Transmission of Mother’s Microflora to the Newborn at Birth

Key Words
Microflora
Vagina
Delivery
Newborn
Colonization

Abstract
Our aim was to study the initial microbial colonization of the newborns by comparing it with their mothers’ vaginal microflora. Nineteen mother-newborn pairs were examined at delivery. We found a close association, both qualitative and quantitative, between the individually different microflora of a mother’s vagina and that of her newborn. The degree of contamination of the newborn significantly correlated with the counts of microorganisms found in the vagina of mothers. In 85% of investigated individual mother-newborn pairs we revealed similar predominant microorganisms. There were no cases of the mothers and their newborns harbouring similar potentially pathogenic prevailing microorganisms.

Introduction

The sudden passage of a sterile fetus through a complex microbial world persistent in vagina makes it crucially important from the point of view of the child’s health what kind of microbes contaminate it first and begin to multiply on it [1–3]. Vaginally delivered newborns evidently acquire their initial microorganisms from vagina, gastrointestinal tract and skin of their mothers [4–6]. Besides the beneficial microorganisms of the indigenous microflora (like mainly lactic acid bacteria), the newborn usually gets also from its mother some opportunistic pathogens, such as β-haemolytic Streptococci, coliforms and Clostridia [3, 7–9]. Unfortunately, there are no data showing what microecological relations exist between the beneficial and opportunistic microorganisms in healthy newborns at the moment of delivery.

Several authors have presumed that the modern obstetrical practice during birthgiving (including treatment of the genital tract with disinfectants) may alter the quantitative composition of mother’s microflora and cause a delayed and deficient colonization of the neonate by indigenous microflora [1, 5]. However, only a few investigators have compared the predominance patterns of a mother’s vaginal microflora with her newborn’s microbiota immediately after birth [3].

The aim of the study was to investigate the initial microbial colonization of a newborn
and compare its qualitative and quantitative parameters with those of its mother's vaginal microflora during birthgiving.

Material and Methods

Subjects
In Tartu University Maternity Hospital, Tartu, Estonia, 19 consecutive mothers (aged 19–35) hospitalized for delivery and their babies were recruited into study. The infants were full-term and vaginally delivered, in 5 cases PROM was observed. At admission and shortly before delivery, vaginal and perineal douching application with a chlorhexidine solution (0.05%) was performed. All mother-newborn pairs were examined immediately after delivery.

Specimens
For newborns, the specimens were obtained from the external ear canal. For the mothers, the material was taken from the lateral side of the internal third part of the vagina. To maintain the viability of fastidious microorganisms, the specimens were collected using blood-thioglycollate-agar-coated cotton-wool swabs [10]. The swabs were put into tubes containing carbon dioxide and sealed with rubber stoppers. The specimens were sent to the laboratory within 2 h of collection.

Culture of the Specimen
The swabs were shaken in 2 ml of prereduced phosphate buffer (pH 7.2) under a gentle stream of oxygen-free CO₂. Serial dilutions 10⁻¹,... 10⁻⁵ of the material were prepared. The dilutions (0.01 ml) were subsequently seeded into different media, such as the prerduced blood-thioglycollate-agar medium, handled as modified roll tubes for anaerobic microorganisms [11], freshly prepared blood agar with 5% human blood for aerobic microorganisms, lactobacilli- and streptococci-selective MRS-4 agar [12], Endo agar for coliforms, and Sabouraud agar for yeasts. The blood agar, Endo and Sabouraud media were incubated aerobically at 37°C for 48–72 h. The MRS-4 medium was incubated in 10% CO₂ for 72 h and the roll tubes at 37°C for 72–120 h.

Identification of Isolates and Enumeration of Microbes
All the aerobes and facultative microorganisms were identified by using standard methods up to the genus level. The anaerobes were identified on the basis of their colony and cellular morphology by Gram stain [13, 14], and their absence of aerotolerance on blood agar and MRS-2 [12]. As a result, we identified Gram-positive anaerobic rods as Bifido-, Propioni- and Eubacteria, or Actinomyces, Gram-positive anaerobic cocci as Pepto- or Peptostreptococci, Gram-negative anaerobic cocci as Veillonella and Gram-negative anaerobic rods as Bacteroides or Fusobacteria. Lactobacilli were identified on the basis of their colony and cellular morphology and absence of catalase production [12]. We were unable to identify 4 of the isolated microbes and they were described as aerobic Gram-positive or Gram-negative coccobacilli. In each sample, the total count of microorganisms per swab was calculated. The density of bacterial growth was characterized by 2*: high and low, i.e. either more or less than 100 CFU/swab. The relative distribution of microorganisms was calculated as percentage of the total count of microbes. For each microbiocenosis in which the density of bacterial growth was high, the predominance pattern was determined by differentiating the predominant and subordinate microbes. We considered as predominant such microbes whose relative proportion in the total count of microorganisms exceeded 10%.

Results

Number of Microorganisms
Various aerobic and anaerobic microorganisms were isolated from all the mothers studied and from 15 babies. In 4 babies we could not detect any of the microorganisms under examination. The distribution of microbial counts in the external ear canal of newborns resembled their mothers' vaginal pattern: most of them – 84% of mothers and 74% of newborns – harboured high numbers of microorganisms (table 1). When the density of microbes in the maternal vagina was low, the infants were never heavily colonized.

Occurrence and Predominance of Different Microorganisms
We isolated 16 different groups of microorganisms from mothers and 13 from newborns (fig. 1). Streptococci were the most frequent microorganisms both in the mothers' vagina

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Table 1. Total counts of microorganisms at delivery

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<th>Count of microbes (CFU/swab)</th>
<th>Number (%) of individuals</th>
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<tr>
<td></td>
<td>mother and newborn</td>
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<td>&lt;100 (low density)</td>
<td>2 (10.5)</td>
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<tr>
<td>&gt;100 (high density)</td>
<td>13 (68.4)</td>
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(47.4%) and the newborns' ear canal (36.8%). Various individually different combinations of up to 8 microbes were observed per sample in mothers and up to 6 microbes per sample in babies. We found that all the microorganisms detected in the ear of a child were present in the samples taken from its mother's perineum, but not all mothers' microbes colonized their newborn.

Comparing the predominance patterns of the microorganisms in the vaginal microflora of mothers and their children, we found Streptococci to be the most frequent predominant microbes: in 6 mothers and 5 newborns (fig. 1). As regards opportunistic microorganisms, we observed that yeasts never predominated either in mothers or in their newborns.

β-haemolytic Staphylococci occurred among the predominant microbes only in unpaired samples (1 mother and 1 newborn), thus comprising less than 25% of the total count of microbes. β-haemolytic Streptococci predominated only in 1 mother (20% of the total count) and never in newborns. We could not find any difference in predominance patterns of the mothers' vaginal or babies' ear canal microorganisms between cases with and without PROM.

Comparison of the Predominant Microorganisms in Mother-Newborn Pairs

We found one or two similar predominant microorganisms in 12 mother-newborn pairs (85%) from among these 14 pairs in which the newborn was heavily contaminated (fig. 2). Streptococci were the most frequent common predominant microorganisms, occurring in 4 mother-newborn pairs. In the remaining pairs, we found different similar predominant microorganisms: Staphylococci, Bacilli, Corynebacteria and anaerobic cocci each in 2 pairs; Bacteroides, Propionibacteria and Eubacteria, each in one pair. In addition to that, up to 4 similar subordinate microorganisms could be detected in the investigated pairs.

Discussion

This study demonstrated that, at birth, there was a great similarity, both qualitative and quantitative, between the individual microflora of mother's vagina and their newborn's ear. The degree of colonization of the newborn significantly correlated with the count of microorganisms in its mother's vagina. In most of the investigated individual mother-newborn pairs we could reveal similar prevailing microorganisms. There were no cases of mothers and their newborns harbouring similar prevailing potentially pathogenic microorganisms.

Pertaining reports offer some conflicting results. Thus, the investigation by Graham [15] has shown that only 25% of healthy neonates harbour microorganisms on their skin immediately after birth and the latter are the same as in the vagina. Ivanov and Kulish [16] claimed that the contamination of healthy neonates with microbes varied in different clinics in a range between 7 and 30%, the most frequent contaminants being epidermic Staphylococci. On the other hand, Sycheva et al. [17] found microorganisms on the conjunctiva of 83% of neonates and the microbes resembled those found on their mothers' skin. It seems that the results may depend, on the
Fig. 1. Occurrence and predominance of microorganisms in vaginal samples of mothers and ear samples of newborns. Lb = Lactobacilli, Str = Streptococci, Bac = Bacilli, Cor = Corynebacteria, Bif = Bifidobacteria, Anco = anaerobic cocci, Eub = Eubacteria, Bact = Bacteroides, Prop = Propionibacteria, Staph = Staphylococci, β-str = β-haemolytic Streptococci, Act = Actinomyces, β-staph = β-haemolytic Staphylococci, Col = coliforms, Unid = unidentified bacteria.

Fig. 2. Similar predominant microorganisms in 12 mother-newborn pairs. 100% = Total count of microorganisms per sample. For abbreviations, see figure 1.
one hand, on what area of the baby's body is studied and, on the other hand, on the methods of sampling and cultivation of microorganisms.

To quantify the mothers' and newborns' microflora, a special method of sampling with blood-thioglycollate-coated cotton-wool swabs was developed [10]. This method helps to avoid the loss of viability of fastidious bacteria. The material from the neonate's external ear canal was considered representative of the transfer of microorganisms from mother to the neonate, because this area seems to be rarely touched and it is thus hardly ever contaminated by the hands of the obstetrician. The predominance pattern of microflora was estimated only in the case of high density (>100) of microbes in a particular sample, to avoid registration of random distribution of microorganisms in scarcely colonized areas.

We revealed a significant correlation between mothers' vaginal microflora and their newborns' external ear canal microflora. At the same time, Lactobacilli frequently found in mothers were quite rarely isolated in newborns immediately after birth; however, this difference appeared to be statistically nonsignificant due to the small number of persons studied. On the one hand, this may be caused by selective attraction of neonates for adherence of Streptococci, as it was described by Long and Swenson [18] in oral mucosa of newborns. On the other hand, the results may depend on the relatively higher resistance of cocci to disinfectants as compared with Lactobacilli [19]. Consequently, the extensive vulvar cleansing during labour does not reduce the number of microorganisms, but leads to a selective transfer of maternal bacteria, unfortunately excluding Lactobacilli. Evidently, the idea of decontamination of the birth canal by modern antisepsics (chlorhexidine) before delivery [20] does not work properly.

We could not find any universal type of microflora or microorganisms characteristic of all mothers. All the microfloras studied were individually different both in qualitative and quantitative terms. Thus, even the previously described increase in counts of Lactobacilli as gestation advanced [3, 21] and similar hormonal status of women during delivery [21] could not reduce the individuality of microflora. This individuality was also common in case of the contaminated newborns and has been demonstrated while screening risk-of-infection newborns due to the contamination with opportunistic pathogens [6, 10, 22, 23]. However, all the numerous investigations have in most cases only registered either the presence or absence of potential pathogens, and their value in predicting the infectious agents is no doubt limited [24].

To overcome this drawback, we determined the predominance pattern of microorganisms in each sample, finding in most cases similar prevailing microbes in mother's vaginal and her neonate's initial microflora. It is interesting to note that we did not find in our study any mother-newborn pairs with opportunistic microorganisms (β-haemolytic Strep-ti- and Staphylococci, coliforms or Candida sp.) as prevailing microbes in both individuals.

We conclude from our study that the predominance pattern of the mother's genital microflora has significant influence on the initial microecological relations of her newborn.

Acknowledgements

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