



LAB #: Sample Report
PATIENT: Sample Patient
ID:
SEX: Female
DOB: 01/01/1993

AGE: 25

CLIENT #: 12345
DOCTOR: Sample Doctor
Doctor's Data, Inc.
3755 Illinois Ave.
St. Charles, IL 60174 U.S.A.

Vaginosis Profile

GRAM STAIN MICROSCOPY			
	Normal	Abnormal	Expected
Lactobacilli		None	Mod - Many
Curved Gram Negative Rods		Many	None
Small Gram Negative Rods		Many	None
Yeast	None		None
RBC's	None		None
WBC's	0		0 - 6
Clue Cells		Present	None
Eosinophils	N/A		None

Eosinophils reported and Wrights Stain performed when WBC's >6

BACTERIAL VAGINOSIS SCORE	
10	score interpretation: 0 - 3 BV not likely 4 - 6 BV indeterminate 7-10 BV highly suggestive

The BV score¹ is calculated based upon the gram stain results and is independent of the yeast, and bacterial cultures.
¹Nugent Scoring System. (Nugent et al. J. Clin. Micro. (1991)29:297-301)

YEAST CULTURE
2+ Candida albicans

Additional Gram Stain Findings:

Rare Gram positive cocci in pairs

BACTERIOLOGY CULTURE		
Expected/Beneficial flora	Commensal (Imbalanced) flora	Dysbiotic flora
NG Lactobacillus spp.	2+ Alpha hemolytic strep 2+ Gamma hemolytic strep 1+ Staphylococcus not aureus	3+ Gardnerella vaginalis 4+ Enterococcus faecalis

NG = No Growth

SPECIMEN DATA

Comments:

Date Collected: 01/14/2019
Date Received: 01/16/2019
Date Completed: 01/23/2019

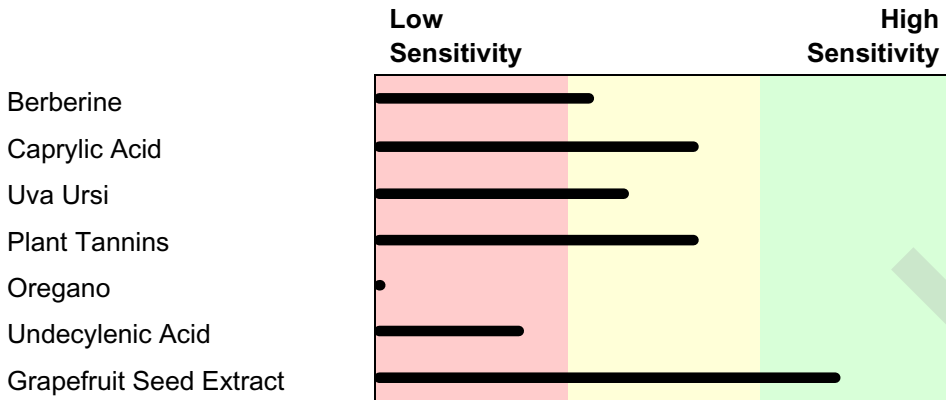


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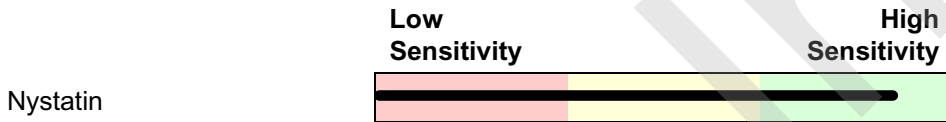
Yeast Susceptibilities: *Candida albicans*

NATURAL ANTIFUNGALS



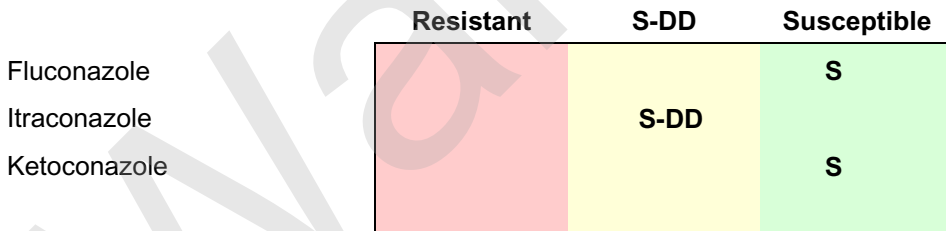
Natural antifungal agents may be useful for treatment of patients when organisms display in-vitro sensitivity to these agents. The test is performed by using standardized techniques and filter paper disks impregnated with the listed agent. Relative sensitivity is reported for each natural agent based upon the diameter of the zone of inhibition surrounding the disk. Data based on over 5000 individual observations were used to relate the zone size to the activity level of the agent. A scale of relative sensitivity is defined for the natural agents tested.

NON-ABSORBED ANTIFUNGALS



Non-absorbed antifungals may be useful for treatment of patients when organisms display in-vitro sensitivity to these agents. The test is performed using standardized commercially prepared disks impregnated with Nystatin. Relative sensitivity is reported based upon the diameter of the zone of inhibition surrounding the disk.

AZOLE ANTIFUNGALS



Susceptible results imply that an infection due to the fungus may be appropriately treated when the recommended dosage of the tested antifungal agent is used.
Susceptible - Dose Dependent (S-DD) results imply that an infection due to the fungus may be treated when the highest recommended dosage of the tested antifungal agent is used.
Resistant results imply that the fungus will not be inhibited by normal dosage levels of the tested antifungal agent.

Standardized test interpretive categories established for *Candida* spp. are used for all yeast isolates.

Comments:

Date Collected: 01/31/2019
 Date Received: 02/01/2019
 Date Completed: 02/07/2019

Yeast antifungal susceptibility testing is intended for research use only.
 Not for use in diagnostic procedures.

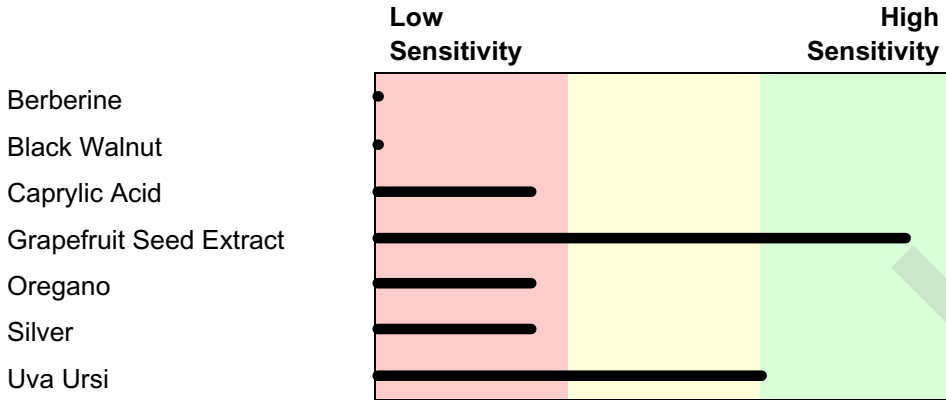


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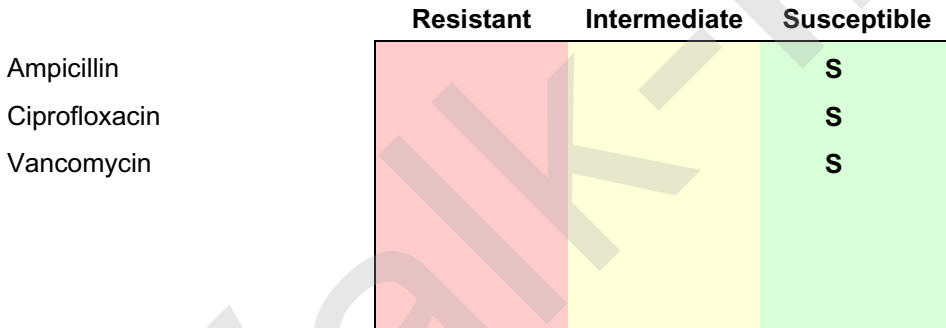
Bacterial Susceptibilities: Enterococcus faecalis

NATURAL ANTIBACTERIALS



Natural antibacterial agents may be useful for treatment of patients when organisms display in-vitro sensitivity to these agents. The test is performed by using standardized techniques and filter paper disks impregnated with the listed agent. Relative sensitivity is reported for each natural agent based upon the diameter of the zone of inhibition surrounding the disk. Data based on over 5000 individual observations were used to relate the zone size to the activity level of the agent. A scale of relative sensitivity is defined for the natural agents tested.

PRESCRIPTIVE AGENTS



Susceptible results imply that an infection due to the bacteria may be appropriately treated when the recommended dosage of the tested antimicrobial agent is used. **Intermediate** results imply that response rates may be lower than for susceptible bacteria when the tested antimicrobial agent is used. **Resistant** results imply that the bacteria will not be inhibited by normal dosage levels of the tested antimicrobial agent.

Comments:

Date Collected: 01/31/2019
 Date Received: 02/01/2019
 Date Completed: 02/07/2019

Natural antibacterial agent susceptibility testing is intended for research use only.
 Not for use in diagnostic procedures.

v10.11

Introductory paragraph

This test was performed to identify the cause(s) of symptoms associated with vaginitis. Infectious vaginitis is the most common type of vaginitis in women of reproductive age and is usually a result of abnormal vaginal microflora. Alterations in normal vaginal flora are antecedent to infections by dysbiotic bacteria, yeast or a pathogenic parasite (*Trichomonas vaginalis*). This comprehensive test utilizes two types of methodology: microscopic evaluation enhanced by standard cell staining techniques and culture. Gram staining differentiates bacterial species into two major groups (Gram-negative and Gram-positive). The microscopic evaluation can also reveal the presence of fungi (yeast) and important cell types that facilitate calculation of the Bacterial Vaginosis Score. Samples are also cultured for yeast to identify the species and perform susceptibility testing against natural and pharmaceutical agents.

Not all bacteria can be definitively classified by the Gram stain so samples are also cultured using standardized microbiological techniques. Expected/Beneficial bacteria (*Lactobacillus* species) are grown under anaerobic and aerobic conditions and their abundance is reported as no growth (NG) to 4+ (most abundant). Imbalanced bacteria are those bacteria that are neither typically harmful nor beneficial to the patient. Some imbalanced bacteria that present at a level of 1-2+, are reported as imbalance and susceptibility testing is not performed. When imbalanced bacteria grow in culture to a level of 3-4+ they are reported as Dysbiotic bacteria and susceptibility testing is performed; the results are reported to assist the attending clinician in developing the most efficacious treatment program. When abnormal test results are reported explanatory paragraphs are provided.

Note: Not all genera or species can be tested for susceptibility in the laboratory due to their specific growth requirements.

Lactobacilli - Abnormal

The level of Lactobacilli assessed by Gram stain or culture is abnormal in this sample. Healthy vaginal flora is composed of members of the *Lactobacillus* genus, which should constitute about 95% of the bacteria normally found in the vagina; they produce a protective biofilm on the mucosa. This predominance begins at the time of puberty, probably because of the effect of estrogens on the glycogen content of vaginal epithelial cells [1]. These bacteria have a beneficial effect by inhibiting growth, adhesion or spread of other microorganisms. The recognized mechanisms include:

- o secretion of organic acids keeping pH <4.5 [2,3]
- o production of antimicrobial substances (hydrogen peroxide, bacteriocins and biosurfactants)
- o competition for nutrients (arginine deaminase)
- o competition for receptors (adhesion on the epithelium)
- o steric exclusion (biosurfactants, adhesion on the epithelium or on the fibronectin)
- o co-aggregation

Lack of H₂O₂ producing lactobacilli predisposes women to bacterial vaginosis by allowing the overgrowth of *Gardnerella* and other anaerobic bacteria. Lactobacilli have also been known to interfere with adherence and colonization of pathogenic bacteria to the cells of the vagina possibly through production of a bacteriocin [2-4]. Three strains of *Lactobacillus* (*acidophilus*, *gasseri*, *jensenii*) were all found to

adhere to epithelial vaginal cells, displacing well-known vaginal pathogens, such as *G. vaginalis* and inhibiting the growth in vitro of *Escherichia coli*, *Streptococcus agalactiae*, and *Prevotella* spp. [5]. Loss of *Lactobacillus* results in bacterial vaginosis or vaginitis and possible urinary tract infections.[1,2] *Lactobacillus* has been known to decline when estrogen levels fall, such as in postpartum or menopausal women [1].

1. Schwebke JR. Gynecologic consequences of bacterial vaginosis. *Obstet Gynecol Clin North Am*, 30(4):685-694, 2003.
2. Faro S. *Vaginitis Differential Diagnosis and Management*. New York: CRC Press, 2004.
3. Tomas MS, Claudia Otero M, Ocana V, Elena Nader-Macias M. Production of antimicrobial substances by lactic acid bacteria in determination of hydrogen peroxide. *Methods Mol Biol*, 268:337-346, 2004.
4. Chan R, Bruce A, Reid G. Adherence of cervical, vaginal and distal urethral normal microbial flora to human uroepithelial cells and the inhibition of adherence of gram-negative uropathogens by competitive exclusion. *J Urol*, 131:596-601, 1984.
5. Boris S, Suarez JE, Vazquez F, Barbes C. Adherence of human vaginal lactobacilli to vaginal epithelial cells and interaction with uropathogens. *Infect Immun*, 66(5):1985-1989, 1998.

Curved Gram Negative Rods -Abnormal

Curved gram-negative rods were detected in this sample. The presence of curved gram negative rods is an indicator of *Mobiluncus* spp., an anaerobic bacteria associated with bacterial vaginosis (see the Bacterial Vaginosis Score). A prime example of the anaerobic bacterium is the *Mobiluncus* spp. *Mobiluncus* produces a toxin that could have an active role in the development of bacterial vaginosis[1]. Sensitivity testing cannot be performed on these anaerobic bacteria since they require a special oxygen-free environment. Metronidazole and Clindamycin are treatments of choice for vaginal anaerobes[2].

1. Taylor-Robinson AW, Borriello SP, Taylor-Robinson D. Identification and preliminary characterization of a cytotoxin isolated from *Mobiluncus* spp., 74(4):357-366, 1993.
2. Schwebke JR. Gynecologic consequences of bacterial vaginosis. *Obstet Gynecol Clin North Am*, 30(4):685-694, 2003.

Small Gram Negative Rods -Abnormal

Small gram-negative rods were detected in this sample. The presence of small gram-negative or gram-variable rods is an indicator of *Gardnerella vaginalis* and other anaerobic gram-negative bacteria that can be seen in bacterial vaginosis [1]. It has not been proven that any of these organisms alone are able to cause bacterial vaginosis, but their numbers are increased in cases of bacterial vaginosis (check the Bacterial Vaginosis Score). The normal *Lactobacilli* are replaced by an overgrowth of a mixed flora of aerobes and anaerobes, including *Gardnerella vaginalis* (in quantities of >105 CFU/mL) and *Prevotella* species [2].

1. Spiegel CA, Amsel R, Holmes KK. Diagnosis of Bacterial Vaginosis by Direct Gram Stain on Vaginal Fluid. *Journal of Clinical Microbiology*, July 1983: 170-177.
2. Washington W, Allen S, Janda W, Koneman E, Procop G, Schreckenberger P, Woods, G. *Color Atlas and Textbook of Diagnostic Microbiology*, 6th edition. Lippincott Williams and Wilkins; 2006. pg 834-838.

Bacterial Score - Abnormal

Gram stain shows grossly altered vaginal flora (7-10), indicative of bacterial vaginosis. Note: Gram stain scoring of vaginal smears for post menopausal women has not been standardized but clinical correlations have been reported.

Dysbiotic Bacteria

Dysbiotic bacteria were detected in culture from this sample. It is very important to refer to the Gram Stain Microscopy, Bacteriology Culture, and the presence or absence of Lactobacilli before considering if these organisms are vaginally pathogenic in this patient. Organisms found in this category may be colonizing without causing harm or they may represent contamination from the gastrointestinal tract. Lactobacillus species are known to be the dominant vaginal bacteria that control the normal flora by keeping potentially harmful bacteria in check [1]. For potential treatment options see the Bacterial Susceptibilities section in this report.

1. Faro S. *Vaginitis Differential Diagnosis and Management*. New York: CRC Press, 2004.

Gardnerella Vaginalis

The bacteria *Gardnerella vaginalis* (*G. vaginalis*) was detected in culture for this sample. It is present in almost all cases of Bacterial Vaginosis at levels 100-1,000 times higher than in non-infected women. However, *G. vaginalis* may be detected in 50-60% of women who do not meet the diagnostic criteria for BV ; carrier rates are as high as 10-40% in asymptomatic women.

For *Gardnerella vaginalis* CDC recommends treatment with metronidazole 500 mg. orally twice daily for seven days, or metronidazole gel 0.75%, one full applicator (5 g.) intravaginally, once a day for five days, or clindamycin cream 2%, one full applicator (5 g.) intravaginally at bedtime for seven days. It is important to advise patients receiving metronidazole as a treatment that they should avoid consuming alcohol during treatment and for 24 hours thereafter [1].

[1] <http://www.cdc.gov/std/treatment/2010/qanda/vaginitis.htm>

Yeast

Yeast was detected by microscopic examination or in culture for this sample. Symptoms of vaginal yeast infections include: itching, inflammation, burning sensation, and white, cheesy discharge.

Vaginal yeast overgrowth, referred to as vulvovaginal candidiasis, is most commonly associated with overgrowth of *Candida albicans* (>85%) but *Candida tropicalis*, *Candida glabrata* and *Candida krusei* have become increasingly prevalent causal factors [1-4]. *Candida* vulvovaginitis is a common problem and it has been estimated that 75% of all women will experience one episode caused by *Candida* [1]. In one study, recurrent vaginal candidiasis was more common in women less than 40 years of age and in those with a history of bacterial vaginosis [5]. Certain voluntary factors such as use of panty liners, panty hose and oral contraceptives have also been associated with recurrent vaginal candidiasis [5,6].

1. Faro S. *Vaginitis Differential Diagnosis and Management*. New York: CRC Press, 2004.
2. Sobel JD. Treatment of vaginal *Candida* infections. *Expert Opin Pharmacother*, 3(8):1059-1065, 2002.
3. Mendoza M, Gonzalez I, Bellorin EJ, Salazar W, Mendoza L, Zambrand EA, de Albornoz MC. Isolation, identification and serotyping of yeasts obtained from the vaginal fluid in patients with clinical vaginitis. *Invest Clin*, 40(1):25-36, 1999.
4. Plourd DM. Practical guide to diagnosing and treating vaginitis. *Medscape Womens Health*, 2(2):2, 1997
5. Patel DA, Gillespie B, Sobel JD, Leaman D, Nyirjesy P, Weitz MV, Foxman B. Risk factors for recurrent vulvovaginal candidiasis in women receiving maintenance antifungal therapy: results of a prospective cohort study. *Am J Obstet Gynecol*, 190(3):644-653,2004.
6. Heidrich FE, Berg AO, Bergman JJ. Clothing factors and vaginitis. *J Fam Pract*, 19(4):491-494, 1984.

Enterococcus species

Microorganisms that are now included in the genus *Enterococcus* were primarily related to the "streptococci of fecal origin" in the recent past. These catalase-negative gram-positive cocci are widespread in nature and can be found in soil, water, food, plants, and in animals including mammals, birds, and insects. They mainly reside in the gastrointestinal tract and are less common in the genitourinary tract and oral cavity [1]. Some important clinical infections caused by *Enterococcus* in immune-compromised individuals include urinary tract infections, bacteremia, bacteria endocarditis, diverticulitis, and meningitis [2].

A vaginal infection caused by *Enterococcus* may present as vulvovaginal itching and burning, erythema and discharge. One particular strain, *Enterococcus faecium* 62-6, was found to inhibit growth of protective *Lactobacilli* which contributed to the establishment and recurrence of bacterial vaginosis [4]. It is more commonplace that bacterial vaginosis is caused by *Enterococcus* strains that are not of the *faecalis* or *faecium* species [3]. *Enterococcus faecalis* and *faecium* represent 90% of isolates however the incidence of other *Enterococcus* isolates is increasing [4].

There is a high level of antibiotic resistance especially with the more common species so that only sensitive strains may be treated with ampicillin and vancomycin [1,3]. *Enterococcus raffinosus* has been shown to be susceptible only to vancomycin, teicoplanin and rifampicin [5]. Refer to the Bacterial Susceptibilities section of this report for possible treatment options.

1. *Enterococcus*. In PR Murray P, Baron E, Jorgensen et al. Manual of Clinical Microbiology, 9th ed. ASM Press, Washington D.C., 2007
2. Fisher K, Phillips C. The ecology, epidemiology and virulence of *Enterococcus*. *Microbiology* 155(6):1749-57, 2009.
3. Savini V, Manna A, D'Antonio F, Talia M, Catavittello C, Balbinot A, Febbo F, Crlino D, Firitoni F, Di Bonaventura G, D'Antonio. First report of vaginal infection caused by *Enterococcus raffinosus*. *J Medical Microbiology* 57, 672-673, 2008.
4. Kelly M, Mequio M, Pybus V. Inhibition of vaginal *Lactobacilli* by a bacteriocin-like inhibitor produce by *Enterococcus faecium* 62-6: potential significance for bacterial vaginosis. *Infec Dis Obstet Gynecol* 11:147-156, 2003.
5. Prakash V, Rao S, Parija S. Emergence of unusual species of enterococci causing infections, South India. *BMC Infect Dis* 5:14, 2005.