INTRODUCTION

Group B streptococcal (GBS) vaginal infection in pregnant women is one of the causes of neonatal infections, leading to increased mortality due to mother-to-child transmission. This transmission almost occur only during labor or rupture of membranes [1].

GBS vaginal infection during pregnancy can cause serious infections for mothers and babies. For mothers, GBS infection increases the risk of membranes infection, premature rupture of membranes, urinary tract infection, sepsis, preterm birth, *postpartum endometritis*. For babies, GBS infection increases the risk of pneumonia, one of the leading causes of neonatal infection and a major cause of perinatal death [1]. Studies show that the prevalence of GBS infection during pregnancy ranges from 7.1% to 48.5%, such as: B. Lu (China), Claudia Reinheimer (Germany) [3], Medugu (Nigieria), K. le Doare (Gambian) ... [4],[5],[6].

In Vietnam, the annual *perinatal mortality* is about 0.95% (accounting for 50-70% of the deaths under one year of age) for many reasons, including GBS neonatal infection [7],[8]. Most studies have not yet gone into the analysis of related factors as well as prophylactic treatment to avoid neonatal infection [9],[10].

There are more than 10,000 births at Nghe An Maternity Hospital each year, therefore we conducted: ***"A study on the situation of group B streptococcal infection in pregnant women, and effectiveness of intrapartum antibiotic prophylaxis in Nghe An Maternity Hospital (2018-2019)"*** with two objectives:

1. To describe the situation, serotype distribution and some related factors of group B streptococcal infection during 35 – 37 weeks of pregnancy at Nghe An Maternity Hospital, 2018 - 2019.

2. To evaluate the antimicrobial susceptibility, and the effectiveness of intrapartum antibiotic prophylaxis against Group B streptococcus.

NOVELTY, SCIENTIFIC AND PRACTICAL SIGNIFICANCE OF THE THESIS

1. Novelty

This is the first time GBS serotype distribution has been identified using PCR and sequencing.

2. Scientific significance

The study was designed by descriptive method with analysis, intervention research and laboratory research with modern PCR techniques and gene sequencing. Data were entered and analyzed using highly reliable softwares.

3. Practical significance

The study revealed the prevalence of GBS infection among pregnant women in Nghe An, and evaluated the effectiveness of antibiotic treatment for mothers with the purpose of preventing mother-to-child GBS transmission, which might be widely applied in the local.

THESIS STRUCTURE

The thesis consists of 124 pages divided into the following sections: Introduction (2 pages), literature review (31 pages), study subjects and methods (30 pages), results (26 pages), discussions (26 pages), conclusions (2 pages), and recommendations (1 page). There are 41 tables, 16 figures, 97 references and 2 appendices.

Chapter 1:
LITERATURE REVIEW

1.1. Overview of Group B Streptococcus (GBS)

Group B Streptococcus (GBS) is an aerobic bacterium, spherical or oval shaped with an average diameter of 1μm (usually 0.5 - 1 x 1 - 2 μm), gram-positive and unmoved. The bacteria in pairs or chains can have ≥ 50 cells per chain, and they divide in a plane perpendicular to the axis of the chain. Although GBS is a gram-positive bacterium, it does not form spores when exposed to adverse environmental conditions. The genome structure of GBS consists of 2,211,485 double bases and encodes for 2,118 proteins. GBS has 10 different serotypes (Ia, Ib, II, III, IV, V, VI, VII, VII, IX) [13].

- Colonization of group B Streptococcus: In humans, Group B Streptococcus mainly colonizes in the vagina and rectum. The rectum serves as the natural reservoir for GBS, from which Group B Streptococcus easily spreads into the vagina [9],[19].

1.2. Diagnosis methods for Group B Streptococcal infection

- Antigen/antibody tests: Antigen/antibody tests are rapid, inexpensive but not highly sensitive.

- Polymerase Chain Peaction – PCR: PCR has a sensitivity of 97%, a specificity of 100%, giving results within 30-40 minutes but the cost is very high so it is difficult to widely apply [21],[22],[23].

- Culture: Culture is the best method to screen GBS [1]. Specimens should be taken in both the vagina and rectum. CDC guidelines will help to effectively screen GBS in pregnant women, avoiding omissions and saving costs [1],[24],[25].

1.3. Studies on Group B Streptococcal infection

1.3.1. Studies in Vietnam

There have not been many publications on GBS infection during pregnancy in Vietnam, especially international publications. The studies mainly focus on determining the prevalence but preventive measures.

Nguyen Thi Ngoc Khanh (2001) with a study on over 602 pregnant women at the *Institute* for *Protection* of *Mother* and *Newborn* found that 4.5% of the women were infected with GBS [50]. In 2006, the study "The prevalence of vaginal and rectal group B Streptococcus in pregnant women and some related factors" conducted on 200 women laboring and giving birth by Do Khoa Nam at Tu Du Hospital showed that 17% of the women were infected with GBS [19]. In the study "The prevalence of mother to child GBS infection and related factors" conducted on 376 pregnant women by Nguyen Thi Vinh Thanh at Tu Du Hospital, specimens were taken from both the vagina and rectum during 35 - 37 weeks of pregnancy, and the prevalence of GBS was 18.1% [9]. Another study conducted on 2,154 pregnant women by Tran Quang Hiep in Bach Mai Hospital in 2011 revealed the prevalence of GBS was 6.5%, and specimens were only taken from the vagina during 34 to 36 weeks of pregnancy, which is earlier than recommended by CDC [10].

1.3.2. Studies in the world

- In the world:

Seal Ac (2015) estimated that 319,000 babies were infected with GBS, including about 205,000 with early onset and 114,000 with late onset. There were 90,000 newborn deaths under 3 months old, and 7,000 with neonatal encephalopathy. About 3.5 million premature births might be due to GBS. The vast majority of pregnant women and newborns infected with GBS came from Africa. Vaccines for mothers were 80% effective. The use of vaccines might prevent stillbirths and neonatal deaths [51].

***Europe*:**

A study conducted on 438 GBS infected babies by C. Joubrel in France in 2015 showed 174 cases of early onset GBS

disease and 264 cases of late onset GBS disease. The women were screened for vaginal cultures at 34 to 38 weeks of pregnancy injected with prophylactic antibiotics. As a result, the prevalence of neonatal GBS infection decreased from 1.8/1,000 live births in the 1990s to 0,26/1,000 live births in recent years [36].

***Asia*:**

In China, Jichang Chen studied 3,434 pregnant women, showing that the prevalence of GBS in pregnant women was 6.1%; the rate of neonatal GBS infection was 0.7%; the rate of mother to child transmission was 7.6%; the rate of early onset GBS infection was 0.58/1,000 live births [53]. The study on 1,328 pregnant women by Mubashir Ahmad Khan (2015) in Saudi Arabia found the prevalence of GBS of 13.4%. All the cases were susceptible to penicillin G, ampicillin and vancomycin [54].

***America*:**

A study on 179,818 live births by Victoria Parente in the US revealed 492 babies with early onset GBS infection, the age of mothers under 18, and black people at a high risk of GBS infection [55].

***Africa*:**

In South Africa, Lucia Matsiane Lekala studied 340 women at 35 - 37 weeks of pregnancy, showing that the prevalence of GBS infection was 48.2%. This rate was higher in low-educated women, women with a history of abortion or stillbirth and with HIV/AIDS [58].

1.4. Factors associated with group B streptococcal infection

About 15 - 40% of women have vaginal and rectal GBS but do not show clinical symptoms [64]. There are many factors affecting the prevalence of GBS in pregnant women, including knowledge, practices, residence, etc.

- Knowledge and practices of GBS prevention during pregnancy is important to prevent neonatal infections [10], [19].

- Residence: There is no consistency between GBS infection in pregnant women and their residence [29],[71]...

- Number of pregnancies: The association between GBS infection and the number of births has still been a controversial issue in some other studies [18].

- Water sources: Tap water is considered hygienic. 65% of the tap water samples meet the standards because it is treated, while other untreated water sources may be a factor to increase the risk of GBS infection [75].

- History of abortion: Pregnant women with a history of abortion are at a higher rate of GBS infection [10].

- Hygiene habits: There are still some unscientific customs in the society such as abstaining from baths during pregnancy and postpartum period, thus increasing the risk for bacteria to develop on the skin as well as in tracts.

- GBS infection in the previous pregnancy: As recommended by CDC, the women infected with GBS from the previous pregnancy will be given a prophylactic antibiotic shot without being screened this time [2].

1.5. **Antibiotic prophylaxis** for GBS infection

- All pregnant women should be screened for culture to detect GBS infection in the vagina and rectum during 35 to 37 weeks of pregnancy. When *rupture* of membranes occurs, they will be administered prophylactic antibiotics. It should be noted that screening is only valid during such pregnancy, which also means that those with GBS infection during previous pregnancies are not always administered intrapartum antibiotic prophylaxis for the current pregnancy [28].

Chapter 2:

STUDY SUBJECTS AND METHODS

2.1. Study method for Objective 1

Describe the situation, serotype distribution and some related factors of group B streptococcal infection during 35 – 37 weeks of pregnancy at Nghe An Maternity Hospital, 2018 - 2019.

2.1.1. Subjects, location, and duration of the study

- ***Study subjects:***

+ Descriptive study: Pregnant women at 35 to 37 weeks of pregnancy who had their pregnancy checked up and managed at Nghe An Maternity Hospital from 2018 to 2019.

+ Laboratory research: culture and serotype samples

Inclusion criteria: Gestational age of 35 to 37 weeks, no vaginal drugs or use of antibiotics within 48 hours before the examination and participation, consent to participate in the study and consent to follow the research process, positive and clean bacterial cultures (+), typical colonies of GBS in the selective medium (Strep B and Todd Hewitt medium).

Exclusion criteria: Gestational age outside the range of 35-37 weeks, use of antibiotics or vaginal medication in the past 48 hours, and unwilling to participate in the study.

- ***Study site:***

+ Descriptive study: At Nghe An Maternity Hospital;

+ Laboratory research: Determination of serotypes and bacterial species was conducted at the high-tech laboratory of Military Medical Academy

- ***Study duration:*** From 3/2018 to 8/2019.

2.1.2. Study method

Descriptive and analytical prospective study, and laboratory research

2.1.2.2. Study contents

- Analytical descriptive study was to identify the situation of GBS infection during 35 to 37 weeks of pregnancy at Nghe An Maternity Hospital, including: The overall prevalence of GBS infection; the prevalence of GBS by age, by occupation, etc.

- After obtaining positive cultures, PCR technique was employed to determine the serotype.

- Some factors related to GBS infection in 35-37 week pregnant women at Nghe An Maternity Hospital were analyzed, including knowledge about genital tract infections; genital hygiene practice; water sources for living….

- ***Sample size:***

+ Sample size for prospective descriptive study: The sample size is calculated using the following formula [76]:

$$n = Z^2_{1-\alpha/2} \frac{(1-p)}{p\varepsilon^2}$$

Where: n: sample size; $z_{(1-\alpha/2)}$: Confidence coefficient, at the confidence level of 95% $z_{(1-\alpha/2)}$ is 1.96; p: expected prevalence of GBS infection (according to Nguyen Thi Vinh Thanh it is 18.1%) [9]. ε: relative error, choose ε = 0.16. Replacing the corresponding values in the above formula, the calculated sample size n = 745.

+ Sample size for laboratory research: 69 positive GBS cultures of vaginal swabs from 750 pregnant women.

- ***Sampling:*** All pregnant women eligible to participate in the study and positive GBS cultures were selected until sufficiency of the sample size.

2.1.2.4. Study variables and indicators

- ***Variables:*** age of pregnant women, residence, occupation, number of births, history of GBS infection, GBS identification results by PCR, serotype, related variables for GBS infection, variables of prophylactic treatment outcomes.

- ***Techniques used in the study:*** Interview technique, clinical examination techniques [34], sample collection technique [26], culture and microbiological identification

technique [15], serotype identification technique by molecular biology [77].

2.1.5. Data processing

Collected data were processed using SPSS 20.0 software to compare the percentages among the research groups to find statistically significant differences.

2.1.6. Errors and error control

To control errors, we followed the research process, had training before getting started, and cleaned the data before processing.

2.1.7. Ethical issues

We complied with all medical ethics regulations in biomedical research. Those pregnant women who did not agree to continue participating in the study would still be checked up for pregnancy and provided guidance on prophylaxis to prevent GBS infection during labor or rupture of membranes.

2.2. Study method for Objective 2

Evaluate the antimicrobial susceptibility, and the effectiveness of intrapartum antibiotic prophylaxis against Group B streptococcus.

2.2.1. Subjects, location, and duration of the study

- ***Subjects:*** For laboratory research, positive GBS cultures were taken for susceptibility testing, and nasopharyngeal swabs were cultured for GBS. For intervention study, those pregnant women with GBS infection and their newborns were selected for intervention.

- ***Study site:*** the Department of Obstetrics, Nghe An Maternity Hospital

- ***Study duration:*** From 3/2018 to 8/2019

2.2.2. Study methods

Non-controlled clinical trial research and laboratory research

- ***Sample size for antibiotic testing and intervention treatment:***

+ **Sample size for antibiotic testing**: 69 positive GBS cultures in Objective 1.

+ **Sample size for intervention study**: 54/69 pregnant women were eligible to be included for evaluation of the effectiveness of antibiotic treatment.

+ **Sample size for antibiotic prophylaxis study:**

A total of 55 babies born from 54 mothers were eligible to participate in the evaluation of antibiotic treatment effectiveness for mothers and their babies.

- **Sampling**: All the mothers and babies who met the criteria were included in the study.

- **Study contents:** The susceptibility, resistance of GBS to each antibiotic, and effectiveness of intrapartum antibiotic prophylaxis.

- **Study variables and indicators:** Sensitivity and resistance to antibiotics; amniotic condition; delivery time (hours); birth weight (grams); neonatal GBS infection; GBS infection in mothers after giving birth; side effects of antibiotics; respiratory infection of babies; etc.

- ***Techniques used in the study:*** Culture technique; antibiotic injection technique; technique for taking nasopharyngeal swabs; technique for antibiotic susceptibility testing [15]

- ***Antibiotics used in the study***

Positive GBS samples would be tested for antibiotic susceptibility and antibiotic use would follow the antibiotic susceptibility pattern.

- **Data processing, and errors in the study:** Collected data were processed using Stata and SPSS 20.0. To control errors, we followed the research process, ensured the sample size, and cleaned the data before processing.

- **Ethical issues:** We complied with all ethical regulations in biomedical research. The study was conducted based on the rights of the mother and newborn.

Chapter 3:

STUDY RESULTS

3.1. Situation, serotype distribution and some related factors of group B streptococcal (GBS) infection in 35 – 37-week pregnant women at Nghe An Maternity Hospital, 2018 - 2019

3.1.1. Situation of GBS infection in 35 – 37-week pregnant women

- Clinical examination results:

Clinical examinations showed the prevalence of vaginal infection in 750 pregnant women as follows:

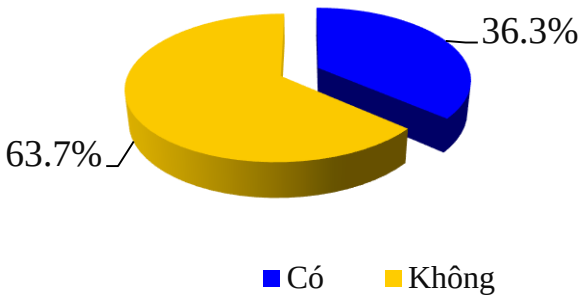

Figure 3.1. Prevalence of vaginal infection during pregnancy

Comments: The prevalence of vaginal infection in pregnant women was 36.3%.

- Clinical manifestations of vaginal infection:

Table 3.2. Clinical manifestations of vaginal infection during pregnancy (n = 750)

Clinical manifestations	**No.**	**Percentage (%)**
Vaginal discharge (1)	193	25.7
Iching (2)	90	12
Burning (3)	15	2
None (4)	452	60.3
Total	750	100
P value	(1: 2; 3) < 0.05	

Comments:

There was a difference in vaginal discharge and iching and burning pain in the vagina (25.7% vs. 12.0% and 2.0%, p < 0, 05).

- Urinary tract infection during pregnancy

Table 3.3. Prevalence of urinary tract infection during pregnancy (n = 750)

Urinary tract infection during pregnancy		No. (n)	Percentage (%)
Yes	Untreated	59	7.9
	Treated	56	7.5
No		635	84.6
Total		750	100

Comments: There were 115 cases of urinary tract infections, accounting for 15.4%.

- History of GBS infection during previous pregnancy:

296/750 pregnant women had given birth before; their history of GBS infection was determined through their medical records.

Table 3.4. History of GBS infection during previous pregnancy (n = 296)

History of GBS infection	No. (n)	Percentage (%)
Yes	6	2.03
No	290	97.97

Total	296	100

Comments: The prevalence of GBS infection during previous pregnancy was 2.03%.

- Prevalence of GBS infection among the study subjects

+ Micro*biological method*: Out of 750 studied pregnant women, 69 women underwent gram stain and CAMP test for positive (+) GBS.

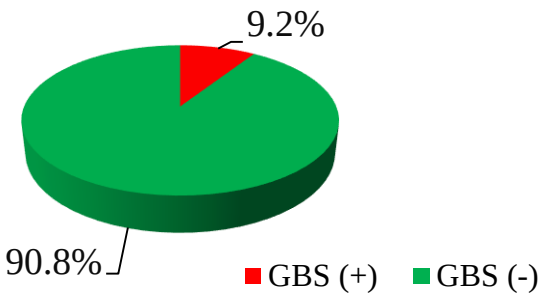

Figure 3.3. Prevalence of GBS infection through culture

Comments: The prevalence of GBS infection through culture was 9.2%.

+ Prevalence of GBS infection using *GBS- specific dltS gene*:

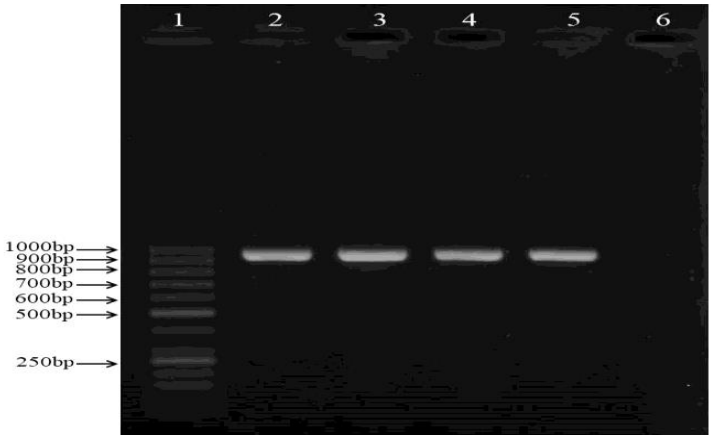

Figure 3.4. PCR products of 952bp segment of dltS gene on 1.5% Agarose gel (Well 1: Standard DNA ladder (50bp); Wells 2 - 5: GBS strains; Well 6: Negative control)

Comments:

DltS gene PCR produced a single and clear band of 952bp in size, which is consistent with the size of GBS.

- Results of GBS gene sequencing and GBS bacterial sequence registration on the genbank

All 69 GBS strains were confirmed by culture, Gram stain, CAMP test, and all carried dltS specific gene of GBS (Figure 3.4). Some representative samples were tested by dltS and 16S gene sequencing, which also resulted in GBS. These sequences were successfully registered on the gene bank with the codes from MK942595 to MK942600 and from MN095196 to MN095199 respectively (Figure 3.) and (Figure 3.6). A total of 69/69 positive GBS cultures were performed PCR to identify serotype with the results as follows:

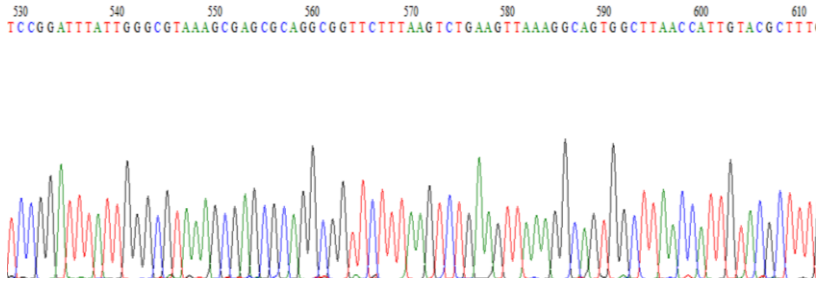

Figure 3.5. The 16S gene sequence obtained using 27F primer

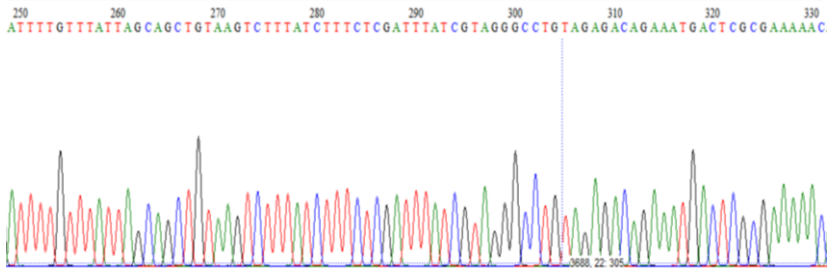

Figure 3.6. The gene segment obtained using dltS-F primer

Table 3.5. List of GBS strains and corresponding codes registered in the gene bank

No.	Strain	Target gene	Fragment size (bp)	Code in the genebank
1	GBS20	16S rRNA	1411	MK942595
2	GBS23	16S rRNA	1405	MK942596
3	GBS25	16S rRNA	1391	MK942597
4	GBS28	16S rRNA	1397	MK942598
5	GBS29	16S rRNA	1425	MK942599
6	GBS31	16S rRNA	1379	MK942600
7	GBS21	*dlt*S	952	MN095196
8	GBS26	*dlt*S	952	MN095197
9	GBS31	*dlt*S	952	MN095198
10	GBS32	*dlt*S	952	MN095199

Comments:

The 10 sequences of GBS were successfully registered and granted a code on the gene bank (Genebank, NCBI).

- Prevalence of GBS by study subjects' characteristics

Table 3.6. Prevalence of GBS infection by age group (n = 750)

Age group (years)	No. of examinations	GBS (+)		**P value**
		No.	Percentage (%)	
< 20 (1)	15	0	0	p (2: 3; 4; 5) > 0.05
20 - < 25 (2)	162	14	8.6	
25 - < 30 (3)	339	32	9.4	
30 - < 35 (4)	171	19	11.1	
≥ 35 (5)	63	4	6.3	
Total	750	69	9.2	

Comments:

There was no difference in the prevalence of GBS among age groups, p (2: 3; 4; 5) > 0.05.

- Prevalence of GBS infection by location

Table 3.7. Prevalence of GBS infection by location (n = 750)

Location	No. of examinations	GBS (+)		P
		No.	Percentage (%)	
Plain (1)	416	28	6.7	(1: 2; 3) < 0.05
Vinh City (2)	235	29	12.3	
Moutainous areas (3)	99	12	12.1	
Total	750	69	9.2	

Comments:

There was a statistically significant difference in GBS infection between women living in the lowland and those living in Vinh City and the mountainous area: 6.7% vs. 12.3% and 12.1%, $p < 0.05$.

- Prevalence of GBS by the number of births

Table 3.8. Prevalence of GBS by the number of births (n = 750)

No. of births	No. of examinations	GBS (+)		P value
		No.	Percentage (%)	
First birth (1)	454	42	9.3	(1: 2; 3; 4) > 0.05
1 (2)	208	22	10.6	
2 (3)	61	4	6.6	
≥ 3(4)	27	1	3.7	
Total	750	69	9.2	

Comments:

The highest prevalence of GBS was in the women who had given a birth before (10.6%), and the lowest was in the group with 3 births or more.

- Prevalence of GBS infection by hygiene habits

Table 3.9. The prevalence of GBS infection by hygiene habits

Hygiene habits	No. of examinations	GBS (+)	
		No.	Percentage (%)
Refraining from baths (1)	16	0	0.0
Vaginal douching (2)	47	2	4.3
Using feminine hygiene products (3)	394	35	8.9
Daily vaginal washing (4)	607	59	9.6
Using unhygienic water sources (5)	503	54	10.7
P value	(1: 2; 3; 4; 5) < 0.05		

Comments:

The prevalence of GBS infection was the highest (10.7%) among the pregnant women using unhygienic water sources.

- Identification of Group B Streptococcus Serotypes

Table 3.10. Distribution of Group B Streptococcus serotypes (+) (n = 69)

Serotype	No.	Percentage (%)
Ia	8	11.6
Ib	2	2.9
II	1	1.4
III	27	39.1
V	22	31.9
VI	8	11.6
VII	1	1.4
Others (IV, VIII, IX)	0	0.0
Total	69	100

Comments: Serotype III accounted for the highest percentage of 39.1%, and the lowest was Serotype II and VII at 1.4% for both.

3.1.2. Some related factors to GBS infection during pregnancy

Related factors to GBS infection during pregnancy included:

- Vaginal hygiene practices:

Table 3.15. Association between vaginal hygiene practices and GBS infection (n = 750)

Vaginal hygiene practices	GBS infection		Total
	Yes	No	
Proper	65	615	680
Improper	4	66	70
Total	69	681	750
OR = 1.74 CI95% (1.16- 4.36), p < 0.05			

Comments:

There was a correlation between improper vaginal hygiene practices and GBS infection (OR = 1.74, CI95% (1.16-4.36), p < 0.05).

- Association between miscarriage, abortion and GBS infection

Table 3.16. Association between miscarriage, abortion and GBS infection (n = 750)

History of miscarriage, abortion	GBS infection		Total
	Yes	No	
Yes	15	130	145
No	54	551	605
Total	69	681	750
OR = 1.177 CI95% (0.64- 2.15), p > 0.05			

Comments:

No association between miscarriage, abortion and GBS infection was found, with OR = 1.177 CI95% (0.64 -2.15), p > 0.05.

- Association between daily vaginal cleaning habit and GBS infection

Table 3.22. Association between daily vaginal cleaning and GBS infection (n = 750)

Daily vaginal cleaning	GBS infection		Total
	Yes	No	
No	25	108	133
Yes	44	573	617
Total	69	681	750
OR = 3.0 CI95% (1.42 – 7.59), p < 0.05			

Comments:

There was a correlation between GBS infection and no habits of daily vaginal cleaning, with OR = 3.0 CI95% (1.42 - 7.59), p < 0.05.

+ Water sources:

Table 3.23. Association between unhygienic water sources and GBS infection (n = 750)

Use of unhygienic water sources	GBS infection		Total
	Yes	No	
Yes	54	449	503
No	15	232	247
Total	69	681	750
OR = 1.86 CI95% (1.36 – 4.59), p < 0.05			

Comments:

GBS infection was related to the use of unhygienic water sources, with OR = 1.86; CI95% (1.36 – 4.59), p < 0.05.

3.2. Evaluation of the antimicrobial susceptibility, and the effectiveness of intrapartum antibiotic prophylaxis against Group B Streptococcus

69 pregnant women having *positive vaginal swab* results for *GBS* were tested for antimicrobial susceptibility; results were as follows:

- Antibiotic susceptibility of penicillins

Table 3.24. Antibiotic *susceptibility of penicillins (n = 69)*

Name	Susceptible		Resistant		Total
	No.	Percentage (%)	No.	Percentage (%)	
Penicillin	69	100	0	0	69
Ampicillin	69	100	0	0	69
Augmentin	69	100	0	0	69

Comments:

100% GBS samples were sensitive to penicillins.

- Antibiotic susceptibility of cephalosphorins

Table 3.25. Antibiotic *susceptibility of cephalosphorins (n = 69)*

Name	Susceptible		Resistant		Total
	No.	Percentage (%)	No.	Percentage (%)	
Cephalothin	69	100	0	0	69
Cefazolin	69	100	0	0	69
Ceftizoxime	69	100	0	0	69

Comments:

100% GBS samples were susceptible to cephlosphorins.

- The effectiveness of intrapartum antibiotic prophylaxis: From the antibiotic susceptibility pattern, we selected tenaphathin 1000mg (cephalothin)

Table 3.29. Prevalence of neonatal GBS infection (n = 55)

No. of newborns followed up	Neonatal GBS infection			
	GBS (+)		GBS (-)	
	No.	Percentage (%)	No.	Percentage (%)
55	0	0	55	100

Comments:

No newborns were infected with GBS.

- Postpartum GBS infection among the women

Table 3.30. Percentage of postpartum GBS infection among the women (n = 54)

Prepartum/ Postpartum follow-up	GBS infection			
	GBS (+)		GBS (-)	
	No.	Percentage (%)	No.	Percentage (%)
Prepartum	54	100	0	0
Postpartum	0	0	54	100

Comments:

No women were infected with GBS after giving birth.

- Side effects of antibiotics in the study

No side effects were recorded.

Chapter 4:
DISCUSSIONS

4.1. Situation, serotype distribution and some related factors of group B streptococcal (GBS) infection in 35 – 37-week pregnant women at Nghe An Maternity Hospital, 2018 - 2019

- Prevalence of GBS infection during 35-37 weeks of pregnancy:

Results obtained through the study of 750 pregnant women at 35-37 weeks showed that the prevalence of vaginal GBS infection was 9.20%. The specimens were only collected from the vagina, not from the rectum, because the vagina is a place directly connected to the uterus containing the fetus. Our culture medium was selective. The recommendations of CDC say that culture in selective media increases the detection of group B streptococci [1]. PCR with *dlt*S-specific gene and sequencing once again confirmed the results of culture, Gram staining and CAMP test which were positive for group B streptococci. According to many international researches, the prevalence of GBS in general ranges from 5% to 48.2%, usually higher in studies on pregnant black women in Africa, and lower in studies in Southeast Asia [51], [71].

- Prevalence of GBS through culture by age group

Our study revealed that the pregnant women aged 30 - <35 accounted for the highest prevalence of GBS at 11.1%, followed by those aged 25 - <30 at 9.5%, and those aged < 20 with no cases of GBS infection; the difference, however, was not statistically significant with $p > 0.05$. The median age of GBS infected pregnant women was 28.0 ± 4.3 (the highest at 44 years old, the lowest at 20 years old). The median age of GBS-free pregnant women was 27.8 ± 4.7 (the highest at 47 years old, the lowest at 18 years old). Thus, there was no association between GBS infection and the age of pregnant women.

- Prevalence of GBS through culture by residence

The prevalence of GBS infection in the study was higher in the group of pregnant women coming from Vinh City (12.3%) and those living in mountainous districts (12.1%). This percentage among the pregnant women coming from lowland districts was lower at 6.7%. This difference was of statistical significance with $p < 0.05$.

- Prevalence of GBS by number of births

Results showed that the prevalence of GBS infection was the highest among pregnant women who had given birth before (10.6%), followed by those of first pregnancy (9.3%), and the lowest in the group of giving birth ≥ 3 times (3.7%), but no significant difference was found.

- Prevalence of GBS by hygiene habits

Our results in Table 3.9 showed a statistically significant difference between factors such as abstaining from bathing, vaginal douching, using feminine hygiene products, daily vaginal washing, using unhygienic water and GBS infection, with the highest prevalence among pregnant women using unsanitary water, $p < 0.05$.

- Prevalence of GBS through microbiological testing by clinical manifestation

For the pregnant women with vaginal burning, the prevalence of GBS infection was 26.7%; with vaginal itching was 17.8%; with much vaginal discharge was 9.8%. There was a difference in GBS infection among groups of clinical manifestation.

- From 69 samples positive for GBS, their serotypes were determined by multiplex PCR. The determination of GBS serotypes not only contributes to identifying epidemiological and molecular epidemiological characteristics of GBS in Vietnam, but also a precondition for other studies to produce vaccines against GBS. In this study, serotype III accounted for

the highest proportion at 39.1%, followed by serotype V at 31.9%, serotypes of Ia, VI at 11.6% both, and serotypes II, VII at the lowest rate of 1.4%. No serotypes IV, VIII, IX were found. Our study may be the first study in Vietnam to analyze serotypes of group B streptococci in pregnant women. The identification of GBS serotypes in clinical laboratories is becoming increasingly important as this is one of the most important virulence factors and antigen determinant factors [17].

4.1.3. Some related factors to GBS infection in pregnant women

Our study results revealed a correlation between hygiene practices and GBS infection. Those women with good hygiene practices were at a lower rate of GBS infection than those with improper hygiene with OR = 1.74; CI95% (1.16 - 4.36), $p < 0.05$.

The number of vaginal washing, time of washing (after urination and defecation), the manner of washing (washing from back to front or from front to back) would affect the infection of bacteria from the urinary tract or digestive tract to the genital tract.

- Relationship between GBS infection and miscarriage, abortion: No correlation between miscarriage, abortion and GBS infection was found , OR = 1.177, CI 95% (0.64 - 2,15), $p > 0.05$. This result showed that miscarriage and abortion were not the cause of GBS infection.

- Relationship between some hygiene habits and GBS infection: No connection between GBS infection and the above mentioned hygiene habits was found. This result is similar to findings of some domestic studies such as: Do Khoa Nam, Tran Quang Hiep. The study by Do Khoa Nam at Tu Du Hospital on the relationship between some daily hygiene habits and GBS infection showed that the habit of using feminine products was not associated to GBS infection in pregnant women. Similarly, the study by Tran Quang Hiep at Bach Mai Hospital also found no connection between hygiene habits such as douching, use of feminine products and GBS infection [10], [19]].

- Relationship between daily vaginal washing and GBS infection: Our study showed the association between GBS infection and daily vaginal washing among pregnant women. Those women with the habit of daily vaginal washing were at a lower rate of GBS infection than those not with OR = 3.0 CI95% (1.42 - 7.59), $p < 0.05$. Daily washing also helps eliminate bacteria from the genital tract, which reduces the risk of genital tract infection.

- Relationship between unhygienic water sources and GBS infection: With OR = 1.86; CI 95% (1.36 - 4.59); $p < 0.05$. Those women using unsanitary water sources (wells, ponds, lakes, streams, etc.) were at a higher risk of vaginal GBS infection than those who used hygienic water sources (tap water). According to the "national standard of domestic water quality" [75] tap water is a water source where the percentage of water samples meeting hygienic standards is 50% (65.2%) higher, while other water sources have the percentage of standard water samples below 50%.

4.2. Evaluation of the antimicrobial susceptibility, and the effectiveness of intrapartum antibiotic prophylaxis against Group B Streptococcus

The results showed that 100% samples were susceptibility to penicillins, namely penicillin, ampicillin and penicillin/clavulanic acid (augmentin). 69/69 (100%) of GBS samples were sensitive to antibiotics such as cephalothin, cefazolin and ceftizoxime. Cephazolin is an antibiotic with a narrow antibacterial spectrum that is commonly used to prevent GBS infection during cesarean surgery [33]. As recommended by the Centers for Disease Control and Prevention, cephazolins

will be a replacement for penicillins when bacteria are resistant to penicillins.

- ***For newborn babies*****:** As recommended by the Ministry of Health, we preferred cephalothin (1st generation cephalosphorins) and it was approved for use by the Ministry of Health.

In our study, 55 newborns were born from 54 GBS-infected pregnant women and none were GBS-infected during postpartum testing.

For postpartum women: In our study, none of the pregnant women had postpartum GBS infection. Retesting also showed negative.

- *Neonatal infection*: In the study, 98.2% of the newborns were in a normal state (no fever, normal eating and sleeping), there was 1 (1.8%) case of fever within 48 hours of birth. We conducted a blood culture test and found that the newborn was infected with *Escherichia coli* (E. *coli*). No other infections such as dermatitis, umbilical cord infection or others were detected.

- *Postpartum infection among the studied women:* In our study, the women in a normal postpartum state (No manifestations of infection) accounted for 92.6%. There were 4/54 women with postpartum fever, accounting for 7.4%, including three cases of urinary tract infections, and one case of mammary blocked duct causing fever.

CONCLUSIONS

1. Situation, serotype distribution and some related factors of group B streptococcal (GBS) infection in 35 – 37-week pregnant women at Nghe An Maternity Hospital, 2018 - 2019

Among 750 pregnant women participating in the study, the prevalence of group B streptococcal infection during 35 to 37 weeks of pregnancy was 9.2%. Serotype III accounted for the highest rate of 39.1%, followed by serotype V at 31.9%, serotypes Ia, Ib, II, VI and VII ranging from 1.4% to 11.6%. There were no serotypes IV, VIII, and IX. Related factors to GBS infection in the pregnant women included using unsanitary water sources (OR = 1.86, CI95%: 1.36 - 4.59, $p < 0.05$); Improper hygiene practices (OR = 1.74, CI95% (1.16 - 4.36), $p < 0.05$); and no habit of daily vaginal washing (OR = 3.0 CI95% (1.42 - 7.59), $p < 0.05$).

2. Evaluation of the antimicrobial susceptibility, and the effectiveness of intrapartum antibiotic prophylaxis against Group B Streptococcus

Over 69 GBS strains isolated from 35-37 week pregnant women, antibiotic testing showed that 100% strains were susceptible to Penicillins and Cephalosphorins. 55 babies born from 54 mothers were followed up for the effectiveness of intrapartum abtibiotic prophylasis against Group B Streptococcus. The results showed that 100% of the newborns were not infected with GBS; 100% of the postpartum women were not infected with GBS; 98.2% of the newborns did not show symptoms of neonatal infection; 92.4% of the mothers had no signs of postpartum infection; and no side effects of prophylactic antibiotic administration were recorded.

RECOMMENDATIONS

1. Screening should be conducted for all women at 35-37 weeks of pregnancy. The women should be administered an antibiotic injection during labor or amniotic rupture for GBS prevention. Tenafathin 1000 (cephalothin) is a preferred choice because of its safety and effectiveness that has been approved by the Ministry of Health.

2. Hygienic water should be used to reduce GBS infection during pregnancy. Health education on how to clean the vagina during pregnancy is also needed.

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The thesis can be found at:

1. The national library
2. The library of the National Institute of Malariology, Parasitology, and Entomology

LIST OF PUBLICATIONS RELATED TO THE THESIS

1 Tran Quang Hanh, Vu Van Du, Pham Thu Hien, Tran Thi Kieu Anh (2020), Prevalence and capsular type distribution of group B *Streptococcus* isolated from vagina of pregnant women in Nghe An province, Vietnam, *Iranian Journal of Microbiology*, Vol.12, pp.11-17.

2. Trần Quang Hanh, Vũ Văn Du, Que Anh Tram, Tran Thi Kieu Anh (2020), Situation and some related factors to group B *Streptococcal* infection during 35 - 37 weeks of pregnancy at Nghe An Maternity Hospital (2018-2019), *the Journal of Practical Medicine, Vol. 4* . PP 20-24.