



LAB#: F000000-0000-0
PATIENT: Sample Patient
ID: PATIENT-S-00550
SEX: Male
AGE: 5

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

Comprehensive Stool Analysis

MICROBIOLOGY

Bacteriology Culture

Beneficial flora		Imbalances		Dysbiotic flora	
Bifidobacter	3+	Bacillus spp.	4+	Citrobacter freundii	4+
E. coli spp.	3+	Gamma strep	4+		
Lactobacillus spp	2+	Beta strep, not Group A or	3+		
Enterococcus spp.	0+				

Mycology (Yeast) Culture

Normal flora	Dysbiotic flora
	Saccharomyces cerevisiae 2+

Beneficial flora: In a healthy balanced state of intestinal flora, the beneficial bacteria make up a significant proportion of the total microflora. The beneficial flora have many health-protecting effects in the gut including manufacturing vitamins, fermenting fibers, digesting proteins and the disaccharide lactose, and propagating anti-tumor and anti-inflammatory factors. Acidophilus, Bifidus, and Enterococcus produce lactic acid and short-chain fatty acids. The fermentation of fibers by beneficial bacteria and subsequent production of short chain fatty acids is crucial in lowering colonic pH and preventing the proliferation of microbial pathogens, including bacteria and yeast. Enterococcus has antibacterial activity against methicillin-resistant S. aureus (MRSA) and food-borne pathogens.

Date Collected: **6/29/2007** Comments:
 Date Received: **7/2/2007**
 Date Completed: **7/13/2007**



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CAMPYLOBACTER CULTURE

	Within	Outside	Ref. Range	
Campylobacter jejuni	Neg		Neg	<p>Campylobacter jejuni is a bacteria and a common cause of diarrheal disease, often accompanied by abdominal cramping, fever, and vomiting. Campylobacter infection is often associated with raw or undercooked poultry, unpasteurized milk, or contaminated water.</p>

DIGESTION / ABSORPTION

	Within	Outside	Ref. Range	
Elastase	467		> 200 µg/mL	<p>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. Meat/Vegetable fibers: The presence of meat and/or vegetable fibers in the stool may be due to a number of factors including, improper mastication, excessive protein intake, a reduction of gastric HCL secretion, or insufficient output of pancreatic enzymes. Carbohydrates: The presence of reducing substances in stool specimens can indicate carbohydrate malabsorption.</p>
Fat stain	None		None - Mod	
Muscle fibers	None		None - Rare	
Vegetable fibers	Rare		None - Few	
Carbohydrates	Neg		Neg	

INFLAMMATION

	Within	Outside	Ref. Range	
Lysozyme*		771	<= 600 ng/mL	<p>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. WBCs: Elevated stool levels of white blood cells occur following an infiltration of leukocytes within the intestinal lumen during an inflammatory process. Mucus in the stool may result from prolonged mucosal irritation or in response to parasympathetic excitability such as spastic constipation or mucous colitis.</p>
Lactoferrin	1.4		< 7.3 µg/mL	
WBC	None		None - Rare	
Mucus	Neg		Neg	

IMMUNOLOGY

	Within	Outside	Ref. Range	
slgA*	87		51 - 204mg/dL	<p>slgA: Secretory IgA is secreted by mucosal-associated lymphoid tissue and represents the first line of defense of the GI mucosa and is central to the normal function of the GI as an immune barrier. Elevated levels of slgA have been associated with an upregulated immune response.</p>

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



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SHORT CHAIN FATTY ACIDS

	Within	Outside	Ref. Range	
Acetate	58		36 - 74	%
Propionate	10		9 - 32	%
Butyrate	30		16 - 39	%
Valerate	2		1 - 8	%
Butyrate	2.2		0.8 - 3.8	mg/mL
Total SCFA's	7.6		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. Lactobacillus and Bifidus 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/g are important for assessing overall SCFA production, and are reflective of beneficial flora levels and/or adequate fiber intake.

INTESTINAL HEALTH MARKERS

	Within	Outside	Ref. Range	
RBC	None		None - Rare	
pH		5.2	6 - 7.8	
Occult Blood	Neg		Neg	
Yeast	Rare		None - Rare	

RBC: Red blood cells 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. **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. **pH:** Fecal pH is largely dependent on the fermentation of fiber by the beneficial flora of the gut. **Yeast:** A positive microscopic yeast level indicates the presence of fungi such as Candida albicans in the stool.

MACROSCOPIC APPEARANCE

	Appearance	Expected	
Color	Brown	Brown	
Consistency	Formed/Soft	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.



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YEAST SUSCEPTIBILITIES

Saccharomyces cerevisiae

Prescriptive agents

Sensitive

Intermediate

Resistant

Fluconazole

S

Itraconazole

R

Ketoconazole

S

Nystatin

S

Natural agents

Sensitive

Resistant

Berberine

S

Caprylic Acid

S

Goldenseal

S

Oregano

R

Tanalbit

R

Undecylenic Acid

R

Uva Ursi

S



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BACTERIAL SUSCEPTIBILITIES

Citrobacter freundii

Prescriptive agents

Amoxicillin

Ampicillin

Augmentin

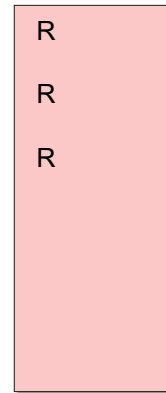
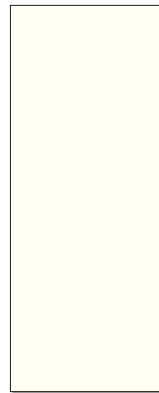
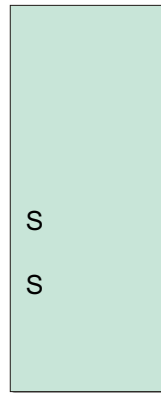
Ciprofloxacin

Trimeth-sulfa

Sensitive

Intermediate

Resistant



Natural agents

Berberine

Black Walnut

Caprylic Acid

Cats Claw

Citrus Seed Extract

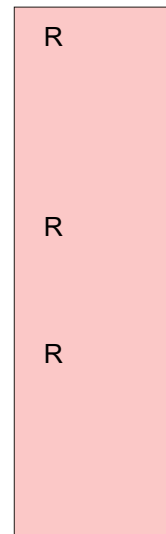
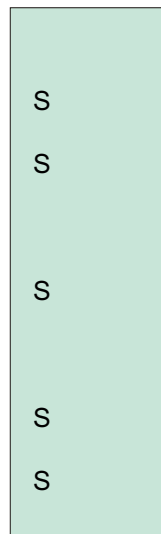
Goldenseal

Oregano

Uva Ursi

Sensitive

Resistant



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 beneficial bacteria are low in this specimen. Beneficial flora include Lactobacillus, Bifidus, Enterococcus sp., and beneficial E. 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 the disaccharide lactose, manufacture of vitamins and essential fatty acids, increasing the number of immune system cells, breaking down bacterial toxins and converting flavinoids into anti-tumor and anti-inflammatory factors [1]. Lactobacillus, Bifidus, and Enterococcus sp. secrete lactic acid as well as other acids including acetate, propionate, butyrate, and valerate. This 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. Lactobacillus acidophilus also secretes the antifungal and antimicrobial agents lactocidin, lactobacillin, acidolin, and hydrogen peroxide [2]. The beneficial flora of the GI have thus been found useful in the inhibition of microbial pathogens [3], prevention and treatment of antibiotic associated diarrhea [4], prevention of traveler's diarrhea [5], enhancement of immune function [6], and inhibition of the proliferation of Candida albicans [7,8].

Enterococcus sp. Are prominent non-anaerobic beneficial bacteria in the gastrointestinal tract. They are fermentative yet not gas producing bacteria that can survive in relatively harsh environments. Most importantly, Enterococcus sp. Provide antimicrobial activity against methicillin-resistant Staphylococcus aureus (MRSA), and impede the growth of food-borne pathogens. S. aureus strains, which are resistant to multiple antibiotics, have dramatically increased hospital associated infections. There is concern that the pharmaceutical industry cannot keep up the MRSA strains, therefore maintenance of healthy levels of Enterococcus sp. is important for antimicrobial activity against MRSA.

In a healthy balanced state of intestinal flora, the beneficial flora make up a significant proportion of the total non-anaerobic 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 [1]. Treatment may include the use of probiotic supplements containing various strains of Lactobacillus, Bifidobacter, and Enterococcus and/or consumption of cultured or fermented foods including yogurt, kefir, miso, tofu, tempen and tamari sauce. Polyphenols in green and ginseng tea have been found to increase the numbers of beneficial bacteria [9]. If dysbiosis is present, treatment may also include the removal of pathogenic bacteria, yeast, or parasites.

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Imbalanced flora

Imbalanced flora are those bacteria that are not pathogenic but are rather commensal. They reside in the host organism (GI tract) and neither injure nor benefit the host [1]. 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 (6.0 - 7.2). It is also not uncommon to find Haemolytic or NLF 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 dysbiosis category.

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 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 [1].

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 [2]. 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 [1]. 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, tofu, tamari sauce) containing strains of *Lactobacillus* and *Bifidus* can help restore healthy flora levels [1]. Polyphenols in green and ginseng tea have been found to increase the numbers of beneficial bacteria [3]. Hypochlorhydria may also predispose an individual to bacterial overgrowth, particularly in the small intestine [4]. 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 [5]. A comprehensive program may be helpful in individuals in whom a dysbiotic condition has caused extensive GI damage.

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Dysbiotic Yeast

Yeast was cultured from this stool specimen and the amount is considered to be dysbiotic. A positive yeast culture and sensitivity to prescriptive and natural agents is helpful in determining which anti-fungal agents to use as part of a therapeutic plan for chronic yeast syndrome. 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 significant amount of yeast cultured. 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 rare or few) may be normal, yeast observed in higher amounts (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 yeast syndrome. However, culturing of yeast is not always possible, due to the fact that yeast does not always survive transit through the intestines. Additionally, yeast colonizes in groups and is not dispersed uniformly throughout the stool. Yeast may therefore appear (microscopically) to be inconsistently concentrated in some stool specimens, even when collected from the same bowel movement.

Lysozyme

The level of lysozyme, a biomarker of inflammation, is elevated in this specimen. Lysozyme is an enzyme that catalyzes the hydrolysis of specific glycosidic bonds in mucopolysaccharides that constitute the cell wall of gram-positive bacteria. Lysozyme is an antibacterial defense present in the G.I. tract and is secreted by granulocytes, macrophages, Paneth cells, and Brunner's Glands as well as normal colonic crypt cells [1]. The main source for fecal lysozyme is the intestinal granulocytes.

Moderate elevations in fecal lysozyme are commonly associated with significant overgrowth of enteropathogens such as yeasts or dysbiotic bacteria. Markedly elevated levels of fecal lysozyme have been identified in colonic inflammatory bowel disease (IBD), such as Crohn's disease and ulcerative colitis as well as other non-IBD G.I. diseases with diarrhea, compared to healthy controls [2,3]. In Crohn's disease, excess lysozyme may be a result of active secretions of macrophages in the lamina propria, and monocytic cells in the granulomas (sites of G.I. inflammation) [4]. In ulcerative colitis, it has been postulated that elevations in fecal lysozyme may be secondary to intestinal loss of granulocytes

and their secretory granules [5]. Additionally, Paneth cell metaplasia, a phenomenon that occurs with various inflammatory conditions of the large intestine, may be a minor contributor to fecal lysozyme elevations [5]. Paneth cells are part of the intestinal epithelial lining found in the deepest part of intestinal crypts which are the crypts of Lieberkühn. Paneth cells contain lysozyme in their secretory granules, and combined with their phagocytic capability, help to regulate intestinal microbial flora [5].

Lysozyme is helpful in the determination of colonic inflammatory activity rather than small bowel disease [2]. Slightly elevated levels of lysozyme may be treated with anti-inflammatory agents or by removing the antagonist, such as enteroinvasive microorganisms or allergens. Moderate to high levels of lysozyme (>2,000) may indicate an active inflammatory bowel condition which often requires further testing such as colonoscopy. To rule out IBD, check fecal lactoferrin levels (elevated with IBD).

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2. Van der Sluys Veer A, Brouwer J, Biemond I, et al. Fecal lysozyme in assessment of disease activity in inflammatory bowel disease. *Dig Dis & Sci.* 1998;43(3):590-5.
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4. Geboes K, Van den Oord JJ, Rutgeerts P, et al. Immunohistochemical identification of lysozyme in pseudopyloric gland metaplasia in Crohn's disease. *Hepatology* 1986;90:1121-8.
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pH low

The pH of this stool sample (<6.0) is too acidic. Ideally, the pH of the stool is slightly acidic. This represents colonic pH, which is largely reflective of bacterial fermentation and putrefaction of intestinal contents. Healthy microflora such as *Lactobacillus* and *Bifidus* generate large amounts of short chain fatty acids (acetic, propionic, butyric, and valeric), which lower colonic pH. Short chain fatty acids are byproducts of the bacterial fermentation process of dietary fiber by beneficial flora in the gut. An acidic pH, below 6.0, is usually reflective of a rapid transit time, e.g. diarrhea or loose stools. Further investigation as to the cause of diarrhea such as food allergy intolerance, viral, bacterial, parasitic infection, irritable bowel syndrome may be warranted. Additionally, research has indicated that an acidic pH (< 6.0) is common in individuals with lactose malabsorption [1]. Unabsorbed lactose in the gut can be hydrolysed by colonic bacteria forming volatile fatty acids which cause the stool to become acidic, often times accompanied by a sweet, sickly stool odor [1]. Hydrolysis of unabsorbed lactose and fermentation by colonic bacteria can also produce hydrogen (and carbon dioxide) which is then absorbed and excreted in the breath. This is the basis for the test for lactose malabsorption (lactose intolerance breath test).

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Citrobacter species

Citrobacter freundii, a gram-negative bacterium and member of the Enterobacteriaceae family, is considered to be an enteropathogen. Other *Citrobacter* species are less commonly identified as pathogens. *Citrobacter* is common in the environment, including food and animal feces. Incidence, per laboratory observation, seems to be higher in Spring and Summer.

C. freundii and, less commonly, *C. koseri* can cause diarrheal disease. *Citrobacter koseri* has also been isolated in rare cases of neonatal meningitis. *Citrobacter* is often asymptomatic, but has been known to cause sepsis and infection in a number of tissues. Symptoms due to *C. freundii* seem to be a result of the elaboration of an *E. coli*-like heat-stable enterotoxin and hydrogen sulfide. *Citrobacter freundii* 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.

C. freundii thrives on FOS, a common ingredient in probiotic formulas. Treatment herbs include garlic and aloe vera. Specific natural or pharmaceutical treatments should be based on susceptibility testing.

Derlet RW, Carlson JR. An analysis of human pathogens found in horse/mule manure along the John Muir Trail in Kings Canyon and Sequoia and Yosemite National Parks. *Wilderness and Environmental Medicine*, 13(2):13-118, 2004.

Morris JG Jr, Lin FY, Morrison CB, Gross RJ, Khabbaz R, Maher KO, Rowe B, Israel E, Libonati JP. Molecular epidemiology of neonatal meningitis due to *Citrobacter diversus*: a study of isolates from hospitals in Maryland. *J Infect Dis*, 154(3):409-14, 1986.

Murray PR, Baron EJ, Pfaller MA, Tenoer FC, Tenover RH. *Manual of Clinical Microbiology*, 6th edition. Washington, DC: ASM Press; 1995.