Original Research Article

Comparative Study of the prevalence of syphilis among pregnant women and prospective blood donors in Ekiti State, South Western Nigeria.

(Running Title: Comparative Study of Syphilis among Pregnant Women & Blood Donors)

Abstract.

Background: Syphilis is a contagious systemic bacterial infection which constitutes a global major public health challenge. The disease is sexually transmitted and can be contacted through blood transfusion as well. This study was to determine the seroprevalence of syphilis among pregnant women and prospective blood donors so as to appropriately assess the epidemiological pattern and population burden of the disease.

Materials and Methods: 300 antenatal enrolees and 370 prospective blood donors were recruited for this comparative study carried out over a period of 12 months. Questionnaire containing the socio demographic information were administered and 5ml of whole blood was collected by venepuncture into EDTA bottle from each participant after obtaining informed consent.

Plasma was immediately separated into a plain container by centrifugation at 2500g for 5minutes.

Treponema pallidum were detected using Diaspot and Lab Acon ELISA – based rapid test devices. Reactivity of each sample to the two test devices were interpreted as positive result while non-reactive results to the two test devices were interpreted as negative.

Results: Seroprevalence of syphilis among pregnant women was 2.0% and 0% in female blood donors. The seroprevalence of syphilis among male blood donors was 2.1% which was not comparatively different from the seroprevalence of 2.0% found in pregnant women.

Conclusion: There is strong association of syphilis infection with pregnancy and blood donors therefore screening of prospective blood donors and pregnant women for syphilis must be mandatory, accessible and affordable as this will enhance early detection of the disease for appropriate treatment.

Author summary

We carried out serological tests on 300 antenatal enrolees and 370 prospective blood donors to determine the seroprevalence of syphilis in pregnant women and prospective blood donors in two tertiary hospitals over a period of 12months.

Our study found the seroprevalence of syphilis among pregnant women to be 2.0% while it was 2.1% among male blood donors. None of the female blood donors tested during the study period was found to be syphilis positive.

The findings in our study buttress strong association of syphilis infection and transmission with pregnancy and blood donors.

We therefore advocate that counselling and public awareness about the mode of transmission and spread of this global health problem, syphilis, must be well established in all Obstetrics clinics and donor section of all blood banks in order to reinforce the need for routine screening of syphilis in all antenatal enrolee and prospective blood donors.

Also, screening of prospective blood donors and pregnant women for syphilis must be mandatory, accessible and affordable as this will enhance early detection of the disease for appropriate treatment.

Keywords: Seroprevalence, Pregnancy, Blood donors and Syphilis.

Introduction.

Syphilis is a systemic bacteria disease caused by Spirochaete, Treponema pallidum. It constitutes a major public health challenge globally [1]. Transmission of syphilis can be vertical through mother to foetus during pregnancy or at birth resulting in congenital syphilis [2]. It can also be horizontal transmission through sexual intercourse with infected person or through parenteral transmission among drug addicts [3]. The disease can also be transmitted through transfusion with infected blood [4]. Treponema infections affect about 36 million people worldwide with an average of 12million new cases yearly [5].

The seroprevalence of the infection in pregnancy is higher in developing countries than in developed countries ranging from 0.02 % in Europe to 4.5% in parts of United States, while 3-18% has been reported in Africa [6,7]. A potential for higher mortality and morbidity with syphilis infection has also been reported through association with HIV infection [8]. WHO reported that about 1million pregnant women have active Treponema infection, while about 1.6 million units of blood were discarded due to transfusion transmissible infections including Treponematoses in 2018[9,10]. The prevalence of Treponema infection in women of reproductive age group in Africa has been put at 0.36% to 3.6%, while the range among blood donors was 0.71% to 20% [11-13].

Blood transfusion even though a major life saving intervention for both medical and surgical conditions is unfortunately inadvertently a route of transmissible infection with varying degrees of severity. A transfusion transmissible infection is any infection that is capable of

transmission from person to person through parenteral administration of blood or its products [14]. Potential blood donors may be asymptomatic for syphilis hence the need for routine screening of all prospective blood donors and not only those that are of high risk behaviours. Window period of the infection is also a major challenge due to absence of the necessary serological markers to detect the infection during this period [15-17]. Provision of constant and safe blood has been a major challenge in the developing countries with high prevalence of transfusion transmissible infections. Lack of adequate voluntary blood donors, poor storage facilities due to irregular power supply contributed adversely to the availability of safe blood in the developing countries like Nigeria [18-20].

Incidence of syphilis infection is found to be higher in pregnant women who did not receive adequate antenatal care [21]. Adverse effects of maternal syphilis infection on the foetus may be fatal ranging from spontaneous abortion, low birth weight, still birth, congenital abnormalities and neonatal deaths [22]. Asymptomatic infected population stands the risk of potential reservoirs of the infection if not detected and treated [23]. Screening for syphilis during antenatal is therefore considered to be very cost effective in improving children's health [24]. In view of the potential risk of congenital syphilis infection and its attendant public health burden, there should be National guideline policy in Nigeria for routine antenatal screening [25,26].

The objective of this comparative study therefore was to determine the epidemiological pattern of this disease under a broader scope by comparing the prevalence of syphilis in pregnant women with the prevalence in prospective blood donors in order not to underestimate the population burden of this public health challenge.

The outcome of this study is expected to reinforce the need for urgent formulation of National guideline policy in Nigeria for the syphilis screening of all blood donors and all pregnant women during antenatal.

METHODOLOGY

Study Locations

The study was carried out in the departments of Haematology and Blood Transfusion as well as Obstetrics and Gynaecology at Federal Teaching Hospital, Ido Ekiti (FETHI), Ekiti State, and Ekiti State University Teaching Hospital (EKSUTH).

Ekiti State is in the south western region of Nigeria. It is located between longitudes 4° 45° and 5° 45° East of the Greenwich meridian and latitudes 7° 15° and 8° 15° North of the equator. The state has three senatorial districts (Ekiti North, Ekiti South and Ekiti Central) and 16 local government areas. Most of the indigenes of the state are Christians with a few Muslims and traditional worshippers. The indigenous people of the state speak Ekiti dialect and Yoruba fluently. Most Ekiti indigenes are farmers while a very few others are public servants and small scale business owners.

Federal Teaching Hospital, Ido Ekiti is one of the two tertiary health institutions in the state and is located at Ido Osi local government Area. Ekiti State University Teaching Hospital is located at the heart and capital of the state, Ado Ekiti which is the urban and industrial centre of the region. The two hospitals have well established departments of Haematology/Blood transfusion and Obstetrics /Gynaecology which cater for the teeming population of the state.

Sampling Technique

Consecutive sampling technique was adopted to enable enough samples to be collected until the desired number for study was reached. That was done to mitigate the negative impacts of COVID-19 pandemic era on recruitment of participants.

Sample Collection

Five millilitres of whole blood samples were collected by venipuncture from 370 blood donors and 300 antenatal patients and dispensed into EDTA blood collection tubes following informed consent. Plasma was immediately separated into a plain container from whole blood by subjecting the sample to centrifugation at 2,500g for 5 minutes.

Inclusion/Exclusion criteria

All prospective blood donors and antenatal attendees who gave informed consent, had no previous history of syphilis, within the 18-65 years and 15-48 years' age brackets for prospective blood donors and antenatal attendees were included in the study. Blood donors who had previous history of syphilis and who did not meet the criteria for blood donation such as < 50kg for 450 ml donation, haemoglobin < 13.5 g/dL and <12.5g/dL for male and female blood donors respectively, and previous history and current evidence of chronic illness were all excluded from the study. Antenatal attendees being treated for syphilis or had previous history of syphilis and non-pregnant women were also excluded from the study.

Ethical Approval

This study was approved by the Human Research and Ethics Committee of the Federal Teaching Hospital, Ido Ekiti and Ekiti State University Teaching Hospital, Ado Ekiti. Confidentiality of all information and Anonymity were ensured.

Serological assays

Treponema pallidium were detected using Diaspot and Lab Acon ELISA-based rapid test devices. Two reactive cases from the two test devices were interpreted as positive results

while two non-reactive results were interpreted as negative. Use of known positive and negative controls served as part of quality control measures to optimize quality data. Analyses were carried out according to the manufacturers' instructions.

Statistical Analyses

Research code was assigned to each questionnaire containing the sociodemographic and analytical data generated for ease of data entry and validation. Statistical Package for Social Sciences version 21 was used to analyze data and results were presented tables and charts. Data were saved indifferent formats and storage media as back-ups to prevent data loss.

RESULTS

Table 1: Demographic Characteristics of Antenatal Patients and Prospective Blood Donors Screened for Syphilis

Demographic Variable	Mean Age (Mean ± SD)	Absolute Number (%)
Total number of Antenatal Attendees	31.3 ± 0.3	300 (100.0)
Study Site:		
EKSUTH		140 (46.7)
FETHI		160 (53.3)
Gravidity		
Primigravida		15 (5.0)
Multigravida		285 (95.0)
Educational Status of Antenatal Attendees:		
At most primary education		11 (3.7)
At least secondary education		289 (96.3)
Total Number of Prospective Blood Donor Screened		**370 (100.0)**
Type of Blood Donors		
Voluntary Blood Donors		17 (4.6)

Replacement Blood Donors		353 (95.4)
Sex: Male Female		 326 (88.1) 44 (11.9)
Educational Status of Antenatal Attendees: At most primary education At least secondary education		 55 (14.9) 315 (85.1)

KEY: EKSUTH = Ekiti State University Teaching Hospital; FETHI = Federal Teaching Hospital, Ido-Ekiti; SD = Standard deviation; % = Percentage

Table 1 shows the demographic characteristics of antenatal patients and prospective blood donors screened for syphilis.

A total number of three hundred (300) antenatal enrolees were screened for syphilis. Fifteen percent (15%) of this population were primigravida while 85% of them were multigravida.

96.3 % of this population had at least secondary education while 3.7 % had primary education.

The prospective blood donors screened for syphilis were 370 and they were mainly replacement blood donors. Replacement blood donors constituted 95.4% while voluntary blood donors constitute 4.6%.

The prospective blood donors were mainly male, 88.1% while the females constituted 11.9 %. Prospective donors with at least secondary school education were 85.1% while the remaining 14.9% had only primary school education.

Table 2: Comparison of the Number of Antenatal and Prospective Blood Donors Participants according to Age Groups

Age Group (In years)	Antenatal Patients Number (Percent)	Blood Donors Number (Percent)	Percentage	P Value
< 20	2 (0.7)	8 (2.2)	1.5	
20 – 24	20 (6.7)	74 (20.0)	13.3	
25 – 29	94 (31.3)	81 (21.9)	- 9.4	

30 – 34	115 (38.3)	80 (21.6)	16.7	**0.006**
35 – 39	54 (18.0)	68 (18.4)	0.4	
40 – 44	13 (4.3)	39 (10.5)	6.2	
45 – 49	2 (0.7)	16 (4.3)	3.6	
50 – 54	0 (0)	3 (0.8)	0.8	
≥ 55	0 (0)	1 (0.3)	0.3	
Total	300 (100.0)	370 (100.0)	42.7	

Table 2 compared the age distribution of the antenatal enrolees with the prospective blood donors.

The highest age distribution for antenatal enrolees was age group 30- 34years which constituted 38.3 % of the total participants while ages less than 20years and 45- 49years constituted the least percentage of 0.7%respectively. The highest percentage of the blood donors were between the ages 25-29 years and it constituted 21.9 % while ages greater than 55years constituted 0.3% of the blood donors. There was a statistically significant difference in the age groups of antenatal attendees and prospective blood donors.

Table 3: Comparison of the Seroprevalence of Syphilis according to Number and Age Groups of Antenatal Attendees and Prospective Blood Donors

Age Group (in years)	Antenatal Attendees N (%)	Antenatal Attendees Seropositive for Syphilis N (%)	Male Blood Donors N (%)	Male Blood Donors Seropositive for Syphilis N (%)	Female Prospective Blood Donors N (%)	Female Prospective Blood Donors Seropositive for Syphilis N (%)
≤ 20	2 (0.7)	0 (0)	4 (1.2)	0 (0)	4 (9.1)	0 (0)
20 – 24	20 (6.7)	0 (0)	68(20.9)	2 (0.6)	6 (13.6)	0 (0)
25 – 29	94 (91.3)	1 (0.3)	74 (22.7)	3 (0.9)	7 (15.9)	0 (0)
30 – 34	115 (38.3)	1 (0.3)	71 (21.7)	1 (0.3)	9 (20.5)	0 (0)
35 – 39	54 (18.0)	4 (1.3)	60 (18.4)	1 (0.3)	8 (18.2)	0 (0)
40 – 45	13 (4.3)	0 (0)	33 (10.1)	0 (0)	6 (13.6)	0 (0)

45 – 49	2 (0.7)	0 (0)	12 (3.7)	0 (0)	4 (9.1)	0 (0)
50 – 54	0 (0)	0 (0)	3 (0.9)	0 (0)	0 (0)	0 (0)
≥ 54	0 (0)	0 (0)	1 (0.3)	0 (0)	0 (0)	0 (0)
Total	300 (100.0)	6 (2.0) †	326 (100.0) ††	7 (2.1) †††	44 (100.0)	0 (0)

Key: † = Approximate syphilis seroprevalence among antenatal patients
†† = Approximate percentage of male blood donors
††† = Approximate Syphilis seroprevalence was predominantly among male replacement blood donors

This table compares the overall syphilis seroprevalence among antenatal enrollees and prospective blood donors.

Overall syphilis seroprevalence among antenatal enrollees was 2.0% and the highest syphilis seroprevalence was reported among antenatal patients within the 35 – 39 years' age group. It was 0% among apparently healthy female blood donors. Comparison of syphilis seroprevalence between antenatal patients and female prospective blood donors (non-pregnant women) showed that infection with syphilis was associated with pregnancy. Overall syphilis seroprevalence among male prospective blood donors was 2.1% and that was not comparatively different from the finding among pregnant women.

DISCUSSION

Syphilis constitutes a global public health problem and hence determination of the epidemiological pattern on a broader scope is required to avoid underestimation of the disease burden.

This comparative study of the prevalence of the disease among pregnant women and prospective blood donors' findings has shed more light on the infection rate and spread of this disease which is strategic to adequate planning towards elimination of the congenital transmission of Treponema infection as proposed by World health organization [27].

Findings in this study corroborated the fact that infection of syphilis is highly associated with pregnancy and blood transfusion [28,29].

Overall syphilis seroprevalence among antenatal enrolees was 2.0%. This finding is relatively lower than the findings in the previous systematic review done in Sub Sahara Africa among pregnant women where the prevalence was reported as 4.5% [30,31]. The value was even found to be higher among incarcerated women and Australia aboriginal populations where

the prevalence was 6.1 and 16.8 respectively [32,33]. However, a study conducted by O.A Olowe et al on the prevalence of syphilis among pregnant women in two Health care facilities in the same south western Nigeria in 2014 recorded a lower value of 1.0% [34]. Seroprevalence of 1.8% was reported among pregnant women in India, a value that is relatively close to our finding in this study [35]. It is of note that the prevalence of syphilis is relative to the degree of risk profiles of the population involved in any study as well as the culture and traditional practices of the people, which may account for this variance in seroprevalence findings. hence the higher values recorded in Australia aboriginal populations and incarcerated women [36].

The seroprevalence of syphilis among female blood donors was found to be zero while it was 2.1 in male blood donors. The zero seropositivity recorded in female donors may be due in part to the participants' gender disproportionality which skewed towards the male population. However, this finding of gender seroprevalence inequality was not different from the study carried out in in Northwest Ethiopia where the overall seroprevalence of syphilis among blood donors was found to be 1.2% with 1.5 % and 0.7% in male and female respectively [37]. The higher seroprevalence finding in male over female have also been reported in previous studies carried out in North Showa [38] and Gondar [39]. This relative higher seropositivity in males may be attributed to increased risk behaviour such as multiple sexual partners and substance abuse among males than females. Our study showed that only 4.6% of the prospective blood donors screened were voluntary blood donors while 95.4% were replacement blood donors of which the bulk are usually commercial donors. Findings showed that some of these commercial donors are subjects of economic crisis with attendant low socioeconomic status and risky practices like sharing of personal care items, unprotected sexual intercourse with multiple sexual partners.

Comparison of syphilis seroprevalence in our study between antenatal patients (2.0) and female prospective blood donors (0) showed that infection with syphilis is associated with pregnancy [40]. The overall syphilis seroprevalence among male prospective blood donors, 2.1% that was found not to be comparatively different from the seroprevalence among pregnant women may insinuate that transfusion transmissible syphilis remains the main cause of seropositivity of syphilis among antenatal patients.

CONCLUSION:

In view of the strong association of syphilis infection with pregnancy and blood donors, screening of prospective blood donors and pregnant women for syphilis must be mandatory, accessible and affordable as this will ensure early detection and treatment of infected individuals and their partners thereby reducing the prevalence or eliminating this global public health problem with all the attendant complications especially in the unborn child.

Advocacy, counselling and public awareness about the mode of transmission and spread of this global health problem, syphilis, must be well established in all Obstetric clinics and donors section of all blood banks in order to reinforce the need for routine screening of syphilis in antenatal women and prospective blood donors.

CONSENT:

Informed consent was obtained from all the participants in this study.

REFERENCES

1. T. Shimelis, K. Lemma, H. Ambachew and E. Tadesse, "Syphilis among people with HIV infection in southern Ethiopia: sero-prevalence and risk factors," BMC infectious diseases, vol. 15, no. 1, pp. 189, 2015.

2. Adesina O, Oladokun A. Routine antenatal syphilis screening in south western Nigeria- a questionable practice. Annals of Ibadan Postgraduate Medicine. 2010; 8:16- 19.

3. O. Loza, T. L. Patterson, M. Rusch, G. A. Martínez, R. Lozada, H. Staines-Orozco, C. Magis-Rodríguez and S. A. Strathdee, "Drug-related behaviors independently associated with syphilis infection among female sex workers in two Mexico–US border cities," Addiction, vol. 105, no. 8, pp. 1448-1456, 2010

4. L. Vera, D. Milka, S.-L. Nurith and S. Eilat, "Prevalence and incidence of syphilis among volunteer blood donors in Israel," Journal of blood transfusion, vol. 2014, 2014

5. G. Elyamany, "Prevalence of Syphilis among Blood and Stem Cell Donors in SaudiArabia: An Institutional Experience," Electronic physician, vol. 8, no. 8, pp. 2747, 2016

6. H. Saloojee, S. Velaphi, Y. Goga, N. Afadapa, R. Steen, and O. Lincetto, “Te prevention and management of congenital syphilis: an overview and recommendations,” Bulletin of the World Health Organization, vol. 82, no. 6, Article ID 008094, pp. 424–430, 2004.

7. E. L. Korenromp, S. G. Mahian´e, N. Nagelkerke et al., “Syphilis prevalence trends in adult women in 132 countries – estimations using the Spectrum Sexually Transmitted Infections model,” Scientific Reports, vol. 8, no. 7, pp. 1–10, 2018.

8. N. Nair, A. D. Urhekar, S. Pachpute, and A. Srivastava, “Original Research Article Incidence of Syphilis among pregnant women attending a tertiary care hospital in Navi Mumbai, India,” International Journal of Current Microbiology and Applied Sciences, vol. 2, no. 8, pp. 79–84, 2013

9. WHO, "Global Health Observatory (GHO) Data-Sexually Transmitted Infections (STIs) ", Ed., 2018.

10. WHO, "Blood Donor Selection-Guidelines on Assessing Donor Suitability for Blood Donation ", Ed., 2012.

11. E. L. Korenromp, G. Mahiané, J. Rowley, N. Nagelkerke, L. Abu-Raddad, F. Ndowa, A. El-Kettani, H. El-Rhilani, P. Mayaud and R. M. Chico, "Estimating prevalence trends in adult gonorrhoea and syphilis in low-and middle-income countries with the SpectrumSTI model: results for Zimbabwe and Morocco from 1995 to 2016," Sex Transm Infect, pp. sextrans-2016-052953, 2017.

12. T. Deressa, W. Birhan, B. Enawgaw, M. Abebe, H. W. Baynes, M. Desta, B. Terefe and M. Melku, "Proportion and predictors of transfusion-transmissible infections among blood donors in North Shewa Zone, Central North Ethiopia," PloS one, vol. 13, no. 3, pp. e0194083, 2018.

13. E. Quintas, A. D. C. Cogle, A. Sebastião, A. da Costa Pereira, A. Sarmento, J. VanDúnem and L. Cordeiro, "Prevalence of syphilis in blood donors in angola from 2011 to 2016," 2018.

14. Ajugwo AO, Erhabor TA, Eledo BO, Eze RI, Digban KA. Prevalence of Transfusion Transmissible Infections in a Nigerian Tertiary Hospital. J Transm Dis Immun.2017; Vol. 1 No. 2:11

15. Grace Bartonjo et al. Prevalence and associated risk factors of transfusion transmissible infections among blood donors at Regional Blood Transfusion Center Nakuru and Tenwek Mission Hospital, Kenya. Pan African Medical Journal. 2019; 34:31. [doi: 10.11604/pamj.2019.34.31.17885]

16.B. Tessema, G. Yismaw, A. Kassu et al. Seroprevalence of HIV, HBV, HCV and syphilis infections among blood donors at Gondar University Teaching Hospital, Northwest Ethiopia: declining trends over a period of five years. *BMC Infectious Diseases*. 2010; vol. 10, article 111, pp. 1–7.

17.Siddiqui FM, Siddiqui N, Oluwatayo O, Jabeen S, Qadir SM et al. Prevalence of Transfusion-Transmissible Infections among Voluntary Blood Donors in Tertiary Health-Care Facility in Islamabad, Pakistan. J Clin Trials. 2019; 9: 383.

18. Kagu MB, Ahmed SG, Askira BH, Utilization of Blood Transfusion Services in North Eastern Nigeria. Highland Med RJ 2007; 5:27-30.

19. Kuliya- Gwarzo. A Survey of Blood Transfusion needs in a tertiary Nigeria Instituite. Sahel Med. J 2007:10:19-23.

20. Enosolease ME, Imarengiaye C, Awodu AO. Donor blood procurement and utilization at the University of Benin Teaching Hospital. Benin City Africa J Reprod Health 2004; 8:59-63.

21. Onwuezobe IA, Ochang EA, Umoiyoho A, Bassey EA, Umoffia EM. Prevalence of syphilis seropositivity in antenatal clinic in a teaching hospital in South-South region of Nigeria. Asian Pacific J Trop. Med. 2011 ;(91):21-23.

22. WHO/2004. Sexually transmitted infections: World health organization fact sheet; 2004

23. CDC, "Sexually Transmitted Disease Surveillance 2017," Ed., 2018.

24. Terris-Prestholt F, Watson-Jones D, Mugeye K, Kumaranayeka K, Ndeki L, Weis H, Changalucha J, Todd J, Lisekic F, Gunodoka B, Mabey D, Hayes R. Is antenatal syphilis still cost effective in sub-sahara Africa? Sex Trans Dis. 2003; 79:375-381

25. Ratnam AV, Din SN, Hira SK, Bhat GJ, Wacha DS, Rukmini A, et al. Syphilis in pregnant women in Zambia. Br J Vener Dis. 1982; 58(6):355-358.

26. Taiwo SS, Adesiji OY, Adekanle AD. Screening for syphilis during pregnancy in Nigeria: A practice that must continue. Sexually Transmitted Infections. 2007; 83(5):357-358. 1

27. WHO, The global elimination of congenital syphilis: rationale and strategy for action, World Health Organization,2007.

28. WHO, "Global Health Observatory(GHO) data – Sexually Transmitted Infections (STIs)", ED.,2018.

29. WHO, "Blood Donor Selection- Guildelines on Assessing Donor Suitabiity for Bloo Donation", Ed.,2012.

30.A. Kuznik, A. G. Habib, Y. C. Manabe, and M. Lamorde, "Estimating the public health burden associated with adverse pregnancy outcomes resulting from syphilis infection across 43 countries in sub-Saharan Africa," *Sexually Transmitted Diseases*, vol. 42, no. 7, pp. 369–375, 2015.

31.R. M. Chico, P. Mayaud, C. Ariti, D. Mabey, C. Ronsmans, and D.Chandramohan, "Prevalence of malaria and sexually transmitted and reproductive tract infections in pregnancy in sub-Saharan Africa: a systematic review," *The Journal of the American Medical Association*, vol. 307, no. 19, pp. 2079–2086, 2012.

32.F. G. Kouyoumdjian, D. Leto, S. John, H. Henein, and S. Bondy, "A systematic review and meta-analysis of the prevalence of chlamydia, gonorrhoea and syphilis in incarcerated persons," *International Journal of STD & AIDS*, vol. 23, no. 4, pp. 248–254, 2012.

33.S. Graham, L. W. Smith, C. K. Fairley, and J. Hocking, "Prevalence of chlamydia, gonorrhoea, syphilis and trichomonas in Aboriginal and Torres Strait Islander Australians: a systematic review and meta-analysis," *Sexual Health*, vol. 13, no. 2, pp. 99–113, 2016.

34. Olowe et al. Prevalence of Syphilis among Pregnant women in two Health Care Facilities in South Western Nigeria. British Journal of Medicine and Medical Research 4(34):5431-5438,2014.

35. Seith S, Sharma K, Dhaliwal LK, Banga SS, Sharma M. Declining trends in syphilis prevalence among antenatal women in Northern India: A 10year analysis from a tertiary health care centre. Sex Trans. Infection. 2007; 83:592-594.

36. S Graham, L. W Smith, C. K. Fairley, and J. Hocking, "Prevalence of chlamydia, gonorrhea, syphilis and trichomonas in Aboriginal and Torres Straint Islander Australians: a systematic review and meta-analysis," Sexual Health, vol. 13, no.2, pp. 99-113,2016.

37. Shiferaw E, Tadilo W, Melkie I, Shiferaw M (2019) Sero-prevalence and trends of transfusion – transmissible infections among blood donors at Bashir Dar district blood bank, northwest Etiopia: A four-year retrospective study. PloS ONE 14(4): e0214755. https://doi.org/10.1371/journal.pone.0214755

38. Deressa T, Birhan W, Enawgaw B, Abebe M, Baynes HW, Desta M et al. Proportion and predictors of transfusion transmissible infections among blood donors in North Shewa Zone, Central North Ethiopia. PLOS ONE 2018. Htts://doi.org/10.1371/journal.pone.0194083.

39.Tessema B, Yismaw G, Kassu A, Amsalu A, Mulu A, Emmrich F, et al.Seroprevalence of HIV, HBV, HCV and syphilis infections among blood donors at Gondar University Teaching Hospital, Northwest Ethiopia : declining trends over a period of five years . BMC Infectious diseases. 2010;10(1):111

40. G.B Gomex, M. L. Kamb, L. M. Newman, J. Mark, N. Broutet, and S. J. Hawkes, "Untreated maternal syphilis and adverse outcomes of pregnancy: a systemic review and meta- analysis" Bulletin of the World Health Organization, vol. 91, no. 3, pp. 217 –226, 2013.