

## A study on microbiome diversity and antibiotic sensitivity pattern of vaginopathogens at gestation time in a tertiary care hospital

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### Abstract

A six-month study was conducted in a tertiary care hospital in Kerala, India, to analyse the microbiome diversity and antibiotic sensitivity pattern of vaginopathogens in 75 pregnant women. Vaginal swabs were collected and sent for microbiological evaluation. Pregnancy causes significant changes in the vaginal microbiota, which plays a crucial role in protecting both the mother and foetus from infections. Case records were reviewed for age, presenting complaints, laboratory results, diagnostic reports, and therapeutic management. Out of 75 vaginal swab samples, 22 tested positives for bacterial growth, identifying 32 bacterial strains, which were classified into 9 distinct species. Klebsiella species were the most prevalent (21.8%), followed by coagulase-negative staphylococci (18.7%), beta-haemolytic streptococci (15.6%), and *E. coli* (12.5%). This study highlights the need for vigilant monitoring and targeted antibiotic therapy to optimize maternal and neonatal outcomes while addressing the growing challenge of antibiotic resistance.

**Keywords:** Pregnancy; Vaginal Microbiota; Antibiotic Sensitivity; Vaginal Swab; Microbiome Diversity

### 1. Introduction

The vagina represents a dynamic environment which is inhabited by diverse organisms that contribute nutrients. These one-of-a-kind surroundings undergo important changes in all levels of life, from start to the age of puberty until menopause. The equilibrium therein can be disturbed by physiological or non-physiological changes, mediated by hormonal status, sexual behavior, vaginal blood, foreign bodies and/or concurrent use of medications. The vaginal microbiota refers to the microorganisms that are living inside the vagina [1].

These microorganisms make contributions to a woman's reproductive and overall health. Lactobacillus species account for 95% of the vaginal flora. The 4 most common types of Lactobacilli dominant vaginal microbiome profiles are characterized by *L. iners*, *L. crispatus*, *L. gasseri*, or *L. jensenii*. Factors along with antibiotic use, sexual activity, and hormonal modifications can disrupt the stability of the vaginal microbiota, along with bacterial vaginosis. [2]

During pregnancy, the vaginal microbiota's composition undergoes significant changes and thereby acting as a protective shield against infection for both the mother and fetus. Hydrogen peroxide produced by Lactobacillus strains plays a critical role in preserving the microenvironment of the vagina and the inhibition of overgrowth of pathogenic microorganism. Lactobacillus and the predominance of anaerobic bacteria are the most common causes of vaginal infections in women of childbearing age. [3]

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Intrapartum antibiotics play a crucial role in preventing Group B Streptococcus (GBS) transmission from pregnant women to their newborns during labor and delivery. GBS is a bacterium commonly found in the vaginal and rectal areas of healthy adults. While it typically does not cause harm in healthy adults, it can pose a significant risk to newborns if they are exposed to the bacteria during childbirth. In newborns, GBS infections can lead to serious complications such as pneumonia, sepsis, or meningitis, which can be life-threatening if not promptly treated. [4,5]

Intrapartum antibiotics plays a crucial role in preventing Group B Streptococcus (GBS) transmission from pregnant women to their newborns during labor and delivery. GBS is a bacterium commonly found in the vaginal and rectal areas of healthy adults. While it typically does not cause harm in healthy adults, it can pose a significant risk to newborns if they are exposed to the bacteria during childbirth. In newborns, GBS infections can lead to serious complications such as pneumonia, sepsis, or meningitis, which can be life-threatening if not promptly treated. To mitigate the risk of GBS transmission to newborns, healthcare providers administer intrapartum antibiotics to pregnant women who are colonized with GBS or who have certain risk factors. [6,7]

The administration of antibiotics during labor helps reduce the concentration of GBS bacteria in the birth canal, thereby lowering the likelihood of newborn exposure during delivery. The timing and duration of intrapartum antibiotics are critical to ensuring their effectiveness while minimizing potential risks associated with antibiotic use. Typically, intrapartum antibiotics are initiated at least four hours before delivery. This time frame allows sufficient time for the antibiotics to reach therapeutic levels in the mother's bloodstream, providing optimal protection against GBS transmission to the newborn. [8]

The antibiotics are usually administered intravenously to ensure rapid absorption and distribution throughout the body. In cases where the mother's GBS status is unknown, or if she has certain risk factors such as preterm labor or prolonged rupture of membranes, healthcare providers may opt to initiate intrapartum antibiotics as a precautionary measure. Additionally, if a pregnant woman tests positive for GBS colonization earlier in her pregnancy but does not receive intrapartum antibiotics during labor due to premature delivery or other circumstances, antibiotics may still be administered at the time of delivery to provide protection to the newborn. While intrapartum antibiotics are highly effective in reducing the incidence of early-onset GBS infections in newborns, they are not without potential drawbacks. Antibiotic use during labor may contribute to the development of antibiotic resistance, both in the mother and in neonatal microbiota. Additionally, maternal antibiotic exposure can impact the newborn's microbiome, potentially affecting their long-term health outcomes. [9,10]

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## 2. Materials and methods

75 cases were collected from the obstetrics and gynecology department.

$$N = \frac{\left(z_{1-\frac{\alpha}{2}}\right)^2 \sigma^2}{E^2}$$

$\sigma$  is the standard deviation and E is the margin of error

Data from a previous study found a standard deviation of 1.1

Here  $\sigma = 1.1$

$z = 1.96$  (95% confidence level)

$E = 0.25$

$$N = \frac{z^2 \sigma^2}{E^2} = \frac{1.96^2 \times 1.1^2}{0.25^2} = 60$$

Anticipating loss to follow-up and missing of data, the minimum sample size is rounded to be **75**.

An observational prospective study, conducted among the pregnant women in the OBG department in a tertiary care hospital, SH Medical Centre, Kottayam, Kerala, India for 6-months period. After obtaining permission from the IEC and Informed consent from individual patients, the data collection was started. The study included all pregnant women with singleton gestation undergoing either vaginal delivery or cesarean section, while excluding those who were allergic to antibiotics, had incomplete medical records, or refused the vaginal swab. Case records are prospectively reviewed from

OPD and IP and the information includes age, presenting complaints, laboratory investigation data, other diagnostic reports, therapeutic management and any added complications. The vaginal swab was collected before the delivery and the swab was cultured at the microbiology lab and reports were collected and the isolated organisms will be noted along with their sensitivity and resistance pattern to the antibiotics. The data will be collected in a specially designed data collection form.

### 3. Results

A total of 75 pregnant women meeting the inclusion criteria were included in the study. Most participants were aged 26–30 years. The primary reasons for admission were safe confinement (28%), complaints of pain (24%), induction of labour (22.7%), and leaking per vagina (13.3%). The majority of the study population were at a gestational age of 38 weeks, followed by 37 weeks, 39 weeks, and 36 weeks. Regarding medical history, gestational diabetes mellitus (29.3%) was the most common condition, followed by hypothyroidism (18.6%), pregnancy-induced hypertension (6.6%), and polycystic ovary syndrome (5.3%). Vaginal delivery was the predominant mode of delivery (62.7%), with 37.3% undergoing cesarean section. Vaginal swab cultures were obtained from all 75 participants, with 22 (29.3%) testing positive and 53 (70.6%) showing no significant flora (NSF). Among full-term pregnancies (63 cases), 18 (28.5%) had positive cultures, and 45 (71.5%) were NSF, while in preterm pregnancies (12 cases), 4 (33.3%) had positive cultures, and 8 (66.7%) were NSF.

#### 3.1. Reason for admission

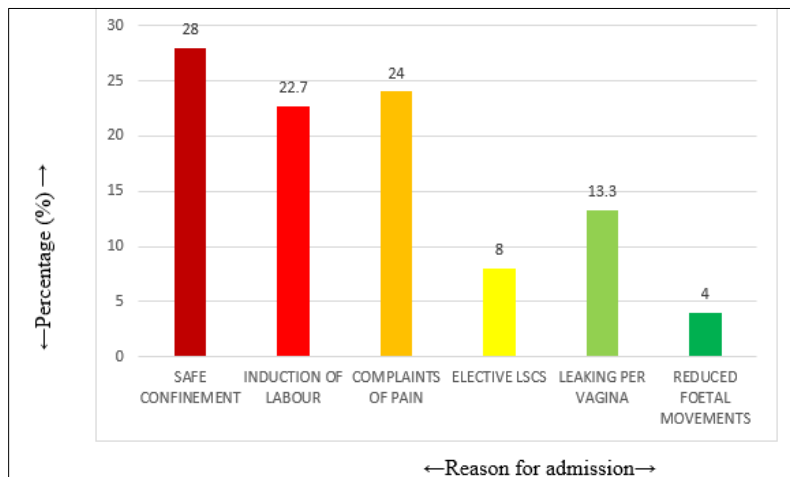


Figure 1 Percentage distribution of reasons for admission

#### 3.2. Gestation age in weeks

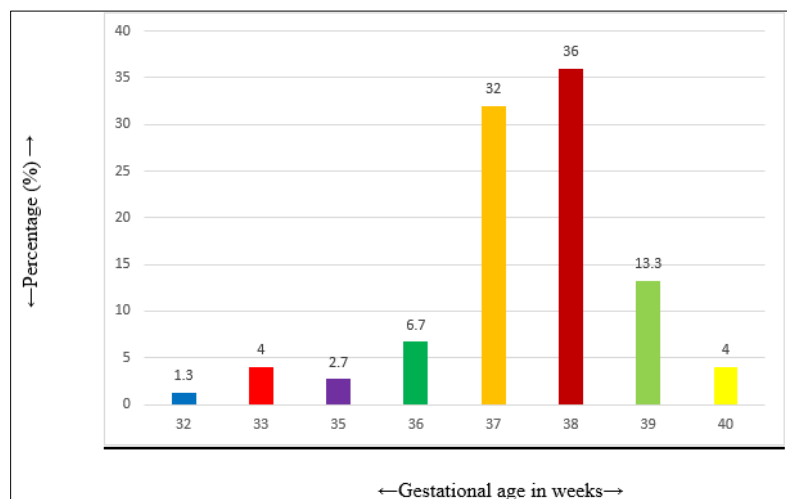
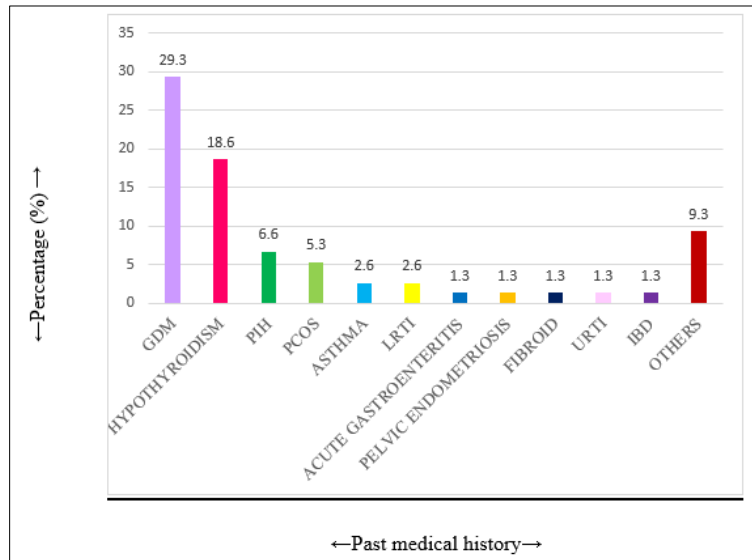


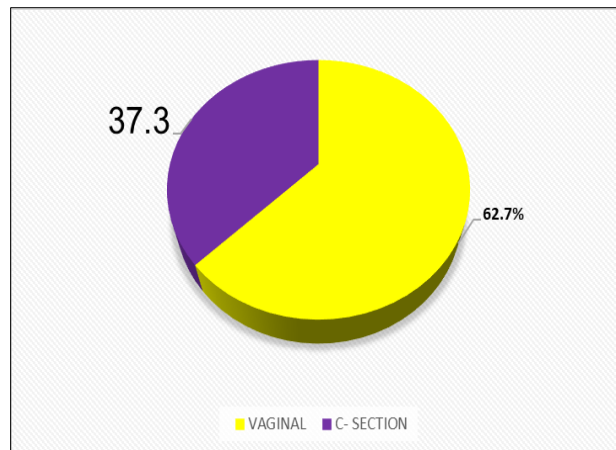
Figure 2 Percentage distribution based on gestational age in weeks

### 3.3. Past medical history



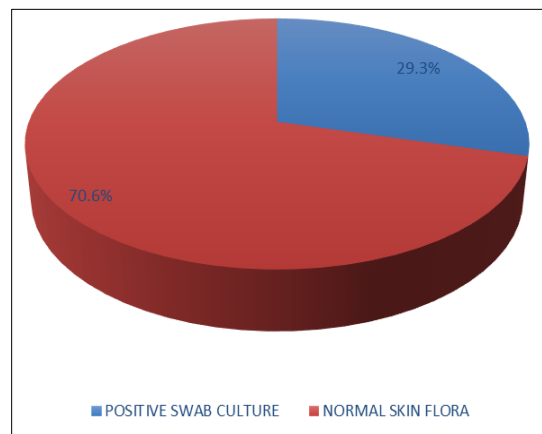
**Figure 3** Percentage distribution of past medical history

### 3.4. Mode of delivery



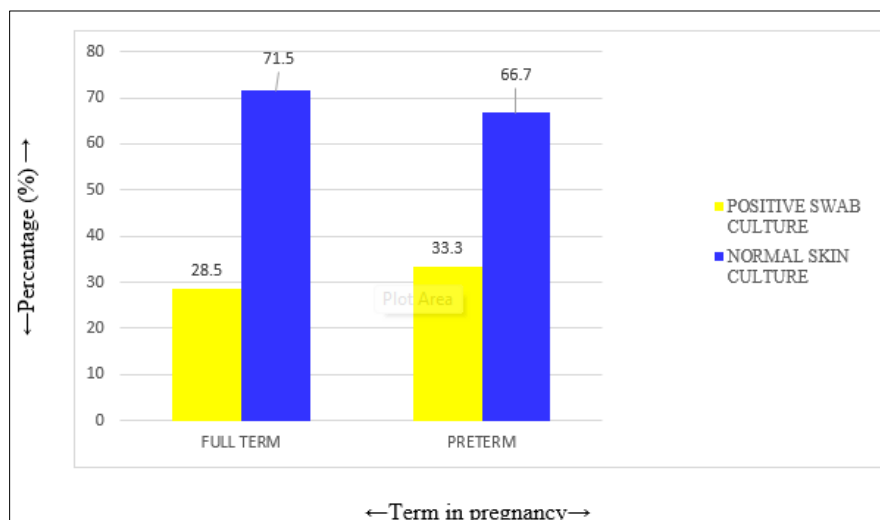
**Figure 4** Percentage distribution based on mode of delivery

### 3.5. Vaginal swab culture



**Figure 5** Percentage distribution of vaginal swab culture

### 3.6. Vaginal swab culture in relation to gestational age



**Figure 6** Percentage distribution of vaginal swab culture based on gestational age

### 3.7. Microbiome diversity analysis

Out of 75 vaginal swab samples, 22 tested positives for bacterial growth. A total of 32 bacterial strains were identified, with some microorganisms appearing in multiple samples. These 32 strains were ultimately classified into 9 distinct bacterial species. *Klebsiella species* were the most prevalent, found in 21.8% of samples, followed closely by *Coagulase negative staphylococci* at 18.7%. *Beta haemolytic streptococci* and *E. coli* were also notable constituents, occurring in 15.6% and 12.5% of samples, respectively.

**Table 1** Frequency distribution of microbiome diversity

| Organisms   | Frequency (n=32) | Percentage (%) |
|---|------------------|----------------|
| <i>Klebsiella species</i>                                     | 7                | 21.8           |
| <i>Coagulase negative Staphylococci</i>                       | 6                | 18.7           |
| <i>Beta-haemolytic Streptococci</i>                           | 5                | 15.6           |
| <i>E coli</i>   | 4                | 12.5           |
| <i>Methicillin resistant coagulase negative Staphylococci</i> | 4                | 12.5           |
| <i>Non-haemolytic Streptococci</i>                            | 2                | 6.2            |
| <i>Staphylococcus aureus</i>                                  | 2                | 6.2            |
| <i>Pseudomonas aeruginosa</i>                                 | 1                | 3.1            |
| <i>Citrobacter diversus</i>                                   | 1                | 3.1            |

### 3.8. Sensitivity and resistance pattern of isolated microorganisms from vaginal swab

*Klebsiella species* demonstrated complete sensitivity to a range of antibiotics including Ampicillin/sulbactam, Meropenem, Imipenem, Cotrimoxazole, Chloramphenicol, and Nitillin, but were entirely resistant to Linezolid and Cefpodoxime.

*Coagulase negative Staphylococci* were found to be highly sensitive to Ampicillin/sulbactam, Cefoperazone/sulbactam, Ceftriaxone, Meropenem, Cefepime, Imipenem, Vancomycin, Nitillin, Methicillin, Cephalexin, Linezolid, and Faropenem, with a notable resistance rate of 67% to Cefpodoxime.

*Beta-haemolytic Streptococci* showed complete sensitivity to Ampicillin/sulbactam and Imipenem, while being completely resistant to Roxithromycin, Erythromycin, Colistin, and Lincomycin.

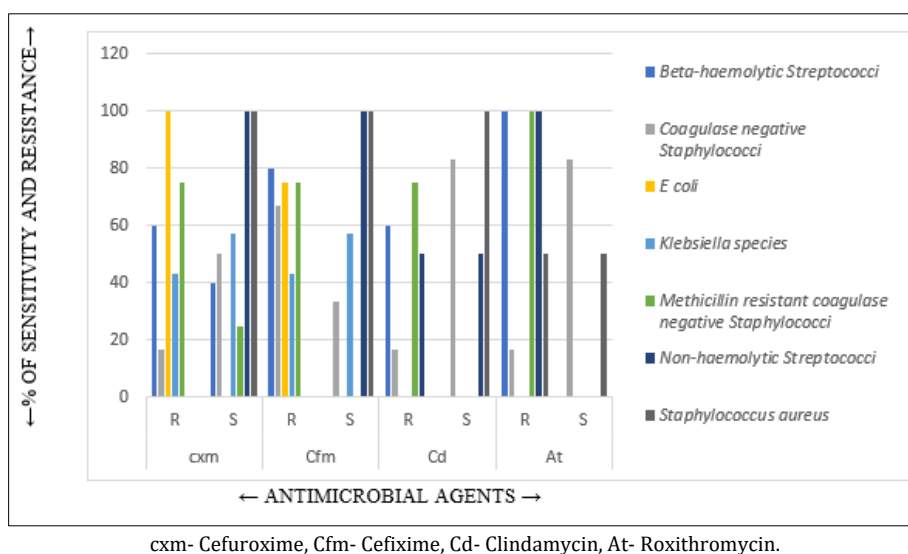
*E. coli* strains exhibited full sensitivity to Meropenem, Gentamycin, Amikacin, Imipenem, Chloramphenicol, Nitillin, Cephalexin, and Linezolid, and complete resistance to Nalidixic acid and Cefpodoxime.

*Methicillin-resistant coagulase-negative staphylococci* were completely sensitive to Imipenem, Cotrimoxazole, Vancomycin, Nitillin, Amoxicillin/clavulanic acid, and Tigecycline, but resistant to Roxithromycin, Erythromycin, Methicillin, Cephalexin, Lincomycin, Faropenem, and Cefpodoxime.

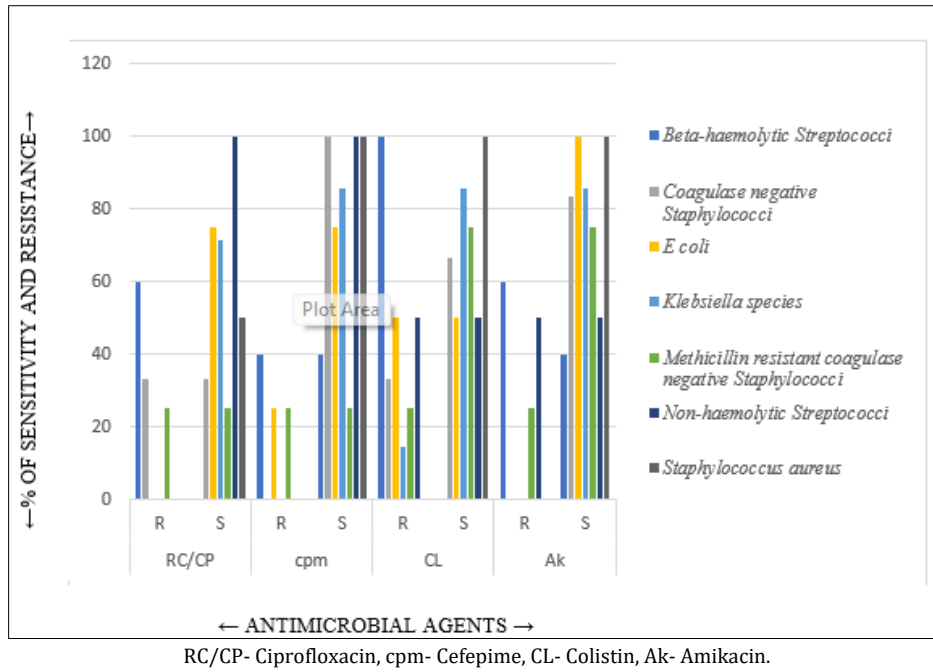
*Non-haemolytic Streptococci* showed complete sensitivity to a broad spectrum of antibiotics, including Cefuroxime, Ampicillin/sulbactam, Cefoperazone/sulbactam, Levofloxacin, Cefixime, Ceftriaxone, Ciprofloxacin, Cefotaxime, Meropenem, Cefepime, Imipenem, Ofloxacin, Cotrimoxazole, Norfloxacin, Vancomycin, Nitillin, Cephalexin, Linezolid, Amoxicillin/sulbactam, Faropenem, Cefpodoxime, and Tigecycline, with complete resistance to Roxithromycin.

*Staphylococcus aureus* was found to be completely sensitive to Cefuroxime, Ampicillin/sulbactam, Cefoperazone/sulbactam, Cefixime, Ceftriaxone, Clindamycin, Cefotaxime, Meropenem, Cefepime, Colistin, Amikacin, Imipenem, Cotrimoxazole, Cloxacillin, Piperacillin/Tazobactam, Nitillin, Methicillin, Cephalexin, Linezolid, Lincomycin, Amoxicillin/clavulanic acid, Faropenem, and Tigecycline.

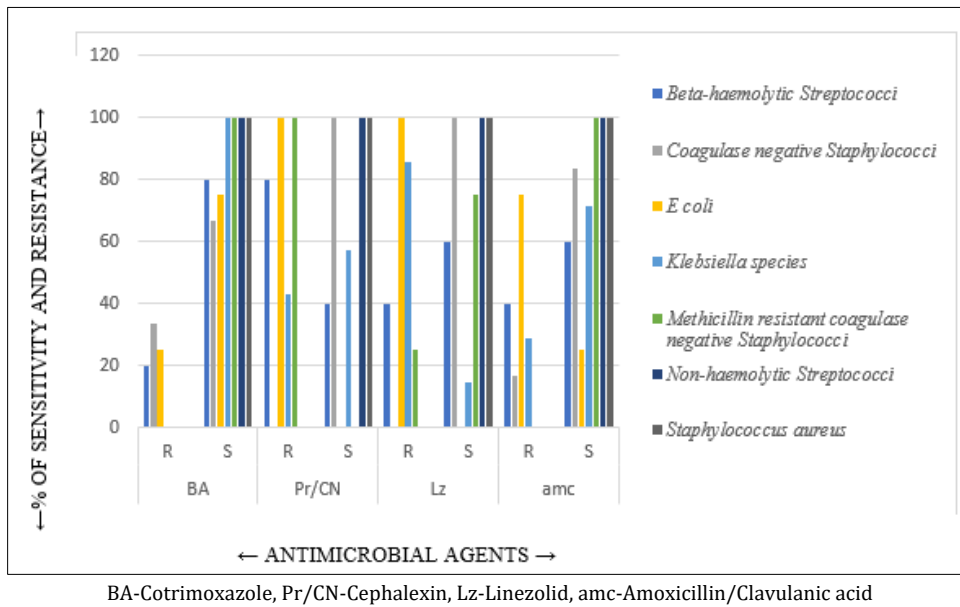
*Pseudomonas aeruginosa* and *Citrobacter diversus* were excluded from the study due to the presence of only a single isolate for each.



**Figure 7** Antibiotic Sensitivity and resistance Pattern of the various isolated microbes to Antimicrobial Agents



**Figure 8** Antibiotic Sensitivity and resistance Pattern of the various isolated microbes to Antimicrobial Agents



**Figure 9** Antibiotic Sensitivity and resistance Pattern of the various isolated microbes to Antimicrobial Agents

First-line antibiotics considered safe in pregnancy include Ampicillin/Sulbactam, effective against Klebsiella, CoNS, beta-haemolytic Streptococci, and Staphylococcus aureus, commonly used for respiratory, UTI, skin, and intra-abdominal infections. Cephalexin (1st Gen) and Cefuroxime (2nd Gen) are also safe, targeting E. coli, Staphylococcus aureus, and Non-haemolytic Streptococci, suitable for UTIs, skin, and respiratory infections. Ceftriaxone (3rd Gen) is recommended for severe infections like pneumonia, meningitis, and sepsis, while Amoxicillin/Clavulanic Acid covers CoNS and Staphylococcus aureus, used for respiratory, UTI, and soft tissue infections. Vancomycin is reserved for serious Gram-positive infections, including MRSA. Antibiotics to avoid or use with caution include Meropenem and Imipenem, which are reserved for resistant cases, and Linezolid, which is used only when necessary. Aminoglycosides like Gentamycin and Amikacin should be used cautiously due to potential nephrotoxicity and ototoxicity.

**Table 2** Antibiotic sensitivity frequency of isolated organisms

| Organisms isolated  | n | As | Pr/CN | cxm | ctr | amc | VA | mep | Imp | Lz | Gm | Ak |
|---|---|----|-------|-----|-----|-----|----|-----|-----|----|----|----|
| <i>Beta-haemolytic Streptococci</i>                           | 5 | 5  | 1     | 2   | 1   | 3   | 1  | 2   | 5   | 3  | 0  | 2  |
| <i>Citrobacter diversus</i>                                   | 1 | 1  | 0     | 0   | 0   | 0   | 0  | 1   | 1   | 0  | 1  | 1  |
| <i>Coagulase negative Staphylococci</i>                       | 6 | 6  | 6     | 3   | 6   | 5   | 6  | 6   | 6   | 6  | 5  | 5  |
| <i>E coli</i>   | 4 | 4  | 0     | 0   | 1   | 1   | 0  | 4   | 4   | 0  | 4  | 4  |
| <i>Klebsiella species</i>                                     | 7 | 7  | 4     | 4   | 6   | 5   | 0  | 7   | 7   | 1  | 6  | 6  |
| <i>Methicillin resistant coagulase negative Staphylococci</i> | 4 | 3  | 0     | 1   | 1   | 4   | 4  | 2   | 4   | 3  | 3  | 3  |
| <i>Non-haemolytic Streptococci</i>                            | 2 | 2  | 2     | 2   | 2   | 2   | 2  | 2   | 2   | 2  | 1  | 1  |
| <i>Pseudomonas aeruginosa</i>                                 | 1 | 1  | 0     | 0   | 0   | 0   | 0  | 1   | 1   | 0  | 1  | 1  |
| <i>Staphylococcus aureus</i>                                  | 2 | 2  | 2     | 2   | 2   | 2   | 1  | 2   | 2   | 2  | 0  | 2  |

n= number of isolates of each organism; As- Ampicillin/Salbactam, Pr/CN- Cephalexin, cxm- Cefuroxime, ctr- Ceftriaxone, amc- Amoxicillin/Clavulanic acid, VA- Vancomycin, mep- Meropenem, Imp- Imipenem, Lz- Linezolid, Gm- Gentamycin, Ak- Amikacin.

**Table 3** Antibiotic sensitivity patterns of isolated organisms in percentage (%)

| Organisms isolated  | n | As   | Pr/CN | cxm  | ctr  | amc  | VA   | mep  | Imp | Lz   | Gm   | Ak   |
|---|---|------|-------|------|------|------|------|------|-----|------|------|------|
| <i>Beta-haemolytic Streptococci</i>                           | 5 | 100  | 40.0  | 40.0 | 20.0 | 40.0 | 20.0 | 40.0 | 100 | 60.0 | 0.0  | 40.0 |
| <i>Citrobacter diversus</i>                                   | 1 | 100  | 0.0   | 0.0  | 0.0  | 100  | 0.0  | 100  | 100 | 0.0  | 100  | 100  |
| <i>Coagulase negative Staphylococci</i>                       | 6 | 100  | 100   | 50.0 | 100  | 83.3 | 100  | 100  | 100 | 100  | 83.3 | 83.3 |
| <i>E coli</i>   | 4 | 100  | 0.0   | 0.0  | 25.0 | 100  | 0.0  | 100  | 100 | 0.0  | 100  | 100  |
| <i>Klebsiella species</i>                                     | 7 | 100  | 57.1  | 57.1 | 85.7 | 85.7 | 0.0  | 100  | 100 | 14.3 | 85.7 | 85.7 |
| <i>Methicillin resistant coagulase negative Staphylococci</i> | 4 | 75.0 | 0.0   | 25.0 | 25.0 | 75.0 | 100  | 50.0 | 100 | 75.0 | 75.0 | 75.0 |
| <i>Non-haemolytic Streptococci</i>                            | 2 | 100  | 100   | 100  | 100  | 50.0 | 100  | 100  | 100 | 100  | 50.0 | 50.0 |
| <i>Pseudomonas aeruginosa</i>                                 | 1 | 100  | 0.0   | 0.0  | 0.0  | 100  | 0.0  | 100  | 100 | 0.0  | 100  | 100  |
| <i>Staphylococcus aureus</i>                                  | 2 | 100  | 100   | 100  | 100  | 100  | 50.0 | 100  | 100 | 100  | 0.0  | 100  |

n= number of isolates of each organism; As- Ampicillin/Salbactam, Pr/CN- Cephalexin, cxm- Cefuroxime, ctr- Ceftriaxone, amc- Amoxicillin/Clavulanic acid, VA- Vancomycin, mep- Meropenem, Imp- Imipenem, Lz- Linezolid, Gm- Gentamycin, Ak- Amikacin.

**Table 4** Antibiotic resistance frequency of isolated organisms

| Organisms isolated  | n | As | Pr/CN | cxm | ctr | amc | VA | mep | Imp | Lz | Gm | Ak |
|---|---|----|-------|-----|-----|-----|----|-----|-----|----|----|----|
| <i>Beta-haemolytic Streptococci</i>                           | 5 | 0  | 4     | 3   | 3   | 1   | 2  | 0   | 0   | 1  | 4  | 3  |
| <i>Citrobacter diversus</i>                                   | 1 | 0  | 1     | 1   | 1   | 0   | 0  | 0   | 0   | 1  | 0  | 0  |
| <i>Coagulase negative Staphylococci</i>                       | 6 | 0  | 0     | 1   | 0   | 1   | 0  | 0   | 0   | 0  | 1  | 0  |
| <i>E coli</i>   | 4 | 0  | 4     | 4   | 2   | 3   | 0  | 0   | 0   | 4  | 0  | 0  |
| <i>Klebsiella species</i>                                     | 7 | 0  | 3     | 3   | 1   | 2   | 0  | 0   | 0   | 6  | 0  | 0  |
| <i>Methicillin resistant coagulase negative Staphylococci</i> | 4 | 1  | 4     | 3   | 1   | 0   | 0  | 1   | 0   | 1  | 1  | 1  |
| <i>Non-haemolytic Streptococci</i>                            | 2 | 0  | 0     | 0   | 0   | 0   | 0  | 0   | 0   | 0  | 1  | 1  |



|                               |   |   |   |   |   |   |   |   |   |   |   |   |
|-------------------------------|---|---|---|---|---|---|---|---|---|---|---|---|
| <i>Pseudomonas aeruginosa</i> | 1 | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 0 | 1 | 0 | 0 |
| <i>Staphylococcus aureus</i>  | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 |

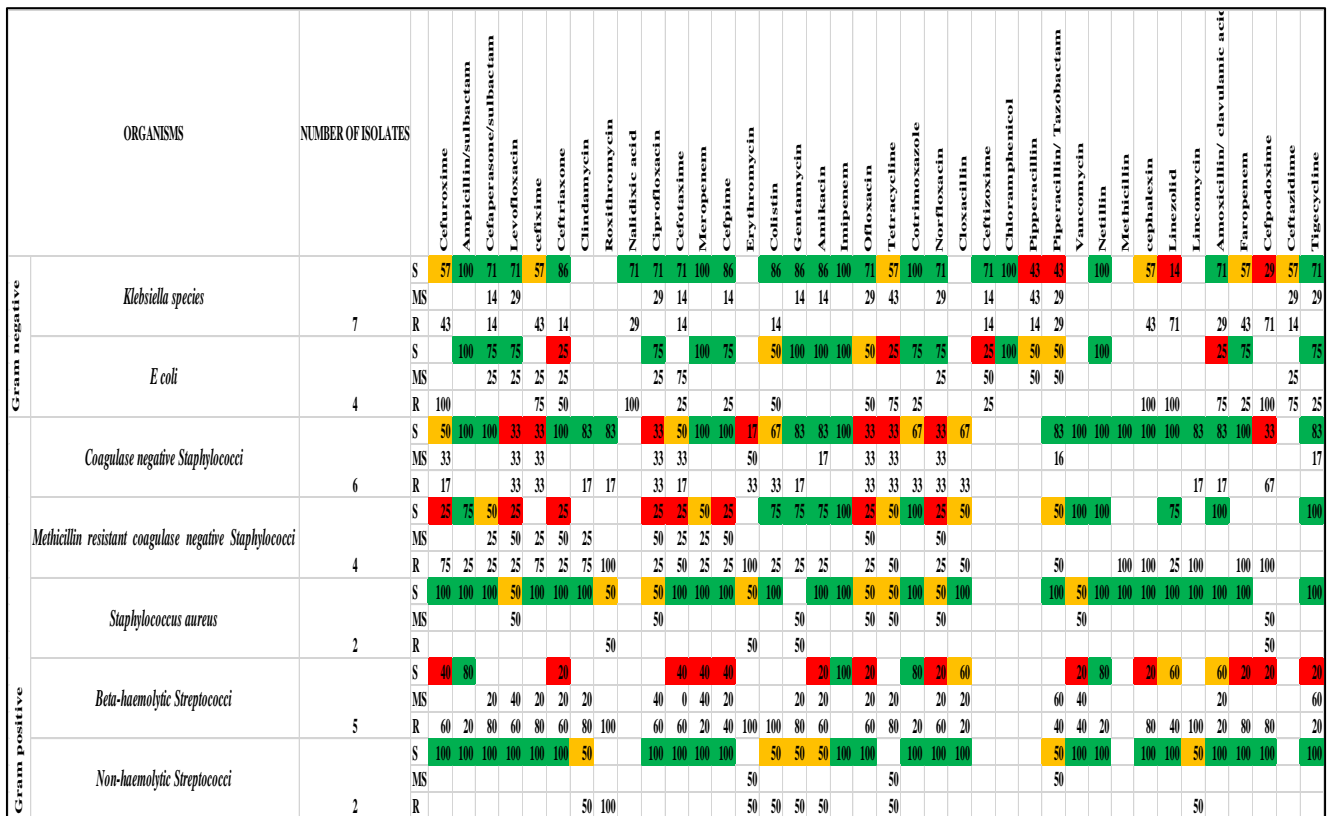
n= number of isolates of each organism; As- Ampicillin/Salbactam, Pr/CN- Cephalixin, cxm- Cefuroxime, ctr- Ceftriaxone, amc- Amoxicillin/Clavulanic acid, VA- Vancomycin, mep- Meropenem, Imp- Imipenem, Lz- Linezolid, Gm- Gentamycin, Ak- Amikacin.

**Table 5** Antibiotic resistance patterns of isolated organisms in percentage (%)

| Organisms isolated                                     | n | As   | Pr/CN | cxm  | ctr  | amc  | VA   | mep  | Imp | Lz   | Gm   | Ak   |
|--|---|------|-------|------|------|------|------|------|-----|------|------|------|
| Beta-haemolytic Streptococci                           | 5 | 0.0  | 80.0  | 60   | 60.0 | 40.0 | 40.0 | 0.0  | 0.0 | 40.0 | 80.0 | 60.0 |
| Citrobacter diversus                                   | 1 | 0.0  | 100   | 100  | 100  | 0.0  | 0.0  | 0.0  | 0.0 | 100  | 0.0  | 0.0  |
| Coagulase negative Staphylococci                       | 6 | 0.0  | 0.0   | 16.7 | 0.0  | 16.7 | 0.0  | 0.0  | 0.0 | 0.0  | 16.7 | 0.0  |
| E coli   | 4 | 0.0  | 100   | 100  | 50.0 | 75.0 | 0.0  | 0.0  | 0.0 | 100  | 0.0  | 0.0  |
| Klebsiella species                                     | 7 | 0.0  | 42.9  | 42.9 | 14.3 | 28.6 | 0.0  | 0.0  | 0.0 | 85.7 | 0.0  | 0.0  |
| Methicillin resistant coagulase negative Staphylococci | 4 | 25.0 | 100   | 75.0 | 25.0 | 0.0  | 0.0  | 25.0 | 0.0 | 25.0 | 25.0 | 25.0 |
| Non-haemolytic Streptococci                            | 2 | 0.0  | 0.0   | 0.0  | 0.0  | 0.0  | 0.0  | 0.0  | 0.0 | 0.0  | 50.0 | 50.0 |
| <i>Pseudomonas aeruginosa</i>                          | 1 | 0.0  | 100   | 100  | 0.0  | 100  | 0.0  | 0.0  | 0.0 | 100  | 0.0  | 0.0  |
| <i>Staphylococcus aureus</i>                           | 2 | 0.0  | 0.0   | 0.0  | 0.0  | 0.0  | 0.0  | 0.0  | 0.0 | 0.0  | 50.0 | 0.0  |

n= number of isolates of each organism.; As- Ampicillin/Salbactam, Pr/CN- Cephalixin, cxm- Cefuroxime, ctr- Ceftriaxone, amc- Amoxicillin/Clavulanic acid, VA- Vancomycin, mep- Meropenem, Imp- Imipenem, Lz- Linezolid, Gm- Gentamycin, Ak- Amikacin.

**3.9. Antibiogram**



**Figure 10** Antibiogram of vaginal pathogens showing susceptibility patterns to commonly used antibiotic

#### 4. Discussion

This observational prospective study, conducted among 75 pregnant women, provides valuable insights into the demographic and clinical characteristics of the study population. The majority of participants were in their late reproductive years, with a mean age of 29 years and a predominant age group of 26 to 30 years.

The primary reasons for hospital admission included safe confinement (28%), pain (24%), induction of labor (22.7%), and leaking per vagina (13.3%). These findings underscore the varied clinical presentations and the importance of individualized management strategies during the peripartum period.

Parity analysis revealed a predominance of primiparous women, followed by those with multiparous statuses such as G2P1L1, G3P1L1A1, and G2A1. This distribution reflects the reproductive patterns and healthcare-seeking behaviors within the population.

The mode of delivery was predominantly vaginal (62.7%), with 37.3% undergoing cesarean sections. This balance highlights the clinical decision-making process aimed at optimizing maternal and fetal outcomes.

Microbiological evaluation of vaginal swab cultures revealed positive results in 29.3% of cases, while 70.6% showed no significant flora (NSF). Among full-term pregnancies, 28.5% had positive swab cultures compared to 71.5% NSF. In preterm pregnancies, the incidence of positive cultures was slightly higher at 33.3%, with 66.7% NSF. These findings suggest that while a substantial proportion of deliveries are associated with positive cultures, most do not exhibit significant bacterial colonization. This has implications for neonatal outcomes and infection control strategies.

A total of nine distinct microorganisms were isolated from the vaginal swabs. *Klebsiella* species, the most prevalent organism, demonstrated complete sensitivity to Ampicillin/Sulbactam, Meropenem, Imipenem, Cotrimoxazole, Chloramphenicol, and Nitrofurantoin but showed complete resistance to Linezolid and Cefpodoxime. This highlights the importance of cautious antibiotic selection to prevent ineffective treatment and resistance development.

Coagulase-negative Staphylococci (CoNS), often regarded as non-pathogenic, demonstrated high sensitivity to commonly used antibiotics but significant resistance to Cefpodoxime. Beta-hemolytic Streptococci exhibited complete sensitivity to Ampicillin/Sulbactam and Imipenem, while showing resistance to macrolides and Colistin, emphasizing the need for alternative therapeutic options.

*E. coli* strains, a common cause of urinary tract infections, were fully sensitive to Meropenem, Gentamycin, Amikacin, Imipenem, Chloramphenicol, Nitrofurantoin, Cephalexin, and Linezolid but resistant to Nalidixic acid and Cefpodoxime, illustrating the challenges posed by emerging resistance.

Methicillin-resistant Coagulase-negative Staphylococci (MRCoNS) exhibited resistance to several antibiotics, including Roxithromycin, Erythromycin, and Methicillin, underscoring the importance of targeted therapy based on culture and sensitivity testing. Non-hemolytic Streptococci and *Staphylococcus aureus* showed broad antibiotic sensitivity, providing reassurance for clinical management, though resistance to Roxithromycin in Non-hemolytic Streptococci raises concerns about macrolide resistance.

First-line antibiotics considered safe in pregnancy include Ampicillin/Sulbactam, Cephalexin, Cefuroxime, and Amoxicillin/Clavulanic Acid for UTIs, respiratory, skin, and intra-abdominal infections. Ceftriaxone is preferred for severe infections, while Vancomycin is reserved for MRSA. Meropenem, Imipenem, Linezolid, and Aminoglycosides (e.g., Gentamycin) require caution due to toxicity risks. For ease of interpretation, an antibiogram of the sensitivity patterns was prepared to guide clinical decision-making.

This study highlights the importance of targeted antibiotic therapy in pregnancy, considering the prevalence of key pathogens and their resistance patterns. Routine microbiological screening and culture-based sensitivity testing are essential for optimizing treatment and minimizing resistance. Further studies with larger sample sizes and a broader range of pathogens are needed to enhance our understanding and improve clinical practices.

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#### 5. Conclusion

This study highlights the microbiome diversity and antibiotic sensitivity patterns of vaginopathogens in pregnant women, suggesting the need for targeted antibiotic therapy to optimize maternal and neonatal health. Among the

identified bacterial species, *Klebsiella* species were the most prevalent, followed by Coagulase-negative *Staphylococci*, *Beta-haemolytic Streptococci*, and *E. coli*. Sensitivity analysis revealed that *Ampicillin/Sulbactam*, *Cephalexin*, and *Cefuroxime* remain effective first-line treatment options in pregnancy, whereas resistance was notably high for *Cefpodoxime* and macrolides such as *Roxithromycin* and *Erythromycin*. The presence of methicillin-resistant CoNS (MR-CoNS) further emphasizes the need for vigilant antimicrobial stewardship.

Given the growing challenge of antibiotic resistance, empirical antibiotic selection should be guided by local susceptibility patterns to ensure both maternal and foetal safety. While broad-spectrum antibiotics like *Meropenem* and *Imipenem* should be reserved for resistant infections, drugs like *Linezolid* and aminoglycosides should be used cautiously due to their potential risks during pregnancy. The findings from this study reinforce the necessity of routine microbiological screening and sensitivity testing in obstetric care to enable precise and effective treatment strategies, ultimately improving pregnancy outcomes

### *Limitations*

The small sample size of 75 pregnant women may not provide enough data to generalize the findings to a larger population, and a larger cohort could yield more robust conclusions. Being a single-centre study conducted in a tertiary care hospital in Kerala, the results may not be applicable to other regions with different demographics, healthcare practices, and environmental factors. Additionally, the six-month duration of the study may not capture seasonal variations in vaginal microbiome diversity and antibiotic resistance trends, and a longer study would provide more comprehensive insights. The study relied on culture-based identification, which may not detect fastidious or low-abundance bacteria, and molecular techniques such as PCR or 16S rRNA sequencing could have provided a more detailed understanding of microbiome composition. Moreover, the study primarily focused on bacterial pathogens, potentially overlooking fungal and viral infections, which could also affect pregnancy outcomes. The potential impact of prior antibiotic use by the participants was not considered, which might have influenced microbiome diversity and resistance patterns. Finally, while the study examined maternal microbiome diversity and resistance patterns, it did not establish a direct correlation with neonatal outcomes, which could provide valuable insights into the long-term impact on both mother and child.

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## **Compliance with ethical standards**

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### *Disclosure of conflict of interest*

The authors declare no conflict of interest in conducting this study, the design, data collection, analysis, interpretation, reporting its findings or publication of this research.

### *Statement of ethical approval*

The study was approved by the Institutional Ethics Committee

### *Statement of informed consent*

Written Informed consent was obtained from all individual participants included in the study.

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