



LAB #: F000000-0000-0
 PATIENT: Sample Patient
 ID: P000000000
 SEX: Male
 DOB: 01/01/2001 AGE: 12

CLIENT #: 12345
 DOCTOR:
 Doctor's Data, Inc.
 3755 Illinois Ave.
 St. Charles, IL 60174 USA

Comprehensive Stool Analysis / Parasitology x3

BACTERIOLOGY CULTURE

Expected/Beneficial flora	Commensal (Imbalanced) flora	Dysbiotic flora
NG Bacteroides fragilis group	2+ Hemolytic Escherichia coli	3+ Citrobacter freundii
NG Bifidobacterium spp.	1+ Staphylococcus aureus	
3+ Escherichia coli		
3+ Lactobacillus spp.		
2+ Enterococcus spp.		
2+ Clostridium spp.		
NG = No Growth		

BACTERIA INFORMATION

Expected /Beneficial bacteria make up a significant portion of the total microflora in a healthy & balanced GI tract. These beneficial bacteria have many health-protecting effects in the GI tract including manufacturing vitamins, fermenting fibers, digesting proteins and carbohydrates, and propagating anti-tumor and anti-inflammatory factors.

Clostridia are prevalent flora in a healthy intestine. Clostridium spp. should be considered in the context of balance with other expected/beneficial flora. Absence of clostridia or over abundance relative to other expected/beneficial flora indicates bacterial imbalance. If *C. difficile* associated disease is suspected, a Comprehensive Clostridium culture or toxigenic *C. difficile* DNA test is recommended.

Commensal (Imbalanced) bacteria are usually neither pathogenic nor beneficial to the host GI tract. Imbalances can occur when there are insufficient levels of beneficial bacteria and increased levels of commensal bacteria. Certain commensal bacteria are reported as dysbiotic at higher levels.

Dysbiotic bacteria consist of known pathogenic bacteria and those that have the potential to cause disease in the GI tract. They can be present due to a number of factors including: consumption of contaminated water or food, exposure to chemicals that are toxic to beneficial bacteria; the use of antibiotics, oral contraceptives or other medications; poor fiber intake and high stress levels.

YEAST CULTURE

Normal flora	Dysbiotic flora
1+ Geotrichum spp	

MICROSCOPIC YEAST

Result:	Expected:
Many	None - Rare

The microscopic finding of yeast in the stool is helpful in identifying whether there is proliferation of yeast. Rare yeast may be normal; however, yeast observed in higher amounts (few, moderate, or many) is abnormal.

YEAST INFORMATION

Yeast normally can be found in small quantities in the skin, mouth, intestine and mucocutaneous junctions. Overgrowth of yeast can infect virtually every organ system, leading to an extensive array of clinical manifestations. Fungal diarrhea is associated with broad-spectrum antibiotics or alterations of the patient's immune status. Symptoms may include abdominal pain, cramping and irritation. When investigating the presence of yeast, disparity may exist between culturing and microscopic examination. Yeast are not uniformly dispersed throughout the stool, this may lead to undetectable or low levels of yeast identified by microscopy, despite a cultured amount of yeast. Conversely, microscopic examination may reveal a significant amount of yeast present, but no yeast cultured. Yeast does not always survive transit through the intestines rendering it unviable.

Comments:

Date Collected: 08/18/2013
 Date Received: 08/21/2013
 Date Completed: 08/28/2013

* *Aeromonas, Campylobacter, Plesiomonas, Salmonella, Shigella, Vibrio, Yersinia, & Edwardsiella tarda* have been specifically tested for and found absent unless reported.





LAB #: F000000-0000-0
 PATIENT: Sample Patient
 ID: P000000000
 SEX: Male
 DOB: 801/02/2001 AGE: 12

CLIENT #: 12345
 DOCTOR:
 Doctor's Data, Inc.
 3755 Illinois Ave.
 St. Charles, IL 60174 USA

Comprehensive Stool Analysis / Parasitology x3

PARASITOLOGY/MICROSCOPY *
<p>Sample 1</p> <p>None Ova or Parasites Many Yeast</p>
<p>Sample 2</p> <p>None Ova or Parasites Mod Yeast</p>
<p>Sample 3</p> <p>None Ova or Parasites Few Yeast</p>
<p>*A trichrome stain and concentrated iodine wet mount slide is read for each sample submitted.</p>

PARASITOLOGY INFORMATION
<p>Intestinal parasites are abnormal inhabitants of the gastrointestinal tract that have the potential to cause damage to their host. The presence of any parasite within the intestine generally confirms that the patient has acquired the organism through fecal-oral contamination. Damage to the host includes parasitic burden, migration, blockage and pressure. Immunologic inflammation, hypersensitivity reactions and cytotoxicity also play a large role in the morbidity of these diseases. The infective dose often relates to severity of the disease and repeat encounters can be additive.</p>
<p>There are two main classes of intestinal parasites, they include protozoa and helminths. The protozoa typically have two stages; the trophozoite stage that is the metabolically active, invasive stage and the cyst stage, which is the vegetative inactive form resistant to unfavorable environmental conditions outside the human host. Helminths are large, multicellular organisms. Like protozoa, helminths can be either free-living or parasitic in nature. In their adult form, helminths cannot multiply in humans.</p>
<p>In general, acute manifestations of parasitic infection may involve diarrhea with or without mucus and or blood, fever, nausea, or abdominal pain. However these symptoms do not always occur. Consequently, parasitic infections may not be diagnosed or eradicated. If left untreated, chronic parasitic infections can cause damage to the intestinal lining and can be an unsuspected cause of illness and fatigue. Chronic parasitic infections can also be associated with increased intestinal permeability, irritable bowel syndrome, irregular bowel movements, malabsorption, gastritis or indigestion, skin disorders, joint pain, allergic reactions, and decreased immune function.</p>
<p>In some instances, parasites may enter the circulation and travel to various organs causing severe organ diseases such as liver abscesses and cysticercosis. In addition, some larval migration can cause pneumonia and in rare cases hyper infection syndrome with large numbers of larvae being produced and found in every tissue of the body.</p>
<p>One negative parasitology x1 specimen does not rule out the possibility of parasitic disease, parasitology x3 is recommended. This exam is not designed to detect <i>Cryptosporidium</i> spp, <i>Cyclospora cayetanensis</i> or <i>Microsporidia</i> spp.</p>

GIARDIA/CRYPTOSPORIDIUM IMMUNOASSAY			
	Within	Outside	Reference Range
Giardia intestinalis	Neg		Neg
Cryptosporidium	Neg		Neg

Giardia intestinalis (lamblia) is a protozoan that infects the small intestine and is passed in stool and spread by the fecal-oral route. Waterborne transmission is the major source of giardiasis.

Cryptosporidium is a coccidian protozoa that can be spread from direct person-to-person contact or waterborne transmission.

Comments:

Date Collected: 08/18/2013
 Date Received: 08/21/2013
 Date Completed: 08/28/2013



LAB #: F000000-0000-0
 PATIENT: Sample Patient
 ID: P000000-0000-0
 SEX: Male
 DOB: 01/01/2001 AGE: 12

CLIENT #: 12345
 DOCTOR:
 Doctor's Data, Inc.
 3755 Illinois Ave.
 St. Charles, IL 60174 USA

Comprehensive Stool Analysis / Parasitology x3

DIGESTION / ABSORPTION

	Within	Outside	Reference Range
Elastase	463		> 200 µg/mL
Fat Stain	None		None - Mod
Muscle fibers	None		None - Rare
Vegetable fibers	Rare		None - Few
Carbohydrates	Neg		Neg

Elastase findings can be used for the diagnosis or the exclusion of exocrine pancreatic insufficiency. Correlations between low levels and chronic pancreatitis and cancer have been reported. **Fat Stain:** Microscopic determination of fecal fat using Sudan IV staining is a qualitative procedure utilized to assess fat absorption and to detect steatorrhea. **Muscle fibers** in the stool are an indicator of incomplete digestion. Bloating, flatulence, feelings of "fullness" may be associated with increase in muscle fibers. **Vegetable fibers** in the stool may be indicative of inadequate chewing, or eating "on the run". **Carbohydrates:** The presence of reducing substances in stool specimens can indicate carbohydrate malabsorption.

INFLAMMATION

	Within	Outside	Reference Range
Lysozyme*	117		<= 600 ng/mL
Lactoferrin	< 0.5		< 7.3 µg/mL
White Blood Cells	None		None - Rare
Mucus	Neg		Neg

Lysozyme* is an enzyme secreted at the site of inflammation in the GI tract and elevated levels have been identified in IBD patients. **Lactoferrin** is a quantitative GI specific marker of inflammation used to diagnose and differentiate IBD from IBS and to monitor patient inflammation levels during active and remission phases of IBD. **White Blood Cells (WBC):** in the stool are an indication of an inflammatory process resulting in the infiltration of leukocytes within the intestinal lumen. WBCs are often accompanied by mucus and blood in the stool. **Mucus** in the stool may result from prolonged mucosal irritation or in a response to parasympathetic excitability such as spastic constipation or mucous colitis.

IMMUNOLOGY

	Within	Outside	Reference Range
Secretory IgA*		17.7	51 - 204mg/dL

Secretory IgA* (sIgA) is secreted by mucosal tissue and represents the first line of defense of the GI mucosa and is central to the normal function of the GI tract as an immune barrier. Elevated levels of sIgA have been associated with an upregulated immune response.

Comments:

Date Collected: 08/18/2013
 Date Received: 08/21/2013
 Date Completed: 08/28/2013

*For Research Use Only. Not for use in diagnostic procedures.



LAB #: F000000-0000-0
 PATIENT: Sample Patient
 ID: P000000000
 SEX: Male
 DOB: 01/01/2001 AGE: 12

CLIENT #: 12345
 DOCTOR:
 Doctor's Data, Inc.
 3755 Illinois Ave.
 St. Charles, IL 60174 USA

Comprehensive Stool Analysis / Parasitology x3

SHORT CHAIN FATTY ACIDS

	Within	Outside	Reference Range
% Acetate	67		36 - 74 %
% Propionate	20		9 - 32 %
% Butyrate	12		9 - 39 %
% Valerate	1.6		1 - 8 %
Butyrate	1.3		0.8 - 3.8 mg/mL
Total SCFA's	10		4 - 14 mg/mL

Short chain fatty acids (SCFAs): SCFAs are the end product of the bacterial fermentation process of dietary fiber by beneficial flora in the gut and play an important role in the health of the GI as well as protecting against intestinal dysbiosis. Lactobacilli and bifidobacteria produce large amounts of short chain fatty acids, which decrease the pH of the intestines and therefore make the environment unsuitable for pathogens, including bacteria and yeast. Studies have shown that SCFAs have numerous implications in maintaining gut physiology. SCFAs decrease inflammation, stimulate healing, and contribute to normal cell metabolism and differentiation. Levels of **Butyrate** and **Total SCFA** in mg/mL are important for assessing overall SCFA production, and are reflective of beneficial flora levels and/or adequate fiber intake.

INTESTINAL HEALTH MARKERS

	Within	Outside	Reference Range
Red Blood Cells	None		None - Rare
pH	6.7		6 - 7.8
Occult Blood	Neg		Neg

Red Blood Cells (RBC) in the stool may be associated with a parasitic or bacterial infection, or an inflammatory bowel condition such as ulcerative colitis. Colorectal cancer, anal fistulas, and hemorrhoids should also be ruled out.

pH: Fecal pH is largely dependent on the fermentation of fiber by the beneficial flora of the gut.

Occult blood: A positive occult blood indicates the presence of free hemoglobin found in the stool, which is released when red blood cells are lysed.

MACROSCOPIC APPEARANCE

	Appearance	Expected
Color	Brown	Brown
Consistency	Loose/Watery	Formed/Soft

Color: Stool is normally brown because of pigments formed by bacteria acting on bile introduced into the digestive system from the liver. While certain conditions can cause changes in stool color, many changes are harmless and are caused by pigments in foods or dietary supplements. **Consistency:** Stool normally contains about 75% water and ideally should be formed and soft. Stool consistency can vary based upon transit time and water absorption.

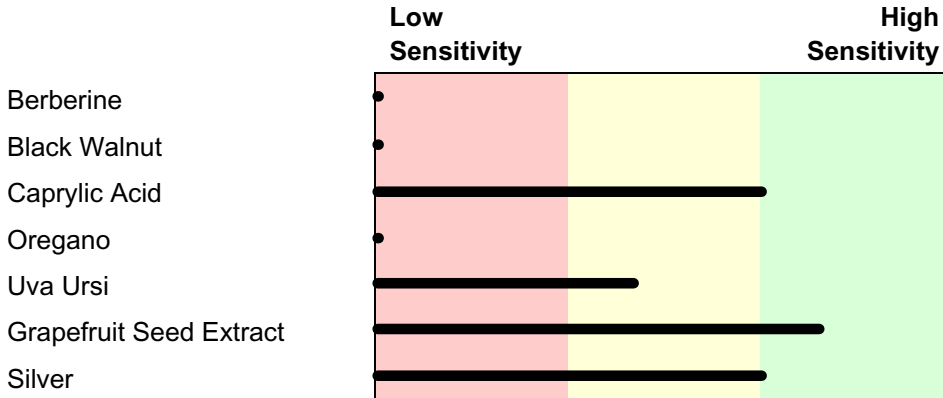


LAB #: F000000-0000-0
 PATIENT: Sample Patient
 ID: P000000000
 SEX: Male
 AGE: 12

CLIENT #: 12345
 DOCTOR:
 Doctor's Data, Inc.
 3755 Illinois Ave.
 St. Charles, IL 60174 USA

Bacterial Susceptibilities: Citrobacter freundii

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

	Resistant	Intermediate	Susceptible
Amoxicillin-Clavulanic Acid	R		
Ampicillin	R		
Cefazolin	R		
Ceftazidime			S
Ciprofloxacin			S
Trimeth-sulfa			S

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: 08/18/2013
 Date Received: 08/21/2013
 Date Completed: 08/28/2013

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

v10.11

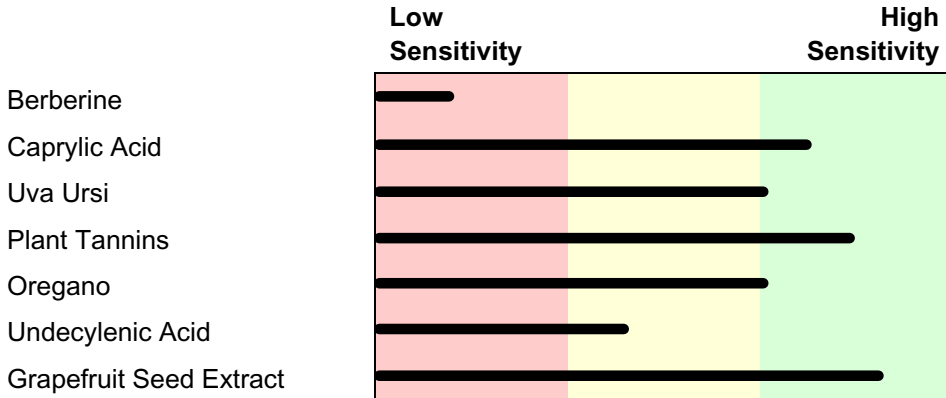


LAB #: F000000-0000-0
PATIENT: Sample Patient
ID: P000000000
SEX: Male
AGE: 12

CLIENT #: 12345
DOCTOR:
Doctor's Data, Inc.
3755 Illinois Ave.
St. Charles, IL 60174 USA

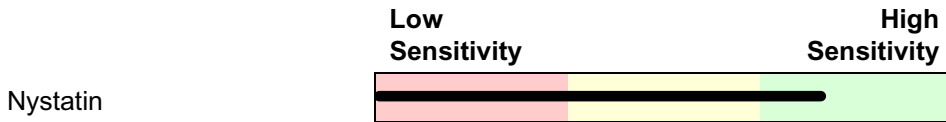
Yeast Susceptibilities: *Geotrichum spp*

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.

Comments:

Date Collected: 08/18/2013
Date Received: 08/21/2013
Date Completed: 08/28/2013

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

v10.11

INTRODUCTION

This analysis of the stool specimen provides fundamental information about the overall gastrointestinal health of the patient. When abnormal microflora or significant aberrations in intestinal health markers are detected, specific interpretive paragraphs are presented. If no significant abnormalities are found, interpretive paragraphs are not presented.

Beneficial Flora

One or more of the expected (beneficial) bacteria are low in this specimen. Beneficial flora include lactobacilli, bifidobacteria, clostridia, *Bacteroides fragilis* group, enterococci, and some strains of *Escherichia coli*. The beneficial flora have many health-protecting effects in the gut, and as a consequence, are crucial to the health of the whole organism. Some of the roles of the beneficial flora include digestion of proteins and carbohydrates, manufacture of vitamins and essential fatty acids, increase in the number of immune system cells, break down of bacterial toxins and the conversion of flavinoids into anti-tumor and anti-inflammatory factors. Lactobacilli, bifidobacteria, clostridia, and enterococci secrete lactic acid as well as other acids including acetate, propionate, butyrate, and valerate. This secretion causes a subsequent decrease in intestinal pH, which is crucial in preventing an enteric proliferation of microbial pathogens, including bacteria and yeast. Many GI pathogens thrive in alkaline environments. Lactobacilli also secrete the antifungal and antimicrobial agents lactocidin, lactobacillin, acidolin, and hydrogen peroxide. The beneficial flora of the GI have thus been found useful in the inhibition of microbial pathogens, prevention and treatment of antibiotic associated diarrhea, prevention of traveler's diarrhea, enhancement of immune function, and inhibition of the proliferation of yeast.

In a healthy balanced state of intestinal flora, the beneficial flora make up a significant proportion of the total microflora. Healthy levels of each of the beneficial bacteria are indicated by either a 3+ or 4+ (0 to 4 scale). However, some individuals have low levels of beneficial bacteria and an overgrowth of nonbeneficial (imbalances) or even pathogenic microorganisms (dysbiosis). Often attributed to the use of antibiotics, individuals with low beneficial bacteria may present with chronic symptoms such as irregular transit time, irritable bowel syndrome, bloating, gas, chronic fatigue, headaches, autoimmune diseases (e.g., rheumatoid arthritis), and sensitivities to a variety of foods. Treatment may include the use of probiotic supplements containing various strains of lactobacilli, bifidobacteria and enterococci and consumption of cultured or fermented foods including yogurt, kefir, miso, tempeh and tamari sauce. Polyphenols in green and ginseng tea have been found to increase the numbers of beneficial bacteria. If dysbiosis is present, treatment may also include the removal of pathogenic bacteria, yeast, or parasites.

Percival M. Intestinal Health. *Clin Nutr In.* 1997;5(5):1-6.

Fuller R. Probiotics in Human Medicine. *Gut.* 1991;32: 439-442.

Siitonen S, Vapaatalo H, Salminen S, et al. Effect of Lactobacilli GG Yoghurt in Prevention of Antibiotic Associated Diarrhea. *Ann Med.* 1990; 22:57-59.

Oksanen P, Salminen S, Saxelin M, et al. Prevention of Travelers' Diarrhea by Lactobacillus GG. *Ann Med.* 1990; 22:53-56.

Perdigon G, Alvarez M, et al. The Oral Administration of Lactic Acid Bacteria Increases the Mucosal Intestinal Immunity in Response to Enteropathogens. *J Food Prot.* 1990;53:404-410.

Valeur, N, et al. Colonization and Immunomodulation by Lactobacillus reuteri ATCC 55730 in the Human Gastrointestinal Tract. *Appl Environ. Microbiol.* 2004 Feb; 70(2):1176-81.

Elmer G, Surawicz C, and McFarland L. Biotherapeutic agents - a Neglected Modality for the Treatment and Prevention of Intestinal and Vaginal Infections. *JAMA.* 1996; 275(11):870-876.

Fitzsimmons N and Berry D. Inhibition of Candida albicans by Lactobacillus acidophilus: Evidence for Involvement of a Peroxidase System. *Microbio.* 1994; 80:125-133

Weisburger JH. *Proc Soc Exp Biol Med* 1999;220(4):271-5.

Clostridium spp

Clostridia are expected inhabitants of the human intestine. Although most clostridia in the intestine are not virulent, certain species have been associated with disease. Clostridium perfringens is a major cause of food poisoning and is also one cause of antibiotic-associated diarrhea. Clostridium difficile is a causative agent in antibiotic-associated diarrhea and pseudomembranous colitis. Other species reported to be prevalent in high amounts in patients with Autistic Spectrum Disorder include Clostridium histolyticum group, Clostridium cluster I, Clostridium bolteae, and Clostridium tetani.

If these disease associations are a concern further testing may be necessary.

Washington W, Allen S, Janda W, Koneman E, Procop G, Schreckenberger P, Woods, G. Koneman's Color Atlas and Textbook of Diagnostic Microbiology, 6th edition. Lippincott Williams and Wilkins; 2006. pg 931-939

Song Y, Liu C, Finegold SM. Real-Time PCR Quantitation of Clostridia in Feces of Autistic Children. *Applied and Environmental Microbiology.* Nov. 2004, 6459-6465.

Parracho H, Bingham MO, Gibson GR, McCartney AL. Differences Between the Gut Microflora of Children with Autistic Spectrum Disorders and That of Healthy Children. *Journal of Medical Microbiology.* 2005;54, 987-991.

Imbalanced flora

Imbalanced flora are those bacteria that reside in the host gastrointestinal tract and neither

injure nor benefit the host. Certain dysbiotic bacteria may appear under the imbalances category if found at low levels because they are not likely pathogenic at the levels detected. When imbalanced flora appear, it is not uncommon to find inadequate levels of one or more of the beneficial bacteria and/or a fecal pH which is more towards the alkaline end of the reference range (6.5 - 7.2). It is also not uncommon to find hemolytic or mucoid E. coli with a concomitant deficiency of beneficial E. coli and alkaline pH, secondary to a mutation of beneficial E. coli in alkaline conditions (DDI observations). Treatment with antimicrobial agents is unnecessary unless bacteria appear under the dysbiotic category.

Mackowiak PA. The normal microbial flora. N Engl J Med. 1982;307(2):83-93.

Dysbiotic Flora

In a healthy balanced state of intestinal flora, the beneficial bacteria make up a significant proportion of the total microflora. However, in many individuals there is an imbalance or deficiency of beneficial flora and an overgrowth of non-beneficial (imbalance) or even pathogenic microorganisms (dysbiosis). This can be due to a number of factors including: consumption of contaminated water or food; daily exposure of chemicals that are toxic to beneficial bacteria; the use of antibiotics, oral contraceptives or other medications; poor fiber intake and high stress levels.

A number of toxic substances can be produced by the dysbiotic bacteria including amines, ammonia, hydrogen sulfide, phenols, and secondary bile acids which may cause inflammation or damage to the brush border of the intestinal lining. If left unchecked, long-term damage to the intestinal lining may result in leaky gut syndrome, allergies, autoimmune disease (e.g. rheumatoid arthritis), irritable bowel syndrome, fatigue, chronic headaches, and sensitivities to a variety of foods. In addition, pathogenic bacteria can cause acute symptoms such as abdominal pain, nausea, diarrhea, vomiting, and fever in cases of food poisoning.

Bacterial sensitivities to a variety of prescriptive and natural agents have been provided for the pathogenic bacteria that were cultured from this patient's specimen. This provides the practitioner with useful information to help plan an appropriate treatment regimen. Supplementation with probiotics or consumption of foods (yogurt, kefir, miso, tempeh, tamari sauce) containing strains of lactobacilli, bifidobacteria, and enterococci can help restore healthy flora levels. Polyphenols in green and ginseng tea have been found to increase the numbers of beneficial bacteria. Hypochlorhydria may also predispose an individual to bacterial overgrowth, particularly in the small intestine. Nutritional anti-inflammatories can aid in reversing irritation to the GI lining. These include quercetin, vitamin C, curcumin, gamma-linoleic acid, omega-3 fatty acids (EPA, DHA), and aloe vera. Other nutrients such as zinc, beta-carotene, pantothenic acid, and L-glutamine provide support for regeneration of the GI mucosa. A comprehensive program may be helpful in individuals in whom a dysbiotic condition has caused extensive GI damage.

Lispki E. Digestive Wellness. New Canaan,CT: Keats Publishing;1996.

Mitsuoka T. Intestinal Flora and Aging. Nutr Rev 1992;50(12):438-446.

Weisburger JH. Tea and Health: The Underlying Mechanisms. Proc Soc Exp Biol Med

1999;220(4):271-275.4.

Pereira SP, Gainsborough N, Dowling RH. Drug-induced Hypochlorhydria Causes High Duodenal Bacterial Counts in the Elderly. *Ailment Pharmacol Ther* 1998;12(1)99-104.

Murray MT. *Stomach Ailments and Digestive Disturbances*. Rocklin, CA: Prima Publishing; 1997.

Citrobacter species

Citrobacter species, a gram-negative bacterium and member of the Enterobacteriaceae family, is considered dysbiotic at 3+ - 4+.

Citrobacter freundii complex, including Citrobacter freundii, Citrobacter youngae, Citrobacter braakii, Citrobacter sedlakii, Citrobacter werkmanii, and less commonly Citrobacter koseri and Citrobacter farmeri, can cause diarrheal disease. Symptoms due to Citrobacter freundii complex seem to be a result of the elaboration of an E. coli-like heat-stable enterotoxin and hydrogen sulfide. Citrobacter freundii complex has been implicated as a cause of gastrointestinal infection and inflammation, acute dysentery, and dyspepsia. Acute symptoms can include profuse, watery diarrhea which is often unaccompanied by abdominal pain, fecal blood, or white blood cells.

Citrobacter species thrive on Fructooligosaccharides (FOS), a common ingredient in artificial or alternative sweetener.

Antibiotics may be indicated if symptoms are prolonged and in systemic infections. Refer to the bacterial sensitivities to identify the most appropriate pharmaceutical or natural agent.

Guarino, A, et al. Production of Escherichia coli Sta-Like Heat-Stable Enterotoxin by Citrobacter freundii Isolated from Humans. *Journal of Clinical Microbiology*. January 1987;110-114.

Washington W, Allen S, Janda W, Koneman E, Procop G, Schreckenberger P, Woods, G. *Koneman's Color Atlas and Textbook of Diagnostic Microbiology*, 6th edition. Lippincott Williams and Wilkins; 2006. pg 686-689.

Murray PR, Baron EJ, Jorgensen JH, Pfaller MA, Tenover FC, Tenover BC. *Manual of Clinical Microbiology*, 8th edition. Washington, DC: ASM Press; 2003. pg 701-704.

Cultured Yeast

Yeast, such as Candida are normally present in the GI tract in very small amounts. Many species of yeast exist and are commensal; however, they are always poised to create opportunistic infections and have detrimental effects throughout the body. Factors that contribute to a proliferation of yeast include frequent use of wide-spread antibiotics/low levels of beneficial flora, oral contraceptives, pregnancy, cortisone and other immunosuppressant drugs, weak immune system/low levels of sIgA, high-sugar diet, and high stress levels.

When investigating the presence of yeast, disparity may exist between culturing and microscopic examination. Yeast grows in colonies and is typically not uniformly dispersed throughout the stool. This may lead to undetectable or low levels of yeast identified by microscopy, despite a cultured amount of yeast. Conversely, microscopic examination may reveal a significant amount of yeast present, but no yeast cultured. Yeast does not always survive transit through the intestines rendering it unviable for culturing. Therefore, both microscopic examination and culture are helpful in determining if abnormally high levels of yeast are present.

Microscopic yeast

Microscopic examination has revealed yeast in this stool sample. The microscopic finding of yeast in the stool is helpful in identifying whether the proliferation of fungi, such as *Candida albicans*, is present. Yeast is normally found in very small amounts in a healthy intestinal tract. While small quantities of yeast (reported as none or rare) may be normal, yeast observed in higher amounts (few, moderate to many) is considered abnormal.

An overgrowth of intestinal yeast is prohibited by beneficial flora, intestinal immune defense (secretory IgA), and intestinal pH. Beneficial bacteria, such as *Lactobacillus* colonize in the intestines and create an environment unsuitable for yeast by producing acids, such as lactic acid, which lowers intestinal pH. Also, *Lactobacillus* is capable of releasing antagonistic substances such as hydrogen peroxide, lactocidin, lactobacillin, and acidolin.

Many factors can lead to an overgrowth of yeast including frequent use of antibiotics (leading to insufficient beneficial bacteria), synthetic corticosteroids, oral contraceptives, and diets high in sugar. Although there is a wide range of symptoms which can result from intestinal yeast overgrowth, some of the most common include brain fog, fatigue, recurring vaginal or bladder infections, sensitivity to smells (perfumes, chemicals, environment), mood swings/depression, sugar and carbohydrate cravings, gas/bloating, and constipation or loose stools.

A positive yeast culture (mycology) and sensitivity to prescriptive and natural agents is helpful in determining which anti-fungal agents to use as part of a therapeutic treatment plan for chronic colonic yeast. However, yeast are colonizers and do not appear to be dispersed uniformly throughout the stool. Yeast may therefore be observed microscopically, but not grow out on culture even when collected from the same bowel movement.

Secretory IgA (sIgA)

The concentration of sIgA is abnormally low in this specimen. Immunological activity in the gastrointestinal tract can be assessed using secretory immunoglobulin A (sIgA). Secretory IgA is the predominant antibody, or immune protein the body manufactures and releases in external secretions such as saliva, tears, and milk [1]. It is also transported through the epithelial cells that line the intestines out into the lumen. Secretory IgA represents the first line of defense of the GI mucosa and is central to the normal function of the GI tract as an immune barrier [1]. As the principal immunoglobulin isotype present in mucosal secretions, sIgA plays an important role in controlling intestinal milieu which is constantly presented with potentially harmful antigens such as pathogenic bacteria, parasites, yeast, viruses, abnormal cell antigens,

and allergenic proteins [1]. Secretory IgA antibodies exert their function by binding to antigenic epitopes on the invading microorganism, limiting their mobility and adhesion to the epithelium of the mucus membrane [2]. This prevents the antigens from reaching systemic circulation and allowing them to be excreted directly in the feces.

Mental and physical stress as well as inadequate nutrition have been associated with low fecal sIgA concentrations. This includes dietary restrictions, excessive alcohol intake, body mass loss, negative moods, and anxiety [3]. One study found depressed levels of sIgA in malnourished children, particularly protein malnourishment, that responded well to nutritional rehabilitation with a significant increase in sIgA [4]. This may be because the synthesis and expression of sIgA requires adequate intake of the amino acid L-glutamine [3]. Animal studies have demonstrated that a glutamine-restricted diet can result in a 50% decrease in sIgA levels [5]. An increase of dietary L-glutamine can restore GI immune function by protection of cells that synthesize sIgA [6]. *Saccharomyces boulardii* is a nonpathogenic yeast that has been used for the treatment of acute infectious enteritis and antibiotic-associated diarrhea [7]. Significantly elevated levels of sIgA and subsequent enhanced host immune response have been found following *S. boulardii* administration in mice and rats [8,9].

References:

1. Crago SS, Tomasi TB. Mucosal Antibodies, Food Allergy and Intolerance. Bailliere Tindall/W.B. Saunders 1987;167-89.
2. Roberts JA. Factors predisposing to urinary tract infections in children. *Ped Neph* 1996;10:517-522.
3. Carins J, Booth C. Salivary immunoglobulin-A as a marker of stress during strenuous physical training. *Aviat Space Environ Med* 2002;73(12)1203-7.
4. Teodosio MR, Oliveira ECM. Urinary secretory IgA after nutritional rehabilitation. *Braz J Med Biolog Res* 1999;32-421-426
5. Alverdy J. Effects of glutamine-supplemented diets on immunology of the gut. *J Parent Enteral Nutr* 1990;14(4):1095-1135.
6. Burke DJ, et al. Glutamine-supplemented total parenteral nutrition improves gut function. *Arch Surg* 1989;24:2396-2399.
7. Alverdy JA. The effect of total parenteral nutrition on gut lamina propria cells. *J Parent. Enteral Nutr* 1990;14(suppl).
8. Qamar A, Aboudola S, Warny M, et al. *Saccharomyces boulardii* stimulates intestinal immunoglobulin A immune response to clostridium difficile toxin A in mice. *Infect Immun* 2001;69(4):2762-5.
9. Buts JP, Bernasconi P, Vaerman JP, et al. Stimulation of secretory IgA and secretory component of immunoglobulins in small intestine of rats treated with *Saccharomyces boulardii*. *Dig Dis Sci* 1990;35(2):251-6.