



Low Prevalence and Risk Factors Related to HIV-1 Mother to Child Transmission under Option B+ Program at 3 Referral Military and Public Hospitals in Cameroon

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Authors' contributions

This work was carried out in collaboration among all authors. Authors JMN, MB, UT and SATC design of the study. Authors JMN, AFE, SATC and ES did the data and sample collection. Authors DSM, SK and SATC did the statistical analysis of the data. Authors SATC, SK and MB did the interpretation of results. Author SATC wrote the protocol of the manuscript. Authors MB, JMN, UT, SK, AFE, AN, AEM, CAMM, CSS, JOE, DSM, JNB and SHR revised of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Immunodeficiency Virus type 1 (HIV-1) Mother-To-Child Transmission (MTCT) prevalence and its predictors. Following the limited success achieved with the previous MTCT prevention programs, the Cameroon's public health ministry adopted in 2014 the *Option B+* program that recommends a systematic lifelong treatment to all HIV positive pregnant woman.

Study Design: A case-control study was conducted within two groups: a reference group constituted of exposed infants from HIV positive mothers undergoing Option B+ program, and a control group of infants from Anti-Retroviral Treatment (ART) naïve HIV positive mothers during pregnancy.

Place and Duration of Study: Douala and Yaounde Military Hospitals (*HMR2* and *HMR1* respectively) as well as the Bertoua Regional Hospital (HRB), From October 2017 to March 2018.

Methodology: This research included infected mother - exposed child pairs. Infected mothers' sociodemographic and clinical characteristics were reported. Infants sampled at six weeks at the HIV MTCT prevention units were tested at the Military Health Research Center for HIV-1 RNA early detection through rtPCR with Abbott m2000sp automated system. Multivariate logistic regression model was built to assess the predictors of MTCT and to compare groups.

Results: Within the study period, the overall HIV-1 prevalence in the 107 six weeks old reference group infants and 23 control group infants was nil and 4.35% (1/23) respectively. Logistic regression showed that predictors of HIV-1 MTCT were: home delivery $p=0.03$ and absence of ART during pregnancy $p = 0.04$.

Conclusion: Vertical transmission of HIV-1 infection is more likely in ART naïve pregnant women as compared to their counterparts established on ART. Hence, implementation of the Option B+ appear to be very essential in eliminating HIV-1 MTCT. Consequently, a systematic enrolment of these pregnant women living with HIV if scaled up, would be very instrumental in eliminating HIV-1 MTCT in Cameroon.

Keywords: Cameroon; case-control study; HIV-1; mother-to-child transmission; option B +; prevalence; risk factors.

1. INTRODUCTION

Human Immunodeficiency Virus (HIV) is a positive sense single stranded Ribonucleic Acid (RNA) virus of the Lentiviruses family responsible for Acquired Immunodeficiency Syndrome (AIDS). HIV infection remains a serious public health problem worldwide, affecting adults, teenagers and children of both sex [1] In 2016, 2.75% (2.1 million) of the global population of individuals living with HIV (estimated at 76.1 million) were children and adolescents under 15 years of age. About 200000 of this group were infected through Mother To Child Transmission (MTCT) [2]. MTCT of HIV1 may occur during pregnancy, labor, delivery, or breastfeeding. In the absence of prophylactic therapy, the risk of HIV-1 MTCT, both in utero and during childbirth is estimated at 15-30%; this risk increases to 20-45% in breastfed children. This vertical

transmission is the highest in resource-constrained countries [3,4].

In Africa, pediatric HIV-1 infection is a common cause of morbidity and mortality, and MTCT is the main route of transmission. Prevention of mother-to-child transmission (PMTCT) of HIV is therefore a priority intervention of HIV/AIDS programs, yet a serious challenge particularly in Sub-Saharan Africa where 90% of the world pediatric HIV-1 infection is recorded [3,5].

To address this public health concern in Cameroon, Option B option was implemented in 2009 following Option A option whose implementation started in 2004. Both were implemented when infected mothers' cluster of differentiation (CD4) cell count was ≤ 350 cells/mm³. In fact, Option A regimen included Azithromycin (AZT) monotherapy starting at 14

weeks gestation followed by single dose Nevirapine (NVP) and 7 days postpartum Azithromycin AZT / Lamivudine (3TC) bitherapy from delivery for mother. For the infant was recommended daily NVP prophylaxis from birth until 1 week after breastfeeding cessation, or 4–6 weeks if no breastfeeding. While Option B included Triple ART at 14 weeks gestation, ending at delivery or 1 week after breastfeeding cessation, as well as infant's daily NVP or twice daily AZT for 4–6 weeks when replacement feeding and daily NVP for 6 weeks when breastfeeding [6,7]. The achieved goals for both programs was very limited; 12.1% in 2011 for Option A [8] and 7.1% in 2014 for Option B [9], indicating a still very high transmission rate. With the support of USA President's Emergency Plan for AIDS Relief (PEPFAR) in Cameroon, the Ministry of Public Health adopted PMTCT program - Option B+ aiming to achieve the ambitious global targets of 95-95-95 set by the Joint United Nations Programme on HIV/AIDS [10]. The 95-95-95 targets is defined as 95% of all people living with HIV to know their HIV status, 95% of all people living with HIV and knowing their status to have access to antiretroviral therapy, and 95% of people on antiretroviral therapy to have an undetectable viral load [6]. These global targets for the infants aimed at eliminating the MTCT of HIV. Option B + recommends a systematic and lifelong treatment of any HIV- positive pregnant or breastfeeding woman regardless of her viral load or CD4 count [2].

Option B+ has been under implementation in Cameroon since 2014 and it is therefore appropriate to evaluate its added value in the PMTCT of HIV-1 in Cameroon with respect to the global targets for infants. Worldwide studies have identified potential risk factors for vertical HIV transmission [8]. These include: maternal factors (maternal immune status, ART regimen), obstetrical factors (traumatic birth, premature rupture of membranes, chorioamnionitis), fetal and infant factors (immune status, prophylaxis, feeding mode) [2,6,11]. As recommended by the World Health Organization (WHO), assessing the implemented PMTCT of HIV-1 programs provides optimal programmatic choices for PMTCT [6]. Thus, this study aimed to determine the prevalence and investigate risk factors related to mother-to-child transmission of HIV-1 in infants at 6 weeks of age in hospitals where the option B+ prevention program is implemented, namely the Military Hospital of Douala (HMR2) and the Military Hospital of

Yaounde (HMR1) in comparison to exposed neonates born from ART naïve HIV positive mothers at the Bertoua Regional Hospital (HRB).

2. MATERIALS AND METHODS

2.1 Study Design, Period and Population

A case control study with two groups; the reference group and the control group was conducted between October 2017 and March 2018. The reference group constituted of six weeks infants born to HIV-1 positive mothers undergoing the Option B+ program, and the control group (no ART group) consisted of six weeks old infants from HIV-1 positive mothers who discovered their HIV status during delivery. Participants of the reference group were sampled at the Douala and Yaounde military hospitals (*HMR2* and *HMR1* respectively), while participants of the control group were sampled at the Bertoua Regional Hospital (BRH). BRH was chosen as study site for the control group because the Eastern region is among the highly HIV affected regions in Cameroon and the cultural practice in that area remains refractory to antenatal care (ANC) with only 49% of pregnant women going for ANC visit [12]. Recruitment was consecutive and non-probabilistic. References and controls were not matched because of the scarcity of no ART HIV positive pregnant women compared to ART HIV positive ones.

2.2 Data Collection and Sample Testing

Socio demographic and clinical information of consenting mothers including mode of delivery, ART regimen and HIV-1 status as well as neonates oral NVP prophylaxis regimen and feeding mode were collected using a questionnaire. Dried blood spots (DBS) samples on Whatmann 903 paper were collected following recommended sampling standards from all six weeks old participating infants [13]. Samples were then tested for early infants' diagnosis of HIV-1 RNA by real time reverse transcriptase polymerase chain reaction (RT-PCR) as described elsewhere, using Abbott m2000 Real Time HIV-1 Qualitative test, at the Military Health Research Center (*CRESAR*), Yaounde-Cameroon [14].

2.3 Quality Assurance

Samples were tested according to the ISO 15189:2012 standard of medical analysis laboratories [13]. External quality control

consisted of samples with known HIV-1 results from an accredited laboratory. The internal processing control (one negative and two positive manufactured controls with high and low levels) consisted of HIV nonspecific primers designed to evaluate the amplification. A test was validated under 3 conditions: internal control should have been amplified in all the samples, negative and positive controls should have been negative and positive respectively.

2.4 Administrative and Ethical Considerations

The present study received Ethical approval N° 006027/LE/MINDEF/01 from the Cameroon's Ministry of Defense as well as the Ethical authorization N° 896/L/MINSANTE/SG/DRSPE/BCASS/BFP from the Bertoua Regional Hospital prior to carry out research. Prior to the sample collection, mothers or caregivers were given an information's notice, afterwards consents for the HIV-exposed infants enrolled in the study were obtained by signing the consent form. All authors hereby declare that experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

2.5 Data Analysis

Statistical analysis was done using the Statistical Package for the Social Sciences (SPSS), software (IBM, Version 25.0. Armonk, NY: IBM corp). Descriptive statistics were done using frequency distribution. Multivariate logistic regression model for multimodal variables and binary logistic regression model for bimodal variables were built to assess the predictors of MTCT and to compare groups. Variables were considered statistically significant for a p-value less than 0.05.

3. RESULTS AND DISCUSSION

3.1 Results

3.1.1 Sociodemographic characteristics of reference and control groups in mothers

During the six months, 141 samples of six weeks old infants received at the PMTCT units with their mothers/caregivers were collected and distributed as follows: 118 in the reference group and 23 in the control group. Out of these, 11 were not included because of lack of suitable information on the infected mother from the caregiver in the reference group (Fig. 1).

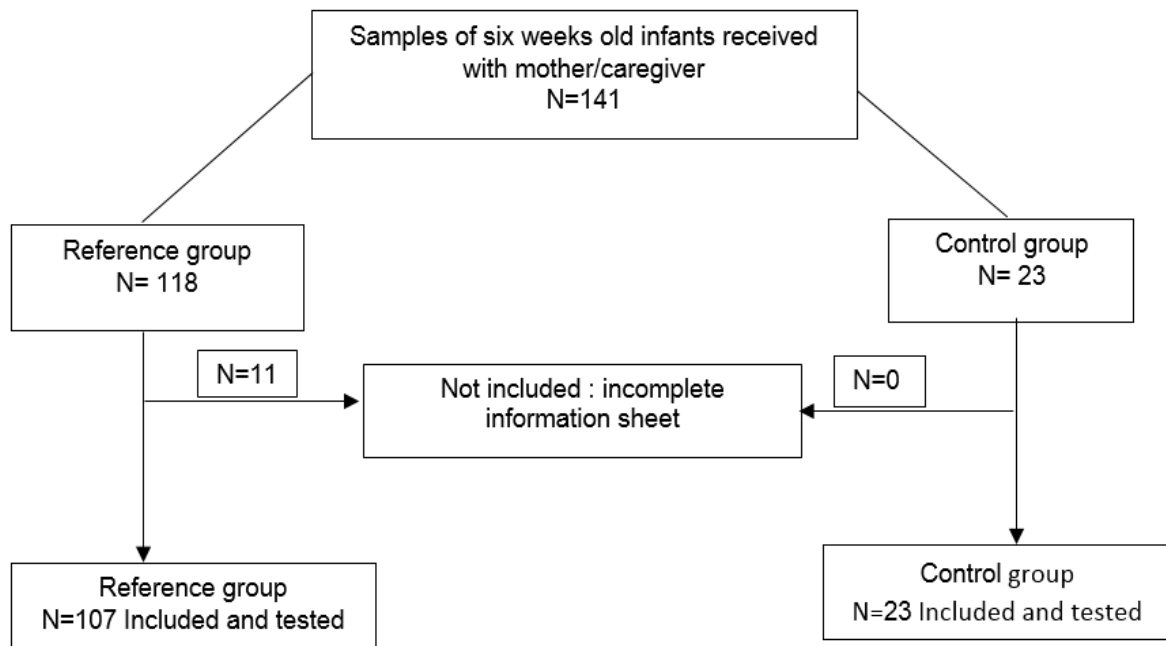


Fig. 1. Flow diagram showing the study population
N: number

Mothers were aged 17 to 42 years with an average of 29 ± 5 years. The most represented age range was 25-34 years old with 70 (83.3) and 14 (16.7) in reference and control groups respectively. According to the results (see Table 1), the later age was a risk for vertical transmission of the virus (p=0.02). Samples were collected in the Center and Littoral regions for the reference group, and in the East region for the control group. Most infected mothers of the reference group resided in Douala 49 (98%) in the Littoral region, and 89,5% of the control group were from the East region. The lowest educational level (primary school) was the most observed in women of the control group (Table 1).

3.1.2 Gender, age, place of birth and mode of delivery in infants

The National algorithm for the follow-up of HIV infected mothers and their children recommends that the first early infant diagnosis should be done about six weeks after the delivery. Accordingly, out of the 130 infants sampled, 69 (76.7%) and 21 (23.3%) were aged less than six weeks in the reference and control group

respectively. A frequency of 84.6% of caesarian was observed in the reference group, while the control group registered 100% of infants born at home with a high risk of HIV MTCT transmission (p=0.03) (See Table 2).

3.1.3 HIV-1 Mother-to-child transmission prevalence

An undetected HIV-1 RNA was considered negative while detected HIV RNA was considered as a positive qualitative HIV-1 rtPCR test. Analysis of exposed newborns and infants taken from DBS gave a MTCT prevalence of 4.35% (1/23) in control group (Fig. 2a) and nil prevalence (0/110) in the reference group under option B+ (Fig. 2b).

3.1.4 Risk factors related to vertical transmission of HIV-1 infection

The Table 3 summarizes the distribution of risk factors of MTCT related respectively to mother and infants according to their infectious status and socio-demographical characteristics, as well as the statistical analysis of maternal and infant's parameters with HIV MTCT (Tab. 3). During the

Table 1. Sociodemographic characteristics of reference and control groups in mothers

Characteristics	Reference group mothers (N=107), n (%)	Control group mothers (N=23), n (%)	p-value
Mean±sd, years	29.60± 5.0	30.87± 6.1	
Age range (years)			
15 - 24	17 (85.0)	3 (15.0)	.06
25 - 34	70 (83.3)	14 (16.7)	
35 - 44	17 (73.9)	6 (26.1)	.02
East Region Origin	2(10.5)	17(89.5)	.9
Douala Residence	49 (98.0)	1 (2.0)	1.0
Primary school education level	6 (31.6)	13 (68.4)	.01
Married status	10 (47.6)	11 (52.4)	.8

Key: N: total number; n: number, % : percentage

Table 2. Demographic characteristics of reference and control groups in infants

Characteristics	Reference infants (N=107) n (%)	Control infants (N=23) n (%)	p-value
Age range (weeks)(reference=1-10)			
1 - 10	69 (76.7)	21 (23.3)	
11 - 20	7 (87.5)	1 (12.5)	.03
21 - 30	14 (93.3)	1 (6.7)	.02
31 - 40	13 (100.0)	0 (0.0)	1
41 - 50	4 (100.0)	0 (0.0)	1
Home birth place	0 (0.0)	2 (100.0)	.03
Caesarean birth	11(84.6)	2(15.4)	.07
Male	44 (75.9)	14 (24.1)	.005

Key: N: total number; n: number, % : percentage

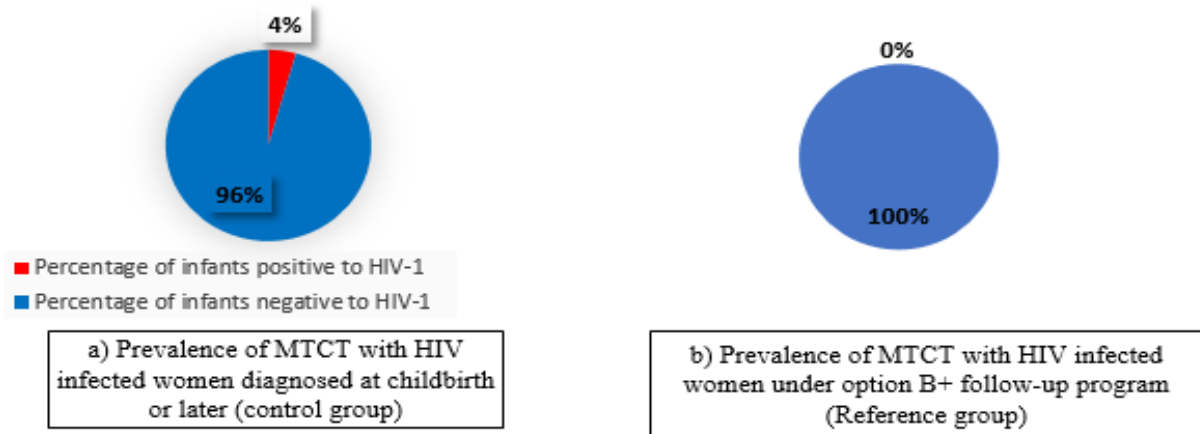


Fig. 2. HIV-1 Mother-To-Child Transmission (MTCT) prevalence among infants of control and reference groups

study period, 1 each, representing 50% of infants were symptomatic at the sampling moment in each group. The following variables : known 11-15 years age range of infection, traditional medicine taken during pregnancy, smoking during pregnancy, no regular physical activities, were not associated with HIV-1 vertical transmission ($p > 0.05$).

On the contrary, the place of delivery was significantly related to MTCT, with home birth place being a major risk factor for vertical HIV transmission ($p = 0.03$), as well as the absence of

mother's ART medication before or at birth ($p = 0.04$).

The two known delivery ways were represented, namely the vaginal delivery and caesarean section; the caesarean section was applied only in 11/107 in reference and 2/23 in control group (Tab. 2). However, the way of delivery was not associated to MTCT in this study ($p > 0.05$). Absence of prophylaxis to the newborn at delivery and the late infants' diagnosis were also not linked to MTCT ($p = 0.9$) (Table 3).

Table 3. Risk factors related to MTCT of HIV-1 in the study population under option B+ and those who were not

Exposure	Reference group	Control group	p-value
Symptomatic infant	1 (50.0)	1 (50.0)	.7
Artificial feeding	11 (78.6)	3 (21.4)	.0
Absence of prophylaxis to the newborn	0(0.0)	2 (100.0)	.9
Home birth place	0 (0.0)	2 (100.0)	.03
Late infant diagnosis	0 (0.0)	1 (100.0)	.9
known age range of infection, 11-15 years	3 (75.0)	1 (25.0)	.9
ART of women before or at birth (reference= HAART (TDF+3TC+EFV))			
AZT+3TC+EFV	10 (100.0)	0 (0.0)	1
AZT+3TC+NVP	4 (100.0)	0 (0.0)	1
HAART (TDF+3TC+EFV)	71 (82.6)	15 (17.4)	
TDF+3TC+NVP	5 (50.0)	5 (50.0)	1
Absence	5 (83.3)	1 (16.7)	.04
Traditional medicine taken during pregnancy	0 (0.0)	3 (100.0)	1
Smoking during pregnancy	0 (0.0)	1 (100.0)	1
Alcohol drinking during pregnancy	8 (40.0)	12 (60.0)	.9
No regular physical activities	44 (71.0)	18 (29.0)	.9

ART: Antiretroviral Treatment; AZT: Azithromycin; 3TC: Lamivudine; EFV: Efavirenz; Nevirapine; TDF: Tenofovir

3.2 Discussion

3.2.1 Socio-demographic and clinical data

Mother-to-child transmission causes about 90% of pediatric HIV-1 infections worldwide. Data on MTCT from various countries and regions are as diverse as the social contexts are not the same. MTCT of HIV may be enhanced or better controlled depending of each local social context, including cultural practices and levels of education, income, access to healthcare, implementation of preventive measure, the prevalence of infection among women in reproductive age in general and in pregnant women in particular [15].

In Cameroon, available data on HIV-1 MTCT are mainly those on the implementation of options A and B in public hospitals and civilian research centers [5]. However, no published data on Option B+ and MTCT of HIV1 are available from military hospitals. The present work is a complement of what has been done so far. Option B + holds two advantages; it reduces infant mortality and morbidity and it increases the mother's life expectancy up to 3.75 years [16]. As one of the WHO recommendations was to determine the best program adapted in context for the elimination of vertical transmission of HIV, it would be important to address the question of associated risk factors, and to identify gaps in the follow-up of HIV-positive pregnant women in Cameroon.

In this study, one infant out of 130 was infected via MTCT, a baby boy. Although this number is not significant enough to infer a gender influence in MTCT of HIV-1, reports from India, (14.2% boys vs. 8.2% girls) and Kenya (23% boys vs. 11% of girls) indicated that baby boys were more vulnerable to MTCT-HIV than baby girls [17,18].

Concerning the study site, it is noted that the present research was carried out for reference group in two military hospitals, HMR1 and HMR2 respectively in Yaounde and Douala. In Cameroon, the general public are not quite aware that non-military patients are welcome in these hospitals. This is an important break in communication that has perhaps prevented the population of the towns of Douala and Yaounde from partaking in the efficient implementation of Option B+ in these hospitals. This may also justify the low sample size within the time period of this study [19].

The maximum frequency of caesarian sections (84.6%) was observed in the reference group. This is in accordance with a Meta-Analysis of 15 Prospective Cohort Studies where it was established that delivery through caesarean section dropped the risk of HIV transmission by about 50% as compared to other types of delivery. That percentage increases if the seropositive female adheres to her ARVs. The combination antiretroviral therapy plus caesarean section before or shortly after membrane ruptures drops the transmission to 87% [17,18].

One major socio-demographic parameter in this study was the home delivery. In fact, two out of twenty three mothers from the control group delivered at home. Home delivery was found to influence HIV-1 MTCT and represents an adverse factor of PMTCT ($p = 0.03$). The non-application of preventive treatment during pregnancy and delivery, and the absence of newborn prophylaxis are important risk factors accounting for more than 60% towards MTCT of HIV1 [2,11]. A study conducted in Zambia showed that it was possible to offer home-based HIV testing and NVP to traditional birth attendants for women who do not have easy access to health facilities in rural areas [20]. Cameroon through the ministry of public health should envisage extension of Option B+ in remote areas where access to healthcare facilities is not easy. This will lower significantly the number of undeclared cases, and the deficiency of follow-up. In France, Frange and Blanche founded that the rare residual cases of MTCT of HIV-1 were not related to the failure of the antiretroviral therapy, but rather associated with deficiencies in factors like; the follow-up of pregnant women, unrecognized screening test, specialized medical follow-up initiated late or stopped early and occurrence of an undiagnosed primary infection during pregnancy or breastfeeding among others [21].

3.2.2 Prevalence of HIV-1 MTCT

HIV-1 MTCT prevalence of 0% was found in the reference group. This prevalence seems encouraging with regard to the UNAIDS global targets of 95-95-95. However, further studies extended to the 10 regions of Cameroon would be necessary to confirm the closeness to the above UNAIDS global target by 2025. This prevalence is close to that of developing and industrialized countries such as Cuba and France where the rate of HIV1 MTCT is lower than 2% [21,22], and is indicative of an improvement if compared to the 0.9% global

prevalence of HIV1-MTCT [10]. Between 2008 and 2014 in Cameroon under the application of option B, the prevalence of MTCT was 7.1% in the rural part of Bamenda subdivision, in the North-West region, 8,7% (197/2254) in 58 health facilities in three regions of varying levels [9]. This observed significant decline in mother-to-child transmission of HIV has been achieved thanks to the consistent increase of awareness and a larger coverage of PMTCT measures [2,11]. Other contributing factors could be the availability of resources with the support of PEPFAR towards the promotion of PMTCT programs implementation in military hospitals [23]. As the matter of fact, the present result in the reference group is lower than that of Njom-Nlend (where the rate of transmission was at 4.20% at 6 weeks), but similar to the control group [24].

3.2.3 Risk factors related to HIV-1 MTCT

The observed significant risk factors associated with the prevalence of HIV MTCT included the lack of protection of the fetus by the mother through not taking the highly active antiretroviral treatment (HAART) during pregnancy ($p = 0.04$). Indeed, vertical transmission of HIV-1 occurs mainly from the end of the second trimester of pregnancy and is account for 15 to 25% of MTCT. It occurs during the passage of the virus from the mother to the fetus via the placenta, amniotic fluid or micro-transfusions [6,25]. The risk of infection in utero varies with maternal viral load, infections of the placenta by other pathogens, and the presence of sexually transmitted diseases. Nevertheless, it can be reduced by early maternal HAART continued throughout the pregnancy. The practice helps via two mechanisms: the reduction of maternal viral load in blood and genital secretions, and the pre-exposure prophylaxis through ART that cross the placenta and produce adequate systemic levels in infants especially during the passage into genital canal [1]. Thus, (as also noted by Ngwej et al) the absence of maternal ART during pregnancy had a 10-fold increased risk $P < 0.0001$ [26].

Absence of Nevirapine (NVP) administration to the newborn was not statistically related to vertical transmission ($p=0.9$). Nevirapine is a non-nucleotidic inhibitor of HIV-1 reverse transcriptase. In case of exposure, it blocks viral replication by binding to the catalytic site of the reverse transcriptase, thus inhibits RNA-dependent DNA polymerase activities. In the

absence of NVP prophylaxis, the process of viral replication remain active, so viral colonization of the newborn remains possible [1,4]. A different result was obtained in Burkina Faso by Ouedraogo et al (2015) where it was found that the risk of vertical HIV transmission was increased with the absence of antiretroviral administered to the newborn immediately after birth [27].

In the control group, the prevalence of MTCT-HIV was 4.35%. Since the global distribution of the MTCT indicates 15-25% of transmission in utero in the absence of ART, the babies from these women would have been more infected than that because of ignorance and lack of awareness [6,11]. In Lubumbashi Democratic Republic of Congo in a similar situation, 25% of the unaware mothers transmitted the virus to their newborns [26]. A contrary result was obtained with the study of Njom-Nlend where infection discovery during the last pregnancy was not a risk factor to the vertical transmission of HIV [24].

3.2.4 Limitations and strength

This study had some limitations. Reference and control group mothers were not paired because of the difficulty in enrolling urban mothers who do not visit antenatal care units during pregnancy (control group). Also, the sample size was not consistent within the collection period because of the exclusion of some samples for which information sheets did not contain reliable data relevant to this research. This could be the reason why there is a probable omission of positive cases. Nevertheless, one of the strengths of this research is that it adds value to previous studies in terms of comparison between the two groups of infected mothers: those on ART and those not on ART during pregnancy, although the study population was not divided into subgroups.

4. CONCLUSION

Transmission of HIV-1 from mother to child was non-existent under option B+ in the military hospitals of Douala and Yaounde: 0% (0/107), while the rate of transmission was 4.35% in women who discovered their seropositivity at or after birth in the BRH (1/23) within the study period. The risk factors associated with MTCT were home delivery ($p = 0.03$) and absence of highly active antiretroviral therapy (HAART) during pregnancy ($p = 0.04$). This prevalence is

encouraging with respect of the ambitious 95-95-95 targets of UNAIDS, which aims to eliminate the MTCT. However significant awareness to raise adherence and compliance to prevention programs are needed to ensure that all pregnant HIV-positive women are enrolled in a management program so that the risk factors associated with the infection of the newborn are brought under control, including the Highly active antiretroviral treatment of HIV positive pregnant women, the respect of the ANC schedule and the delivery at an appropriate health facility. We recommend a nationwide implementation of Option B+ to considerably reduce MTCT of HIV1 in Cameroon.

CONSENT AND ETHICAL APPROVAL

The present study received Ethical approval N° 006027/LE/MINDEF/01 from the Cameroon's Ministry of Defense as well as the Ethical authorization N° 896/L/MINSANTE/SG/DRSPE/BCASS/BFP from the Bertoua Regional Hospital prior to carrying out the research. Prior to the sample collection, mothers or caregivers were given an information's notice, afterwards consents for the HIV-exposed infants enrolled in the study were obtained by signing the consent form. All authors hereby declare that experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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