COMMON BACTERIAL CAUSES OF NEONATAL SEPSIS AND ANTIMICROBIAL SUSCEPTIBILITY AT UNIVERSITY TEACHING HOSPITAL, LUSAKA, ZAMBIA.

BY

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DECLARATION

I hereby declare that this dissertation represents my own work not been presented either wholly or in part for the degree at the University of Zambia, School of Medicine or any other University.

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ABSTRACT

Background: Prescribing effective preventative antibiotics and treating infections in mothers, providing a clean birth environment, and delivering the baby within 24 hours of rupture of membranes, where possible, can all help lower the chance of neonatal sepsis. However, neonatal sepsis remains one of the leading causes of morbidity and mortality among neonates, responsible for 30- 50% of the total neonatal deaths in low-and middle-income countries. The incidence and mortality of neonatal sepsis in Zambia is quite high. In 2007, the neonatal mortality rate in Zambia was 38/1,000 live births. The present study sought to identify the common bacterial causes of neonatal sepsis and antimicrobial susceptibility at the University Teaching Hospital in Lusaka, Zambia.

Methods: This was a descriptive cross sectional study with convenience sampling for a period of three months of 142 neonates at the University Teaching Hospital (UTH) Paediatric Department. Data was obtained from mothers with neonates clinically diagnosed with sepsis at the UTH from October 2013 to January, 2014. Data was collected by reviewing obstetric and neonatal files while laboratory studies done were HIV rapid test (for HIV exposure), random blood sugar, and full blood count and blood culture to establish common blood pathogens. All tests were done after consent from the mothers or a guardian for blood to be collected from neonates was obtained. There was a questionnaire administered for each patient for a more detailed antenatal and delivery history of the neonate.

Results: Positive blood culture results were at 30% and most common bacteria isolated causing neonatal sepsis in A-block, UTH were gram positive *staphylococcus aureus* and *coagulase negative staphylococcus*, which together accounted to 85% of the positive blood culture tests. *Staphylococcus aureus* and *coagulase negative staphylococcus* were highly susceptible to Ciprofloxacin (100%), Chloramphenicol (84%), Cefotaxime (70%), Oxacillin (69%) and Gentamycin (70.6%). Penicillin (65%) showed high resistance to *staphylococcus aureus* and *coagulase negative staphylococcus*. The most common clinical presenting features were fever, irritability and poor feeding. Neonates born from the UTH had on average 4.5 increased odds for early onset sepsis compared to neonates born from other health centers (OR: 4.48, CI: 1.85 - 10.85, $p < 0.01$).

Conclusions: Gram positive *staphylococcus aureus* is the common cause neonatal sepsis in A-block UTH. The first line Penicillin treatment is highly resistance and careful consideration should be made by UTH management to introduce third generation cepholosporins and macrolides as first line to effectively improve treatment and prevent morbidity. Infection prevention in delivery and nursery wards should be held as highest priority to prevent loss of life**.**

Key words: Neonate, early onset sepsis, late onset sepsis, University Teaching Hospital (UTH)

DEDICATION

To my dear mum and dad, who have always believed in me and supported my career choices. Their love and support have kept me going.

To my loving husband and son the two Benjamins in my life, your patience and understanding of my absence has made this journey possible. Love you always.

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ABBREVIATIONS

AIDS	Acquired Immunodeficiency Syndrome
C/S	Caesarean Section
CRT	Capillary Refill Time
DHS	Demographic and Health Survey
DWC	Differential White Cell Count
EOS	Early Onset Neonatal Sepsis
ESR	Erythrocyte Sedimentation Rate
FBC	Full Blood Count
HIV	Human Immunodeficiency Virus
HR	Heart Rate
LOS	Late Onset Neonatal Sepsis
MOH	Ministry of Health
NICU	Neonatal Intensive Care Unit
PMTCT	Prevention of Mother to Child Transmission
PROM	Premature Rupture of Membranes
BREC	Biomedical Research Ethics Committee
ROM	Rupture of Membranes
SIRS	Systemic Inflammatory Response Syndrome
SVD	Spontaneous Vaginal Delivery
UNZA	University of Zambia
UTH	University Teaching Hospital
WBC	White Blood Cell
WHO	World Health Organisation

1.0 INTRODUCTION

Neonatal sepsis is a clinical syndrome characterised by the presence of generalised systemic inflammatory response to invading pathogens and is associated with growth of organisms from one or more sites (blood, cerebral spinal fluid, urine) and occurs in babies less than 28 days old (Edmond and Zaidi, 2010). The infections in neonates include sepsis, meningitis, pneumonia and deep seated infections. The neonates are susceptible to various infections due to the immature immune system and other factors such as maternal health and environment in which there are born in (Edmond and Zaidi, 2010).

Neonatal sepsis is a major cause of morbidity and mortality among the newborns in Zambia. Globally about 1.6 million neonates die from sepsis which accounts for 38% of deaths of children under five years old (WHO, 2006). The majority of children die in developing countries accounting for over 99% of deaths in the world and Zambia is no exception (Chomba et al, 2009 and Seale et al, 2009). The neonatal mortality rate is 38/1,000 live births in Zambia (DHS, 2007) and which is still very high. Sepsis is one of the top three causes death in neonates in the first 24hours coupled with birth asphyxia and prematurity/low birth weight (Chomba et al, 2009).

The Microbiology Department at the University Teaching Hospital in Zambia, periodically swabs the ward equipment in the nurseries, labour ward and theatres and found *Klebsiella* and *E.Coli* species on some equipment which was resistant to Penicillin and Gentamycin (Lukwesa, 2011).

Developed countries such as the United States of America, Australia and Britain have low incidence of neonatal sepsis as low as 1.5 to 3.5 per 1000 live births (Vergnano et al (2005).
Between the years 2009-2011 University Teaching Hospital data for admissions in Department of Paediatrics in-patient (A-block) ranged from 15,000- 17,500 children per year and 12-15% of these were neonates. The admissions due to neonatal sepsis were between 25-46% and neonatal mortality rate due to sepsis in A-block and neonatal intensive care unit (NICU) was 18.3% and 22-35% respectively.

The University Teaching Hospital is located in the capital city, Lusaka, Zambia and is the only tertiary hospital in Zambia catering for a population of two million in Lusaka province. Majority of newborns needing critical medical care in Lusaka are admitted at University Teaching Hospital in the Paediatric Department.

The pathogens causing early (<7days) and late (>7days) onset neonatal sepsis according to WHO sepsis criteria differ at national and global levels. In Asia early onset sepsis (EOS) is defined as sepsis within 48hours while late onset sepsis (LOS) is sepsis developing more than 48hours of delivery (Tiskumara et al, (2008). The neonatal mortality due to sepsis accounts for 47% in Zambia and high prevalence (14.3%) of HIV-AIDS has a massive effect on the newborn's outcome in cases of sepsis (Engmann et al, 2011 and MOH, 2009).

The ill neonates vary in clinical presentation with symptoms such as fever, poor feeding, irritability, abdominal distension and reduced activity, while signs range from pallor, jaundice, grunting respiration, tachypnoea, tachycardia, lethargy, hypothermia, seizures and unconsciousness. (Speck et al, 1986 and Kayange et al, 2010).

It is therefore important to carry out laboratory studies to establish the common blood pathogens and complete blood count investigations to associate the type of sepsis with presentation and outcome (Aletayeb et al, 2011). The laboratory studies help identify which infections are causing EOS and LOS with trends in Asia showing 10.3% positive cultures for EOS and 46.6% bacteria causing LOS (Tiskumara et al, (2008).

The last study on the common pathogens causing neonatal sepsis was last carried out about 14 years ago (1998/9) and no other study has been done to identify new pathogens and susceptibility of antimicrobials at UTH, Lusaka (Mulenga, 2000). The study was also done at the time when routine HIV testing was not done so some effects of HIV on outcome were not looked at. Studies have shown that it difficult to compare antibiotic resistance between countries but easier to compare antibiotic susceptibility over time in the same unit and department (Vergnano et al (2005).

1.1 STATEMENT OF THE PROBLEM

Neonatal sepsis remains one of the major disease conditions causing high numbers of morbidity and mortality in the developing nations (WHO, 2006). . Zambia is no exception with neonatal rate up to 40-50% being attributed to EOS sepsis (UTH, 2011).

In Zambia guidelines on appropriate clinical diagnosis and empirical antibiotic treatment given is followed though pathogens vary from different places. The laboratory services for identification of organisms causing sepsis are there at UTH but diagnosis of sepsis is usually made clinically because of lack of culture bottles, shortage of human resource and other challenges in resources in the laboratory. The antibiotic sensitivity and resistance patterns of microbes keep evolving making management difficult. Resource constraints in material used in isolation of bacteria, overcrowded, unsafe delivery rooms and lack of staff contribute to the poor outcome of sepsis. HIV infection has increased the burden of disease in the neonates and making the organisms becoming more atypical.

Hospitals in neighbouring countries and globally frequently carry out prevalence studies on common aetiological organisms and sensitivity patterns to insure the best possible care for the newborns (Kairavi et al, 2010 and Thaver et al, 2009).

1.2 RATIONALE OF STUDY

The identification of pathogens in patient's bodily fluids has great diagnostic and prognostic value. Frequent information on common microorganisms and the antimicrobial susceptibility patterns is essential in formulating treatment guidelines for patients.

Knowing the common type of bacteria gram positive or negative is essential in developing treatment protocols in hospital. Recently there has been changing trends of gram negative bacteria causing EOS (*E.coli and Klebsiella*) with gram positive bacteria causing LOS (*staphylococcus sp*) in most Asian hospitals and some hospitals in Latin America and Africa (Tiskumara et al, 2008).

The study will help identify bacterial organisms and resistance patterns to commonly used antibiotics at UTH in treatment of neonatal sepsis. The study will also help determine whether gram positive organisms are still the leading cause of neonatal sepsis at UTH. The study will help determine if neonatal sepsis is community acquired infection or nosocomial

infections. The study will also set a baseline to many surveillance studies that will follow in the department.

1.3 SIGNIFICANCE OF STUDY

The study will contribute to information often looked for in developing countries on which antibiotic to use with emerging resistant strains (Thaver et al, 2009). And evidence shows that there is no data in Zambia (Vergnano et al, (2005).

Findings of the study will help improve the prevention and management of sepsis in the neonates using newer, sensitive and less resistance antibiotics at UTH.

1.4 RESEARCH QUESTIONS

1. What are the common bacteria that cause neonatal sepsis at UTH and do the same bacteria cause early and late onset neonatal sepsis?
2. What is the sensitivity patterns of first line antibiotics and other antibiotics used at UTH in treatment of neonatal sepsis?
3. What clinical, laboratory, obstetrics (maternal fever, PROM, HIV infection, mode of delivery, place of delivery) and neonatal (HIV exposure) features that increase the risk of neonatal sepsis?

2.0 STUDY AIMS

The primary aim of this study was to determine the common bacterial organisms, clinical features and antimicrobial susceptibility of neonatal sepsis at the University Teaching Hospital (UTH) in Lusaka, Zambia.

2.1 SPECIFIC AIMS

1. Investigate the common bacteria that cause early and late onset neonatal sepsis at the UTH.
2. Estimate the sensitivity and resistance patterns to first line antibiotics and other antibiotics used at the UTH in treatment of neonatal sepsis.
3. Describe the clinical, laboratory, obstetrics (maternal fever, PROM, HIV infection, mode of delivery, place of delivery) and neonatal (HIV exposure) features that increase the risk of neonatal sepsis.

2.2 STUDY HYPOTHESIS

There is no difference between bacterial organisms causing early and late onset neonatal sepsis and the antimicrobial susceptibility is the same in both types of organisms.

3.0 LITERATURE REVIEW

Sepsis is the presence of viable bacteria in the blood stream (McKenzie and Furr, 2001). World Health Organisation (WHO) formulated criteria for initial diagnosis of neonatal sepsis which has made early diagnosis faster but it varies in babies as sepsis has no pathognomonic features (Kayange et al 2010).

When there is hypotension, hypo-perfusion (delayed CRT) and organ dysfunction then there is severe sepsis. This may be reversed or may progress to septic shock with persistent hypotension and hypo-perfusion regardless of resuscitative measures. Though the definitions of severe sepsis and septic shock have been revised by the International Paediatric Sepsis Consensus Conference (IPSCC) they help in differentiating probable sepsis with those who have definite sepsis (Goldstein et al, 2005 and Maramba-Lazarte et al, 2011).

Systemic inflammatory response syndrome (SIRS) criteria is currently being used to define septicaemia as presence of at least two or more of the following of which one should be abnormal temperature or white blood cell count (WBC):

1. Hyperthermia (>38.5 degrees C) or Hypothermia (<36 degrees C)
2. Tachycardia (HR>165) or Bradycardia (HR <90) at rest
3. Tachypnoea (respiratory rate more than 60)
4. Elevated or depressed white cell count (<4,000 or > 12,000) or more than 10% immature neutrophils (Goldstein et al, 2005 and Maramba-lazarte et al, 2011).

SIRS is the end stage of inflammatory response which is started by the physiological response to tissue injury by pathogens. The inflammatory response to microbes by the body is through activation of the immune and complements systems from the blood. The phagocytes, neutrophils and monocytes, injured cells release inflammatory mediators (histamine) and eicosanoids, cytokines and tumour necrosis factor at the site of damage. The Gram positive and negative bacteria initiate different defence mechanisms in the body there by having different clinical presentation and outcome of sepsis (Aletayeb et al, 2011).

Based on the timing of the infection neonatal sepsis has been classified into early-onset sepsis (EOS) and late-onset sepsis (LOS). This classification helps to guide antibiotic therapy as it implies differences in the presumed mode of transmission and predominant organisms. EOS is defined as onset of sepsis in the first seven days and is mostly the result of vertical

transmission of bacteria from mothers to infants during the intrapartum period. LOS is defined as infection occurring after 1 week of life is attributed to the horizontal transmission of pathogens acquired postnatally and is often more insidious in onset (WHO, 2011). One investigative group classified neonatal sepsis into early-onset (≤4 days), late-onset (5–30 days), and late, late-onset (>30 days) according to the infant's age when positive blood culture obtained. Very low birth weight (VLBW) preterm infants are at particularly high risk for both EOS and LOS in part because of immaturity of the immune system, prolonged mechanical ventilation, prolonged hospitalization, use of indwelling catheters, endotracheal tubes, and other invasive procedures. (Published online 2013 Nov 1.doi 10. 4161/viru. 26906) At UTH, EOS is defined as sepsis in the first week of life and LOS as infection obtained after one week of life of a baby, this is according to treatment guideline protocol at neonatal intensive care unit (NICU/UTH, 2011). Most of the symptoms and signs for EOS appear in the first 48 hours and organisms are mainly gram positive bacteria such as *staphylococcus aureus, streptococcus, E.coli* as also indicated in study by Freeman and others. (Freeman et al, 1981).

The increasing global incidence of neonatal sepsis in developing countries has been attributed to resistant strains of bacteria emerging against the empirical WHO first line antibiotics (Vergnano et al, 2005 and Thaver et al, 2009). The incidence of neonatal sepsis still remains high in Asia and Africa with reports indicating 7.1 to 38 per 1000 live births and 6.3 to 23 per 1000 live births respectively (Vergnano et al, (2005).

A study done at UTH in Zambia in 1997-1998 showed that 32.6% of positive blood cultures and gram positive bacteria 47% (*Staphylococcus* species) being the most prevalent for early onset sepsis and gram negative bacteria 53.3% (*Klebsiella pneumoniae* and *E.coli*) responsible for late onset sepsis (Mulenga, 2000). Overall gram positive bacteria caused neonatal species with all species resistant to penicillin and gentamycin but sensitive to Cefotaxime (Mulenga, 2000).

Information obtained from the Microbiology laboratory shows 12.3% blood culture positive for neonates in NICU for a period of 6 months and antibiotic resistance of more than 90% to UTH first line penicillin and gentamycin (Lukwesa, 2011).

In recent studies done in Tanzania and Nigeria positive blood culture in neonates yielded 47.1% and 54.1% respectively with gram negative *Klebsiella* species and *E.coli* species commonly isolated with high resistance to third generation cephalosporin and penicillins (Kayange et al, 2011 and Bode- Thomas et al, 2004).

The same trend of gram negative bacteria as dominating pathogens causing sepsis in neonates was noted in Iran with 10.4% positive blood and Philippines with 50% positive blood cultures with *Enterobacter , Klebsiella* and *E.coli* the main microbial agents (Karambin et al, 2010 and Maramba-lazarte et al, 2011). India and Pakistan have shown 80% gram negative bacteria (*Klebsiella* species) emerging as neonatal sepsis causing morbidity and mortality with high resistance to penicillins, third generation cephalosporin and carbapenems (Mane et al, 2010 and Waseem et al, 2005).

A survey done in Asia neonatology units showed 89% sepsis caused by multi resistant gram negative bacilli with major resistant to ampicillin and third generation cepholosporins (Isaacs, 2005). In the same study there is evidence showing gram positive bacteria encoding vancomycin resistant *enterococci* and methicillin- resistant *Staphylococcus aureus* (MRSA) narrowing the range of broad spectrum antibiotics to use (Isaacs, (2005).

Host susceptibility, socioeconomic factors, obstetric and nursery practices, and health and nutrition of the mother are all important in the pathogenesis of neonatal sepsis. Infants who develop particularly EOS usually have a history of risk factors associated with the pregnancy and delivery. Maternal Risk Factors: Although the reason is not well defined, the rates of early-onset GBS infection are higher among blacks than in other racial groups.

The rates of prematurity and LBW, which both predispose to neonatal infection, are inversely related to socioeconomic status. Asymptomatic bacteriuria has been associated with premature birth. Other risk factors include premature birth, LBW, premature rupture of membranes (PROM), prolonged time of rupture of membranes (ROM), maternal peripartum infection, and septic or traumatic delivery. Uncomplicated ROM lasting longer than 24 hours has been associated with a 1% incidence of neonatal sepsis above the baseline rate of 0.1% to 0.5%. The risk of infection increases four- fold if chorioamnionitis and prolonged ROM coexist. Newborns in developing countries are also exposed to external risk factors that put

them at a greater risk for infections compared with neonates in the industrialized countries. (The Turkish Journal of Pediatrics 2012.54:449-457).

Late-onset sepsis is associated with the following risk factor; prematurity, central venous catheterization (duration >10 days), nasal cannula or continuous positive airway pressure (CPAP) use, H_2 -receptor blocker or proton pump inhibitor (PPI) use and GI tract pathology. Neonates born by vaginal and instrumental delivery also have a high risk of infections and those born to mothers with hemorrhage, maternal anaemia and peripartum fever (Davies, 1971).

In sub-Sahara Africa, immune-suppression due to HIV-AIDS complicates the presentation of neonatal sepsis with high mortality attributed to co-infections of opportunistic infections such as Candida, Cytomegalovirus, herpes simplex and toxoplasmosis. In the study done by Mulenga in 1997-1998 babies with HIV exposure had poor outcome with sepsis complicating to meningitis (Mulenga, 2000).

The only intervention proven to decrease the incidence of early-onset neonatal sepsis is maternal treatment with intrapartum intravenous antimicrobial agents for the prevention of GBS infections. Adequate prophylaxis is defined as penicillin (the preferred agent), ampicillin, or cefazolin given for ≥4 hours before delivery. Erythromycin is no longer recommended for prophylaxis because of high resistance rates. In patients who have a non-serious penicillin allergy, cefazolin is the drug of choice. For patients with a history of serious penicillin allergy (anaphylaxis, angioedema, respiratory compromise, or urticarial rash), clindamycin is an acceptable alternative agent, but only if the woman's rectovaginal GBS screening isolate has been tested and documented to be susceptible. If the clindamycin susceptibility is unknown or the *Group B Streptococcus* isolate is resistant to clindamycin, vancomycin is an alternative agent for prophylaxis. However, neither clindamycin nor vancomycin has been evaluated for efficacy in preventing early-onset *Group B Streptococcus* sepsis in neonates (American Academy of Pediatrics May 2021, Vol. 129/ Issue 5).

With early diagnosis and treatment, term infants are not likely to experience long-term health problems associated with neonatal sepsis; however, if early signs or risk factors are missed, mortality increases. Residual neurologic damage occurs in 15-30% of neonates with septic meningitis.

Mortality from neonatal sepsis may be as high as 50% for infants who are not treated. Infection is a major cause of fatality during the first month of life, contributing to 13-15% of all neonatal deaths. Low birth weight and gram-negative infection are associated with adverse outcomes.

Neonatal meningitis occurs in 2-4 cases per 10,000 live births and contributes significantly to mortality from neonatal sepsis; it is responsible for 4% of all neonatal deaths. In preterm infants who have had sepsis, impaired neurodevelopment is a concern. Proinflammatory molecules may negatively affect brain development in this patient population.

In a large study of about 6000 premature infants who weighed less than 1000 g at birth, preterm infants with sepsis who did not have meningitis had higher rates of cognitive deficits, cerebral palsy, and other neurodevelopmental disabilities than infants who did not have sepsis (Anderson-Berry A.L. 2015).

4.0 METHODOLOGY

4.1 STUDY DESIGN

This was descriptive cross sectional study conducted over a period of three months i.e. from October 2013 to January 2014.

4.2 STUDY VENUE

The study was conducted in the Department of Paediatrics and Child Health at UTH, emergency ward AO1 and admission wards, Lusaka, Zambia.

4.3 STUDY POPULATION

All neonates presenting with features of sepsis at UTH were targeted for the study. Sample size of 142 neonates was included in the study upon meeting the inclusion criteria. All statistical calculations were done with help and guidance of a statistician

4.4 SAMPLE SIZE

The sample size was obtained using Open Epi version 2 for frequency in a population at 95% confidence level. The sample population was every neonate meeting the inclusion criteria.

The sample size was derived for frequency in a population which was two hundred and thirty eight and the hypothesized percentage frequency of outcome factor was 35% +/- 5 in the population. The confidence limits as percentage of hundred was 5%. Therefore the sample size at 95% confidence level was 142.

4.5 INCLUSION AND EXCLUSION CRITERIA

4.5.1 Inclusion Criteria

1. Neonates from birth to 28 days of age who presented with symptoms of sepsis including fever, pallor, tachypnoea, tachycardia, abnormal skin colour (cyanosis, rash, jaundice), poor feeding, abdominal distension, lethargy, hypotonia, delayed CRT and convulsions
2. Term (>37weeks) and preterm <37weeks

4.5.2 Exclusion Criteria

1. Neonates who have received more than 48hrs of antibiotics
2. Newborns with multiple congenital abnormalities and needed multi-disciplinary approach to care.
3. Babies whose parents did not consent.

4.5.3 Case Definitions

Early onset sepsis: neonatal sepsis within seven days after birth.

Late onset sepsis: neonatal sepsis after seven days after birth up to 30 days of life**.**

4.6 STUDY PROCEDURES

Babies brought in the emergency ward and are less than one month, who presented with features of sepsis were identified. The history was obtained from mother or guardian and physical examination was done by principal investigating doctor and if they meet the inclusion criteria they were selected for study. A structured questionnaire was administered after informed consent was signed and procedures for sepsis work up were explained.

The blood was collected from the index finger after swabbing with methylated spirit (70% ethanol) and a drop of blood put on a rapid HIV Abbot Determine strip after a pre-test counselling was done. Then random blood sugar test was done by bedside using Accu Check glucometer and strips with blood from the same finger. If a newborn was found to be hypoglycaemic they were immediately treated with a bolus of 10% dextrose fluid. The results for HIV test were communicated to the mother or guardian after post-test counselling and appropriate action taken according to the results.

The sepsis work up for this study included collection of 2ml of blood for full blood count (FBC), differential white cell count (DWC), Erythrocyte sedimentation rate (ESR), and put in an EDTA bottle, 1-2ml of blood put in a blood culture bottle (BD BACTEC Paeds) for microscopy, culture and sensitivity.

4.7 COLLECTION OF BLOOD

The collection of blood was done in aseptic conditions to avoid blood contaminants. The investigator washed their hands thoroughly with soap and water. Then using sterile gloves cleaned the area of the femoral vein in inguinal region with iodine solution and methylated

spirit. Then the venepuncture was made with a sterile needle and minimum 3ml of blood was collected in sterile syringe and the needle was changed upon depositing 1ml of the blood in BD BACTEC Paeds bottle for blood culture and 2ml in the EDTA blood for FBC/DWC/ESR.

4.8 LABORATORY STUDIES

The samples were then taken to the laboratory within two hours of collection. The laboratory tests were done at UTH microbiology and haematology laboratories which are under routine laboratory tests. The BD BACTEC Paeds bottle has high recovery rate of organisms. The sample was then inoculated on sheep blood agar, chocolate agar and MacConkey agar then incubated at 35-37 degrees Celsius. The incubation takes up to 10 days for some organisms to grow but the fast growing bacteria alarm in the first 48 hours. The microorganisms isolated from positive culture were identified using Standard Biochemical Techniques.

The susceptibility of the organisms to antibiotics was done using a disc with different antibiotics tested especially the antibiotics used in routine treatment of sepsis at UTH.

The data collected was coded with serial numbers and entered on a spread sheet for analysis.

4.9 STUDY VARIABLES

The following variables were considered for this study:

Dependent variables: Blood culture results, isolated bacteria and antibiotic sensitivity patterns. Neonates with early and late onset sepsis.

Independent variables: HIV exposure of neonate, age and sex of neonate, birth weight, gestational age, place of delivery (name of clinic, hospital, home), maternal age and parity, mode of delivery (SVD, C/S, instrumental), HIV status of mother, duration of PROM, maternal PMTC measures, clinical signs and symptoms (fever, difficulty in breathing, tachypnoea, poor feeding, vomiting, lethargy, seizures etc.).

4.10 STATISTICAL ANALYSIS

All statistical tests were at 5% significance level. T-test was used to compare mean values between groups and the Chi-squared test or Fisher’s exact test was used for comparison of

proportions between groups. The relationship between study variables and positive blood culture results was examined using logistic regression. The selection for entry into the logistic regression model was considered at level $p<0.21$ or known clinical significance.

For the final multivariate logistic regression model, significant patient level factors of mother HIV status, place of birth, referring clinic, and neonate HIV status were fitted into the model. The p-value, odds ratio, and 95% confidence interval were reported. Both bivariate and multivariate analysis odds ratios with their corresponding 95% CI were calculated.

Statistical analyses were performed using SAS version 9.3, and for some preferred descriptive graphs using IBM SPSS Statistics version 21.0

5.0 ETHICAL APPROVAL

Ethical approval for the study was obtained from the University of Zambia Biomedical Research Ethics Committee (UNZABREC) and permission for the study was granted by the UTH management. All the procedures were done privately and results of the test were kept confidential and communicated to the mother or guardian.

Informed consent was obtained for all study participants and no treatment was denied for any patient presenting with sepsis. In any patient whose guardian denied consent to the study and the baby received treatment as per hospital standard of care and management.

All research results and study participants were given a serial number to avoid breaching confidentiality. Only the research team had access to the study data.

6.0 RESULTS

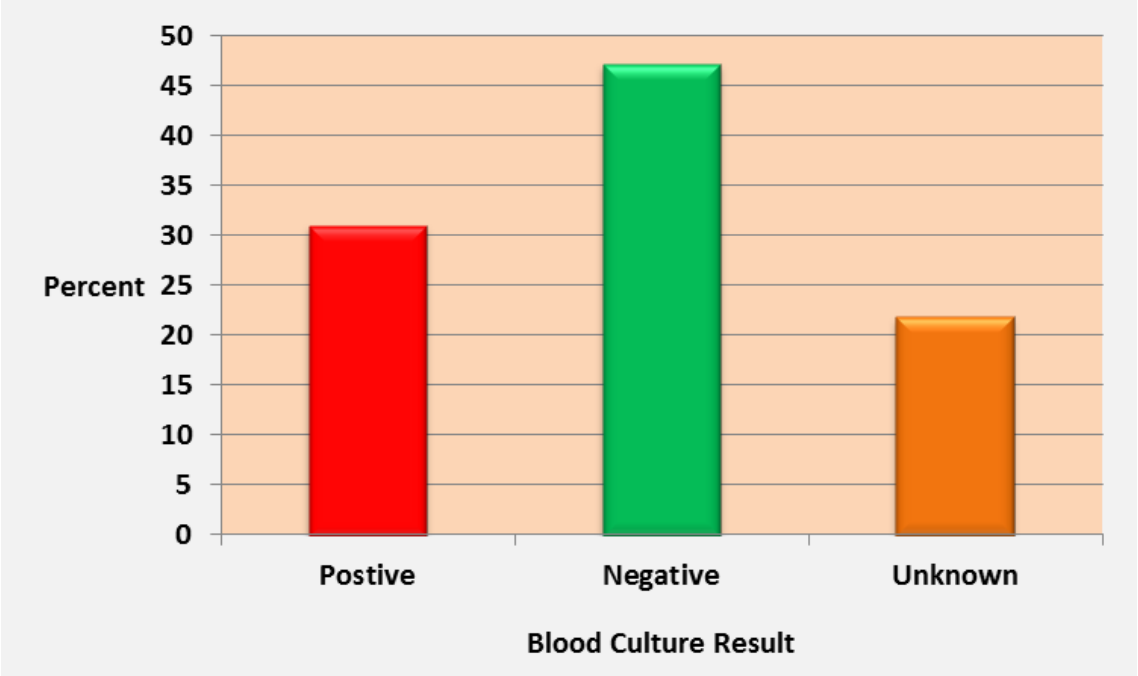
6.1 NEONATAL RESULTS

A total of one hundred and forty two neonates were recruited in the study. Seventy one were female and seventy one males giving an equal proportion of babies seen. From the babies seen the mean age was four days with about fifty percent of the neonates presenting with symptoms at the age of less than three days. The earliest a neonate presented is two hours with those that presented within 24 hours being in the majority.

The gestational age of the neonates ranged from 30 weeks to 42 weeks and the mean gestational age was 38 weeks. The birth weight was normally distributed with the mean weight being 2.9 kilograms.

The blood culture tests were positive in 31% of the cases and most common admission diagnosis was early onset sepsis (EOS) at 69% as shown in figure (1). The negative test results were found in 47% of the laboratory results and 22% of the results were pending or missing. The two most common culture organisms were Staphylococcus Aureus (61.4%) and Coagulase Negative Staphylococcus (25%) as shown in table 1.

Figure 1: Neonatal Blood Culture Results



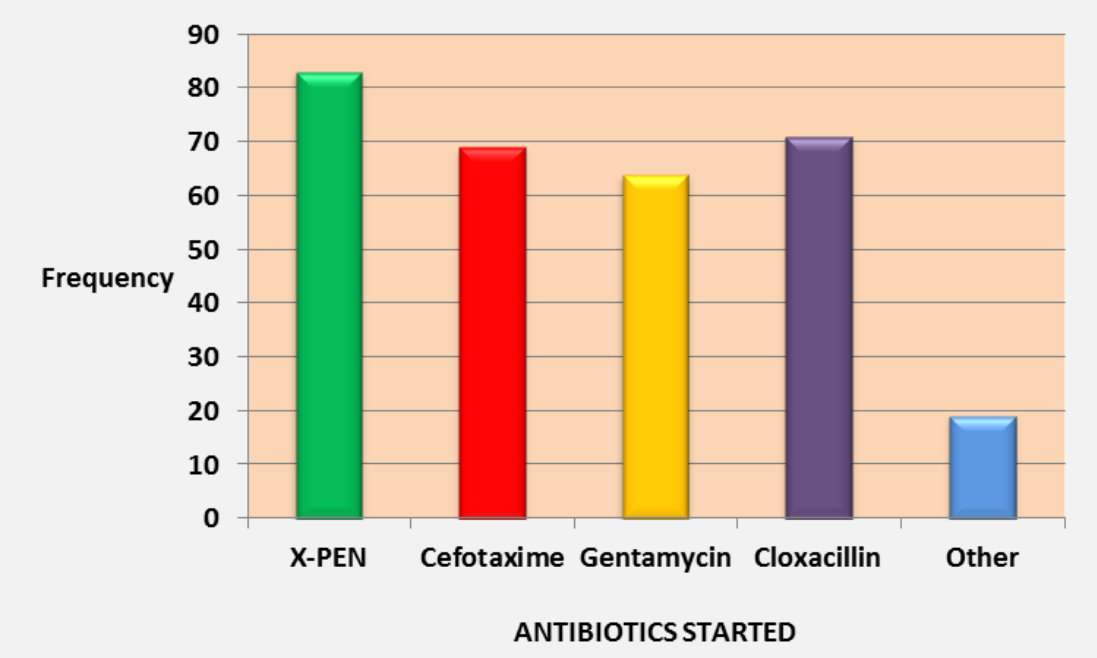
Majorities (77.3%) of isolates were from babies with EOS and 22.7% of isolates were from babies with LOS. The neonatal HIV exposure showed that 14% of babies were born to HIV positive mothers while 80% were born to HIV negative mothers. The remaining 6% either refused consent and HIV status remained unknown. These HIV tests were done as bedside rapid tests and the positive tests were later taken for DNA PCR confirmation tests of HIV infection at the laboratory and prevention of mother to child transmission (PMTCT) was started on the HIV exposed neonates as part of the treatment protocol.

Table 1: Organisms Isolated From Positive Blood Cultures

Organism	Frequency	Percent
STAPHYLOCOCCUS AUREUS	26	61.4
COAGULASE NEGATIVE STAPHYLOCOCCUS	11	25.0
ESCHERICHIA COLI	2	4.5
KLEBSIELLA SPECIES	2	4.5
CITROBACTER DIVERSUS	1	2.3
ENTERBACTER	1	2.3
Total	**44**	**100.0**

The majority of the organisms causing EOS and LOS were gram positive bacteria at UTH.
The most common presenting features upon admission were fever (94%), irritability (47%), and failure/poor feeding (45%). Additionally, the most frequent treatment prescribed at clinics was penicillin (17%) and paracetamol (13%). Over 50% of neonates did not receive any treatment for sepsis at the primary health care centre. At the UTH, most neonates were started on Penicillin (58%), Cloxacillin (50%), Cefotaxime (48%), and Gentamycin (45%). The tables 2 of summary of neonatal characteristics show most common presenting features on admission, treatment commonly given at clinics, other diagnoses alongside sepsis on admission, and antibiotics started at the UTH. Jaundice and being HIV exposed was most commonly diagnosed with neonatal sepsis.

Figure 2: Antibiotics Started at UTH



The mean axillary body temperature on admission was 38.6 +/- 1.39 SD degrees Celsius and the temperature was taken using a standardized thermometer. Majority of neonates had normal blood sugar at admission ranging from 4.75 +/- 2.50 with about 15% presenting with hypoglycemia. Haemoglobin levels were normally distributed with mean 15.87+/-2.67 and about 10% were found to be anaemic. The neutrophil count from the DWC showed that less than 5% had neutropenia and about 10% showed neutrophilia. The FBC showed that the white cell count showed about 6% leucopenia and 30% showed leukocytosis on admission, these features are also indicative of neonatal sepsis.

Table 2: Summary of Neonatal Characteristics

Variable Descriptive Category	Frequency (N = 142)	%
Child Age Group		
≤ 3 days	70	49.3
4 – 7	28	19.7
≥ 8	44	31.0
Mean, Median, Std. Deviation	6.4, 4, 5.93	
Range (Min, Max)	26 (1, 27)	
Sex		
Female	71	50.0
Male	71	50.0
Gestation Age (Weeks)		
N	137	
Mean, Median, Std. Deviation	38.0, 39, 2.50	
Range (Min, Max)	12 (30, 42)	
Birth Weight (Kg)		
N	141	
Mean, Median, Std. Deviation	2.9, 3, 0.54	
Range (Min, Max)	2.9 (1.2, 4.1)	
Time Onset of Illness (Hours)		
N	135	
Mean, Median, Std. Deviation	19.0, 13, 18.83	
Range (Min, Max)	94 (2,96)	
Admission Diagnosis		
Early Onset Sepsis	98	69.0
Late Onset Sepsis	44	31.0
HIV Rapid Test		
Negative	114	80.3
Positive	24	16.9
Missing	4	2.8
Blood Culture Results		
Positive	44	31.0
Negative	67	47.2
Missing	31	21.8
Culture Organism		
Coagulase Negative Staphylococcus	11	25.0
Staphylococcus Aureus	27	60.1
Other	7	15.9

Variable	Descriptive Category	Frequency (N = 142)	%
Presenting Features			
Failure/Poor Feeding		64	45.1
High Temperature		133	93.7
Irritability		67	47.2
Tachynoeaic		25	17.6
Difficulty Breathing		34	23.9
Chest In drawing		13	9.2
Convulsions		7	4.9
Lethargy		18	12.7
Umbilical Discharge		16	11.3
Jaundice		24	16.9
Pallor		6	4.2
Cyanosis		11	7.7
Abdominal Distension		6	4.2
Other Symptoms		40	28.2
Treatment Given at Clinic			
X-Pen		24	16.9
Gentamycin		14	9.9
Paracetamol		18	12.7
Other		7	4.9
None		79	55.6
Antibiotics Started at UTH			
X-PEN		83	58.5
Cefotaxime		69	48.6
Gentamycin		64	45.1
Cloxacillin		71	50.0
Other		19	13.4
Temperature on Admission			
N		135	
Mean, Median, Std. Deviation		38.6, 38.7, 1.39	
Range (Min, Max)		9.6 (32.4, 42)	
Random Blood Sugar			
N		133	
Mean, Median, Std. Deviation		4.75, 4.3, 2.50	
Range (Min, Max)		19.1 (0.9, 20)	
Variable	Descriptive Category	Frequency (N = 142)	%
HGB			
N		124	
Mean, Median, Std. Deviation		15.87, 15.85, 2.67	
Range (Min, Max)		16 (8.4, 24.4)	
PLT			
N		124	
Mean, Median, Std. Deviation		298.88, 293.5, 142.16	
Range (Min, Max)		764 (16, 779)	

WBC	
N	124
Mean, Median, Std. Deviation	13.19, 11.65, 6.42
Range (Min, Max)	46.6 (0.1, 46.7)
NEU %	
N	81
Mean, Median, Std. Deviation	34.01, 34, 12.79
Range (Min, Max)	64 (9, 73)
LYM %	
N	123
Mean, Median, Std. Deviation	37.12, 36.8, 10.80
Range (Min, Max)	55.8 (12.5, 68.3)

6.2 MATERNAL CHARACTERISTICS

From the 142 women interviewed whose babies were enrolled in the study, the mean age of the mothers at delivery was 24.8 years ± 5.7 SD. About 60% of the mothers were aged between 20 – 29 years.

A majority (89%) of the mothers were married as per their self-belief and report no action was taken to proof marital status. And 34.5% were prim gravidas. About 70% of the mothers tested negative for Syphilis, less than 3% had tested positive and the rest had no test done during antenatal. And approximately 15% were seropositive for HIV. The majority of the mothers delivered their babies through normal vaginal delivery (97%) and about 91% of the mothers gave birth at a health centre rather than at home. Of the delivery health centres, the UTH was the most common health centre that most mothers delivered from (27%), UTH being a tertiary hospital for complicated deliveries and for mothers with high risk pregnancies. Kanyama clinic was the second common at 17% as a delivery site.

The clinics with most referrals to the UTH were Kanyama, Chipata, and Chawama. The residential areas for the referred mothers were Kanyama and John Laing compounds these are densely populated areas with low socioeconomic status.

The median time of rupture of membranes prior to onset of labour was 30 minutes. Rupture of membranes occurred within 1 hour in about 65% of the mothers, and more than 1 hour to 24 hours in 30% of the mothers. Preterm premature rupture of membranes (PPROM) occurred in about 9% of the mothers and 23% of the mothers had premature rupture of membranes (PROM).

Table 3: Summary of Maternal Characteristics

Variable	Descriptive Category	Frequency (N = 142)	%
Mother Age Group (Years)			
< 20		25	17.6
20 - 29		87	61.3
30 - 39		29	20.4
40+		1	0.7
Mean, Median, Std. Deviation		24.8, 24, 5.70	
Range (Min, Max)		26 (15, 41)	
Marital Status			
Married		127	89.4
Unmarried		15	10.6
Community			
Kanyama		16	11.3
John Laing		13	9.2
Other		113	79.6
RPR			
Positive		3	2.1
Negative		101	71.1
Unknown		38	26.8
HIV Status			
Positive		21	14.8
Negative		121	85.2
Variable	Descriptive Category	Frequency (N = 142)	%
Parity			
One		49	34.5
Two		37	26.1
Three		26	18.3
Four		18	12.7
Five		9	6.3
Six		2	1.4
Seven		1	0.7
Mean, Median, Std. Deviation		2.37, 2, 1.37	
Range (Min, Max)		6 (1, 7)	

Mode of Delivery		
Spontaneous Vaginal Delivery	138	97.2
Caesarian Section	4	2.8
Duration of Rupture of Membranes (hours)		
≤ 1 hour	92	64.8
More than 1 - 24 hours	43	30.3
More than 24 hours	4	2.8
Unknown	3	2.1
Mean, Median, Std. Deviation	12.3, 0.5, 68.58	
Range (Min, Max)	719 (0.017, 720)	
PROM		
< 37 weeks	13	9.2
≥ 37 weeks	33	23.2
Place of Delivery		
Health Centre	129	90.8
Home	13	9.2
Referring Clinic		
Kanyama	34	23.9
Chipata	19	13.4
Chawama	18	12.7
Other	71	50.0

It was observed that out of 142 cases of neonatal sepsis, 46 (32%) had PROM and 68% developed sepsis in the absence of PROM. Of the PROM cases, 12 (26%) had positive blood culture test results of which 6 (50%) was *staphylococcus aureus* organism.

6.3 DISTRIBUTION OF COMMON BACTERIA CAUSING NEONATAL SEPSIS

It was noted that *Staphylococcus Aureus* was frequently isolated in both EOS and LOS as shown the table below.

Table 4: Distribution of Bacteria Causing Early and Late Onset Neonatal Sepsis

Organism	EOS (%)	LOS (%)
STAPHYLOCOCCUS AUREUS	18 (40.9)	9 (20.5)
COAGULASE NEG STAPHYLOCOCCUS	10 (22.7)	1 (2.3)
ESCHERICHIA COLI	2 (4.5)	0 (0)
KLEBSIELLA SPECIES	2 (4.5)	0 (0)
CITROBACTER DIVERSUS	1 (2.3)	0 (0)
ENTERBACTER	1 (2.3)	0 (0)
Total	34 (77.3)	10 (22.7)

6.4 ORGANISM ISOLATED AND THEIR SUSCEPTIBILITY TO ANTIBIOTICS

All the isolated organisms were studied for their susceptibility to antibiotics and are presented in Table 6.3.2. The two most common organisms of *Staphylococcus Aureus* and *Coagulase Negative Staphylococcus* were highly susceptible to Ciprofloxacin and Chloramphenicol. Gentamycin, Oxacillin, and Cefotaxime also showed fairly good activity against staphylococcus aureus.

Table 5: Susceptibility Patterns of Bacteria Isolated From Neonatal Sepsis

	CAUSATIVE BACTERIA (%)					
Antibiotic	*Staphylococcus Aureus*	*Coagulase Negative Staphylococcus*	*Escherichia Coli*	*Klebsiella Species*	*Citrobacter Diversus*	*Enterbacter*
Chloramphenicol	21 (84)	7 (87.5)	2 (100)	1 (100)	-	1 (100)
Cefotaxime	16 (69.6)	8 (72.7)	2 (100)	2 (100)	1 (100)	1 (100)
Ciprofloxacin	14 (100)	8 (100)	2 (100)	2 (100)	-	1 (100)
Oxacillin	18 (69.2)	5 (45.5)	-	-	-	0 (0)
Gentamycin	12 (70.6)	5 (71.4)	2 (100)	1 (100)	1 (100)	-
Erythromycin	14 (66.7)	5 (62.5)	-	-	-	0 (0)
Cotrimoxazole	0 (0)	0 (0)	0 (0)	0 (0)	-	-
Tetracycline	5 (71.4)	2 (100)	-	1 (100)	-	-
Penicillin	5 (35.7)	2 (33.3)	-	-	-	-
Vancomycin	3 (100)	1 (100)	-	-	-	-
Imipenem	1 (100)	-	-	-	-	-

6.5 ORGANISMS ISOLATED AND ANTIBIOTIC RESISTANCE PATTERNS

The two most common bacteria *Staphylococcus Aureus* and *Coagulase Negative Staphylococcus* showed high resistance to Cotrimoxazole and Penicillin Table 5 shows the resistance patterns of the isolated organisms from neonatal sepsis.

6.6 DISTRIBUTION OF PATIENT ATTRIBUTES IN ASSOCIATION WITH EOS AND LOS IN POSITIVE BLOOD CULTURES

A bivariate examination of the association of various mother and neonate attributes, and EOS and LOS is presented in Table 6.5.1.

There was significant association between mother HIV status (neonatal HIV exposure) and EOS and LOS ($p < 0.01$).

Place of birth was highly associated with EOS and LOS occurrence ($p < 0.01$).

Neonatal HIV exposure was highly associated with EOS and LOS occurrence ($p = 0.01$).

Table 6: Mother and Neonatal factors Associated with Neonatal Sepsis

Variable	Early Onset Sepsis		Late Onset Sepsis		P-value
	N	%	N	%	
Sex					
Male	49	50.0	22	50.0	0.99
Female	49	50.0	22	50.0	
Total	98	100.0	44	100.0	
Marital Status					
Married	89	90.8	38	86.4	0.56
Single	9	9.2	6	13.6	
Total	98	100.0	44	100.0	
Mother HIV Status					
Positive	9	9.2	32	72.7	< 0.01
Negative	89	90.8	12	27.3	
Total	98	100.0	44	100.0	
Mother Age					
< 20 years	18	18.4	7	15.9	0.74
20 -29 years	61	62.2	26	59.1	
30+ years	19	19.4	11	25.0	
Total	98	100.0	44	100.0	

Parity					
one	33	33.7	16	36.4	0.99
two	26	26.5	11	25.0	
three	18	18.4	8	18.2	
four or more	21	21.4	9	20.5	
Total	98	100.0	44	100.0	
Duration of Rupture of Membranes					
< 1 hour	64	66.7	28	65.1	0.86
1 hour or more	32	33.3	15	34.9	
Total	96	100.0	43	100.0	
Place of Birth					
Other	79	80.6	25	56.8	< 0.01
UTH	19	19.4	19	43.2	
Total	98	100.0	44	100.0	

Table 6: (Continued) Factors Associated with Neonatal Sepsis

Variable	Early Onset Sepsis		Late Onset Sepsis		P-value
Referring Clinic					
Kanyama	25	25.5	9	20.5	0.21
Chipata	10	10.2	9	20.5	
Chawama	15	15.3	3	6.8	
Other	48	49.0	23	52.3	
Total	98	100.0	44	100.0	
Birth Weight					
≥ 2500	82	83.7	33	76.7	0.33
< 2500	16	16.3	10	23.3	
Total	98	100.0	43	100.0	
Gestation Age					
< 37 weeks	26	28.0	15	34.9	0.41
≥ 37 weeks	67	72.0	28	65.1	
Total	93	100.0	43	100.0	
Neonate HIV Status					
Positive	83	88.3	31	70.5	0.01
Negative	11	11.7	13	29.5	
Total	94	100.0	44	100.0	

6.7 MULTIVARIATE LOGISTIC REGRESSION ANALYSIS OF PREDICTORS OF EARLY NEONATAL SEPSIS

Based on standard practice methods for logistic regression analysis, attributes from the bivariate analysis in section 4.5 with p-values ≤ 0.21 were selected and fitted into a logistic regression analysis model. Table 6.6.1 shows the multivariate logistic regression analysis results predicting neonatal sepsis.

In the bivariate analysis (unadjusted odds ratios), mothers with positive HIV status had significant increased odds for neonatal sepsis compared to HIV negative status mothers (OR: 3.71, CI: 1.43 - 9.63, p = 0.007). This relationship, however, was not significant after adjusting for place of birth, referring clinic, and neonatal HIV status (OR: 3.55, CI: 0.23 - 55.67, p = 0.37).

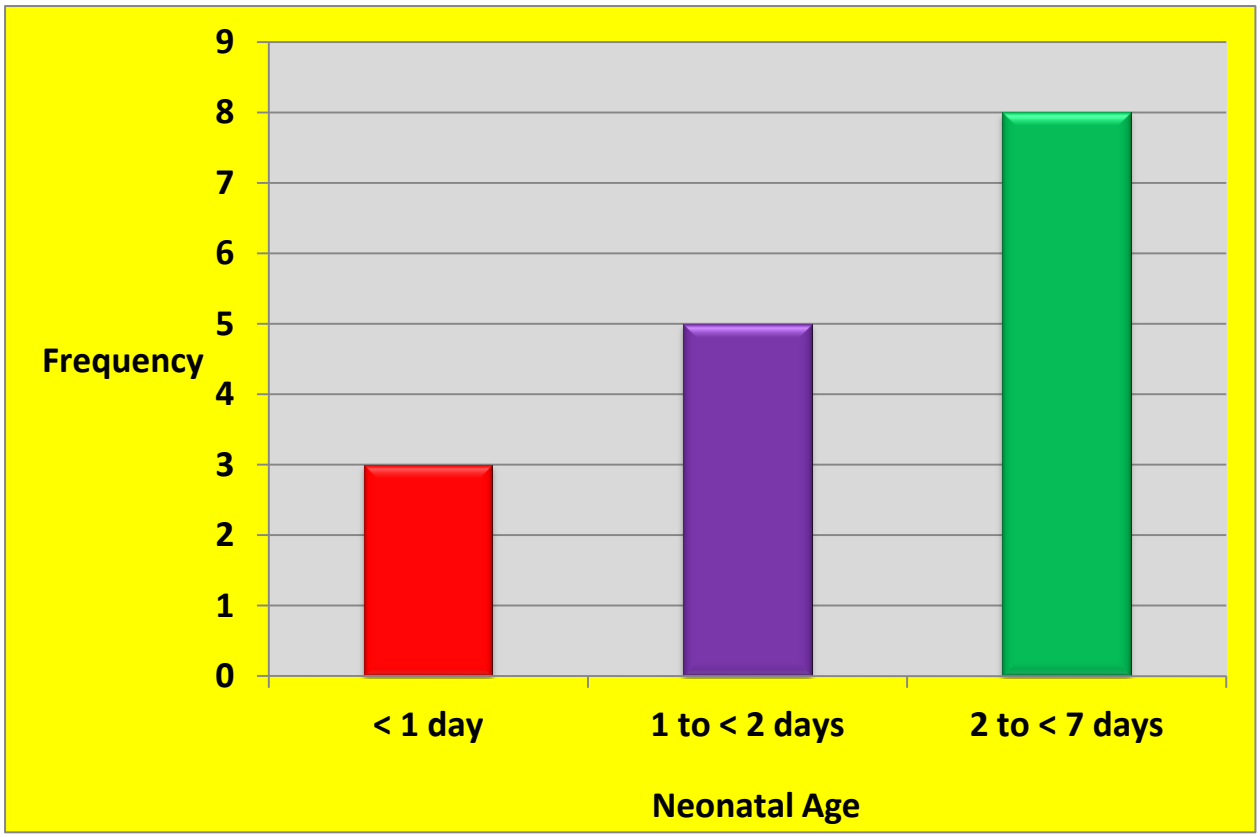
After adjusting for mother HIV status, referring clinic, and neonatal HIV exposed neonates that were born from the UTH had on average 4.5 increased odds for neonatal sepsis compared to neonates born from other health centers (OR: 4.48, CI: 1.85 - 10.85, p < 0.01).
HIV exposed neonates also showed increased odds for neonatal sepsis in the bivariate analysis but this relation was not significant after adjusting for the other factors. Referring clinic was not significant in predicting neonatal sepsis.

Table 7: Multivariate Logistic Regression Analysis of Risk Factors Associated with Neonatal sepsis

Variable	Unadjusted Odds Ratios (95% CI)			Adjusted Odds Ratios (95% CI)			P-Value
Mother HIV Status							
Negative	1.00			1.00			
Positive	3.71	1.43	9.63	3.55	0.23	55.67	0.37
Place of Birth							
Others	1.00			1.00			
UTH	3.16	1.45	6.88	4.48	1.85	10.85	< 0.01
Referring Clinic							
Others	1.00			1.00			

Kanyama	0.75	0.30	1.87	0.88	0.33	2.38	0.80
Chipata	1.88	0.67	5.25	2.37	0.73	7.68	0.15
Chawama	0.42	0.11	1.59	0.30	0.07	1.27	0.10
Neonate HIV exposure							
Negative	1.00			1.00			
Positive	3.16	1.28	7.80	1.18	0.09	16.14	0.90

Figure 3: Mortality of Neonate from EOS by Age



The number of neonates that died from early onset sepsis was 16 out of the 44 positive isolates. This number accounted for 36.4% mortality. The deaths in 50% of the babies occurred in the first two days and the other 50% within a week of admission shown in figure above. Seventy five percentage of male babies died from EOS compared to 25% of female babies. No neonates died from LOS in this study.

7.0 DISCUSSION

The study yielded 31% positive blood cultures which compares to the findings of some of the studies done in Nigeria, Tanzania and Asia where the yield ranges from 26.8 to 65% (Mane et al, 2010, Maramba-Lazarte et al 2011, Desai et al, 2010, Kayange et al, 2010 and Mokuolu et al, 2002).

There is still controversy in the definition of early and late onset sepsis, so the definition is based on what the institution decides to use whether 72hours or 7 days. Early onset (within first week of life) neonatal sepsis is generally acquired from pathogens of maternal genital tract, whereas late onset sepsis (after first week till 28 days of life) has its origin either from the community or from hospital (Karambin et al, 2011, Rad et al, 2004, Waseem et al 2005, Vergnano et al 2005). Some studies in literature defined EOS as less than 72 hours and LOS more than 72hours (Zaidi et al, 2005, Mane et al, 2010, Maramba-lazarte et al, 2011).

Our study reported that majority of the bacterial isolates caused EOS(77.3%)and LOS was at 22.7% this information is similar to what has been reported in some studies indicating the EOS is the common cause of morbidity in the neonates as reported by Zaidi and others in 2005 in a review paper(Zaidi et al, 2005). Other studies done in Asia, Africa and Australia have shown that majority of neonatal infections are early onset as 64% , 81.6% and 61% respectively (Waseem, et al, 2005, Mane et al, 2010, Maramba-Lazarte et al, 2011). There have been some studies that have found a high rate of LOS such as one done in Tanzania were 60% of sepsis was due to LOS and 80% of bacterial isolates in a study in Asia caused LOS (Kayange et al,2010 and Tiskumara et al, 2009).

This study has demonstrates that gram positive bacteria are still the most frequent organisms causing neonate sepsis in babies referred from the community to UTH. This information corresponds with that from a previous study by Mulenga V in 1997-1998 though her study was done in NICU (Mulenga, 2000). Studies done elsewhere are in support of gram positive bacteria such as *Staphylococcus aureus* and *Coagulase negative staphylococcus (CONS)* been majority agents causing neonatal sepsis by 60% (Mokuolu et al, 2002). Our study showed more than 85% gram positive bacteria and about 15% gram negative bacteria such as *Klebsiella* species and *Escherichia coli.*

Emerging information from hospital based cases in NICU at UTH has seen the emergence of gram negative bacteria been the cause of neonatal sepsis (NICU/UTH clinical audits, 2014). Unpublished data from maternity ward and NICU/UTH study by Bates et al 2014 is showed alarming figures of multidrug resistance *Klebsiella* species and other studies and data from Africa, Asia, Australia and Latin America are showed gram negative bacteria such as *Klebsiella species, Escherichia coli, Pseudomonas species and Enterobacter species* causing devastating sepsis in the neonates (Engmann et al,2011, Kayange et al,2010, Waseem et al, 2005, Isaacs, 2006, Rad et al, 2004, Aletayeb et al,2011).

The susceptibility of Staphylococcus aureus and CONS to WHO recommended antibiotics in treatment of neonatal sepsis penicillin and Gentamycin showed low sensitivity of 35% and 70% respectively. Ciprofloxacin, chloramphenicol and Cefotaxime showed higher sensitivity of more than 80%. Even the gram negative *Klebsiella* and *E.coli* showed sensitivity of more than 90% to cefotaxime, ciprofloxacin and Vancomycin. This is different compared to many studies which show high resistance to penicillin, aminoglycosides such as Gentamycin , third generation Cepholosporins and macrolides, resistance ranges from 50-100% in some cases. Studies done by Zaidi and friends showed resistance of common bacteria both gram positive and negative to penicillin and Gentamycin of more than 71% , Thaver and others also showed resistance of bacteria to WHO recommended antibiotics of more than 70% too (Zaidi et al2005 and Thaver et al, 2009). Other studies in Asia and Africa have shown gram negative *Klebsiella* species and *E.coli* being resistance to third generation cepholosporins, ciprofloxacin, penicillin and vancomycin ranging from 60-100% (Mane et al, 2011, Maramba-Lazarte, 2011, Desai et al, 2004, Kayange et al 2010, Isaacs, 2006 and Rad et al 2004).

Our study documented that mortality secondary to early onset sepsis was 38.8% and 50% of the neonates died within the first two days of admission. Engmann and others in their study mentioned that 80% of neonates die in the first three days of life and 49% are due to infection (Engmann et al, 2011). Other literature also indicates that 40.3% of deaths occur in neonates out of the 64% of deaths in children fewer than 5 years that are due to infections (Liu et al, 2012). Most of the deaths occur in the developing countries and can be prevented by simple hand washing and hygiene awareness (Isaacs, 2006, Liu et al, 2012, Zaidi et al 2005, Tiskumara et al, 2009).

The maternal HIV status and period of PROM had a significant association with development of early onset sepsis in our study though little has been documented in literature and other studies showed no significance of the two variables (Bang et al, 2005). A study done in 1986 at UTH showed that vaginal delivery had increased risk of neonatal sepsis, in our study over 97% of mothers delivered vaginally (Lisambo, 1986).

This study found increased risk for early onset neonatal sepsis of between 2 – 11 times higher (5 times higher on average) for neonates born from the UTH compared to other health centers. This may be attributed to high numbers of patients (low staff to patient ratios) poor infection control and prevention among the health workers. The hygiene standards of the mothers may also be questionable. The study done in Tanzania by Kayange and others found significant relationship with babies delivered at home and development of EOS 31% and LOS 42% and it also showed that PROM was a predictor of positive blood culture (Kayange et al, 2010).

This study at UTH, identified fever as being the most frequent clinical feature of neonatal sepsis. Irritability and poor feeding were some of the presenting complaints in babies with probable sepsis. Other features of note were jaundice and skin rash (Staphylococcus pustules).

A study done in India identified reduced sucking, cold baby and weak cry as some of the important neonatal features of sepsis and the Tanzanian study also identified poor feeding, lethargy, convulsions and cyanosis as indicators of neonatal sepsis (Bang et al, 2005 and Kayange et al, 2010).
Another neonatal feature which came out was the HIV exposure of neonate being a strong association to development of sepsis. The high white cell count was indicative of sepsis in majority of patients with an average of 64%. The average time for onset of illness was 13 hours that is most neonates presented to hospital within a day of onset of illness.

There is high burden of neonatal sepsis in health centres catering for densely populated areas of Kanyama, Chawama, Chipata and Matero Clinics referred the most patients and also the places of delivery. The other clinics such as Bauleni, Chelston, Mtendere and Kalingalinga recorded a low referral of patients.

Of major concern was data showing that over 50% of neonates referred to UTH despite having features of sepsis were not started on antibiotics at local clinic. We believe this intervention would have saved some babies that died from neonatal sepsis.

8.0 CONCLUSION

Gram positive organisms such as *Staphylococcus aureus* are still the leading cause of neonatal sepsis in babies referred from local clinics. EOS is more common at >75% of all cases of neonatal sepsis.

Penicillin has high resistance (>65%) as first line antibiotics to these organisms.

Neonatal sepsis contributed to 36.4% of neonatal mortality in this study and fever remains a common presenting clinical feature in a neonate.

Neonates born at UTH have an increased 4.5 odds (OR: 4, 48, Cl: 1.85 – 10.85, $p<0.01$) getting neonatal sepsis compared to those born at other places of delivery in Lusaka.

8.1 STUDY LIMITATIONS

Laboratory sample missing (22%) at UTH Laboratory despite careful transportation and labelling.

8.2 RECOMMENDATIONS

In view of the findings and conclusions of the this study, we recommend that

- Infection prevention and infection control become a priority in all delivery and nursery wards and health workers to wash hands before handling neonates
- UTH management to consider changing Penicillin and Gentamycin as first line antibiotics in treating neonatal sepsis to another alternative to save lives
- MOH/WHO revise the guidelines on prevention and treatment of neonatal sepsis
- Community health education to mothers on safe, hygienic and infective prevention such hand washing be intensified
- Recommend improved services at the clinics that refer to UTH with capacity building
- Drastic interventions are needed at NICU/UTH to save babies lives

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APPENDICES

APPENDIX I: DATA COLLECTION SHEET ON NEONATAL SEPSIS

DATE.....

SERIAL NUMBER..... .....UTH NUMBER.....

INITIALS.....

DATE OF BIRTH..... AGE IN DAYS.....

SEX: FEMALE/MALE

ANTENATAL HISTORY

AGE OF MOTHER.....

MARITAL STATUS.....

COMMUNITY.....

PARITY.....

CHILDREN ALIVE.....

CHILDREN DIED AND CAUSE.....

1.....

2.....

3.....

4.....

5.....

L.M.P.....

E.D.D.....

ILLNESSES.....

TREATMENT.....

RPR.....

HIV STATUS.....

LABOUR DATA

DURATION OF RUPTURE OF MEMBRANES.....

MODE OF DELIVERY.....

TIME OF DELIVERY.....

PLACE OF DELIVERY.....

REFERING CLINIC.....

NEONATES DATA ON ADMISSION

GESTATION AGE.....

BIRTH WEIGHT.....

TIME OF ONSET OF ILLNESS IN HOURS.....

PRESENTING FEATURES (TICK)

FAILURE/POOR FEEDING.....

TEMPERATURE.....

IRRITABILITY.....

TACHYNOEAIC.....

DIFFICULTY BREATHING.....

CHEST INDRAWING.....

CONVULSIONS.....

LETHARGY.....

UMBILICAL DISCHARGE.....

JAUNDICE.....

PALLOR.....

CYANOSIS.....

ABDOMINAL DISTENSION.....

OTHER SYMPTOMS.....

TREATMENT GIVEN AT CLINIC.....

ADMISSION DATA

DATE OF ADMISSION.....

DIGNOSIS 1.....

2.....

3.....

TEMPERATURE ON ADMISSION.....

INVESTIGATIONS (INICATE RESULT FOR RAPID TESTS AND DATE)

BLOOD CULTURE.....

FBC/DWC/ESR.....

HIV RAPID TEST.....

RANDOM BLOOD SUGAR (RBS).....

DATE TREATMENT STARTED AND NAME OF ANTIBIOTICS.....

.....

.....

BLOOD CULTURE RESULTS (TICK)

POSITIVE.....NEGATIVE.....

DATE OF DEATH.....

APPENDIX II: INFORMATION SHEET

Common bacterial causes of neonatal sepsis and antimicrobial susceptibility at the University Teaching Hospital, Lusaka, Zambia

The purpose of the study is to identify the common germs that cause sickness in newborn babies and find how the medicine given to them is working. The information gathered will be used to improve the prevention, care and management of infections in babies in Lusaka and Zambia as a whole.

We would like your baby to take part with your permission in this important study. The study will include answering some questions about your personal life during pregnancy and labour. The questions include your age, marital status, number of children, residence, antenatal and maternal medical history and events surrounding the birth of the baby and what kind of symptoms lead your baby to come to UTH.

There will be medical procedures such as drawing blood for tests and needle prick on finger or toe for the rapid tests. All procedures will be carried out in a safe environment with care not to do harm.

All the information collected will be kept in secret (confidential) and will not be communicated to anyone except the research team. The interview will be done in private.

Your participation in the study will be appreciated and this study is voluntary and you are free to withdraw your baby at any time. Your participation will not affect the quality of care that you will receive at our institution.

If you are ready to participate and join the study, kindly sign the document on the next page to give consent and that you are agreeable with the procedures of the study.
If you have any concerns and questions regarding the study feel free to contact us on following address below.

Thank you for your cooperation and your support is highly appreciated.

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APPENDIX III: CONSENT FORM

I acknowledge that I have been given all the information regarding the study and the data will be treated with confidentiality and privacy and my baby's name will not be disclosed to the public. I and my baby are not under any pressure to contribute to the study and I am satisfied with the information given.

I agree to participate in the study ( )

Name.....Signature.....Thumb print.....

Date.....Witness.....Signature.....

Name (researcher).....Signature.....

Note: For any information regarding study contact us using the address below.

Dr Sophia .T. Msiska-Simpungwe

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