REVIEW ARTICLE

The Threat of Aerobic Vaginitis to Pregnancy and Neonatal Morbidity

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Abstract

Aerobic vaginitis (AV) is an endogenous opportunistic infection brought about by the disruption of the normal vaginal microbiota. Its early diagnosis and treatment during pregnancy may reduce the risk of negative pregnancy outcomes. The aim of this review was to report on the aerobic bacteria most prevalent in AV and to provide evidence of the threat of untreated AV on pregnancy outcomes. More than 300 papers on preterm delivery were extracted from several research domains and filtered to include only AV-associated bacteria such as *Staphylococcus aureus, Enterococcus faecalis, Escherichia coli* and Group B streptococci and their association with adverse pregnancy outcomes. Due to the diverse sample groups, study techniques and outcomes, a meta-analysis was not conducted. The review revealed that the association of AV with adverse pregnancy outcomes has not been as widely researched as bacterial vaginosis (BV) and needs further investigation. Furthermore, the frequent misdiagnosis of AV coupled with the emerging antimicrobial resistance associated with bacteria implicated in AV and neonatal nosocomial infections pose a problem for prophylaxis and treatment to reduce the risk of maternal and neonatal morbidity and mortality. *(Afr J Reprod Health 2017; 21[2]: 109-118).*

Keywords: Aerobic vaginitis; pregnancy risks; antimicrobial resistance

Résumé

La vaginite aérobie (VA) est une infection opportuniste endogène provoquée par la perturbation du microbiota vaginal normal. Son diagnostic précoce et son traitement pendant la grossesse peuvent réduire le risque de résultats négatifs de grossesse. L'objectif de cette revue était de faire un rapport sur les bactéries aérobies les plus répandues en VA et de prouver la menace de la VA non traitée sur les résultats de la grossesse. Plus de 300 articles sur l'accouchement prématuré ont été tirés de plusieurs domaines de recherche et filtrés pour inclure uniquement des bactéries associées à la VA telles que *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli* et streptocoques du groupe B et leur association avec des résultats de grossesse indésirables. En raison des divers groupes d'échantillons, des techniques d'étude et des résultats, une méta-analyse n'a pas été menée. L'examen a révélé que l'association de la VA avec les résultats de grossesse défavorisés n'a pas été aussi largement étudiée que la vaginose bactérienne (VB) et doit faire l'objet d'une enquête plus approfondie. En outre, le diagnostic erroné de la VA y compris la résistance antimicrobienne émergente associée à une bactérie impliquée dans les infections à vent et non-néomatologiques constitue un problème pour la prophylaxie et le traitement afin de réduire le risque de morbidité et de mortalité maternelle et néonatale. *(Afr J Reprod Health 2017; 21[2]: 109-118).*

Mots clés: vaginite aérobie, risques de grossesse, résistance antimicrobienne

Introduction

Despite the advances in prenatal care and increased public awareness, adverse pregnancy outcomes still present a major public health problem worldwide. The human vagina is colonised by a very diverse microbiota which plays a protective role in health but when disrupted, can impact negatively on the reproductive health of women, particularly during pregnancy[1]. The increasing incidence of preterm delivery[2] and other complications of pregnancy such as intra-amniotic infection[3] and chorioamnionitis[4] not only affect the mother, but her foetus as well[2]. Although vulvovaginal conditions such as *Candida* vaginitis, bacterial vaginosis and trichomoniasis are commonly identified and diagnosed, less is known about

aerobic vaginitis (AV). AV has been described as a disruption of the normal vaginal microbiota with a reduction in hydrogen peroxide-producing lactobacilli in a mainly facultatively anaerobic microbiota, accompanied by an increased vaginal pH, signs of inflammation with leukocyte infiltration[5-9]. AV differs distinctly from bacterial vaginosis (BV) in its clinical features and host response with BV being a condition brought about by an imbalance and shift of the normal vaginal flora in favour of anaerobic bacteria with no signs of inflammation[6].

Although the association of BV with pregnancy outcomes has been extensively studied, not much has been published pertaining to the epidemiology and aetiology of AV and its influence on pregnancy outcomes. Little is known about treatment success in reducing adverse pregnancy outcomes in mothers with AV infection although it is suspected that treatment failure may be largely attributed to the emerging drug resistance of AV-associated bacteria.

The aim of this review is to report on the frequency of AV-associated bacteria in pregnancy and to provide evidence of the influence of untreated AV infection on pregnancy outcomes and neonatal morbidity.

Methods

Using PubMed, WSI, Research Gate, Google Scholar and World of Science, > 300 papers were screened using the keywords "preterm, vaginal endogenous infections, maternal and neonatal morbidity and mortality". For this review, only papers published between the years 2000 and 2016 were included which described bacteria commonly associated with AV (*Escherichia coli,* Group B streptococci, *Staphylococcus aureus* and *Enterococcus faecalis)* and known to pose a risk for adverse pregnancy outcomes and subsequent neonatal infection. Papers published before the year 2000 were excluded on the basis that AV was an observed but uncategorized condition in 2000 with the diagnostic criteria for AV only formally proposed in 2002 by Donders *et al*[6]. Excluded from the search were adverse maternal and neonatal outcomes due to exogenous infections and other endogenous infections such as BV.

Because of the diverse sample groups, techniques and outcomes, a meta-analysis was not conducted.

Diagnosis and treatment of AV

The appropriate diagnosis and distinction between AV and BV is crucial to their treatment and may reduce preterm birth[11]. The diagnosis of AV is based on wet mount microscopy, showing vaginal smears deficient in lactobacilli, with the presence of cocci or coarse bacilli, parabasal epithelial cells, and toxic leucocytes[9,12]. Similar to the Nugent scoring system used for grading BV, a composite AV score is obtained by grading each of the above as normal or slight (0), moderate (1) or severe (2)[5,9]. The number of points establishes the composite AV score, with the maximum score being 10[5]. Furthermore, lactobacillary grades (LBG) may range from I (increased lactobacilli, decreased cocci), through II (reduced lactobacilli with a mixed background microbiota) and III (mixed bacteria in the absence of lactobacilli)[5,13]. Parabasal cells are considered a sign of severe epithelial inflammation encountered only in moderate or severe forms of AV such as in desquamative inflammatory vaginitis[12]. This, inflammatory reaction along with the thinned vaginal mucosa are thought to contribute to the symptoms of AV[5].

Although widely accepted as efficient in diagnosing AV, the above method is time-consuming, only employs a microscopic examination and is often adapted according to available resources[14]. A commercial kit developed in China (Beijing ZhongSheng JinYu Diagnosis Technology Co., LTD, Beijing, China) demonstrated an ability to diagnose AV with accuracy and sensitivity of 90% and has been proposed as a rapid alternative for the diagnosis of AV in China[15]. AV is diagnosed if the diagnostic strip tests indicate a colour change to measure hydrogen peroxide concentration (reflecting the status of lactobacilli), leucocyte esterase activity (presence of inflammation), β-glucuronidase activity (the presence of *E. coli* and GBS) and coagulase (detecting *S. aureus*) thereby eliminating the use of microscopy in the often overcrowded outpatient clinics.

Because of differences in their microbial

aetiology, the prophylaxis and treatment of AV and BV differ. Metronidazole is commonly used for the treatment of BV[12] and because AV does not respond well to metronidazole, clindamycin is considered to be a better choice for treating pregnant women with an unconfirmed diagnosis of abnormal vaginal microbiota because it is able to eradicate both AV and BV-associated bacteria[16]. Topical application of kanamycin has also been reported[17] as well as the use of probiotics[18]. Conventional treatment for AV consists of a course of broad-spectrum antibiotics that target anaerobic and aerobic bacteria such as the aminoglycoside, kanamycin[5,19] and the quinolone, moxifloxacin[19], both of which are proven to be effective.

Maternal and neonatal risk posed by AV infection during pregnancy

The presence of an abnormality in the vaginal microbiota in early pregnancy is considered to be one of the most significant risk factors for preterm delivery, preterm pre-labor rupture of membranes (PPROM) and low birth weight[5,11]. The inability to diagnose subclinical ascending uterine infection may result in foetal infection with a neonatal mortality rate of 25%-90% due to congenital neonatal sepsis[10].

During pregnancy the vaginal biofilm, conditioned by high estrogen levels, has a good supply of glycogen and an elevated proportion of lactobacilli, reported to constitute > 90% of the microorganisms present in the vagina during childbearing age[19]. The increased concentrations of circulating estrogens, accompanied by increased numbers of parabasal cells yields infrequent reports of AV in pregnancy[5]. However, there is a clear indication that, even when asymptomatic, AV may represent a risk factor for preterm delivery[20,21] and other complications of pregnancy such as ascending chorioamnionitis and PPROM[5,22,23].

In AV, the concentrations of pro-inflammatory cytokines, interleukin – 1b (IL-1b), interleukin – 6 (IL-6) and interleukin – 8 (IL-8), are inversely proportional to LBG[5], and because these cytokines have been associated with adverse pregnancy outcomes, patients with AV may similarly be at increased risk of delivering

preterm[5,13]. IL-1b demonstrates an eight fold increase in AV compared to bacterial vaginosis (BV), while IL-6 increases fivefold in those diagnosed with AV[7,13] and is a well-recognized marker for bacterial amnionitis and imminent term and preterm delivery[13]. IL-8 is considered to play a significant role in neonatal infections with elevated serum concentrations of IL-8 observed within the first 24 hours in severe cases of necrotizing enterocolitis (NEC)[24]. NEC is described as the most serious gastrointestinal disorder in newborn infants with an incidence of 10 – 15% in those with very low birth weight[25]. Thus, the host response to AV contributes significantly to both maternal and neonatal morbidity.

At birth, an infant's gastrointestinal tract is sterile. However it rapidly becomes colonized with organisms from the mother and the local environment, with enterococci and members of the family *Enterobacteriaceae* being the predominant organisms found in the stools during the first days of life in full-term infants[26,27]. In the preterm neonates, especially those with a very low birth weight, other factors such as the underdeveloped crucial functions, the mode and environment of delivery, as well as feeding and drug therapy regimens may influence the biodiversity of the intestinal flora and increase the risk of gastrointestinal disease such as NEC[27].

Early onset neonatal sepsis (EOS) occurs within the first few days of life and remains an uncommon but significant cause of morbidity and mortality among very low birth weight (VLBW) infants[28,29], while late onset neonatal sepsis (LOS) occurs between 4-7 days of life and is associated with the morbidity and mortality of hospitalized neonates worldwide due to invasive interventions, prolonged ventilation and prolonged stay in neonatal intensive care units as a consequence of prematurity and VLBW[30].

Organisms commonly implicated in AV and associated with negative pregnancy and neonatal outcomes include *Escherichia coli*, Group B streptococci (also known as *Streptococcus agalactiae* and referred to as GBS for the remainder of this review), *Enterococcus faecalis* and *Staphylococcus aureus*[18,31,32], and when acquired during the perinatal period, may be

associated with a high neonatal mortality due to septicemia, meningitis or pneumonia[32,33].

Escherichia coli (E. coli)

E.coli accounts for 80 to 90% of AV infections in pregnant women[31] and is frequently isolated as the sole organism[5]. *E. coli* has also been more frequently reported in women who deliver preterm than those delivering full term[34,35]. The physiologic changes along with reduced immunological status during pregnancy are believed to predispose women to urinary tract infections (UTI) with *E. coli* being the most commonly associated pathogen. Specific virulence determinants in uropathogenic strains of *E. coli* are associated with invasive infection and pyelonephritis in pregnancy by facilitating their adherence to uroepithelial cells, thereby preventing bacteria from urinary lavage and allowing for multiplication and tissue invasion[31].

Some studies have reported an emerging trend of *E. coli* predominating in early neonatal sepsis following prophylaxis for GBS colonization during pregnancy[36-41]. Even though the role of *E. coli* in vaginitis is particularly controversial, it is known to alter the micro-environment[19] and remains one of the most common causes of neonatal sepsis, often being recovered from placental tissues in chorioamnionitis[5]. Neonates born preterm are more likely to develop neonatal sepsis and meningitis caused by *E. coli* compared with neonates born at term[28,29].

A study by Bizzaro *et al.*[42] revealed that there were significant increases in both EOS and LOS which could be attributed to *E. coli*. A significant increase in ampicillin-resistant *E. coli* was reported in very low birth weight infants with *E. coli* EOS, while in *E. coli* LOS, no significant change in antimicrobial susceptibility was reported[42].

Group B Streptococci (GBS)

GBS exists as normal microbiota in the female genital tract and anal areas of healthy adults[43-46], with an estimated one out of three women being a carrier of GBS. The gastrointestinal tract serves as the natural reservoir for GBS and is expected to be the source of vaginal colonization[23,43].

Pregnant women who are colonized with GBS might develop infections of the urinary tract, bacteremia, chorioamnionitis, and postpartum endometritis[23,47,48], thus increasing the risk of PTD, PPROM and perinatal transmission[49-51], resulting in neonatal sepsis and meningitis[45,52,53]. A prevalence of 7 - 25% GBS colonization in AV has been reported in women between 35 and 37 weeks of gestation[54], with intrauterine infection associated with the ability of GBS to ascend from the lower genital tract and colonize the upper genital tract[54-56]. In the newborn, GBS infection may be congenital or acquired[22, 57] and remains the foremost cause of neonatal mortality and morbidity in the world despite a recent decline in occurrence[48,52,58,59].

GBS early-onset disease (EOD) occurs at the age of 0-6 days and has been associated with the presence of GBS in the vagina of the mother with transmission thought to take place vertically through aspiration of infected amniotic fluid or passage through the birth canal[60-62], while late-onset disease (LOD), occurs at age between 7 and 90 days[48,62,63] with the source of infection attributed to community or nosocomial acquisition[62]. However, there is evidence that in several infants with LOD, the GBS causing the infection share identical serotypes as the GBS isolated from their mothers, suggesting a maternal source[48,62].

Intrapartum colonization is strongly associated with approximately 4% of neonatal fatalities and serious morbidities including sepsis, pneumonia, meningitis, illness and death in infants, as well as long-term disabilities such as loss of hearing, impaired vision, developmental problems, cerebral palsy, osteomyelitis and septic arthritis[22,58].

Staphylococcus aureus (S. aureus)

The rate of vaginal carriage of *S. aureus* has been reported to be 4% – 22% of the vaginal microbiota of pregnant women[64,65], with the presence of lactobacilli known to reduce *S. aureus* virulence[8]. The risk factors for *S. aureus* colonization in pregnancy and the association between maternal colonization and infant infections are not very well

defined[64].

S. aureus accounts for > 90 % of late-onset sepsis in neonates. Late-onset sepsis is noted to be four times higher in very low birth weight (VLBW) infants[66]. Imperative factors that are considered to increase the risk for neonates due to S. *aureus* colonization include breastfeeding, number of household members, prematurity, low birth weight and the duration of antibiotic or ventilator days[67-69].

Traditional antibiotic therapy, even though highly effective, has led to the emergence of antibiotic-resistant strains, such as methicillin resistant *S. aureus* (MRSA)[70]. Community-acquired MRSA infections have been reported to occur in neonates and pregnant women or postpartum women, as a result of vaginal colonization of S. *aureus* during pregnancy[70], with a prevalence of 0.5% - 10% and in some areas, higher rates of colonization have been reported[64,68,71].

The factors that contribute to *S. aureus* rectovaginal colonization in pregnant women are not well understood, but it is believed to include black race[64,72]. MRSA is noted as a significant pathogen in neonatal intensive care units[67], with one study reporting 1.8% of neonates colonized with MRSA and 16 of 50 (32%) of these colonized neonates developing MRSA infection[69].

Enterococcus faecalis

Lancefield group D streptococci include the enterococci now classified in a separate genus from other streptococci and non-enterococcal group D streptococci. Group D enterococci and non-enterococci are part of the genital tract, but are a common cause of urinary tract infections, subacute bacterial endocarditis, abdominal abscesses, and wound infections[73,74]. The predominance of asymptomatic genital tract infection in pregnant women has been reported in the literature[75], with enterococci causing life threatening infections in preterm infants and other immuno-compromised patients[73].

The enterococcal species which are considered to be significant pathogens for humans are *E. faecalis* and *E. faecium*. These organisms are likely to affect patients who are elderly or whose normal microbiota has been altered by antibiotic treatment[73]. Furthermore, in neonates, *E. faecalis* is associated with 6% mortality rate in early onset septicaemia (EOS) which increases to 15% in late-onset (LOS) infections, whilst in general it is implicated in 7% to 50% of fatal cases[74].

Enterococci have previously been reported to be harmless inhabitants of the gastrointestinal tract flora initially, and are included among the main causes of health care–associated infections[76], being recorded as the second most common cause of nosocomial urinary tract infections[77]. *E. faecalis* is considered to increase *tst* expression leading to increased production of toxic shock syndrome toxin-1 (TSST-1) thus increasing the virulence of *S. aureus.* Enterococci are believed to be difficult to treat because of their intrinsic resistance to antibiotics including beta – lactams and aminoglycosides which are frequently used to treat infections due to Gram-positive cocci[74,78]. Resistance to trimethoprim, gentamycin and vancomycin have also been reported[79,80].

An association was found between increased enterococcal colonization and prematurity[73], with preterm birth neonates who carry *E. faecalis* being more likely to be suffering from NEC than neonates born at term[24, 25]. The presence of *E. faecalis* in the amniotic fluid considerably increases the risk of histological inflammation of the placenta (OR 10.7, 95% CI 1.27–89.9) and also increases the risk for bronchopulmonary dysplasia (BPD) and NEC[81].

E. faecalis is described as one of the first colonizers that could suppress the inflammatory responses and also shape the immune system of neonates[24]. In the neonates, the intestines are likely to undergo acute inflammation when exposed to Gram-negative bacteria[24]. The presence of *E. faecalis* is believed to help the intestines maintain the immune balance in response to such challenges[24]. However, the therapeutic effects of *E. faecalis* must be examined thoroughly, especially as *E. faecalis* is a recognized opportunistic pathogen in hospital infections[24,78].

Conclusion

Preterm birth is a critical priority in healthcare throughout the world because of medical, social,

and economic reasons. Maintaining a healthy vaginal biofilm during pregnancy is vital to the control of opportunistic infection which may result in poor pregnancy outcomes. Optimal treatment includes antibiotics that have little consequence on the normal beneficial microbiota such as *Lactobacillus* species, while eradicating *E. coli*, GBS, *S. aureus*, and *E. faecalis* all of which have been implicated in the aetiology of AV[19,82,26]. The administration of probiotic lactobacilli employed in recent studies restores the disrupted microflora of the vagina and offers a safer alternative to the prolonged prophylactic use of antibiotics which may present with side effects[83,84].

As with most endogenous infections, species prevalence differs among the different geographical regions with some reporting *S. aureus* as the most prevalent[85,19,9,] followed by *E. coli*[85,86] *Enterococcus*[85,87] and GBS[86]. Others report *E. coli* followed by *E. faecalis*[88,89], GBS and Enterobacteriaceae[90]. There is currently no standardized treatment for AV infected mothers to reduce the rate of adverse pregnancy outcomes[18], although treatment during the second trimester of pregnancy has been advocated[10].

The effects of screening and treatment for abnormal vaginal microbiota in order to reduce preterm delivery (PTD) remain controversial[21] with low prevalence of AV reported in some studies[11,91] and others showing a very high prevalence[92,93]. Interventions to prevent pre-term birth have been largely ineffective[1,2] and may be attributed to both the increasing antimicrobial resistance amongst opportunistic pathogens which complicates adequate administration of prophylaxis and treatment as well as the often misdiagnosis of AV as BV, thus leading to treatment failures and thereby contributing to and increasing the risk of maternal and neonatal morbidity.

The paucity of studies relative to AV and pregnancy outcomes emphasizes the need for a better understanding of the epidemiology, aetiology and pathogenesis of AV in both pregnant and non-pregnant women particularly in Africa where AV has been poorly investigated and has often not been considered as a risk factor for poor pregnancy outcomes which may contribute to

maternal and neonatal morbidity.

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Contribution of Authors

E. Kaambo prepared the first draft. CWJ Africa wrote the final paper and revised the manuscript. Both authors approved the manuscript.

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