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Maternal Vagina Colonization with Extended Spectrum B-Lactamase Producing *Enterobacteriaceae* in Pregnancy: Any Correlation with ESBL-Positive Early Onset Neonatal Sepsis?

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Abstract
Background: *Species of Enterobactericeae elaborate extended spectrum β-lactamases(ESBL), which areenzymes that confer resistance to third generation cephalosporins and aztreonam but inhibited by β-lactamase inhibitors, like clavulanic acid. Studies have reported association betweenESBL- productionand multidrug resistance to other classes of antibiotics like aminoglycosides and ampicillins.This development is disturbing, given that cefotaxime– gentamicinor ampicillin-gentamicin combinations are readily the empirical regimens for suspected neonatal sepsis. Treatment of ESBL- infections, posed serious therapeutic challenges, resulting in increasing treatment failures.*
The purpose of this study is to establish if correlation exists between maternal vagina colonization with ESBL-producing Enterobacteriaceae in pregnancy and ESBL- early onset neonatal sepsis (EONS) using E.coli and K .pneumoniae as surrogates.
Materials and Methods*: The study was conducted in May, 2015. Maternal high vagina swabs (HVS) and baby's blood cultures were evaluated for surrogates of E. coli, Enterobacteriaceae and Gram negative bacilli respectively. Antibiogram, ESBL confirmation and plasmid DNA analysis were performed on these isolates. Mother-to-newborn transmission of sepsis was evaluated ,by comparing antibiogram and plasmid DNA profiles of mother and her baby's isolates. A similarity in these parameters between a mother and her newborn was considered a mother- to-newborn transmission of blood borne infection.*
Results: *ESBL was detected in 16.0% of the maternal vagina isolates and 19.3% of the babies suspected of sepsis. Percentage Mother-to-newborn transmission of ESBL-sepsis was 0.6%. This was not statistically significant to establish a correlation between maternal ESBL- colonization in pregnancy and ESBL- early onset neonatal sepsis.*
Conclusion*: There is no correlation between maternal vagina colonization with ESBL-producing Enterobacteriaceae in pregnancy and ESBL-early onset neonatal sepsis.*
Key words*: MaternalVagina, Pregnancy, ESBL, Hospital, Nigeria*

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I. Introduction

Rising prevalence of *Enterobacteriaceae* over *Group B Streptococcus* (GBS) as predominant cause of early onset neonatal sepsis has been reported, resulting from antenatal antibiotic prophylaxis for GBS.[1] Aside from GBS prophylaxis, intrapartum antibiotics use appears to be unavoidable because, in the events of threatened premature deliveries, antibiotic administration may apply, sometimes as a measure to address other potential risks.[2] Prolonged maternal exposure to broad spectrum antibiotics in pregnancy permits changes in maternal vagina flora including emergence of potentially antibiotic resistant organisms. These may colonize the newborn in the course of vaginal delivery with risks of infections or outbreaks.

Members of *Enterobacteriaceae*are known to elaborate extended spectrum β-lactamases (ESBL), which are enzymes that confer resistance to third generation cephalosporins but which are inhibited by β-lactamase inhibitors likeclavulanic acid[3,4]. Organisms producing ESBLs have also been identified with multidrug resistance to other classes of antibiotics including aminoglycosides, ampicillins and fluoroquinolones.[5,6] This development is worrisome, given that ampicillin-gentamicin or cefotaxime-gentamycin

combinations are readily the empirical antibiotic regimens for the treatment of suspected bacterial neonatal sepsis. Treatment of infections arising from ESBL-producing organisms pose serious therapeutic challenges as it is limited to small number of expensive drugs.[7] Colonization of newborn babies by ESBL-producing gram-negative bacteria and associated increasing rate of progression to neonatal sepsis have been reported in community based studies.[8-10] Disturbing global reports of mother to newborn transmission of ESBL- producing organisms during deliveries are documented.[11,12]In Abuja, Nigeria, an outbreak of high mortality nosocomial septicaemia caused by a single ESBL-producing strain of *K. pneumoniae* in a neonate referred to the hospital was reported.[13] Studies have identified prevailing risk factors for early onset neonatal sepsis in developing countries to include prematurity, low birth weight, prolonged rupture of membrane, maternal fever, foul smelling liquor, multiple vaginal examination, prolonged labor, aspiration of meconium and intrapartum antibiotics use.[14-16]These agreed with findings in developed countries, but whereas there is established culture of laboratory based risk follow-up of the exposed in developed economies, this is often not the case in developing economies. This missing gap may probably account for the high neonatal mortality rates in developing world.

The general objective of this study is to establish if there is a correlation between maternal vagina colonization with ESBL-producing *Enterobacteriaceae* in pregnancy and ESBL- early onset neonatal sepsis using *Escherichia coli* and Klebsiella *pneumoniae* as surrogates. The specific objectives include determining the carriage rate of ESBL-producing *Enterobacteriaceae* amongst pregnant women in LUTH, determining the antibiotic susceptibility profiles of maternal vagina and newborn blood isolates, evaluating the risk factors for maternal vagina ESBL carriage and determining the percentage mother-to-newborn transmission of ESBL-EONS in the first 7 days of postnatal life.

II. Materials and Methods

Study Design: This was a self-sponsored prospective cohort study which was conducted amongst pregnant women in labour, gestational ages, ≥28 weeks and their newborn babies, suspected of sepsis within the first 7 days of postnatal life. Simple structured questionnaires were administered to obtain socio- demographic data. Maternal High vagina swabs (HVS) and blood of the babies suspected of sepsis were cultured. Antibiogram, ESBL-testing and paired plasmid profiling of mother and newborn isolates were conducted.

Study Duration: The study was carried out between August 2014 and September 2015.

Study Location: The study was carried out in Lagos University Teaching Hospital, Lagos, Nigeria, a 761 bed tertiary health facility.[17] It is patronized by inhabitants of Lagos and adjoining states, as well as occasional referrals from bordering countries.

Sample Size :A total of 350 pregnant women, gestational ages ≥28 weeks and 57 of their babies suspected of sepsis were enlisted.

Sample Size Calculation: Sample size was calculated on the basis of the formular[18] :$N = Z^2 pq/d^2$, where N = sample size, p = 20.8% (the local ESBL prevalence[19]), Z = critical value at 95% confidence level, set at 1.96 and q = 1- p, d = precision, at 5%.

When calculated, N = 253, however, a sample size of 350 was used to increase the validity of the study and make up for non-response cases.

Ethical consideration: The study was reviewed and approved by the Health Research Ethical Committee of LUTH. Informed consents were obtained from participating mothers for selves and their newborn babies.

Inclusion criteria:

1. Pregnant women receiving antenatal care in LUTH
2. Gestational ages ≥28 weeks
3. Safe vaginal delivery(SVD) only.
4. Subjects who gave written informed consent.
5. Newborn babies who showed signs and symptoms of sepsis within first 7 days of postnatal life

Exclusion criteria:

1. Parturient women with intrapartum hemorrhage.
2. Pregnant women who delivered by Caesarian section (CS).
3. Still birth deliveries.

Samples collection

HVS were collected from the women in first stage of labor and submitted for processing. A milliliter of venous blood was collected for culture, from each participating baby, suspected of sepsis.

Samples processing

All samples were processed in a level 2 biosafety cabinet.

Culture

Maternal HVS samples were inoculated on MacConkey agar plates and incubated for 18 hours at 37^0C in ambient air. Colonies suspected to be those of *E.coli* and *K. pneumoniae*, based on morphological appearance and Gram staining reactions were identified to species using Microbact biochemical identification kits (Oxoid ltd, Basingstoke Hants, UK).

Blood samples of the babies suspected of sepsis were inoculated into BACTEC PEDS/F paediatric culture bottles and incubated aerobically at 37^0c using BD bactec 9050 incubator (Becton Dikinson Inc. New Jeysey, USA). Incubated samples were monitored daily for growth for 7 days. About 0.5ml of samples that flagged positive was aspirated and Gram stained. Those that yielded Gram negative organisms were sub-cultured on MacConkey, blood and chocolate agar plates and incubated at 37^0c in ambient air for 18 hours. Gram negative isolates were identified to species using Microbact biochemical identification kits (Oxoid ltd, Basingstoke Hants, UK).

Antibiotic susceptibility test

Antibiotic susceptibility testing was by the Kirby Bauer disk diffusion technique according to Clinical Laboratory Standard Institute (CLSI) guidelines.[20]

Gram negative bacteria with zones of inhibition to ceftazidime or cefotaxime or both less than 22mm and 27mm respectively were suspected to be ESBL- producing. Suspected isolates were confirmed to be ESBL-producing by the Combined Disks Synergy Technique using *E.coli* ATCC35218 and *E.coli* ATCC25922 as positive and negative controls respectively.

PCR amplifications of ^{bla}TEM, ^{bla}SHV, ^{bla}CTX-M and ^{bla} OXA genes.

PCR amplifications for TEM, SHV, CTX-M and OXA plasmid genes were performed on a thermocycler (A&E Laboratories UK model cyl- 005-1) using primer pairs.[21] The reaction volume of 25µl was used consisting of 10X PCR buffer, 10mM $MgCl_2$, 10 mMdNTP mixture, 5 U/µL of Taq DNA polymerase (Fermentas, USA), 10pmol of each primer set and 5ng of extracted plasmid DNA from samples, negative (*E.coli ATCC25922*) and positive (*ATCC 35218)* controls. Amplifications was done following an initial denaturation at 96^OC for 5minutes, 35 cycles at 96^OC for 60S, 60 ^{O}C for 60S (SHV), 58^OC for 60S (TEM& OXA) and 50 ^{O}C for 6Os (CTX-M), followed by 72^OC for 60S and a final period of extension at 72^OC for 10 minutes. Amplified gene products (10µL) along with controls were separated using 1.5% agarose gel electrophoresis stained with 0.5µg/ml of ethidium bromide. Stained gel was examined under ultraviolet (UV) light transilluminator (Clinix Japan, model 1570). Major bands corresponding to the major band sizes for the genes tested were considered for the profiling. A DNA ladder digest of 1kilobase pair (Fermentas USA) was used as molecular weight marker.

Relatedness

An agent of sepsis is said to be transmitted from mother to her newborn if the maternal vagina isolate has the same plasmid DNA arrays and antibiotic susceptibility pattern with the blood isolate from her baby.

Data analysis

SPSS software version 19.0 (SPSS Inc. Chicago, IL. USA) was used for dataanalysis.Continuous variables were represented as mean ± deviation(SD). Categorical variables were represented as actual numbers or percentages or bar or pie charts. Categorical data were compared using chi square and p- value < 0.05 were considered significant for all tests. Multivariate logistic regression analysis was used to determine the individual contributions of clinical factors to prediction of risk of ESBL-colonization of pregnant woman and her newborn baby.

III. Results

Socio -demographic data

The pregnant women were ages,16 – 46 years (average,29.3± 1.69 years). Gestational ages of the women were of the range 28-42 weeks (average, 38.5 ±0.410weeks). 200 (57.1%), had tertiary education, 148 (42.3%) had secondary education and 2 (0.6) had primary education. They were mostly civil servants, 156 (44.6%), 70(20%) were of the business class, 113 (32.2%) were house-wives and 11(3.3%) belonged to other professions.

Booked cases were 304 (86.9%) while 46 (13.1%) wereemergency referrals from peripheral hospitals that were not booked for antenatal care. No case of multiple pregnancy was enrolled, as they were delivered by caesarian section.

There was history of previous hospital admission in 40(11.4%), fever in 183 (52.3%), previous antibiotics use in 103 (29.4%), vaginal discharge in 89 (25.4%) of cases.

Fifty seven (57) babies suspected of sepsis, all were term, gestational ages 37-42 weeks, 24 (42.1%) were males and 33 (57.9%) were females. Body weights at birth were all normal (2.5 – 4.0kg). Total hours spent in labor was normal for all deliveries (average 8.2 ± 0.54 hours).

Percentage ESBLcolonization in pregnancy.

A total of 243 (69.4%) pregnant women had vagina colonization with *Enterobacteriaceae. Escherichia coli* constituted 184 (52.6%) and Klebsiella pneumoniae 59 (16.9%). There were co-colonization with *E. coli* and *K. pneumoniae* in 23 (6.6%) subjects. The percentage maternal vagina ESBL-colonization was 16% (56/350) with *Escherichia coli* contributing 30(8.6%) and *Klebsiellapneumoniae* 26(7.4%). The predisposition to ESBL-production was significantly higher in *K. pneumoniae* than E.coli ($X^2 = 358.653$ *df =4, p=0.01)*.

Fifty seven (16.2%, n = 350) were suspected of sepsis in the first 7 days of postnatal life based on defined criteria. Twelve (21.1%, n= 57) of the babies suspected of sepsis, grew Gram negative bacilli . Eleven (19.3%, n=57) were ESBL-positive while 1(1.8%, n=57) was ESBL-negative.

Table no. 1 shows the risk Factors for vagina ESBL-Colonization in the Pregnant women. Four factors were statistically significant as risk factors for vagina ESBL- colonization in pregnancy using univariate regression analysis ($p \leq 0.05$). These included previous antibiotic use in pregnancy of ≥ 4 weeks prior to sample collection, fever in pregnancy of ≥ 4 weeks prior to sample collection, malodourous vaginal discharge in pregnancy of ≥ 4 weeks prior to sample collection, and positive HIV status. These potential factors from the univariate regression analysis were entered into a forward step-wise multivariate logistic
regression analysis. Only malodorous vaginal discharge of ≥ 4 weeks prior to sample collection (p=0.001) was statistically significant as independent risk factor for vagina ESBL- colonization in pregnancy.

Table no 1: Shows the results of univariate / multivariate regression analysis of risk factors for maternal vagina ESBL-colonization in pregnancy**.**

Risk factors	Univariate analysis		Multivariate analysis	
	OR	P-value	OR	P- value
Antibiotic use in pregnancy	3.76	0.00	2.27	0.13
Fever in pregnancy	2.28	0.009	0.38	0.54
Malodorous vag. discharge	5.20	0.00	6.24	0.001
HIV	0.18	0.00	0.00	0.99
Occupation	1.14	0,057	0.15	0.7
Maternal age	6.74	0.44	-	-
Higher education	1.37	0.630	-	-
Previous hospital admission	3.47	0.302	-	-
Antenatal care	0.65	0.287	-	-
Steroidal use in pregnancy	0.00	0.58	-	-

Table no 2 shows the antibiogram of isolates of maternal vagina colonizing *Enterobacteriaceae* in pregnancy

The antibiotic susceptibility of the isolates was tested against twelve antibiotics. The antibiotic susceptibility rates of *Enterobactericeae* isolates amongst the pregnant women were as followed: Imipenem (99.2%), followed by piperacillintazobactam (94.7%) and meropenem (93.4%). Amoxicillin clavulanate with 53.1% was least sensitive.

Table no 2: Shows antibiogam of maternal vagina colonizing *Enterobacteriaceae* in pregnancy.

Antibiotics	Sensitivity (%)	Intermediate sensitivity (%)	Resistance (%)	Total (%)
Amox. clav.(20 + 10µg)	129 (53.1)	27 (11.1)	87 (35.8)	243 (100)
Ceftazidime (30µg)	190 (78.2)	9 (3.7)	44 (18.1)	243 (100)
Cefotaxime (30µg)	187 (77.0)	7 (2.9)	49 (20.1)	243 (100)
Cefepime (30µg)	214 (88.1)	9 (3.7)	20 (8.2)	243 (100)
Cefoxitin (30µg)	213 (87.6)	6 (2.5)	24 (9.9)	243 (100)
Levofloxacin	188 (77.4)	11 (4.5)	44 (18.1)	243 (100)
Azstreonam	183 (75.3)	5 (2.1)	55 (22.6)	243 (100)
Piperacillintazo. (110µg)	230 (94.7)	3 (1.2)	10 (4.1)	243 (100)
Gentamicin (10µg)	190 (78.2)	12 (4.9)	41 (16.9)	243 (100)
Meropenem (10µg)	226 (93.4)	5 (5.1)	11(4.5)	243 (100)
Imipenem (10µg)	241 (99.2)	0 (0)	2 (0.8)	243 (100)
Ertapenem (10µ)	213 (87.6)	5 (2.1)	25 (10.3)	243 (100)

Key: Piperac/Tazo = piperacillintazobactam
Amox/clav = Amoxicillin clavaulanate

Table no 3 shows the antibiogram of the blood isolates from the newborn babies with sepsis within the first 7 days of postnatal life.
The antibogram of these blood isolates demonstrated widespread resistance. Amoxicillin clavulanate, cefotaxim and Azstreonam had percentage susceptibilities of 8.3%, 8.3% and 25% respectively. Imipenem was 100% susceptible, Piperacillin tazobactam (91.7%) and Ertapenem (66,7%).

Table no 3: Shows theantibiogram of Gram negative blood isolates from the newborn babies with sepsis.

Antibiotics	No(%) sensitivity	NO(%) intermediate sensitivity	No(%) resistance	No(%) total
Amoxicillin clavulanate	1(8.3)	0 (0)	11(91.7)	12 (100)
Ceftazidime	2(16.7	1 (8.3)	9 (75.0)	12 (100)
Cefotaxime	1 (8.3)	1 (8.3)	10 (83.4)	12 (100)
Cefepime	6 (50.0)	1 (8.3)	5 (41.7)	12 (100)
Cefoxitin	12(100)	0 (0)	0 (0)	12 (100)
Levofloxacin	6 (50.0)	1 (8.3)	5 (41.7)	12 (100)
Azstreonam	3 (25.0)	1 (8.3)	8 (66.7)	12 (100)
Piperacillintazobactam	11 (91.7)	1 (8.3)	0 (0)	12 (100)
Meropenem	6 (50)	0 (0)	6 (50.0)	12 (100)
Imipenem	12 (100)	0 (0)	0 (0)	12 (100)
Ertapenem	8 (66.7)	0 (0)	4 (33.3)	12 (100)

Figure no 1 shows the percentage co-existence of ESBL- producction with resistance to other classes of antibiotics among the Gram negative blood isolates from the babies with sepsis.Extended spectrum β-lactamases production co-existed with resistance to other classes of antibiotics as follows, amoxicillin clavulante (90.9%%), aztreonam (72.7%), gentamicin (63.6%) and levofloxacin (45.5%).

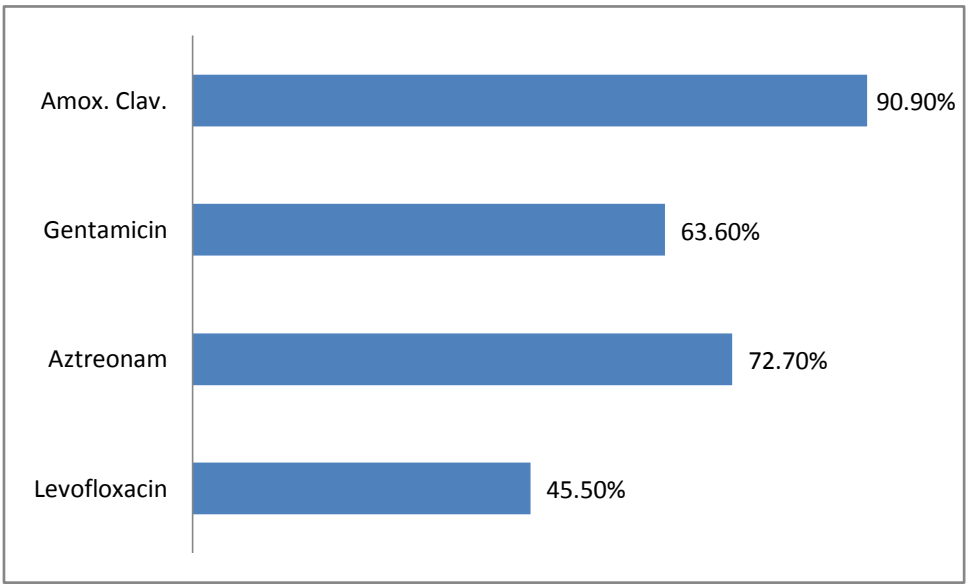


Figure no1:S hows Percentage co-existence of ESBL-production with resistance to other classes of antibiotics among Gram negative blood isolates from the babies with sepsis
Key :Amox. Clav. = Amoxicillin clavulanate.

Figure no2 : shows mother-newborn TEM1 plasmid gene comparative electrophoresis of a segment of the sample lanes 19-24, where lane no 19 represents sample 105M, 19(105M), 20(105B), 21(125M), 22(125B), 23(274M) and 24(274B). Lane numbers 20,21,22 showed positive amplification band of 517 bp corresponding to the expected band size for TEM gene. Lane numbers 19, 23 and 24 showed no positive bands for TEM gene.

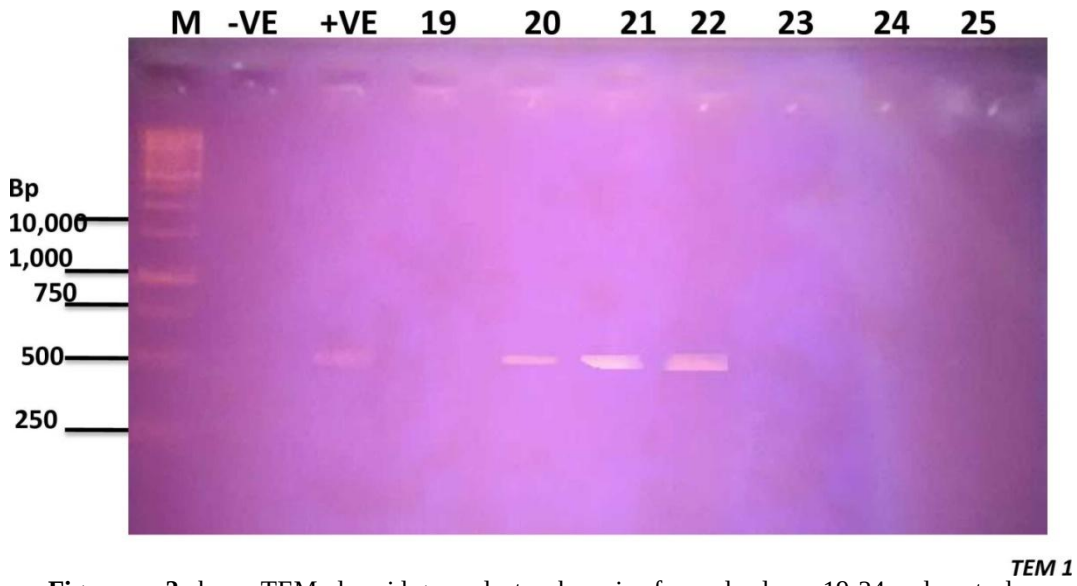
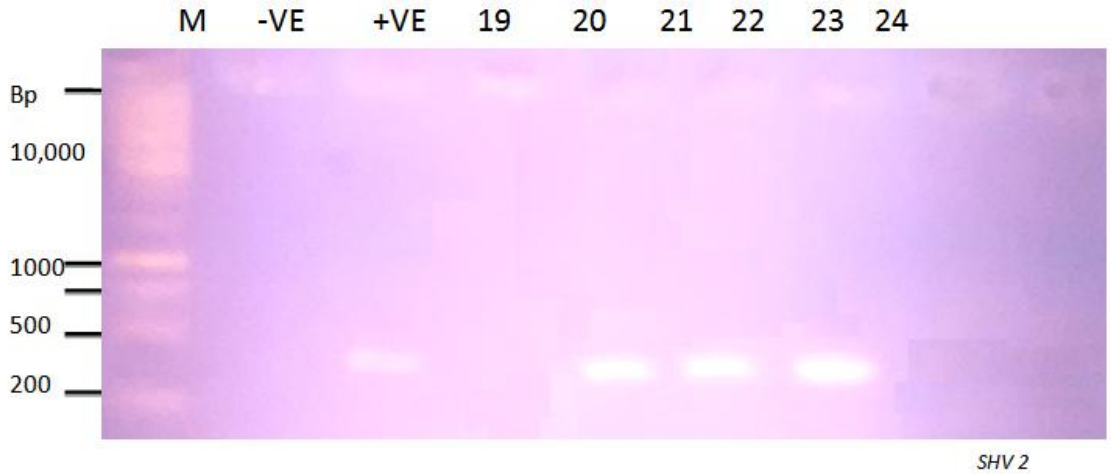


Figure no 2:shows TEM plasmid gene electrophoresis of samples lanes 19-24 and controls.
Lane M shows bands for 1Kb molecular ladder. Lanes + VE and– VE represent the positive and negative controls respectively.

Fgure no3 shows mother-newborn plasmid SHV2 gene comparative electrophoresis for sample lanes 19-24where lane no 19 represents sample 105M, 19(105M), 20(105B), 21(125M), 22(125B), 23(274M) and 24(274B). Lanes 20, 21 and 22 showed positive amplification band of 393 bp corresponding to the expected band size for SHV gene in the *isolate* tested. Lane numbers 19, 23 and 24 showed no positive bands for SHV gene in the isolate tested.

Figure no 3: shows SHV2 plasmid gene electrophoresis of samples lanes 19-24, standard molecular ladder and controls.
Lane M shows bands for 1Kb molecular ladder. Lanes + VE and – VE represent the positive and negative controls respectively.



Fgure no 4 shows mother-newborn plasmid CTX-M gene comparative electrophoresis for sample lanes 19-24, where lane no 19 represents sample 105M, 19(105M), 20(105B), 21(125M), 22(125B), 23(274M) and 24(274B).Lanes 21 and 22 showed positive amplification band of 585 bp corresponding to the expected band size for CTX-M gene in the isolates tested. Lane numbers 19,20, 23-25 showed no positive bands for CTX gene in the isolates tested.

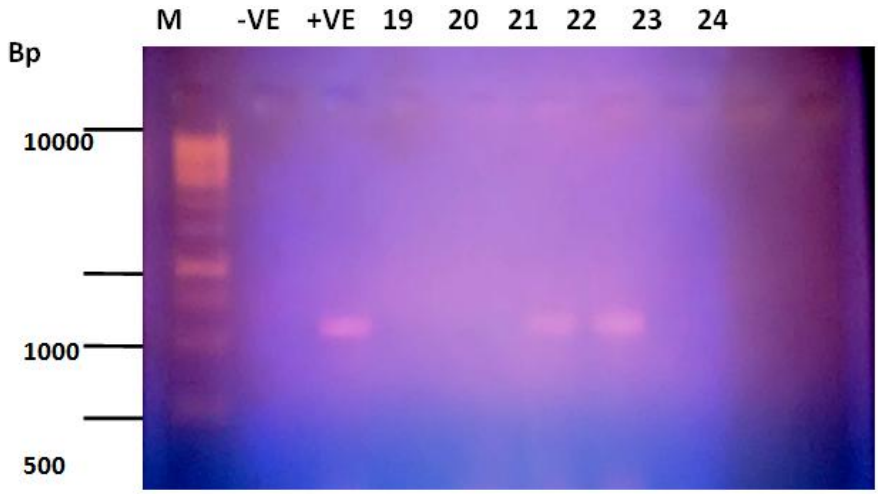


Figure no 4: shows CTX-M plasmid gene electrophoresis of samples lanes 19-24, standard molecular ladder and controls.

Lane M shows bands for 1Kb molecular ladder. Lanes + VE and ‾ VE represent the positive and negative controls respectively.

Figure no 5: shows mother-newborn plasmid OXA gene comparative electrophoresis for sample lanes 19-24, where lane no 19 represents sample 105M, 19(105M), 20(105B), 21(125M), 22(125B), 23(274M) and 24(274B). .lane numbers 21, 22 shows positive amplification band of 620 bp corresponding to the expected band size for OXA gene in the isolates tested. Lane numbers 19, 20, 23 - 25, showed no positive bands for OXA gene in the isolates tested.

Figure no 5:shows OXA plasmid gene electrophoresis of samples lanes 19-24, standard molecular ladder and controls.

Lane M shows bands for 1Kb molecular ladder. Lanes + VE and ‾ VE represent the positive and negative controls respectively.

Table no 4 **:**shows the Percentage mother to newborn transmission of blood borne infections within first 7 days of postnatal life by comparative analysis of the antibiograms and the plasmid DNA profiles of the Gram

negative maternal vagina colonizers in pregnancy and blood isolates from her respective newborn with sepsis in the first 7 days of postnatal life.

Three isolates (25%, n=12) demonstrated similarities in antibiogram and ESBL - DNA profile with the vagina isolates of their respective mothers representing 0.9% (n=350) mother to newborn transmission of blood borne infections in the first 7 days of postnatal life.Two of the 3 isolates were ESBL-positive thus translating to 0.6% (n=350)mother to newborn transmission of ESBL-positive blood borne infectionswithin the first 7 days of postnatal life. Pearson's regression analysis was not statistically significant to establish a correlation between maternal vagina colonization with ESBL- producing *Enterobacteriaceae* in pregnancy and ESBL- early onset neonatal sepsis (*p=0.861*).

Lab no.	Sample no	Isolates	Antibiogram	Plasmid DNA genes				Plasmid DNA profile	Remarks
				TEM	SHV	CTX-M	OXA		
1	5M	K. pneu.	At variance	+*	-	+	+	At variance	
2	5B	E.agglo		+	-	+	+		NVTI
3	347M	E. coli	At variance	-	-	+	+	At variance	
4	347B	E.coli		+	-	+	+		NVTI
5	279M	K. pneu.	At variance	+*	+	-	+	At variance	
6	279B	K. pneu.		+	+	+	+		NVTI
7	131M	K. pneu.	At variance	+	+	+	-	At variance	
8	131B	E. agglo		+*	+	+	-		NVTI
9	133M	E. coli	At variance	-	-	-	-	At variance	
10	133B	E. coli		+	-	+	+		NVTI
11	155M	K. pneu.		+	+	+	-		
12	155B	K. pneu.	Similar	+	+	+	-	Similar	VTI
13	236M	E. coli		+	+	+	+		
14	236B	E. coli	At variance	-	-	-	-	At variance	NVTI
15	333M	K.pneu.		+	+	+	+	Similar	
16	333B	K. pneu.	Similar	+	+	+	+		VTI
17	244M	E. coli		-	+	+	-		
18	244B	P. staurti	At variance	+	+	-	-	At variance	NVTI
19	105M	E. coli		-	-	-	-		
20	105B	P. aerugino.	At variance	+	-	-	-	At variance	NVTI
21	125M	K. pneu.		+	+	-	+		
22	125B	K. pneu.	At variance	+	+	+	+	At variance	NVTI
23	274M	E. coli		-	-	-	-		
24	274B	E. coli	Similar	-	-	-	-	Similar	VTI

Table4: Comparison of the antibiogram and plasmid DNA profile between maternal vaginal Gram negative colonizers in pregnancy and the Gram negative blood isolates of their respective newborn babies with sepsis.

KEY: M= respective mother, B= respective baby, Pos.= positive, Neg.= Negative, E. agglo.= *Enterobacter agglomerans*, P. aerugi= *Pseudomonas aeruginosa*, K. pneu = *Klebsiella pneumonia*, + = detected, - = not detected, +* = detected with a secondary band, MNT = Mother to newborn transmission, NMNT = Non mother to newborn transmission.

IV. Discussion

This study recorded a total vagina *Enterobacteriaceae* colonization of 243 (69.4%, n=350). This value is greater than 14% reported in Washington DC[22] but similar to the value of 65.3% reported in New Delhi, India.[23] These differences in findings might reflect individualized country's disposition to infection prevention and control and cultural differences to achieving vagina health in pregnancy. A maternal vagina ESBL-colonization rate of 16% (56/350) was reported in this study indicating a gradual introduction of community based ESBLs to our hospitals. This value is greater than the value of 5.4% reported in Argentina.[24] It is however similar to the figure of 15.3% reported in Central India[25] and42.5% by another study in India.[26] This study reported an ESBL prevalence of 19.3% amongst the babies with sepsis. This is higherthan the prevalence reported amongst their mothers suggesting acquisition of organisms from the hospital environment.

Malodourous vagina discharges in pregnancy was statistically significant (OR=6.24, p= 0.001) as an independent risk factors for vaginal ESBL- colonization. This finding is at variance with results of community based study in Israel that reported previous antibiotics use as an independent risk factor for colonization with ESBL-producing *Enterobacteriaceae.*[27] The reason for this variance could not be ascertained.This finding emphasized the need for a probe into whether there is a correlation between bacterial vaginosis and vaginal ESBL-colonization in pregnancy. Prompt management of bacterial vaginosis and other conditions presenting with malodourous vaginal discharges would hence, remarkably reduce the chances of vagina ESBL-colonization in pregnancy.

This study established a 0.6% (n=56) mother-to-newborn transmission of ESBL- positive blood borne infections during the first 7 days of postnatal life. This percentage transmission was not statistically significant to establish a correlation between maternal vagina colonization with ESBL-producing *Enterobacteriaceae* in pregnancy and ESBL-positive early onset neonatal sepsis, (*p=0.861*). There is however need for antenatal screening for vagina ESBL-colonization in endemic areas.

V. Conclusion

This study did not establish a correlation between maternal vagina colonization with ESBL-producing *Enterobacteriaceae* in pregnancy and ESBL-positive early onset neonatal sepsis to warrant antenatal screening. However, in ESBL endemic areas, antenatal screening for maternal vagina ESBL-colonization is recommended.

Conflict of interest: No conflict of interest about this work.

Contribution of authors

We declare that this study was done by the authors named in this article. All liabilities pertaining to the content will be borne by us. It was conceptualized and designed by Godwin I. Ogban, Onyinola O. Oduyebo and IretiolaB.Fajolu. Manuscript was written by Godwin I. Ogban and vetted by Onyinola O. Oduyebo, IretiolaB.Fajolu, Patrick O.Oshun, Thomas U. Agan,Ubleni E. Emanghe and Simon N. Ushie. Samples collection and processing were carried out by Godwin I. Ogban, Anthony A. Iwuafor and Simon N. Ushie.

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