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Bacterial Vaginosis a "Broad Overview"

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ABSTRACT. Bacterial vaginosis (BV) was first reported in 1995 by Gardner and Dukes, who described the unique clinical signs and symptoms and the distinctive nature of the vaginal discharge associated with it. They also described a "new" causative organism, which they named "*Haemophilus vaginalis*", subsequently renamed *Gardnerella vaginalis*. BV is currently the most prevalent cause of infectious vaginitis among women attending for genitourinary diseases. BV has a complex microbiology. *Lactobacillus* populations, which are usually dominant in healthy women, are replaced by a polymicrobial group of organisms that includes *G. vaginalis*, anaerobic Gram-negative rods such as *Prevotella* species, *Peptostreptococcus* species, *Mycoplasma hominis, Ureaplasma urealyticum*, and often *Mobiluncus* species. Anaerobic bacteria produce enzymes, aminopeptidases, that degrade protein and decarboxylases that convert amino acids and other compounds to amines. Those amines contribute to the signs and symptoms associated with the syndrome, raising the vaginal pH and producing a discharge odor. The excessive amounts of bacteria characteristic of the syndrome attach to epithelial cell surfaces, resulting in "clue cell". Nearly half the patients report no noticeable symptoms, but many develop a characteristic copious, malodorous discharge if untreated. Results from epidemiologic studies have associated BV with serious upper genital tract infections and adverse pregnancy outcome. In particular, the presence of BV in pregnant women increases the risk of preterm delivery, and evidence is now compelling that BV is a cause of preterm delivery. The interest in potential invasiveness of *G. vaginalis* has increased. However, virulence determinats have not been studied enough. The most important therapy includes clindamycin and metronidazole.
Key words: Bacterial vaginosis, *Gardnerella vaginalis*.

RESUMEN. La vaginosis bacteriana (VB) fue reportada por primera vez en 1955 por Gardner y Dukes, quienes describieron los signos y síntomas de esta condición. También describieron al organismo causal el cual llamaron originalmente *Haemophilus vaginalis*. En 1980, Greenwood and Pickett propusieron que la bacteria fuese llamada *Gardnerella vaginalis* en honor a Gardner. Actualmente, la VB es la principal causa de infección vaginal en mujeres en edad reproductiva. Presenta una microbiología compleja en la cual los lactobacilos son reemplazados por un grupo de microorganismos que incluyen *G. vaginalis*, bacilos Gram-negativos anaerobios como *Prevotella*, *Peptostreptococcus* y otros. Las bacterias anaerobias producen aminopeptidasas que degradan proteínas y decarboxilasas que convierten los aminoácidos en diaminas. Estas últimas contribuyen a la aparición de los signos y síntomas asociados con el síndrome, como la elevación del pH vaginal y la producción de una descarga vaginal maloliente. Cerca de la mitad de las pacientes con VB no reportan síntomas, sin embargo, pueden llegar a presentar la descarga maloliente característica si no son tratadas. Estudios epidemiológicos recientes han asociado a la VB con transtornos ginecológicos como salpingitis y enfermedad pélvica inflamatoria y con complicaciones en el embarazo. El interés en el potencial de *G. vaginalis* se ha incrementado, sin embargo sus factores de virulencia no han sido bien estudiados. Los principales regímenes terapéuticos incluyen clindamicina y metronidazol.
Palabras clave: Vaginosis bacteriana, *Gardnerella vaginalis*.

INTRODUCTION

Bacterial vaginosis (BV) is currently the most prevalent infectious cause of vaginitis in women attending genitourinary medicine clinics in different countries.[41,64,96,107] The condition is characterized by a profuse, malodorous vaginal discharge. Despite this, more than one half of patients showing signs, do not have symptoms.[8, 141]

This syndrome could be the most important vaginal infection because of its recent association with serious upper genital tract infection.[57,82]

Bacterial vaginosis has a complex microbiology. *Lac-*

tobacillus populations, which are usually dominant in healthy women, are replaced by a polymicrobial group of organisms that includes *Gardnerella vaginalis*, anaerobic Gram-negative rods such as *Prevotella* species, *Peptostreptococcus* species, *Mycoplasma hominis*, *Ureaplasma urealyticum*, and often *Mobiluncus* species.[148] Overall, concentrations are 100- and 1000-fold greater for aerobes and anaerobes, respectively, than levels measured in women without BV.[134] However, the factor(s), either endogenous or exogenous, that initiate the shift in the ecology of the vagina and result in the massive overgrowth of these microbial populations are incompletely understood.[69,120]

A review of the history of BV allows not only a historical perspective but provides a better future understanding.

BRIEF HISTORY OF BACTERIAL VAGINOSIS

In 1894 Döderlein[35] published drawings of what he named "unhealthy vaginal flora" that accurately represent Gram's staining of vaginal secretions seen in patients with BV. In 1914 Curtis[31] associated black pigmented *Bacteroides*, a curved rod now known as *Mobiluncus* and anaerobic cocci to abnormal vaginal discharge.

The syndrome was virtually ignored until 1953, when Leopold[90] isolated a nonmotile, nonencapsulated pleomorphic Gram negative rod from men's urethral discharge with prostatitis and from women's vagina having cervicitis.

Until 1954, any vaginal discharge that was not due to gonorrhea, trichomonads or yeast was named "non specific vaginitis". In 1955 however; Gardner and Dukes claimed to have found the etiologic agent of the syndrome.[49] They described a new microorganism, which named "*Haemophilus vaginalis*", and a new infection was named "*Haemophilus vaginalis* vaginitis" They also described the clinical features of this syndrome that nowadays forms the diagnosis basis.

The findings of Gardner and Dukes were important to define the clinical disease and the association of at least one organism, *H. vaginalis* to the syndrome, however, they failed to appreciate the complex microbiology of BV. Now, it seems that a variety of anaerobic microorganisms together with *H. vaginalis* are associated to BV.

Gardner and Dukes originally assigned the organism could cause nonspecific vaginitis to the genus *Haemophilus*. The name was widely accepted and remained indisputable until 1961; when Lapage[87] demostrated that neither X (hemin) nor V (nicotinamide adenine dinucleotide) factors were essential for its growth, and suggested the microorganism might belong to the genus *Corynebacterium*. In 1963, Zinnemann and Turner,[150] according to their methods, concluded that *H. vaginalis* was Gram positive and proposed a specific name "*Corynebacterium vaginale*". Dunkelberg in 1969[37] added further evidence, the organism is not of the genus *Haemophilus* and endorsed the name of *Corynebacterium vaginale*.

Greenwood and Pickett[59] clarified the taxonomy of the organisms by using 104 biochemical growth test as well as a microscopic and DNA characterization test performed on many *Haemophilus* strains. They proposed the organism was renamed *Gardnerella vaginalis* in Gardner's honor.

It soon became apparent that the organism was part of the normal vaginal flora. In 1977 McComarck claimed that it was not even a marker of the syndrome[98] as verified in 1982 by Totten's group, who showed that the organism could be recovered in high numbers from a significant proportion of the normal population.[144]

The nature of the cells wall of *G. vaginalis* has been a dispute subject: the first study by electron microscopy was reported by Reyn et al.[122] They reported that in thin osmium-fixed cell sections, cells wall and septa of this organism resembled to those of Gram positive organisms. However, Criswell et al[28,29] worked with the same strain used by Reyn, determine that the fine walls structure was more typical of Gram negative organism. Their cells wall analysis revealed that only 20% of cell-wall overall weight was peptidoglycan, rather than the Gram positive organism, and also many aminoacids were found, and any diaminopimelate (DAP) were present in the peptidoglycan; therefore, they described the organism as a Gram-negative bacterium.

Their results have been refuted by Piot et al,[112] Harper and Davis,[63] and O'Donnell et al,[106] who, in contrast, found simple aminoacid profiles that including lysine. DAP, common to many Gram negative cell walls, and found in a few Gram-positive species,[63] was not found. Subsequently, the diamino acid in the *G. vaginalis* wall was confirmed as lysine,[106] thus explaining the absence of DAP.

Finally, Sadhu et al[127] concluded that *G. vaginalis* cell wall was unequivocally Gram positive in its ultrastructural characteristics and chemical composition. Their examination showed absence of an outer membrane or any other lamellar structure; therefore, cell-wall extracts made by methods specific for LPS gave negative reactions by silver staining and for endotoxin in the limulus amoebocyte lysate assay 2-Keto-3-deoxy-D-manno-2-octonoic acid (KDO), heptose and hidroxy fatty acids specific for LPS were not detected in the extracts.

They also concluded, that at the ultrastructural level the cell walls of *G. vaginalis* show Gram-positive organization, but that these structures are unusually thin in most cells thereby contributing to the mistaken assumption that they are Gram-negative.

INCIDENCE AND RISK FACTORS

The BV incidence varies in different populations studied. The lowest reported prevalence is 4% found in a

symptomatic college populations.[41] Older studies showed rates of 30% to 45% in reproductive age women in varied clinical settings.

The highest rate, 45% was reported by Gardner et al.[50] Modern reviews show a wide diversity of incidence, probably resulting from more exacting objective criteria used to diagnose the condition in women without symptoms. Rates in sexually transmitted disease clinics varies from 33% to 64%.[27,39,41,66,68] Rates in prenatal or obstetric clinics vary from 10% to 26%.[58,65,96,126] Rates of 23% to 29% have been reported in other gynecology or family planning clinics.[6,123,141] Intrauterine devices have been considered like a major BV risk factor.[2,13,60,62,73,85,89] The number of different sexual partners within the month before examination was also related directly to the ocurrence.[7,67] Other probable risk factors like; age, smoking status, abnormal Papanicolaou smears, menstrual flow days, menarche age, diaphragm use, and lifetime number of sexual partners have not been associated with BV.[2,73,75,79,107,118]

SEXUAL TRANSMISSION

The issue of weather BV is a sexually transmitted infection has not been resolved. The literature provides evidence supporting both sides of the argument. The age of women involved, history of previous genital infections, and previous sexual experience suggested that BV is sexually transmitted,[111,134] furthermore other authors have shown that the bacteria can be collected from urine and urethral scrapings from the women's male copule having BV.[38,73,111,113,143] Arguments against sexual transmission are supported by authors like Bump et al[20] that detected BV in 12% of virgin women; Linaldi et al,[91] who isolated *G. vaginalis* in 16.6% of 114 girls and teenagers, and Hammerschlag et al[61] who found *G. vaginalis* in 13.5% of 59 patients ranged in age from 1 to 15 years.

Another argument for sexual transmission is the finding by Briselden and Hillier,[17] who showed that longitudinal biotyping of *G. vaginalis* reveals women who acquire bacterial vaginosis are more likely to have *Gardnerella* strains with different biotypes than women who still had normal vaginal flora at their follow-up visits. This suggests that the *G. vaginalis* isolates recovered from women represent newly acquired strains rather than overgrowth of previously colonizing biotypes. Unequivocal sexual transmission of bacterial vaginosis remains to be proved, and niether the isolation of *G. vaginalis* nor the diagnosis of BV should be estimated absolute evidence of sexual activity or abuse.

THE ANAEROBIC BACTERIA ROLE

Facultative Lactobacilli maintain the vagina pH acid by glucose metabolism generated by glycogenolysis. Low pH inhibits the growth of some organisms, including anaerobic organisms, maintaining a higher oxidation-reduction potential. Hydrogen peroxide produced by facultative Lactobacilli also might control the growth of catalase negative organisms like anaerobes.[14] In patients with BV, lactobacilli are replaced by *G. vaginalis* and a mixed predominantly of anaerobic bacteria. Recently, in 1989, Eschenbach et al[42] reported that Lactobacilli were not only reduced but the Lactobacilli isolated in BV tend to be of species other than those found in normal women and include organisms incapable of producing hydrogen peroxide.

Mobiluncus is a fastidious curved anaerobic motile rod more useful as a marker for disease than is *Gardnerella.*[138] The organism is highly specific for BV but can be difficult to identify in wet mount examination of vaginal secretions because of its physical size.[140] The genus is divided into two species. *M. curtissi* and *M. mulieris*, representing small and large morphotypes, respectively. Because of the difficulty to isolate the organism by culture techniques, genetic probe and monoclonal antibody immunofluorescent methods of identification have been developed.[47,78,125]

The combination of *G. vaginalis* and anaerobes produces organic acids other than lactic acid as well as several amines,[33] that is, decarboxylation of ornithine yields putrescine; decarboxylation of lysine yields cadaverine and decarboxylation of choline yield trimethylamine.[15,34]

The latest has been suggested as the substance primarily responsible for the fishy odor associated to BV.[15] Furthermore, other authors have suggested that trimethylamine production results from the higher concentrations of trimethylamine oxide present in women suffering BV. In the presence of *Mobiluncus* sp. which provides the specific decarboxylase enzyme, trimethyamine production continous.[30] The polyamines may contribute to the abnormal discharge by causing exfoliation of vaginal mucosal epithelial cells.[15]

SEQUELAE

Although BV frequently produces few patients symptoms, serious infectious sequelae occur in women who have this disease. The following bacteria associated with BV are known to be potential pathogens: *Prevotella bivia*; *Prevotella disiens*, *Prevotella melaninogenica*, *G. vaginalis*, and *M. hominis*. In addition, patients with BV have greatly lowered vaginal tissue redox potential and elevated vaginal pH, both conditions known to be associated with increased infective potential.[72] There is, increasing evidence that the microorganisms may be isolated from extra-vaginal.[72,86]

G. vaginalis has been isolated from the bloodstream in obstetric patients, which indicates that the microorganism is not intrinsically virulent, but is an opportunistic pathogen, spreading to the bloodstream following trauma to genital tract tissues.[32,40,121]

In patients with gynecologic disease, microorganisms associated to BV are related to laparoscopically proved pelvic inflammatory disease, urinary tract infections, endometritis and postoperative vaginal cuff infections.[54,81,88,108,135]

In patients with obstetric disease, BV is related to preterm labor, premature rupture of membranes, chorioamnionitis and postcesarean and postpartum endometritis.[26,44,70,104,117]

The mechanism involved in maternal genital infection and onset labor preterm and chorioamnionitis and preterm premature rupture of membranes are not clear. Bacterial lipase and protease could reduce the chorioamniotic membrane stregth, leading to rupture. High phospholipase A_2 production was detected to *Bacteroides*, anaerobic streptococci, *Fusobacterium* and *G. vaginalis*.[10,14] Phospholipase A_2 initiates prostaglandin production by releasing arachidonic acid from its esterified from *B. fragilis*. May increase also, the synthesis of prostaglandins in membranes. Membrane colonization chorioamnionitis, and an inflammatory response also may be precipitating factors in prostaglandin synthesis and labor initiation.[10] Altough the relation between BV in pregnancy and premature labor requieres confirmation.

PATHOGENICITY

Despite the great number of works done on *G. vaginalis* during the last 30 years, its pathogenic and epidemiology role remains confusing and controversial. Several studies have been carried out to look for epidemiological markers, using biotyping in order to clarify this confusion. Among all these works, those of Benito,[11,12] Ison,[77] Pandit,[109] and Piot[115] are the most relevant. They concluded that there were no differences in biotypes between strains isolated from patients with and without BV. In México, using the scheme proposed by Benito et al[12] and doing a modification in the interpretation González P. et al[55] determined a group of biotypes associated with BV.

Microbial adherence to epithelial cells is necessary for successful colonization both by members of the flora and bacterial pathogens,[9,51] selectivity of adherence may explain pathogens tropism for certain tissues.[52]

The finding of epithelial cells coated with bacteria in the vaginal discharge of BV was first reported by Gardner and Dukes.[49] These cells were called clue cells. The finding of clue cells has since been a common feature of many studies of BV.[16,137] The presence of cells coated with Gram variable bacilli seems to indicate an association with adhesive strains of *Gardnerella* spp.[129]

The adhesive properties of *G. vaginalis* has been investigated using human red blood cells[76] and vaginal epithelial cells.[95,133,146]

Nevertheless, Scott et al[130] showed that separated adhesin receptor systems were involved in the attachment of *G. vaginalis* strains to human red cells and to an epithelial cell line (McCoy). The same author suggested that the adherence of *G. vaginalis* to the epithelial cell line seems to be mediated by an outer fibrillar coat while adherence to red cells appeared to be mediated by fimbriae.[131]

In recent years, the interest in potential invasiveness of *G. vaginalis* has increased. However, little is know its virulence determinants. The hemolysin (Gvh), released in the culture broth and responsible for the beta-hemolysis on human blood agar plates, is likely to represent an important factor in the pathology.[83]

Cauci et al,[23] demostrated that Gvh is a pore-forming protein and its damaging action depends on the amount of cholesterol and of negatively charged phospholipids in the target lipid bilayer. Moreover, a specific immune response against the toxin was documented in the vaginal mucosal fluid of patients with recurrent BV[25] and the functional properties of Gvh have been compared with those of *Clostridium perfringens* theta toxin (pfo) and *Escherichia coli* haemolysin (Hlya).[24]

McGregor et al[99] and Briselden et al,[18] associated the mucinase and sialidase activities (neuroaminidases) with BV. These enzymes may directly injure intrauterine tissues and mediate preterm labor or membranes rupture. Bacterial sialidases also decrease collagen synthesis in fibroblasts.[1]

DIAGNOSIS

The symptoms of BV include a profuse, milk like, homogeneous discharge that loosely adheres to the vaginal walls and an odor that is most evident after sexual intercourse.[2,41,142] After the original description of BV,[49] others determined the clinical criteria for diagnosis.[2] At least three of the following four criteria must be fulfilled to establish the diagnosis: A grey-white homogenous discharge; a pH 4.5; a fishy amine odor on mixing the discharge with 10% potassium hydroxide; and clue cells.[2] More recently, it has been demonstrated that the use of two of the four criteria, like clue cells and positive amines, were sensitive in the BV diagnosis.[142]

Leopold[90] and Gardner and Dukes,[49] first isolated *G. vaginalis* using Casman's media. Dunkelberg et al[37] demonstrated that the microorganism required five beta-vitamins and both purines and pyrimidines. The organism can also be isolated from brain heart infusion broth, rabbit blood, or chocolate blood agar.[147]

In 1982 Totten et al,[144] developed a selective and differential human blood bilayer agar media with Tween 80 (HBT medium).

Vaginal fluid Gas-liquid chromatography analysis for the BV diagnosis was performed by Spiegel et al[136] to detect organic acids. Each genus has an organic acids production typical pattern, that can be used to identify specific organisms. About this, Krohn et al[84] reported 78% sensitivite and 81% specificity for diagnosing women with BV clini-

cal signs. Others have reported higher specificities; 90 to 98%,[2,19] and both higher[2] and lower[19] sensitivities.

Several studies have demonstrated that Gram's stain of vaginal fluid correlated well to the clinical diagnosis of BV.[41,80,84,137]

A new scoring system using the most reliable morphotypes from the vaginal smear was proposed by Nuggent et al[105] for BV diagnosing. In these scoring system, three morphotypes were used to create a total score of 0 to 10. These three morphotypes are large Gram-positive rods (*Lactobacillus*), small Gram-negative or Gram-variable rods (*Bacteroides* or *Gardnerella*), and curved gram-negative to Gram-variable rods (*Mobiluncus* spp). The total scores were computed by adding the weighted quantitation (0 to 4 +) of the three morphotypes.

A score of 7 to 10 was considered to indicate BV and a score to 0 to 6 was considered to indicate no BV. Gram staining is particularly useful to exclude BV because it has a high negative predictive value.[4,14,56,128]

Cano et al[21] developed an indirect immunofluorescent-antibody test; they observed an incidence of 24.2% in patients with nonspecific vaginitis. Hansen et al[62] used direct immunofluorescence technique on vaginal specimens obtaining 21% of the patients with *G. vaginalis*. Pao et al[110] developed a DNA-based technique for *G. vaginalis* direct identification by the presence of its DNA sequences in clinical specimens, detecting a single copy, with 89% of sensitivity and a specificity of 95%. Later, Sheiness et al[132] developed an oligonucleotide probe that hybridizes specifically with *G. vaginalis* 16S rRNA with the advantage of detecting a highly abundant cellular component (rRNA). They obtained 95% of sensitivity, and a specificity of 98.6%. The use of nucleic acid probes or DNA-based technologies is becoming increasingly more commonplace for identifying infections agents and probably will continue to do so as improved methods become available.[45,149] Sequences of several cloned *G. vaginalis* are being determined now in an attempt to apply polymerase chain reaction (PCR) techniques to the *G. vaginalis* identification.[145]

Clue cells and changes in bacterial flora can be found in the Papanicolaou smear (pap smear), which normally would be an incidental finding and has limited diagnostic potential in comparison with other methods.[14,43] However, Platz-Christensen[116] found a sensitivity of 90% and specificity of 97%, when compared the pap smear with clinical BV diagnosis. The positive and negative predictive values of the method tested were 94% and 95% respectively.

TREATMENT

Is important to determine which patients with BV should receive medical treatment. It is generally agreed that patients who do have symptoms should be treated to alleviate discomfort, but more than one half of patients with BV do not have symptoms. The Centers for Disease Control recommendation that patients without symptoms should not be treated does not consider the implications of the more recently published studies documenting serious infectious sequelae.[143] Risk from therapy versus benefit to patients must always be weighed.

From the various antibiotics tested for BV treatment metronidazole was found to be the most effective.[100,103,114,119] Neither the treatment duration nor the administration mode are relevant to the cure rates[48,93,101,124]

Drug use during the first pregnancy trimester has been discouraged because of suspected mutagenicity.[36] However, a more recent report, states that the accumulated data suggests that metronidazole is probably safe throughout pregnancy.[22] Other antibiotic options are cephradine,[53] intravaginal clindamycin,[46,71] amoxicillin,[139] and niridazole[5]; having different cure rates and none has shown 100% success. Ampicillin is active against *G. vaginalis*[97] but has a low cure rate (43%) in the BV treatment,[94] possibly because of its activity against lactobacilli.[92] sulfonamides, tetracyclines and erytomycins are ineffective.[94]

Today there is a tendency for patients, especially those who are pregnant; to prefer natural products in the disease treatment. There have been several attempts to treat BV with such alternative substances as acid gel or lactobacilli containing products.[3,74,102] The authors have indicated that alternative regimens are useful and effectives.

CONCLUSION

The pathophysiology of BV remains inexact. The effects of specific antibiotic therapy on the vaginal bacterial population are largely unknown. Serious sequelae documenting the predisposition to infectious morbidity in women with this disease have only been identified recently. Trials so show the value of therapeutic intervention in women with and without symptoms can now be justified. Further studies will be needed to verify who patients are at increased risk of complications (The major one being preterm delivery of low-weight infants, still the main cause of morbidity and mortality among new-borns) and so identify women who really need treatment during pregnancy. Epidemiologic studies will indicate if eradication of BV could become a preventive strategy against adverse pregnancy outcomes, such as preterm delivery, chorioamnionitis, amniotic fluid infection and post-cesarean endometritis.

REFERENCES

1. Aalto, M., T. Ronnemaa and E. Kulonen. 1974. Effect of neuroaminidase on the synthesis of collagen and other proteins. Biochim. Biophys. Acta. 342:247-253.
2. Amsel, R., P. Totten and C. Spiegel. 1983. Nonspecific vaginits: diagnostic criteria and microbial and epidemiologic associations. Am. J. Med. 74:14-22.

3. Andersch, B., L. Forssman, K. Lincoln and P. Tostensson. 1986. Treatment of bacterial vaginosis with an acid cream: a comparison between the effect of lactate gel and metronidazole. Gynecol. Obstet. Invest. 21:19-25.
4. Ankirskaia, A., V. Murevéva, T. Akopian and G. Bairamova. 1997. Evaluation of the sensitivity and specificity of methods of rapid diagnosis in bacterial vaginosis. Klin. Lab. Diagn. 7:41-45.
5. Bannatyne, R. M. and J. Jackowski. 1987. Antimicrobial effects or niridazole on *Gardnerella vaginalis*. Infection. 15:128.
6. Barberis, I., M. Pajaro, S. Godino, L. Pascual and L. Albesa. 1996. Evaluación clínica y microbiológica de vaginosis bacteriana en mujeres de un hospital de Rio Cuarto, Argentina. Enfer. Infecc. Microbiol. Clin. (Spain). 14:611-613.
7. Barbone, F., H. Austin, W. Louv and W. Alexander. 1990. A follow up study of methods of contraception, sexual activity, and rates of trichomoniasis, candidiasis and bacterial vaginosis. Am. J. Obstet. Gynecol. 163:510-514.
8. Bazarra, A. and A. Castro. 1998. Bacterial vaginosis: where are we? Med. Clin. (Barc). 110:213-214.
9. Beachey, E. H. and I. Ofek. 1976. Epithelial cell binding of group A streptococci by lipoteichoic acid on fimbriae denuded of M protein. J. Exp. Med. 143:759-771.
10. Bejar, R., V. Curbelo, C. Davis and L. Gluck. 1981. Premature labor II: bacterial sources of phospholipase. Obstet. Gynecol. 57:479-482.
11. Benito, R., J. A. Sáenz-Nieto, S. Berron, J. Vázquez and A. Fenoll. 1986. Esquemas de identificación y biotipia de *Gardnerella vaginalis*. Infectologia 6:54-60.
12. Benito, R., J. Vázquez, S. Berron, A. Fenoll and J.A. Sáenz-Nieto. 1986. A modified scheme for biotyping *Gardnerella vaginalis*. J. Med. Microbiol. 21:357-359.
13. Bhalla, P., N. Rewari and P. Chadha. 1989. *Gardnerella vaginalis* in cu T 20 users. Ind. J. Med. Res. 89:80-86.
14. Biswas, M. K. 1993. Bacterial vaginosis. Clin. Obstet. Gynecol. 36:166-176.
15. Brand, J. and R. P. Galask. 1986. Trimethylamine: the substance mainly responsible for the fishy odor often associated with bacterial vaginosis. Obstet. Gynecol. 63:682-685.
16. Brewer, J. I., B. Hayern and G. Thomas.1957. *Haemophilus vaginalis* vaginits. Am. J. Obstet. Gynecol. 74:834-842.
17. Briselden, A. and S. Hillier. 1990. Longitudinal study of the biotypes of *Gardnerella vaginalis*. J. Clin. Microbiol. 28:2761-2764.
18. Briselden, A. M., B. J. Monlla, C. E. Stevens and S. H. Hillier. 1992. Sialidases (neuroaminidases) in bacterial vaginosis and bacterial vaginosis-associated microflora. J. Clin. Microbiol. 30:663-666.
19. Bump, R. C., F. P. Zuzpan, W. J. Buesching, L. W. Ayers and T. J. Stephens. 1984. The prevalence six-month persistence, and predictive values of laboratory indicators of bacterial vaginosis (nonspecific vaginitis) in asymptomatic women. Am. J. Obstet. Gynecol. 150:917-924.
20. Bump, R. and J. Buesching. 1988. Bacterial vaginosis in virginal and sexually active adolescent females: evidence against exclusive sexual transmission. Am. J. Obstet. Gynecol. 158:935-939.
21. Cano, R. J., M. A. Beck and D. V. Grady. 1984. Detection of *Gardnerella vaginalis* on vaginal smears by immunofluorescence. Can. J. Microbiol. 29:27-32.
22. Carvajal, A., A. Sánchez and G. Hurtarte. 1995. Metronidazole during pregnancy (letter). Int. J. Gynecol. Obstet. 48:323-324.
23. Cauci, S. R., R. Monte, F. Quadrifoglio, M. Ropele and G. Menestrina. 1993. Ionic factors regulating the interaction of *Gardnerella vaginalis* hemolysin with red blood cells. Biochim. Biophys. Acta. 1153:53-58.
24. Cauci, S., R. Montr, M. Ropele, C. Missero, T. Not and Q. Franco. 1993. Pore-forming and haemolytic propierties of the *Gardnerella vaginalis* cytolysin. Mol. Microbiol. 9:1143-1155.
25. Cauci, S., M. Rosella, S. Driussi, P. Lanzafame and F. Quadrifoglio. 1998. Impairment of the mucosal immune system: IgA and IgM cleavage detected in vaginal washings of a subgroup of patients with bacterial vaginosis. J. Infec. Dis. 178:1698-1706.
26. Cito, G. 1996. Screening for bacterial vaginosis in a population of pregnant women reporting to cardiotocographic test. Minerva. Ginecol. 48:481-484.
27. Cristiano, L., S. Rampello, C. Noris and V. Valota. 1996. Bacterial vaginosis: prevalence in an Italian population of asymptomatic pregnant women and diagnostic aspects. Eur. J. Epidemiol. 12:383-390.
28. Criswell, B., J. Marston, W. Stenback, S. Black and H. Gardner. 1971. *Haemophilus vaginalis* 594, a gram negative organism? Can. J. Microbiol. 17:865-869.
29. Criswell, B. S., W. A. Stenback, S. H. Black and H. L. Gardner. 1972. Fine structure of *Haemophilus vaginalis* J. Bacteriol. 109:930-932.
30. Cruden, D. L. and R. Galask. 1988. Reduction of trimethylamine oxide to trimethylamine by *Mobiluncus* strains isolated from patients with bacterial vaginosis. Microbiol. Ecology In Health and Disease. 1:95-100.
31. Curtis, A. 1914. On the etiology and bacteriology of leucorrhoea. Surg. Gynecol. Obstet. 18:299-306.
32. Chaudhuri, M. and B. D. Chatterjee. 1996. Pathogenic potential of *Gardnerella vaginalis* on the female urogenital system. J. Indian. Med. Assoc. 94:11-16.
33. Chen, K. C., P. S. Forsyth, T. Buchanon and K. K. Holmes. 1979. Amine content of vaginal fluid from untreated and treated patients with non-specific vaginits. J. Clin. Invest. 63:825-835.
34. Chen, K. C., R. Amsel and D. Eschenbach. 1982. Biochemical diagnosis of vaginitis: determination of dia-

mines in vaginal fluid. J. Infect. Dis.145:337-345.
35. Döderlein, A. 1894. Die scheidensekretuntersuchugen. Zentralb. Gynakol. 18:10-14.
36. Drinkwater, P. 1987. Metronidazole. Aust. Nz.. J. Obstet. Gynaecol. 27:228-230.
37. Dunkelberg, W. and I. Mc Veigh. 1969. Growth requirements of *Haemophilus vaginalis*. Antonie Van Leeuwenhoek. 35:129-145.
38. Elsner, P. and A. Hartmann. 1987. *Gardnerella vaginalis* in the male upper genital tract: a possible source of reinfection of the female partner. Sex. Transm. Dis. 14:122-123.
39. Embree, J. J. Caliando and W. Mc Cormack. 1984. Nonspecific vaginitis among women attending a sexually transmitted diseases clinic. Sex. Transm. Dis. 11:81-84.
40. Eschenbach, D. 1984. History and review of bacterial vaginosis. Am. J. Obstet. Gynecol. 169:441-445.
41. Eschenbach, D., S. Hillier and C. Critchlow. 1988. Diagnosis and clinical features associated with bacterial vaginosis. Am. J. Obstet. Gynecol. 158:819-828.
42. Eschenbach, D. A., P. Davick and B. Williams. 1989. Prevalence of hydrogen peroxide producing *Lactobacillus* species in normal women and women with bacterial vaginosis. J. Clin. Microbiol. 27:251-256.
43. Fan, S., Y. Ke and Q. Li. 1996. Detection of bacterial vaginosis in gram stained vaginal smears and papanicolaou stained cervical smears. Chung. Hua. I. Hsueh. Tsa. Chih. 76:284-286.
44. Faro, S., L. E. Phillips and M. G. Martens. 1988. Perspectives on the bacteriology of postoperative obstetric-gynecologic infections. Am. J. Obstet. Gynecol. 158 (suppl):694-700.
45. Ferris, D. G., J. Hendrich, P. H. Payne, A. Getts, R. Rassekh, D. Mathis and H. S. Litake. 1995. Office laboratory diagnosis of vaginitis clinician-performed test compared with a rapid nucleic acid hibridization test. J. Fam. Pract. 41:575-581.
46. Ferris, D. G., M. S. Litaker, L. Woodward, D. Mathis and J. Hendrisch. 1995. Treatment of bacterial vaginosis a comparison of oral metronidazole, metranidazole vaginal gel, and clindamycin vaginal cream. J. Fam. Pract. 41:443-449.
47. Fohn, M. J., S. A. Lukehart and S. Hillier. 1988. Production and characterization of monoclonal antibodies to *Mobiluncus* species. J. Clin. Microbiol. 26:2598-2603.
48. Fredricsson, B., B. Hagström, A. K. Möller and C. E. Nord. 1986. Bacterial vaginosis treated with metronidazole: effects on the vaginal microbiology flora by a single dose versus a five day regimen. Zentralbl. Gynakol. 108:799-804.
49. Gardner, H. and C. Dukes. 1955. *Haemophilus vaginalis* vaginitis: a newly defined specific infection previously classified "nonspecific vaginitis". Am. J. Obstet. Gynecol. 69:962-976.
50. Gardner, H., T. Dampeer and C. Dukes. 1957. The prevalence of vaginitis: A study in incidence. Am. J. Obstet. Gynecol. 73:1080-1087.
51. Gibbons, R. and J. Van Houte. 1975. Bacterial adherence in oral microbial ecology. Annu. Rev. Microbiol. 29:19-45.
52. Gibbons, R., D. M. Spirell and Z. Skobe. 1976. Selective adherence as a determinant of the host tropisms of certain indigenous and pathogenic bacteria. Infect. Immun. 13:238-246.
53. Goldstein, E. J., Y. Y. Kwok and V. L. Sutter. 1983. Susceptibility of *Gardnerella vaginalis* to cephradine. Antimicrob. Agents Chemother. 24:418-419.
54. González-Pedraza, A. A., M. C. Ortiz and E. R. Morales. 1990. Frecuencia de aislamiento de *Gardnerella vaginalis* en vías urinarias. Rev. Lat-Amer. Microbiol. 32:157-161.
55. González-Pedraza, A. A., M. A. Inzunza, M. C. Ortiz, E. R. Morales and R. Ponce. 1995. Biotipos de *Gardnerella vaginalis* modificación de un esquema propuesto. Rev. Lat-Amer. Microbiol. 37:19-26.
56. González-Pedraza, A. A., M. A. Inzunza, M. C. Ortiz, R. Ponce and C. A. Irigoyen. 1997. Comparación de 2 métodos de laboratorio clínico en el diagnóstico de la vaginosis bacteriana. Aten. Primaria. 19:357-360.
57. Govender, L., A. Hossen, J. Moodley and A. W. Sturm. 1996. Bacterial vaginosis and associated infections in pregnancy. Int. J. Gynaecol Obstet. 55:23-28.
58. Gravett, M., H. Nelson, T. De Roven, C. Critchlow, D. Eschenbach and K. K. Holmes. 1986. Independent association of bacterial vaginosis and *Chlamydia trachomatis* infection with adverse pregnancy outcome. JAMA. 256:1899-1903.
59. Grenwood, J. and M. Pickett. 1980. Transfer of *Haemophilus vaginalis* Gardner and Dukes to a new genus *Gardnerella: G.vaginalis* (Gardner and Dukes). Comb. Nov. Int. J. Syst. Bacteriol. 30:170-178.
60. Gupta, B., R. Kumar, R. Sofat and S. Deepinder. 1998. The role of *Gardnerella vaginalis* in nonspecific vaginitis in intra uterine contraceptive device users. Indian. J. Pathol. Microbiol. 41:67-70.
61. Hammerschlag, M. R., M. Cummings, B. Doraiswamy, P. Cox and W. M. Mc Cormack. 1985. Nonspecific vaginitis following sexual abuse in children. Pediatrics. 75:1028-1031.
62. Hansen, W., B. Vray, K. Miller, F. Crokaert and E. Yourassowski. 1987. Detection of *Gardnerella vaginalis* in vaginal specimens by direct inmunofluorescence. J. Clin. Microbiol. 25:1934-1937.
63. Harper, J. and G. Davis. 1982. Cell wall analysis of *Gardnerella vaginalis (Haemophilus vaginalis)*. Int. J. Syst. Bacteriol. 32:48-50.
64. Hay, P., D. Taylor and R. Lamont. 1992. Diagnosis of bacterial vaginosis in a gynecology clinics. Br. J. Obstet. Gynaecol. 99:63-66.
65. Hay, P., R. Lamont, D. Taylor-Robinson, D. Morgan,

C. Ison and J. Pearson. 1994. Abnormal bacterial colonization of the lower genital tract as a marker for subsequent preterm delivery and late miscarriage. Br. Med. J. 308:295-298.
66. Hay, P. and R. Lamont. 1994. A longitudinal study of bacterial vaginosis during pregnancy. Br. J. Obstet. Gynaecol. 101:1048-1053.
67. Hay, P., A. Ugwumadu and J. Chowns. 1997. Sex, Trush and bacterial vaginosis. Int. J. STD. Aids. 8:603-608.
68. Hill, L., H. Ruparelia and J. Embil. 1983. Non specific vaginitis and other genital infections in three clinic populations. Sex. Transm. Dis. 10:114-118.
69. Hill. G. 1993. The microbiology of bacterial vaginosis. Am. J. Obstet. Gynecol. 169: 450-454.
70. Hillier, S. L., J. Martius, M. Krohn, N. Kiviat, K. K. Holmes and D. A. Eschenbach. 1988. A case-control study of chorioamnionic infection and histologic chorioamnionitis in prematurity. N. Eng. J. Med. 319:972-978.
71. Hillier, S., M. A. Krohn and D. H. Watts. 1990. Microbiologic efficacy of intravaginal clindamycin cream for the treatment of bacterial vaginosis. Obstet. Gynecol. 76:407-413.
72. Holmes, K., K. Chen and C. Lipinski. 1985. Vaginal redox potential in bacterial vaginosis (nonspecific vaginitis). J. Infec. Dis. 152:379-382.
73. Holst, E., B. Wathne and B. Hovelius. 1987. Bacterial vaginosis: microbiologic and clinical findings. Eur. J. Clin. Microbiol. 6:536-541.
74. Holst, E. and A. Brandberg. 1990. Treatment of bacterial vaginosis in pregnancy with a lactate gel. Scand. J. Infect. Dis. 22:625-628.
75. Hooten, T., S. Fihn, J. Johnson, P. Roberts and W. Stamm. 1989. Association between bacterial vaginosis and acute cystitis in women using diaphragms. Arch. Intern. Med. 149:1932-1936.
76. Ison, C. A. and C. S. Easmon. 1985. Studies on the mechanism of adhesion of *Gardnerella vaginalis* to human erythrocytes. Scan. J. Nephron (supplement). 86:190-193.
77. Ison, C. A., D. G. Harvey, A. Tanna and C. S. Easman. 1987. Development and evaluation of a scheme for serotyping *Gardnerella vaginalis*. Genitourin. Med. 63:196-201.
78. Ison, C. A., B. Kolator, J. H. Reid, J. Dermottclark and C. S. F. Easmon. 1989. Characterization of monoclonal antibodies for detection of *Mobiluncus* sp. in genital specimens. J. Med. Microbiol. 30:129-136.
79. Jenny, C., T. Hooten and A. Bowers. 1990. Sexually transmitted diseases in victims of rape. N. Engl. J. Med. 322: 713 -716.
80. Joesoef, M. R., S. L. Hillier, S. Josodiwondo and M. Linnan. 1991. Reproducibility of a scoring for gram stain diagnosis of bacterial vaginosis. J. Clin. Microbiol. 129:1730-1731.
81. Johnson, A. P. and L. Boustouller. 1987 Extravaginal infection caused by *Gardnerella vaginalis*. Epidem. Inf. 98:131-137.
82. Kimberlin, D. F. and W. W. Andrews. 1998. Bacterial vaginosis: association with adverse pregnancy outcome. Semin. Perinatol. 22:242-250.
83. Kretzchmar, U. M., R. Hammann and H. J. Kutzner. 1991. Purification and characterization of *Gardnerella vaginalis* hemolysin. Curr. Microbiol. 23:7-13.
84. Krohn, M., S. L. Hillier and D. A. Eschenbach. 1989. Comparison of methods for diagnosing bacterial vaginosis among pregnant women. J. Clin. Microbiol. 27:1266-1271.
85. Krohn, M, S. Hillier and R. Nugent. 1995. The genital flora of women with intraamniotic infection. J. Infect. Dis 171:1475-1480.
86. Krohn, M. A., S. L. Hillier, R. P. Nugent, M. F. Cotch, J. C. Carey, R. S. Gibbs and D. A. Eschenbach. 1996. The genital flora of women with intraamniotic infection. Vaginal infection and prematurity study group. J. Infect. Dis. 171:1475-1480.
87. Lapage, S. 1961. *Haemophilus vaginalis* and its role in vaginitis. Acta Pathol. Microbiol. Scand. 52:34-54.
88. Larsson, P. G., B. Bergman, U. Forsum and C. Panlson. 1990. Treatment of bacterial vaginosis in women with vaginal bleeding complications or discharge and harboring *Mobiluncus*. Gynecol. Obstet. Invest. 29:296-300.
89. Lefebre, J., S. Averous, R. Bariaud, C. Blanc, M. Bartrand and M. Lareng. 1988. Lower genital tract infections in women: comparison of clinical and epidemiologic findings with microbiology. Sex. Transm. Dis. 15:110-113.
90. Leopold, S. 1953. Heretofore undescribed organisms isolated from the genitourinary system. U.S. Armed. Forces. Med. J. 4:263-266.
91. Linaldi, A., J. R. Urbina. and J. Castañeda. 1988. Vaginitis por *Gardnerella vaginalis* en niñas y adolescentes. Bol. Med. Hosp. Infant. Mex. 45:101-103.
92. Lossick, J. G. 1990. Treatment of sexually transmitted vaginosis/vaginits. Rev. Infect. Dis. 12 (suppl): S665-S681.
93. Lugo-Miro, V., M. Green and L. Mazur. 1992. Comparison of different metronidazole therapeutic regimens for bacterial vaginosis. JAMA. 268:92-95.
94. Malouf, M., M. Fortier, G. Morin and J. L. Dube. 1981. Treatment of *Haemophilus vaginalis* vaginitis. Obstet. Gynecol. 57:711-714.
95. Märdh, P. A. and L. Weström. 1976. Adherence of bacteria to vaginal epithelial cells. Infect. Immun. 13:661-666.
96. Martínez de Tejeda, B., O. Coll, M. de Flores, S. Hillier and D. V. Landers. 1998. Prevalence of bacterial vaginosis in an obstetric population of Barcelona. Med. Clin. (Barc). 110:201-204.
97. Mc Carthy, L. R., P. A. Mickelsen and E. G. Smith. 1979. Antibiotic susceptibility of *Haemophilus*

vaginalis (Corynebacterium vaginale) to 21 antibiotics. Antimicrob. Agents. Chemother. 16:186-189.

98. McCormack, W., C. H. Hayes and B. Rosner. 1977. Vaginal colonization with *Corynebacterium vaginale (Haemophilus vaginalis)*. J. Infect. Dis. 136:740-745.

99. McGregor, J. A., J. I. French and W. Jones. 1994. Bacterial vaginosis is associated with prematurity and vaginal fluid mucinase and sialidase: results of a controlled trial of topical clindamycin cream. Am. J. Obstet. Gynecol. 170:1048-1060.

100. McLean, N. W. and J. A. McGroarthy. 1996. Growth inhibition of metronidazole susceptible and metronidazole resistant strains of *Gardnerella vaginalis* by Lactobacilli in vitro. Appl. Environ. Microbiol. 62:1089-1092.

101. Morales, E. R., A. A. González-Pedraza, M. C. Ortiz, M. P. Ramírez and J. S. Barajas. 1992. Estudio comparativo de cinco diferentes regimenes de metronidazol en el tratamiento de la vaginosis por *Gardnerella vaginalis*. Rev. Lat-Amer. Microbiol. 34:157-164.

102. Neri, A., G. Sabah and Z. Samra. 1993. Bacterial vaginosis in pregnancy treated with yoghurt. Acta Obstet. Gynecol. Scand. 72:17-21.

103. Neri, A., D. Rabinerson and B. Kaplan. 1994. Bacterial vaginosis, Drugs versus alternative treatment. Obstet. Gynecol. Surv. 49:809-813.

104. Newton, E. R., T. J. Prihoda and R. S. Gibbs. 1990. A clinical and microbiologic analysis of risk factors for puerperal endometritis. Obstet. Gynecol. 75:402-406.

105. Nuggent, R. P., M. A. Krohn and S. L. Hillier. 1991. Reliability of diagnosing bacterial vaginosis is improved by a standardized method of gram stain interpretation. J. Clin. Microbiol. 29:297-301.

106. O'Donnell, A., D. Minnikin, M. Goodfellow and P. Piot. 1984. Fatty acid, polar lipid and wall amino acid composition of *Gardnerella vaginalis*. Arch. Microbiol. 138:68-71.

107. Ortiz, M., A. González Pedraza, R.. Morales, M. Camorlinga and S. Giono. 1990. Frecuencia de aislamiento de *Gardnerella vaginalis* y su relación con probables factores de riesgo en la vaginosis bacteriana. Rev. Lat-Amer. Microbiol. 32:1-5.

108. Paavonen, J., K. Teisal and P. K. Heinonen. 1987. Microbiological and histopathological findings in acute pelvic inflammatory disease. Br. J. Obstet. Gynecol. 94:454-460.

109. Pandit, D. V., S. M. Varoe and L. P. Deodhar. 1989. Biotypes of *Gardnerella vaginalis* isolated from nonspecific vaginitis patients in Bombay. Indian. J. Med. Res. 89:435-438.

110. Pao, C. C., S. S. Lin and T. T. Hsieh. 1990. The detection of *Gardnerella vaginalis* DNA sequences in uncultured clinical specimens with cloned *G. vaginalis* DNA as probes. Moll. Cell. Probes 4:367-373.

111. Pheifer, T., P. Forsyth, M. Durfee, H. Pollock and K. K. Holmes. 1978. Nonspecific vaginitis: role of *Haemophilus vaginalis* and treatment with metronidazole. N. Engl. J. Med. 298: 1429-1434.

112. Piot, P., E. Van Dyk, H. Goodfellow and S. Falkow. 1980. A taxonomic study of *Gardnerella vaginalis (Haemophilus vaginalis)* Gardner and Dukes. J. Gen. Microbiol. 119:373-396.

113. Piot, P. and J. Vanderheyden. 1984. *Gardnerella vaginalis* and nonspecific vaginitis in: Sexually transmitted diseases (Holmes, K. K., Märdh, P. A., Sparling, P. F., and Wiesner, P. J., Ed). 1984 Vol. 1, pp. 412-427. Mc. Graw Hill. New York.

114. Piot, P. 1984. Bacterial vaginosis: an evaluation of treatment. Scand. J. Urol. Nephrol. Suppl. 86:229-235.

115. Piot, P., E. Van Dyck, M. Peeters, J. Hale, P. A. Totten and K. K. Holmes. 1984. Biotypes of *Gardnerella vaginalis*. J. Clin. Microbiol. 20:677-679.

116. Platz-Christensen, J. J., P. G. Larsson, C. T. Sundstrom and L. Bomdeson. 1989. Detection of bacterial vaginosis in Papanicolaou smears. Am. J. Obstet. Gynecol. 160:132-133.

117. Priestley, C. and G. Kinghorn. 1996. Bacterial vaginosis. Br. J. Clin. Pract. 50:331-334.

118. Priestley, C., B. Jones, J. Dhar and L. Goodwin. 1997. What is normal vaginal flora? Genitourin. Med. 73:23-28.

119. Pulkkinen, P., M. Saranen and R. Kauja. 1993. Metronidazole combined with nystatin (vagitories) in the prevention of bacterial vaginosis after initial treatment with oral metronidazole. Gynecol. Obstet. Invest. 36:181-184.

120. Pybus, V. and A. Onderdonk. 1997. Evidence for a commensal, symbiotic relationship between *Gardnerella vaginalis* and *Prevotella bivia* involving ammonia: Potential significance for bacterial vaginosis. J. Infect. Dis. 175: 406-413.

121. Reimer, L. and G. B Reller. 1984. *Gardnerella vaginalis* bacteremia: A review of thirty cases. Obstet. Gynecol. 64:170-172.

122. Reyn, A., A. Birch-Andersen and S. Lapage. 1966. An electron microscope study of thin sections of *Haemophilus vaginalis* (Gardner and Dukes) and some possibly related species. Can. J. Microbiol. 12:1125-1136.

123. Riordan, T., M. Macaulay and J. James. 1990. A prospective study of genital infections in a family planning clinic 1. Microbiological findings and their association with vaginal symptoms. Epidemiol. Infect. 104:47-53.

124. Rivera, L., M. Trenado, A. Valdez and C. González. 1996. Prevalence of bacterial vaginitis and vaginosis: association with clinical and laboratory features, and treatment. Ginecol. Obstet. Méx. 64: 26-35.

125. Roberts, M. C., S. L. Hillier, F. D. Schoenknecht and K. K. Holmes. 1985. Comparison of Gram stain, DNA probe and culture for the identification of species of *Mobiluncus* in female genital specimens. J. Infect. Dis. 152:74-77.

126. Rosenstein, I., D. Morgan, M. Sheehan, R. Lamot and D. Taylor Robinson. 1996. Bacterial vaginosis in pregnancy:

distribution of bacterial species in different gram-stain categories of the vaginal flora. J. Med. Microbiol. 45: 120-126.

127. Sahdu, K., P. Domingue and A. Chow. 1989. *Gardnerella vaginalis* has a gram-positive cell-wall lipopolysaccharide. J. Med. Microbiol. 29:229-235.
128. Schwebke, J., S. Morgan and H. Weiss. 1997. The use of sequential self-obtained vaginal smears for detecting changes in the vaginal flora. Sex. Transm. Dis. 24: 236-239.
129. Scott, T. G., C. J. Smyth and C. T. Keane. 1987. In vitro adhesiveness and biotype of *Gardnerella vaginalis* strains in relation to the occurrence of clue cells in vaginal discharges. Genitourin. Med. 63:47-53.
130. Scott, T. G. and C. J. Smyth. 1987. Haemagglutination and tissue culture adhesion of *Gardnerella vaginalis*. J. Gen. Microbiol. 133:1999-2005.
131. Scott, T. G., B. Curran and C. J. Smyth. 1989. Electron microscopy of adhesive interactions between *Gardnerella vaginalis* and vaginal epithelial cells, Mc Coy cells and human red blood cells. J. Gen. Microbiol. 135:475-480.
132. Sheines, D., K. Dix, S. Watanabe and S. L. Hillier. 1992. High levels of *Gardnerella vaginalis* is detected with an oligonucleotide probe combined with elevated pH as a diagnostic indicator of bacterial vaginosis. J. Clin. Microbiol. 30:642-648.
133. Sobel, J. D., J. Schneider, D. Kaye and M. Levison. 1981. Adherence of bacteria to vaginal epithelial cells at various stages in the menstrual cycle. Infect. Immun. 32:194-197.
134. Sobel, J. 1989. Bacterial vaginosis- an ecologic mystery. Ann. Inter. Med. 111:551-553.
135. Soper, D. E., R. C. Bump and W. G. Hurt. 1990. Bacterial vaginosis and trichomoniasis vaginitis are risk factors for cuff cellulites after abdominal hysterectomy. Am. J. Obstet. Gynecol. 163:1016-1023.
136. Spiegel, C. A., R. Amsel, D. Eschenbach, P. Schenecht and K. K. Holmes. 1980. Anaerobic bacteria in nonspecific vaginitis. N. Engl. J. Med. 303:601-607.
137. Spiegel, C. A., R. Amsel and K. K. Holmes. 1983. Diagnosis of bacterial vaginosis by direct gram stain of vaginal fluid. J. Clin. Microbiol. 18:170-177.
138. Spiegel, C. and M. Roberts. 1984. *Mobiluncus* gen. nov., *Mobiluncus curtisii* subsp. *holmesii* subsp. nov., and *Mobiluncus mullieris* sp. nov., curved rods from the human vagina. Int. J. System. Bacteriol. 34:177-184.
139. Symonds, J., and A. K. Biswas. 1986. Amoxycillins, augmentin and metronidazole in bacterial vaginosis associated with *Gardnerella vaginalis* (letter). Genitourin. Med. 62:136.
140. Thomason, J. L., P. A. Schreckenberger, W. N. Spellacy, L. J. Riff and L. J. LeBeau. 1984. Clinical and microbiological characterization of patients with nonspecific vaginosis having motile curved anaerobic rods. J. Infect. Dis. 149:801-809.
141. Thomason, J., S. Gelbart and L. Wilcoski. 1988. Proline amino-peptidase activity as a rapid diagnostic test to confirm bacterial vaginosis. Obstet. Gynecol. 71:607-611.
142. Thomason, J. L., S. H. Gelbart and F. Brockhuizen. 1989. Advances in the understanding of bacterial vaginosis. J. Rep. Med. 34:581-587.
143. Thomason, J., S. Gelbart and N. Scaglione. 1991. Bacterial vaginosis: current review with indications for asymptomatic therapy. Obstet. Gynecol. 165:1210-1217.
144. Totten, P. A., R. Amsel, J. Hale and K. K. Holmes. 1982. Selective differential human blood bilayer media for isolation of *Gardnerella (Haemophilus) vaginalis*. J. Clin. Microbiol.15:141-147.
145. van-Belkum, A., A. Koeken, P. Vandamme, M. van-Esbroeck, H. Goosens and J. Koopmans. 1995. Development of a species-specific polymerase chain reaction assay for *Gardnerella vaginalis*. Mol. Cell. Probes. 9:167-174.
146. Villegas Castrejon, H., M. González, S. Fuentes, G. Casanova, M. Beltrán and E. Flores. 1995. *Gardnerella vaginalis* infection in heterosexual couples. Ultrastructural study of desquamation cells of the stratified epithelium. Ginecol. Obstet. Méx. 63:139-146.
147. Vontver, L. A. and D. A. Eschenbach. 1981. The role of *Gardnerella vaginalis* in nonspecific vaginitis. Clin. Obstet. Gynecol. 24:439-462.
148. Westrom, L., G. Evaldson and K. K. Holmes. 1984. Taxonomy of vaginosis: bacterial vaginosis a definition. Scand. J. Urol. Nephrol. Suppl. 86:259-264.
149. Wu, S. R., S. L. Hillier and K. Nath. 1996. Genomic DNA fingerprint analysis of biotype *Gardnerella vaginalis* from patients with and without bacterial vaginosis. J. Clin. Microbiol. 34:192-195.
150. Zinnemann, K., and G. Turner. 1963. The taxonomic position of *Haemophilus vaginalis* (*Corynebacterium vaginale*). J. Pathol. Bacteriol. 85:213-219.