## นีพนก์ต้นฉบับ

# Aerobic Microbial Flora and Fungi of the Cervix in Asymptomatic Late Pregnancy

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ได้ทำการเพาะ เชื้อซึ่งค้องการ อ๊อกซิ เจน จากบริเวณ ปากมดลูก ของหญิง คั้งครรภ์ แก่ที่ไม่มีอาการโรคปากมดลูกอักเสบจำนวน 897 ราย แยกได้ เชื้อทั้งหมดจำนวน 1104 สายพันธ์ เป็นเชื้อแกรมบวกรูปกลม 502/1104 สายพันธ์ (ร้อยละ 45.5) แกรมบวก รูปแท่ง 357/1104 สายพันธ์ (ร้อยละ 32.2) แกรมลบรูปแท่ง 151/1104 สายพันธ์ (ร้อยละ 13.7) ยีสต์ 94/1104 สายพันธ์ (ร้อยละ 8.5)

เชื้อแกรมบวกรูปกลมที่พบมากที่สุดได้แก่สแตพีโลคอคคัส อิพิเดอร์มิดิสจำนวน 402/897 ราย (ร้อยละ 44.9) แกรมบวกรูปแท่งพบมากที่สุดเป็นเชื้อที่วิเคราะห์ ไม่ได้จำนวน 183/897 ราย (ร้อยละ 20.5) แกรมลบรูปแท่งพบมากที่สุดได้แก่ ฮีโมพีลัส วาจัยนาลีส จำนวน 95/897 ราย (ร้อยละ 10.6) ยีสต์ที่พบมากที่สุดได้แก่ แคนดิด้า แอลบิแคนส จำนวน 74/897 ราย (ร้อยละ 8.2) และพบเชื้อสเตรปโตคอคคัส กรุ๊ปบี 23/897 ราย (ร้อยละ 2.6) ได้ทำซีโรไทฟของสเตรปโตคอคคัส กรุ๊ปบี 20/23 สายพันธ์ เป็นไทฟ Ib=5, III=4, III R=4, II R=2, Ic=1 และ นอน-ไทพาเบิล=4

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#### Introduction:

Published information regarding microbial flora of the cervix is rare in Thailand. The study reported here in preliminary form was undertaken to determine the range of organisms present in cervix during the third trimester of pregnancy. All of them were asymtomatic. No attempt was made to find anaerobic bacteria.

The flora of vagina and cervix has been implicated in causing recurrent infections of the urinary tract. In pregnancy it is also important. Passage through an infected birth canal, premature rupture of membranes with retrograde infection, and tranplacental infection are possible mechanisms of infection of the infant. In many recent studies group B streptococcus has been reported to be a common causative agent. (5-8)

It is necessary to determine the normal flora before one can evaluate what is abnormal. There are studies that have assessed the normal bacterial flora, but their results have differed (9-14) Determination of the vaginal and cervical flora would aid in the evaluation of

genital, neonatal and urinary tract infections. (15-18)

#### Materials and Methods:

897 pregnant women, age 15 through 45 years from the Department of Obstetrics and Gynecology Antenatal Clinic, Chulalongkorn Hospital Bangkok were chosen. All women were in third trimester of pregnancy and asymtomatic, Culture material was obtained by a cervical swab. A clean sterile metal speculum was inserted into vagina without any lubricant, and material for culture was taken from cervical os. The sterile charcoal cotton swab was inoculated immediately on a 5 per cent horse blood agar plate and a chocolate agar plate. Both of them were incubated in a CO<sub>2</sub> incubator at 35°C. The plates were examined at 24 and 48 hours respectively. and the organisms were identified.

#### Results:

Of the 897 cultures, 653 cultures were positive (72.8%). Pure culture made up 40.0% of the total culture and no growth was 27.2% (Table I). Classified bacterial groups were demonstrated in Table II

**Table 1:** The distribution of aerobic bacteria from 897 specimens in asymtomatic late pregnancy.

	No. of positive isolated		per <b>c</b> ent of	No growth	percent of
	Pure isolates	mixed isolates	positive isolates		no growth
No. of patients	359	294	72.8	244	27.2

**Table 2:** Bacteria isolated from the cervical os of asymptomatic pregnant women in third trimester.

(From 897 specimens 1104 isolates were obtained)

Organisms found	No. of	Total	
Organisms found	Mixed	Pure	Total .
gram positive cocci	318	184	502
gram positive bacilli	276	81	357
gram negative rod	99	52	151
yeast	_50	44	94
Total	743	361	1104

502 strains of aerobic gram-positive cocci were isolated (Table 3) Staph. epidermidis (or staph. albus) was the most common. Streptococci could also

be isolated both from mixed and pure isolates. In addition serotyping of Group B streptococci was identified (Table 4).

Table 3: The distribution of aerobic gram-positive cocci of 502 isolates.

organisms found:	No. of Mixed isolated	percent	No. of Pure isolated	percent
Staphylococcus albus (epidermidis)	252	79.2	150	81.6
Micrococci	23	7.2	2	1.0
Non-hemolytic streptococci	21	<b>6</b> .6	10	5.4
Group B streptococci	10	3.2	13	7.1
Staphylococcus aureus	10	3.2	9	4.9
Group A streptococci	1	0.3	0	0
Streptococcus pneumoniae	1	0.3	0	0
Total	318	100	184	100

Table 4: Serotype of group B streptococci (20/23 strains).

Serotype	No. of strains	percent
Ib	5	25
lc	1	5
II R	2	10
III	4	20
III R	4	20
Non typable	4	20
Total	20	100

Table 5: The distribution of aerobic gram-positive bacilli of total 357 isolates.

organisms found:	No. of Mixed isolates	percent	No. of Pure isolates	percent
Diphtheroid	129	46.8	54	66.7
Gram+ve rod	126	45.6	22	27.1
Bacillus sp.	21	7. <b>6</b>	5	6.2
Total	276	100.0	81	100.0

Table 6: The distribution of gram negative bacilli of 151 isolates.

Bacteria	No. of Mixed isolates	percent	No. of Pure isolates	percent
Hemophilus vaginalis	58	58.6	37	71.2
(Corynebacterium vaginale)	Ì			
Acinetobacter sp.	19	19.2	11	21.2
Escherichia coli	15	15.2	2	3.8
Pseudomonas sp.	2	2.0	1	1.9
Klebsiella sp.	2	2.0	-	-
Alkaligenes	2	2.0	-	-
Pseudomonas aeruginosa	1	1.0	-	-
Enterobacter			1	1.9
Total	99	100.0	52	100.0

Table 7: Candida of total 94 isolates (9.5% of total specimens)

Yeast	No. of Mixed isolates	percent	No. of Pure isolates	percent
Candida albicans	39.	78	35	79.5
Candida species	11	22	9	20.5
Total	50	100	44	100.0

#### Discussion:

Most pregnant women have a vaginal discharge which may be physiological or pathological. Normally viscoelastic secretions of the cervical glands act as a mechanical and biological barrier against intruding organisms (19,20) Many studies suggest that the gastrointestinal tract may be the primary site of colonization in pregnant women who habor the organisms in the vagina (21,22). In general, there are less microbial types isolated from the cervix than from the vagina (28), but the flora were similar both in vagina and cervix qualitatively and quantitatively as reported by Bartlett and co-workers (24)

It has been found that the aerobic gram positive cocci are the most prevalent organism isolated (Table II). From Table III. 81.6% and 79.2% are Staph. albus (epidermidis) isolated in pure and mixed culture respectively. The high prevalence of Staph. albus has also been noted in several studies and it is definitely not contaminated from skin (10,18,24) Staph. aureus in this study is found in pure culture 4.9% whereas 3.2% in mixed culture. In Corbishley's study 17% of S. aureus are found at the cervix but stated that the origin of all the staphylococci is probably the flora of the skin (28).

Streptococci could be isolated in high number in normal pregnant women. Only 7.1% of colonization of group B streptococci are isolated in

the present series. If selective media were used in this study the recovery rate of group B streptococci should be doubled. (29) Incidence of group B streptococci have been reported to vary from 2-30% in healthy pregnant women. Serotyping of group B Streptococci in Table IV showed the serotype III strains accounted for 40% of the isolates and it is the predominant serotype isolated, whereas Type Ib, Ic and IIR accounted for 25, 5 and 10% respectively. 20% were non-typable. Serotype III strains cause the "late onset" type of infection in newborn.

Other streptococci isolated are in small number. Usually their significance are in postpartum and post-abortal sepsis similar to group B streptococci.

From Table V it is evident that gram positive rods are predominant (66.7% and 46.8% in pure and mixed culture). No facilities to identify further, Among the gram positive rods lactobacilli mostly predominate in the flora. Other surveys show incidences of lactobacilli varying between 50%-92% (10, 12, 13, 42) Diphtheroids isolated are 27.1%. It was reported between 13% to 75% from other series (10,13,43)

For gram negative bacilli, the majority of the organisms isolated is corynebacterium vaginale (Hemophilus vaginalis) as demonstrated in Table VI. 71.2% of cases are colonized with C. vaginalis in pure culture and 58.6% in mixed culture. McCormack and associates

noticed that women who had been pregnant were more likely to be colonized with C. vaginale than non-pregnant women, and C. vaginale did not appear to be associated with an abnormal vaginal discharge. However Gardner. described the clinical characteristics of C. vaginale vaginitis that it is the most benign of the three common vaginal infections: Candidiasis, trichomoniasis and C. vaginale vaginitis. It is a venereal disease and one of the most contagious.

Acinetobacter sp. are isolated in 21.2%. It is a potentially pathogen. Coliforms (lactose-fermenting) i.e. E. coli and enterobacter are obtained in 3.8% and 1.9%. They can be found in higher number. Furthermore enteric organisms are found most frequently in the younger age group.

Candida albicans and candida species, considered not to be in normal flora, (48,49) are positive in 94 of 897 subjects (9.5%). 74 from 94 cultures (78.7%) grown Candida albicans. Candidiasis is also venereally transmitted and as mentioned before its primary site of colonization is in the intestinal tract. Abortion associated with intrauterine infection by candida albicans has been reported recently.

#### Conclusion

Similar studies have been carried out in many countries in the past and

have provided the medical communities of those countries with the list of bacteria prevalent in the cervix in the third trimester of pregnancy. These bacteria can then be considered as the most likely candidates for study when investigating maternal, prenatal and neonatal infections. It was decided to perform this study in Chulalongkorn Hospital to make Thai physicians familiar with these significant strains of bacteria as they appear in pregnant women in our country. In the end it was discovered that the composition of cervical flora in the Thai pregnant women studied was extremely similar to that found in women in other parts of the world.

Finally, one aspect of the results which should recieve special attention here is the discovery of group B streptococci. Although this identification has been made in previous studies it takes on an added significance today due to the recent knowledge that this strain is responsible for neonatal septicaemia and meningitis.

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