**Multitarget Polymerase Chain Reaction Testing for Diagnosis of Bacterial Vaginosis**

(204127)

**Medical Benefit**
- Effective Date: 04/01/15
- Next Review Date: 01/18

**Preauthorization**
- No
- Review Dates: 01/15, 01/16, 01/17

**Preauthorization is not required.**

The following protocol contains medical necessity criteria that apply for this service. The criteria are also applicable to services provided in the local Medicare Advantage operating area for those members, unless separate Medicare Advantage criteria are indicated. If the criteria are not met, reimbursement will be denied and the patient cannot be billed. Please note that payment for covered services is subject to eligibility and the limitations noted in the patient’s contract at the time the services are rendered.

<table>
<thead>
<tr>
<th>Populations</th>
<th>Interventions</th>
<th>Comparators</th>
<th>Outcomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Individuals:</td>
<td>Interventions of interest are:</td>
<td>Comparators of interest are:</td>
<td>Relevant outcomes include:</td>
</tr>
</tbody>
</table>
| • With signs or symptoms of bacterial vaginosis | • Multitarget polymerase chain reaction testing | • Clinical and microscopic evaluation, including scoring systems such as the Nugent score | • Test accuracy
|                         |                                        |                                                  | • Test validity
|                         |                                        |                                                  | • Symptoms
|                         |                                        |                                                  | • Change in disease status

**Description**

Bacterial vaginosis (BV) is a common medical condition resulting from an imbalance in the normal vaginal flora. Although identification of *Gardnerella vaginalis* has traditionally been associated with BV, there is no single etiologic agent. Most cases are asymptomatic, and most symptomatic cases can be diagnosed using clinical and microscopic evaluation. Multitarget polymerase chain reaction (PCR) testing is proposed as an alternative to currently available laboratory tests to diagnosis BV. This test may improve outcomes if it is a more accurate and reliable method to diagnose BV, especially in symptomatic women with an indeterminate diagnosis.

**Summary of Evidence**

The evidence for multitarget PCR testing in patients who have signs or symptoms of BV includes several prospective studies on the diagnostic accuracy of PCR assays of individual markers or combinations of markers, and several prospective studies validating the diagnostic accuracy of multitarget tests. Relevant outcomes are test accuracy and validity, symptoms, and change in disease status. None of the studies evaluated a multitarget PCR test that is commercially available in the United States. The available studies suggest that the multitarget PCR tests that were evaluated may have high sensitivity and specificity for diagnosing BV, but it is not possible to determine the true diagnostic accuracy with certainty due to limited research and heterogeneity in methodology (e.g., differences in the included markers, scoring systems, and/or reference tests). However, studies of diagnostic accuracy alone in unselected populations of women with BV are inadequate, because most symptomatic women can be diagnosed with a standard workup and/or a trial of empirical therapy. Studies have not been conducted in the most clinically relevant target population, symptomatic women with indeterminate diagnoses after standard workup. Furthermore, there is a lack of evidence on the clinical utility of PCR testing for BV, i.e., studies showing that testing leads to better patient management decisions and/or better health outcomes than current approaches. The evidence is insufficient to determine the effects of the technology on health outcomes.
**Policy**
Multitarget polymerase chain reaction (PCR) testing for diagnosis of bacterial vaginosis is considered investigational.

**Policy Guidelines**
The most common diagnostic approach to BV is use of the Amsel criteria. The Amsel criteria require three of the following four to be present in order for a diagnosis of BV to be confirmed:

- vaginal discharge that is homogeneous, thin and whitish gray discharge;
- presence of clue cells on microscopic examination. These are squamous epithelial cells that normally have a sharply defined cell border but in BV, have bacteria adherent to their surfaces and appear to be “peppered” with bacteria;
- pH of vaginal fluid greater than 4.5;
- a fishy odor of vaginal discharge before or after addition of 10% KOH

For patients who cannot be diagnosed by the Amsel criteria, there are other scoring systems used in conjunction with Gram stain for the laboratory diagnosis of BV:

**Nugent criteria:**
Levels of three types of bacteria in vaginal discharge samples are estimated, Lactobacillus, Gardnerella/Bacteroides and Mobiluncus. Levels of Lactobacillus and Gardnerella/Bacteroides are rated on a scale of zero to four based on the number of cells per field at 100X magnification, and levels of Mobiluncus are rated on a scale from zero to two. A composite score is then calculated by summing the three subscores, as follows:

- Not consistent with BV:
  - Score of zero to three; or
  - Score of four to six with clue cells not present

- Consistent with BV:
  - Score of four to six with clue cells present; or
  - Score of at least seven

Some clinicians include a third, middle category in Nugent scoring, with a total score of zero to three considered normal, four to six as intermediate/equivocal and seven to ten as definite BV.

**The simplified Ison and Hay criteria** are as follows:

- **Grade 1 (Normal):** Lactobacillus morphotypes predominate;
- **Grade 2 (Intermediate):** Flora are mixed with some Lactobacillus morphotypes and some Gardnerella or Mobiluncus morphotypes present;
- **Grade 3 (BV):** Gardnerella and/or Mobiluncus morphotypes predominate. Lactobacilli morphotypes few or absent.
Background

Disease

BV is a condition caused by an imbalance in the normal bacteria vaginal flora. It is a common disorder, especially in women of reproductive age. While there is no single known etiologic agent, there is a shift in vaginal flora that involves a depletion of Lactobacillus species and overgrowth of other bacteria, including Gardnerella vaginalis, Mycoplasma hominis, Peptostreptococcus, Mobiluncus species, and various other anaerobic gram-negative rods. Prevalence of the condition is high, and it is asymptomatic in most cases. According to data from a nationally representative sample of women surveyed in 2001 to 2004, the prevalence of BV among women ages 14 to 49 in the United States is 29%. BV is often confused with nonbacterial causes of vaginitis, including Candida (i.e., yeast infection, caused by a fungus) and Trichomonas (caused by a parasite).

When symptomatic, BV is associated with characteristic signs and symptoms. The most common sign of BV is an abnormal grayish white vaginal discharge, generally with an unpleasant (often “fishy”) smell. Some women experience mild itching. In addition, BV may be a risk factor for conditions such as preterm delivery and spontaneous abortion in pregnant women, pelvic inflammatory disease, HIV and other sexually transmitted diseases. However, causality is difficult to demonstrate, especially in this type of situation where these associations may be spurious due to confounding, because both BV and HIV infection are related to multiple sexual partners. Because of potential risks during pregnancy, treatment of BV is indicated for symptomatic pregnant women. However, national organizations do not recommend routine screening for BV among pregnant women, and national guidelines do not address screening of nonpregnant women.

BV resolves spontaneously in a high percentage of women. Treatment for symptomatic BV is usually a course of oral antibiotics, either metronidazole or clindamycin. Antibiotic treatment results in a high rate of remission of symptoms, but recurrences are common within the first year after treatment. Probiotics, alone or in conjunction with antibiotics, are also used but their efficacy in improving cure rates or preventing recurrences is not well-characterized.

Laboratory- and Examination-Based Methods of Diagnosis

BV can be diagnosed in the primary care setting based on patient-reported symptoms, clinical findings during vaginal examination and analysis of vaginal discharge. Office-based analysis of vaginal discharge includes a wet mount preparation using saline, an odor (“whiff”) test to detect amines before or after the addition of 10% potassium hydroxide (KOH) and a test of the pH level. Clinical diagnosis generally involves applying the Amsel criteria, which requires three of the following four to be present in order for a diagnosis of BV to be confirmed:

- vaginal discharge that is homogeneous, thin and whitish gray discharge;
- presence of clue cells on microscopic examination. These are squamous epithelial cells that normally have a sharply defined cell border but in BV, have bacteria adherent to their surfaces and appear to be “peppered” with bacteria;
- pH of vaginal fluid greater than 4.5;
- a fishy odor of vaginal discharge before or after addition of 10% KOH

In most cases of uncomplicated BV, clinical and microscopic examination of the discharge is sufficient to make a presumptive diagnosis using the Amsel criteria. For patients with a moderate to high probability of BV following clinical and microscopic exam, an empiric treatment trial can be prescribed. Patients who respond to empiric treatment do not require further workup.

A subset of women may require more definitive tests to determine whether BV is present. These include women with unusual or unexpected signs and symptoms and those in whom it is not possible to exclude other etiologies.
with certainty. In these cases, laboratory tests are available to assist with making a definitive diagnosis. Gram staining of vaginal discharge samples is the conventional laboratory method of BV diagnosis, and many experts consider it to be the criterion standard for diagnosing BV. Samples are analyzed using criteria such as the Nugent criteria, or a modified version by Ison and Hay.

A limitation of both of the above diagnostic methods (i.e., clinical diagnosis using Amsel criteria and laboratory diagnosis using Nugent, or Ison and Hay criteria) is that they have subjective components and therefore may be imprecise. Gram stain examination, moreover, is time-consuming and requires substantial training, and it is difficult to determine an appropriate clinical response for intermediate scores. The two methods of diagnosis can also be used in combination to increase diagnostic accuracy.

Various commercial tests are also available to provide rapid and accurate pH evaluation and amine detection. For example automated devices that measure the volatile gases produced from vaginal samples and a colorimetric pH test are commercially available.

Vaginal culture is not an appropriate diagnostic method to identify BV because it is not caused by the presence of a particular bacterial species.

**Nucleic Acid Probes**

DNA probes have been developed and are now available to directly detect and quantify the bacteria in vaginal fluid samples. Bacterial DNA is extracted and amplified by polymerase chain reaction (PCR) methods, using either universal or specific primers. Bacteria are then identified by characterizing their ribosomal DNA (rDNA) sequences. The specific target is typically the ribosomal subunit of the $16S_rRNA$ gene, which is present in all bacteria. The $16S_rRNA$ genes can be amplified by PCR using universal and/or specific primers. The amplified product is then quantified to give an assessment of how many microorganisms are present. In addition to being able to more accurately diagnose health conditions, use of these new techniques has resulted in the identification of previously unrecognized cultivation-resistant organisms in vaginal fluid.

**Proposed Multitarget PCR Test**

At least one commercially available product measures multiple organisms using PCR technology for the diagnosis of BV. This product, SureSwab (Quest Diagnostics) tests for *Lactobacillus* species, *G. vaginalis*, *Atopobium vaginae*, and *Megasphaera* species. *A. vaginae* is a bacterium species named in 1999 and subsequently, using molecular-based techniques, has been found to be more common in women with BV than women with normal flora.\(^2\)

The SureSwab Total test involves obtaining vaginal swab specimens and extracting total DNA. Next, real-time PCR is used to quantitate the four types of bacteria. Results are reported as log cells per mL for each organism (concentrations of all *Lactobacilli* species are reported together).

In addition, the company provides summary interpretive information based on the findings from all tests. Interpretive information accompanying test results classify findings into one of the following three categories:

Not supportive of BV diagnosis:

- Presence of *Lactobacillus* species, *G. vaginalis* levels < 6.0 log cells/mL and absence of *A. vaginae* and *Megasphaera* species; or
- Absence of *Lactobacillus* species, *G. vaginalis* levels < 6.0 log cells/mL and absence of *A. vaginae* and *Megasphaera* species; or
- Absence of all targeted organisms.
Equivocal:

- Presence of *Lactobacillus* species, plus *G. vaginalis* at least 6.0 log cells/mL and/or presence of *A. vaginae* and/or *Megasphaera* species.

Supportive of BV diagnosis:

- Presence of *Lactobacillus* species, *G. vaginalis* levels at least 6.0 log cells/mL and presence of *A. vaginae* and/or *Megasphaera* species.

Quest Diagnostics also offers a SureSwab bacterial vaginosis/vaginitis test that includes the bacterial vaginosis test, previously described, and tests for *Trichomonas vaginalis* and four *Candidiasis* species.

**Regulatory Status**

Clinical laboratories may develop and validate tests in-house and market them as a laboratory service; laboratory-developed tests (LDTs) must meet the general regulatory standards of the Clinical Laboratory Improvement Act (CLIA). No multitarget quantitative polymerase chain reaction tests for bacterial vaginosis are available under the auspices of CLIA. Laboratories that offer LDTs must be licensed by CLIA for high-complexity testing. To date, the U.S. Food and Drug Administration has chosen not to require any regulatory review of this test.

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Services that are the subject of a clinical trial do not meet our Technology Assessment Protocol criteria and are considered investigational. For explanation of experimental and investigational, please refer to the Technology Assessment Protocol.

It is expected that only appropriate and medically necessary services will be rendered. We reserve the right to conduct prepayment and postpayment reviews to assess the medical appropriateness of the above-referenced procedures. Some of this protocol may not pertain to the patients you provide care to, as it may relate to products that are not available in your geographic area.

**References**

We are not responsible for the continuing viability of web site addresses that may be listed in any references below.