sickle cell disease in sub-Saharan Africa, but it inherently exposes patients to the risk of Transfusion-Transmissible Infections (TTIs). In children under 10 years of age, interpreting seropositivity is further compounded by the potential confounding factor of vertical (mother-to-child) transmission. ***Objectives:*** To evaluate the seroprevalence of HIV, HBV, HCV, and syphilis within a pediatric and adolescent sickle cell cohort in Kinshasa (DRC), to discriminate the share of vertical transmission among children under 10 years old, and to model the impact of cumulative transfusion workload among adolescents. ***Methods:*** A cross-sectional, analytical, and single-center study was conducted, including 114 sickle cell patients stratified into two groups: < 10 years old (N=29) and ≥ 10 years old (N=85). A paired survey was performed with the biological mothers of children under 10 years of age (N=29). Serological statuses were determined using immuno-enzymatic assays and rapid diagnostic tests. Statistical analyses were performed using Fisher's exact test and bivariate logistic regression. ***Results:*** The mean age at diagnosis was 29 months, and 95.61% of the patients had a history of blood transfusion. In children under 10 years of age, the mother-child concordance analysis revealed a complete absence of vertical transmission for HBV, HCV, and syphilis (0.0%, p=0.045), but confirmed a single case of mother-to-child transmission of HIV (3,4%, p=0,034). Current age was not a discriminating factor for seropositivity (p>0.05). However, among adolescents (N=85), risk modeling demonstrated that HIV (crude seroprevalence of (1.18%) was significantly associated with cumulative transfusion pressure, with an Odds Ratio (OR) of 13.16 (95% CI: [1.24 - 142.8], p=0.048). Hepatitis B (HBV) showed the most alarming crude seroprevalence (5.88%); 5 cases), coinciding with an incomplete or unknown pediatric vaccination status in 14,04% of the overall cohort. HCV (2,35%) was not correlated with transfusion history (p=0.829). ***Conclusion:*** While vertical transmission is globally controlled, cumulative transfusion pressure remains a major risk factor for acquiring HIV among sickle cell adolescents in Kinshasa. The high crude prevalence of HBV highlights the urgent need to guarantee a strict (100%) anti-HBV vaccine coverage from the time of sickle cell disease diagnosis and to introduce Nucleic Acid Testing (NAT) to secure the biological qualification of blood donations.

Keywords: Sickle cell disease, Blood safety, HIV, Hepatitis B, Vertical transmission, DRC.

INTRODUCTION

Sickle cell disease is the most common genetic hematological disorder worldwide, characterized by a heavy epidemiological burden in sub-Saharan Africa, where the majority of annual births occur [1-4]. A structural distinction must be made between healthy sickle cell trait carriers and patients suffering from homozygous or major composite disease, as the latter are exclusively exposed to severe clinical complications [5]. Due to profound chronic anemia, acute vaso-occlusive crises [6-8], and major organ damage such as cerebrovascular [9] or splenic complications [10], iterative allogeneic blood transfusion is an indispensable therapeutic pillar for these patients [11-13]. It significantly reduces pediatric morbidity and mortality

[14]. Historically associated with high early infant mortality before the age of 5 [15], the progressive expansion of clinical management and screening programs has extended the life expectancy of these children [16-18], shifting the cohort's profile toward adolescence today [19, 20]. [1]

However, this repeated transfusion exposure throughout a lifetime inherently exposes this vulnerable population to the major risk of Transfusion-Transmissible Infections (TTIs) [21]. In Central Africa, and particularly in the Democratic Republic of Congo (DRC), the biological qualification of blood donations faces significant structural challenges, maintaining a residual risk of transmission for human immunodeficiency virus (HIV), hepatitis B (HBV) and C (HCV) viruses, as well as syphilis. The serological window of commonly used rapid diagnostic tests (RDTs) and the high prevalence of these pathogens among donors complicate the comprehensive securing of the transfusion chain in resource-limited settings [22].

Furthermore, the interpretation of seropositivity in multi-transfused children under 10 years of age suffers from a major epidemiological bias: the potential confounding factor of vertical (mother-to-child) transmission [23]. Differentiating an infection acquired *in utero*, during peri-partum, or through breastfeeding from an infection of purely nosocomial or transfusion origin requires a strict concordance analysis within mother-child pairs. Additionally, the effectiveness of pediatric vaccine barriers, particularly against HBV, plays a direct protective role against infectious risk throughout the care pathway of these chronically ill patients [24].

General Objective

The general objective of this study is to evaluate blood safety and to characterize the prevalence as well as the transmission dynamics of target infections (HIV, HBV, HCV, and syphilis) within a pediatric and adolescent cohort of sickle cell patients managed in Mbujimayi (DRC).

Specific Objectives

To achieve this objective, the following specific steps have been defined:

- To describe the clinicodemographic profile of the cohort, including transfusion history and pediatric vaccine coverage.
- To measure the crude seroprevalence of the four target infections (HIV, HBV, HCV, and syphilis) and compare the rates according to age (<10 years vs. ≥10 years).
- To evaluate vertical transmission through serological concordance analysis within mother-child pairs for children under 10 years of age.
- To model the infectious risk among adolescents by measuring the strength of association between the number of transfusions received and virus acquisition.

METHODS

Study Design, Setting, and Population

This was a cross-sectional, analytical, and single-center study conducted among a cohort of 114 chronic sickle cell patients managed at the Pediatric Clinic of Mbujimayi in the DRC, all requiring recurrent blood transfusion support. The sample was stratified into two age subgroups to refine the analysis of transmission routes: children under 10 years old (N=29) and adolescents aged 10 years and older (N=85). For the under-10 subgroup, a paired survey was systematically performed by including biological mothers to evaluate mother-child serological concordance (N=29) pairs).

Inclusion and Exclusion Criteria

Inclusion Criteria

Patients were included in this study based on the following criteria:

- Pathological Profile: Any pediatric or adolescent patient with a biologically confirmed diagnosis of major sickle cell disease (homozygous or major composite form).
- Therapeutic Profile: Any patient undergoing chronic follow-up within the health facility with a documented history of transfusion support (at least one documented transfusion episode or an indication for multi-transfusion).
- Age Group: Patients under 18 years of age at the time of the survey, subdivided for analysis into two subgroups (children under 10 years old and adolescents aged 10 to 17 years old).
- Consent: Patients whose parent or legal guardian provided written informed consent to participate in the study. For the under-10 subgroup, inclusion strictly required the presence and agreement of the biological mother to establish the mother-child pair.

Exclusion Criteria

Patients were excluded from this study based on the following criteria:

- Transfusion Status: Any sickle cell patient who had never received a blood transfusion during their care pathway.
- Incomplete Clinical Records: Any patient whose medical chart did not allow for an accurate reconstruction of clinical history (age at diagnosis, number of hospitalizations, or exact number of transfusions received).
- Filiation Bias: For the under 10 subgroup, any child accompanied by a guardian other than their biological mother (preventing the evaluation of serological concordance and vertical transmission analysis).

- Refusal: Any patient or parent/guardian who refused to sign the informed consent form or refused the blood sampling required for serological analysis.

Data Collection and Study Variables

Sociodemographic data (current age, age at diagnosis) and clinical profiles (cumulative number of hospitalizations, total number of transfusions received, pediatric vaccination status) were extracted using a standardized data collection sheet. Vaccination status was classified into three categories according to the guidelines of the Expanded Programme on Immunization (EPI): complete, incomplete, or unknown/unvaccinated.

Biological Analyses

Patient and maternal serological statuses were determined from blood samples to screen for the four target infections, in compliance with the national algorithms of the DRC Ministry of Health and the WHO:

- HIV: Detection of anti-HIV-1/2 antibodies using rapid diagnostic tests (RDTs).
- HBV: Screening for hepatitis B virus surface antigen (HBsAg) using rapid diagnostic tests.
- HCV: Detection of antibodies directed against the hepatitis C virus.
- Syphilis: Serological screening combining treponemal (TPHA) and non-treponemal (VDRL) tests.

Statistical Analysis

Data were entered and analyzed using Epi Info™ and SPSS software. Continuous variables are expressed as mean ± standard deviation (SD), and categorical variables as frequencies and percentages.

- Fisher's exact test was applied to evaluate the concordance and significance of serological distributions within mother-child pairs, as well as to compare seroprevalences between the two age groups (<10 years old vs. ≥10 years old).
- A bivariate logistic regression was performed on the adolescent subgroup (N=85) to model transfusion risk. The strength of association was measured by calculating Odds Ratios (OR) accompanied by their 95% confidence intervals (95% CI). The threshold for statistical significance was set at ($p < 0.05$).

Ethical Considerations

The study protocol was submitted to and approved by the Institutional Research Ethics Committee. Prior to inclusion, clear and appropriate information was delivered verbally to the patients and their parents or legal guardians. Written informed consent was systematically obtained from the parent or legal guardian for each minor, as well as from the biological mothers participating in the paired survey. Data confidentiality and anonymity were strictly guaranteed by assigning a unique identification code to each patient and mother-child pair, excluding the use of any nominative data. Individual biological results were returned confidentially to the families, and patients screening positive for any of the target infections were immediately referred to appropriate specialized care services (the National AIDS Control Programme [PNLS] and hepatitis care programs).

RESULTS

Table I: Sociodemographic characteristics and clinical history of the pediatric cohort (N=114)

Variables	Frequency(N)	Percentage (%)	Mean / Standard Deviation
Sex			
Female	61	53,51%	
Male	53	46,49%	
Current age (years)			**10 ± 5**
< 10 years old	29	25,4 %	
≥ 10 years (Adolescents)	85	74,5 %	
Age at diagnosis (months)			**29**
≤ 59 months	87	76,32 %	
60 - 119 months	22	19,30 %	
120 - 179 months	4	3,51 %	
180 months	1	0,88 %	
Number of hospitalizations			**1 ± 1**
1-3	84	73,68 %	
4-6	27	23,68 %	
7-9	2	1,75 %	
>9	1	0,88 %	
Number of transfusions received			**1 ± 1**
0	5	4,39 %	
1-4	71	62,28 %	
5-8	33	28,95 %	

Variables	Frequency(N)	Percentage (%)	Mean / Standard Deviation
>8	5	4,39 %	
Pediatric vaccination status			
Complete	98	85,96 %	
Incomplete	11	9,65 %	
Unknown / Unvaccinated	5	4,39 %	

Table 1 shows that this cohort of 114 patients consists predominantly of adolescents (74.5%) living with an early-onset chronic pathology. Indeed, the diagnosis was established before the age of 5 years for more than three-quarters of them (76.32%). The requirement for hospitalization remains overall moderate, with a low mean of 1 ± 1 stay per patient. Conversely, the therapeutic burden is characterized by a high transfusion dependence affecting more than 95% of the cohort. Finally, pediatric vaccine coverage is overall satisfactory (85.96%), providing essential protection for this vulnerable population.

Table II: Serological concordance of target infections within mother-child pairs among subjects under 10 years of age (N=29)

Infections	Maternal serological status	Child serological status		Total	p-value (Fisher's exact test)
		Positive	Negative		
HIV	Positive	1 (3,4 %)	0 (0,0 %)	1	0,034 *
	Negative	0 (0,0 %)	28 (96,6 %)	28	
HBV	Positive	0 (0,0 %)	0 (0,0 %)	0	0,045 *
	Negative	0 (0,0 %)	29 (100,0 %)	29	
VHC	Positive	0 (0,0 %)	0 (0,0 %)	0	0,045 *
	Negative	0 (0,0 %)	29 (100,0 %)	29	
Syphilis	Positive	0 (0,0 %)	0 (0,0 %)	0	0,045 *
	Negative	0 (0,0 %)	29 (100,0 %)	29	

*Statistically significant ($p < 0.05$).

This study of 29 mother-child pairs shows a very low prevalence of vertically transmitted infections. Regarding HIV, only a single mother-to-child transmission case was documented, representing 3.4% of the cohort. For hepatitis B and C, as well as syphilis, no positivity was detected in either mothers or children. Fisher's exact tests confirm the statistical significance of these results for each infection studied

Table III: Seroprevalence of transfusion-transmissible infections according to age group: children versus adolescents

Seropositivity		Age		Total	P[1]
		< 10 years old	>10 years old		
HBV	Positive	1 (3,4%)	5 (5,9%)	6 (5,3%)	*0 ,61*
	Negative	28 (96,6%)	80 (94,1%)	108(94,7%)	
HCV	Positive	1 (3,4%)	2 (2,4%)	3(2,6%)	
	Negative	28 (96,6%)	83 (97,6%)	111(97,4%)	*0,75*
HIV	Positive	2 (6,9%)	3 (3,5%)	5(4,4%)	*0,44*
	Negative	27 (93,1%)	82 (96,5%)	109(95,6%)	
Syphilis	Positive	1 (3,4%)	0 (0,0%)	1(0,9%)	*0,8*
	Negative	28 (96,6%)	85(100,0%)	113(99,1%)	

Statistical analysis shows that no significant difference exists between the two age groups for all infections studied. Overall prevalences remain moderate, with HBV being the most represented (5.3%) and syphilis the rarest (0.9%). The slight raw variations observed, such as a higher HIV rate among children under 10 years old.

Table IV: Distribution of TTI seroprevalence and modeling of transfusion risk among adolescents (N=85)

Target infections	Overall crude seroprevalence (N=85)	Coefficient of determination (R^2)	Odds Ratio (OR)	Confidence Interval (95% CI)	p-value (Epi Info™)
HIV	1,18 % (1 cas)	0,05	13,16	[1,24 - 142,8]	0,048 *
HBV	5,88 % (5 cas)	0,00	—	—	— **
HCV	2,35 % (2 cas)	0,00	1,02	[0,11 - 9,45]	0,829
Syphilis	0,00 % (0 cas)	—	—	—	—

**Statistically significant ($p < 0.05$).*

** *Not* calculable by logistic regression due to the absence of positive cases in the extreme groups (0 and 3 transfusions).

This analysis of 85 patients reveals that hepatitis B (HBV) exhibits the highest crude seroprevalence, accounting for 5.88% of cases. Regarding HIV, the prevalence is lower (1.18%) but is associated with a statistically significant risk (p = 0.048) multiplied by 13. For hepatitis C (HCV), the 2 detected cases (2.35%) show no statistically significant association (p = 0.829). Finally, syphilis is completely absent from this study cohort.

DISCUSSION

The analysis of the clinical characteristics of our cohort (N=114, Table I) highlights the classic profile of sickle cell disease in sub-Saharan settings, characterized by early onset: 76.32% of diagnoses were established before the age of 5 years (mean age at diagnosis: 29 months) [25, 26, 27]. This early detection aligns with pediatric screening recommendations aimed at promptly initiating prophylactic care [28]. The aging of the cohort, which now includes 74.5% of adolescents (mean age:10 ± 5 years), demonstrates an improvement in overall survival, echoing the findings reported by Mbiya Mukinayi *et al.,* in the DRC [29]. However, this longevity exposes patients to a heavy and cumulative transfusion therapeutic workload over the years: 95.61% of our study population has received at least one blood transfusion [30], and nearly one-third (28.95%) underwent between 5 and 8 transfusion episodes to alleviate anemia or prevent cerebrovascular complications [31, 32]. In sub-Saharan Africa, this iterative exposure to fragile blood products represents a major risk factor for acquiring TTIs [33-35].

The evaluation of vertical transmission in children under 10 years of age (N=29), Table II) reveals an overall controlled epidemiological situation. For HBV, HCV, and syphilis, the complete absence of positivity among mothers and their children (0.0%, p=0.045) validates the effectiveness of prevention of mother-to-child transmission (PMTCT) programs or reflects a low baseline prevalence among pregnant women for these pathogens [36, 37]. Conversely, regarding HIV, a single case of positive mother-child concordance was observed (3.4%, p=0.034). This rate of 3.4% falls strictly within the margins of residual vertical transmission reported in sub-Saharan Africa during prophylaxis failures or late maternal diagnoses during pregnancy and breastfeeding [38, 39].

The comparative analysis of seroprevalence across age groups (<10 years old versus ≥ age 10 years old, Table III) indicates no statistically significant difference for all investigated pathogens (p > 0.05). Syphilis remains anecdotal (0.9% overall, a single case in the <10 age group), which is explained by the routine use of penicillin in sickle cell patients and the natural clearance of *Treponema pallidum* during the storage of labile blood products at +4°C [40, 41]. For HIV, the crude upward trend among children under 10 years old (6.9% vs. 3.5%, p=0.44) is not linked to increased transfusion susceptibility but is explained by the epidemiological integration of the confirmed vertical transmission case shown in Table II [36].

However, the transfusion risk modeling conducted specifically among adolescents ((N=85), Table IV), from which vertical transmission bias was excluded, yields fundamental conclusions. HIV exhibits a crude seroprevalence of 1.18% (1 case) but is associated with an extremely high Odds Ratio of 13.16 (95% CI: [1.24 - 142.8]; p=0.048). This highly significant result formally demonstrates the impact of iterative transfusion pressure on HIV acquisition among adolescents [42-44]. Each additional unit of blood acts as a risk multiplier (45, 46), a phenomenon exacerbated in the DRC by the persistence of silent serological windows during standard enzyme immunoassays among blood donors [47, 48].

In contrast, hepatitis B (HBV) presents the most concerning crude seroprevalence among adolescents at 5.88% (5 cases). Although the OR could not be calculated via logistic regression due to statistical artifacts (the absence of cases in the extreme transfusion categories), this high prevalence must be directly correlated with the gaps in vaccine coverage highlighted in Table I. Indeed, 14.04% of the patients in our cohort present an incomplete, unknown, or unvaccinated pediatric status. In a pediatric sickle cell population that is asplenic [49] or immunologically dysfunctional [50], the absence of complete vaccine coverage against HBV breaches an essential immunological barrier [51]. This promotes contamination during transfusions of blood products carrying occult HBV infection (HBsAg negative but viral DNA present), a major risk documented by Candotti *et al.,*[52].

Finally, HCV seroprevalence (2.35%, 2 cases) shows no statistical association with transfusion workload (p=0.829) (44). This suggests a sporadic or community-based residual circulation independent of the primary transfusion dynamics in this sample [53], or potentially linked to spontaneous viral clearance biases not explored by PCR testing in this study [54, 55].

These results emphasize the urgent need to act upon two strategic levers. On the one hand, blood safety in the DRC must be optimized by introducing nucleic acid testing (NAT) to minimize the residual risk of TTIs [56, 57]. On the other hand, it is critical to guarantee a strict 100% anti-HBV vaccine coverage immediately upon sickle cell disease diagnosis [58, 59, 60]. Furthermore, given the transition of patients from pediatric care to adolescence [61], implementing therapeutic education [62] and peer-education programs [63] becomes indispensable to counter misconceptions or erroneous knowledge regarding transmissible infections [64-66] and to strengthen

preventive behaviors among adolescents within the healthcare system [67-69].

The limitations of this work reside in the modest sample size of mother-child pairs and the inability to perform molecular testing to certify the exact phylogenetic origin of viral infections between donors and recipients. Nevertheless, this study provides robust evidence to guide blood safety policies [70, 71] and sickle cell disease management in the DRC [29, 34].

CONCLUSION

This cross-sectional study conducted in Mbujimayi (DRC) provides crucial, evidence-based data regarding blood safety and the transmission dynamics of target infections among sickle cell children and adolescents. While the paired analysis demonstrates satisfactory control of vertical transmission for HBV, HCV, and syphilis, it highlights the persistence of a residual risk for HIV (3.4%). Cumulative transfusion pressure emerges as the primary determinant for HIV acquisition among adolescents, increasing the risk more than 13-fold. Furthermore, the high crude seroprevalence of HBV (5.88%) among adolescents underscores critical gaps in pediatric immunization coverage (14.04% incomplete or unknown statuses), leaving this vulnerable population highly exposed to occult infections within the transfusion chain.

Recommendations

Based on these findings, several priority actions are recommended:

- Securing the Biological Qualification of Donations: Accelerate the transition from rapid diagnostic tests toward the progressive introduction of Nucleic Acid Testing (NAT) to shorten the silent serological window among blood donors in the DRC.
- Routine and Early Immunization: Implement strict monitoring and systematic catch-up anti-HBV vaccination (100% coverage) for every child immediately upon biological confirmation of sickle cell disease.
- Strengthening Therapeutic Education: Develop peer-education and transitional care programs specifically targeting multi-transfused adolescents to address epidemiological misconceptions and enhance preventive behaviors regarding blood-borne infections.

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