Dr Henry Butt MSc PhD



Specialist Microbiological Laboratory

ABN: 87 682 058 987

5 Little Hyde Street Yarraville, Victoria, 3013 Ph: +61 3 9687 3355 Fax: +61 3 9687 3377 admin@bioscreenmedical.com

Report of Faecal Microbiology

Patient Name: Janine WADE

Address: 46 Sunshine St

Manly Vale NSW 2093

Date of Birth: 14/01/1982

Name of Requesting Practitioner: Robyn COSFORD

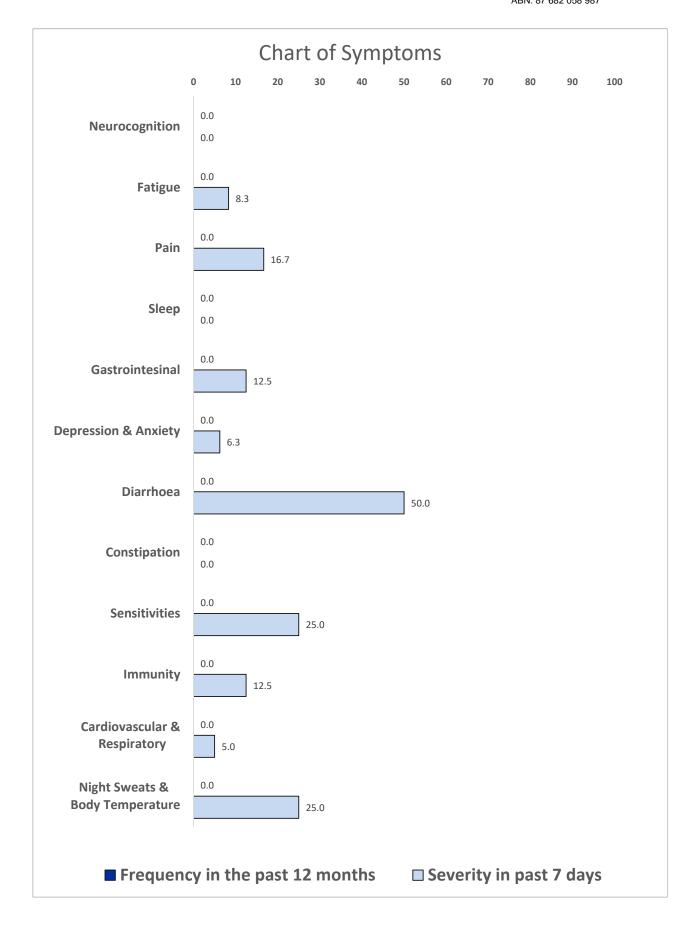
Laboratory Number:158756Date of Sample Collection:7/05/2024Date of Sample Processing09/05/2024Date of Report Issued:16/05/2024

Dear Dr Robyn COSFORD,

Thank you for referring your patient to Bioscreen.

The following is a summary of our faecal microbiota testing from your patient's sample.







Detailed Report, Faecal Microbiology

Bacterial Count (Total)
Facultative Anaerobe
[Aerobe] Counts
Anaerobe Count

Count cfu/g	Counts Reference Range cfu/g	Comment	Distribution % Total Count	Distribution Reference Range
3.6 x 10 ⁹	1.0 x 10 ⁹ - 1.0 x 10 ¹²	Within Ref Range		
2.1 x 10 ⁷	1.0 x 10 ⁷ - 1.0 x 10 ⁸	Within Ref Range		
3.6 x 10 ⁹	1.0 x 10 ⁸ - 1.0 x 10 ¹²	Within Ref Range		

Aerobe: Anaerobe Ratio:

5.9 (Reference Range is 0.5 - 4.0)

Facultative Anaerobe [Aerobe] Counts

Aerobe Count (total) Escherichia coli coliform (Total)

Escherichia coli

Non-E.coli coliforms

Klebsiella pneumoniae

Enterococcus (Total)

Enterococcus faecium

Streptococcus (Total)

Streptococcus parasanguinis

Staphylococcus (Total)

Staphylococcus epidermidis

Staphylococcus hominis

Count cfu/g	Counts Reference Range cfu/g	Comment	Distribution % Total Count	Distribution Reference Range
2.1 x 10 ⁷	1.0 x 10 ⁷ - 1.0 x 10 ⁸	Within Ref Range		
2.1 x 10 ⁴	7.0 x 10 ⁶ - 9.0 x 10 ⁷	Low		
2.1 x 10 ⁴			0.1%	70-90%
2.1 x 10 ⁵	<5.0 x 10 ⁵	Within Ref Range		
2.1 x 10 ⁵			1%	<5%
1.6 x 10 ⁷	<5.0 x 10 ⁵	High	74%	<5%
1.6 x 10 ⁷			74%	
5.2 x 10 ⁶	<3.0 x 10 ⁵	High	24.7%	<5%
5.2 x 10 ⁶			24.7%	
5.2 x 10 ⁴	<2.0 x 10 ⁵	Within Ref Range	0.2%	<5%
4.2 x 10 ⁴	<2.0 x 10 ⁵		0.2%	
1.0 x 10 ⁴	<2.0 x 10 ⁵		< 0.01%	

Faecal Fungi (Total)

Rhodotorula mucilaginosa

Count cfu/g	Counts Reference Range cfu/g	Comment	Distribution % Total Count	Distribution Reference Range
1.0 x 10 ²	<1.0 x 10 ⁴	Within Ref Range		
1.0 x 10 ²				



Specialist Microbiological Laboratory

ABN: 87 682 058 987

Anaerobe Counts

Anaerobe Count (total) Bacteroides and related genera (Total)

Bacteroides xylanisolvens

Bacteroides salyersiae

Alistipes finegoldii

Eubacterium and related genera (Total)

Collinsella aerofaciens

Eggerthella lenta

Lactobacillus and related genera (Total)

Ligilactobacillus ruminis

Lactobacillus acidophilus

Bifidobacterium and related genera (Total)

Bifidobacterium adolescentis

Count cfu/g	Counts Reference Range cfu/g	Comment	Distribution % Total Count	Distribution Reference Range
3.6 x 10 ⁹	1.0 x 10 ⁸ - 1.0 x 10 ¹²	Within Ref Range		
<9.0 x 10 ⁷	5.0 x 10 ⁸ - 9.5 x 10 ¹¹	Low	1.2%	85-95%
1.0 x 10 ⁷			0.3%	
2.1 x 10 ⁷			0.6%	
1.0 x 10 ⁷			0.3%	
1.4 x 10 ⁹	1.0 x 10 ⁸ - 1.0 x 10 ⁹	High	37.6%	<15%
1.2 x 10 ⁹			34.7%	
1.0 x 10 ⁸			2.9%	
2.2 x 10 ⁷	5.0 x 10 ⁵ - 1.0 x 10 ⁷	High	0.6%	0.5-2%
2.1 x 10 ⁷			0.6%	
1.0 x 10 ⁶			< 0.01%	
2.2 x 10 ⁹	5.0 x 10 ⁵ - 5.0 x 10 ⁸	High	60.7%	5-11%
2.2 x 10 ⁹			60.7%	



Summary Report, Faecal Microbiology

Total Aerobe Count: 2.1 x 10⁷ cfu/g (colony forming units/g)

Total Anaerobe Count: 3.6 x 10° cfu/g

Aerobe: Anaerobe Ratio is: 5.9. The Reference Range is 0.5 to 4.

Comment: This ratio is above the reference range.

Aerobe: Anaerobe Ratio - High

 A high aerobe:anaerobe ratio is usually due to a low count of anaerobes or the absence some genera of anaerobes. Growth of anaerobes is promoted by a diet that includes plentiful meat and /or bone broth, fish and soybean protein.

Faecal Aerobes

Enterococcus sp.: Overgrowth Streptococcus sp.: Overgrowth

E.Coli: Undergrowth

Comments

E. coli

- The reason for the low E.coli percentage distribution/ total count in the sample is unclear. However, recent exposure to antipyretics and/or analgesia (eg. paracetamol) may cause a marked change in the faecal ecology resulting in a significant alteration of the E.coli viable count (Bioscreen data, 2001). Recent supplementation with fructo-oligosaccharide (FOS) may also have suppressed growth of this organism.
- Undetectable levels of E.coli. Oral application of E.coli probiotics may be beneficial. Changing and normalizing the colonic aerobic microbial flora with the E. coli probiotic has shown to be safe and beneficial in patients with Ulcerative Colitis^{1,2} and Crohn's Disease³. The probiotic, once ingested and if adhere to the mucosal wall, will colonize the colon within a few days, and will remain colonized after oral administration ceased. Oral application of the E.coli probiotic has shown to stimulate and enhance immune responses and induces nonspecific natural immunity^{4,5}.
- E.coli is an important intestinal micro-organism responsible for the synthesis of essential amino acids (eg. trytophan, phenylalanine, tyrosine)^{6,7,8} vitamins (folic acid, vit K2)^{9,10}, and coenzymes (CoQ10)¹¹ important for cellular metabolism and reproduction. Determination into the levels of these essential amino acids in patients with persistent and chronic low levels of E.coli may be beneficial. Acute depletion of tyrosine and phenylalanine has shown to have selective effect on decision-making in depressive patients¹². Tyrosine depletion has also shown to have recognition and working memory impairment¹³.
- Consider supplementing oral sugars (eg galactose, fucose) to increase the densities of current intestinal coliforms (eg E.coli)^{14,15} as opposed to adding a different strain with probiotics. Health professionals can contact Bioscreen for further information.
- Consider checking the folate, vitamin K2, CoQ10 levels and supplement if indicated.
- Consider checking the levels of the following essential amino acids: tryptophan, tyrosine, phenylalanine, and supplement if indicated.

Streptococcus/Enterococcus

 Both Enterococcus and Streptococcus spp. are Gram positive, facultative anaerobic organisms and are classified as homofermenative, producing only lactic acid from glucose catabolism and generally regarded as potent D- and L-lactic acid producers (Bioscreen data).



- Increased distribution of lactic acid bacteria (Streptococcus, Enterococcus sp.) may lower the colonic pH¹⁶ and has been reported to :(1) modify faecal microbial metabolism particularly the Bacteroides and Bifidobacterium spp, resulting in a decreased production of volatile fatty acids¹⁷, and (2) alter intestinal epithelial barrier function increasing passive intestinal permeability to small and large molecules. However, this consideration requires further study.
- High colonization of faecal lactic acid bacteria (Streptococcus, Enterococcus sp.) significantly and
 positively correlate with cognitive dysfunctions (nervousness, memory loss, forgetfulness,
 confusion, mind going blank)^{18,19,20,21}, and sleep patterns (Bioscreen data).
- Increased proportion of lactic acid may result in a change in the distribution of the anaerobic microbial flora. This change of the fecal flora may affect the production of primary bile acids and influencing the bile acid composition in both the bile and the intestine²². The possibility of fat malabsorption may occur. However, this consideration requires further study.
- If indicated, ampicillin/amoxycillin may assist in the suppression of the faecal Enterococcus and Streptococcus spp.. If the patient has not recently been exposed to ampicillin/amoxycillin, the organism Enterococccus should be susceptible to the antibiotic. Bacitracin may be a suitable alternative against both organisms if patient is reported to have adverse reactions to the penicillins. Bacitracin is a non-absorbable antibiotic; however, potential nephrotoxicity and allergic reactions may occur²³.

Faecal Anaerobes

Eubacterium sp.: Overgrowth Bifidobacterium sp.: Overgrowth Lactobacillus sp.: Overgrowth Bacteroides sp.: Undergrowth

Comments

Bacteroides sp.

Undergrowth of Bacteroides spp.

- Low and limited distribution of Bacteroides spp may affect the production of volatile fatty acids. Volatile fatty acids are important for the growth of colonocytes.
- Consider the supplement of menaquinones (vitamin K2), and protoporphyrin IX (hemin) to improve the growth of anaerobes. High concentration of vitamin K2 (up to 300µg/kg) is found in natural yogurt, cheese, and butter²⁴.
- Consider the supplement of biotin, bicarbonate, sodium and/or potassium to assist the production of volatile fatty acids by anaerobes.
- A general diet consisting of meat/ bone broth, fish protein or soybean protein may assist the growth of these organisms.
- The cellulolytic Bacteroides (B.vagatus, B.ovatus, Parabacteroides distasonis, B.fragilis, B. thetaiotamicron, B. uniformis, B. intestinalis, B.cellulosilyticus) are generally regarded as the most prevalent anaerobic micro-organisms in human intestinal microbial flora responsible for the degradation of the insoluble components of dietary fibers.
- This change of the fecal flora may affect the production of primary bile acids and influencing the bile acid composition in both the bile and the intestine²².

Bifidobacterium/Lactobacillus sp.

- Members of the genera Lactobacillus and Bifidobacterium are Gram positive bacilli and lactic acid producing bacteria. A few members of both genera can grow in a microaerophilic environment; but most are obligate anaerobes.
- High levels of Bifidobacterium spp. in the anaerobic microbial flora. Increased level of Bifidobacterium may stimulate amine production²⁵. Similarly, increased levels of this organism may also lower the colonic pH¹⁶, modifying faecal microbial metabolism particularly the Bacteroides spp, resulting in a decreased production of volatile fatty acids¹⁷, and altering intestinal epithelial barrier function increasing passive intestinal permeability to small and large molecules.



- High levels of Lactobacillus spp. in the anaerobic microbial flora.
 Metabolic acidosis and neurological dysfunction (depressed conscious state, confusion, aggressive behaviour, slurred speech and ataxia) have been reported in patients with increased level of lactobacilli in the anaerobic faecal flora²⁶.
- Cease all oral supplementation of lactic acid probiotics if indicated.

Eubacterium sp.

- Eubacterium sp is generally regarded as one of the most frequently recovered organisms in the gastrointestinal tract, second only to Bacteroides spp.
- The increased distribution of this organism in the gastrointestinal tract is unclear, however, the cell wall of the organism has shown to be proinflammatory and arthritogenic²⁷.

Faecal Fungi

Faecal fungi were recovered and the % abundance was within the normal range.

If you require further assistance please contact Bioscreen and arrange a consultation.

Report authorised 20th May, 2024 by Dr Henry Butt.

Bioscreen Pty Ltd.

References

- 1. Rembacken BJ, Snelling AM, Hawkey PM, Chalmers DM, Axon AT. Non-pathogenic Escherichia coli versus mesalazine for the treatment of ulcerative colitis: a randomised trial. Lancet. 1999 Aug 21;354(9179):635-9.
- Kruis W, Schütz E, Fric P, Fixa B, Judmaier G, Stolte M. Double-blind comparison of an oral Escherichia coli preparation and mesalazine in maintaining remission of ulcerative colitis. Aliment Pharmacol Ther. 1997 Oct;11(5):853-8.
- 3. Malchow HA. Crohn's disease and Escherichia coli. A new approach in therapy to maintain remission of colonic Crohn's disease? J Clin Gastroenterol. 1997 Dec;25(4):653-8.
- 4. Cukrowska B, Lodlnová-Zádnlková R, Enders C, Sonnenborn U, Schulze J, Tlaskalová-Hogenová H. Specific proliferative and antibody responses of premature infants to intestinal colonization with nonpathogenic probiotic E. coli strain Nissle 1917. Scand J Immunol. 2002 Feb;55(2):204-9.
- 5. Hockertz S. Immunomodulating effect of killed, apathogenic Escherichia coli, strain Nissle 1917, on the macrophage system. Arzneimittelforschung. 1991 Oct;41(10):1108-12.
- 6. Dosselaere F, Vanderleyden J. A metabolic node in action: chorismate-utilizing enzymes in microorganisms. Crit Rev Microbiol 2001;27.
- 7. Gerigk M, Bujnicki R, Ganpo-Nkwenkwa E, Bongaerts J, Sprenger G, Takors R. Process control for enhanced L-phenylalanine production using different recombinant Escherichia coli strains. Biotechnol Bioeng 2002;80.
- 8. Polen T, Kramer M, Bongaerts J, Wubbolts M, Wendisch VF. The global gene expression response of Escherichia coli to L-phenylalanine. J Biotechnol 2005;115.
- Roux B, Walsh CT. p-Aminobenzoate synthesis in Escherichia coli: mutational analysis of three conserved amino acid residues of the amidotransferase PabA. Biochemistry 1993;32:3763-68.



- Burg AW, Brown GM. The biosynthesis of folic acid. 8.
 Purification and properties of the enzyme that catalyzes the production of formate from carbon atom 8 of guanosine triphosphate. J Biol Chem 1968;243.
- 11. Nichols BP, Green JM. Cloning and sequencing of Escherichia coli ubiC and purification of chorismate lyase. J Bacteriol. 1992;174:5309-16.
- 12. Roiser JP, McLean A, Ogilvie AD, Blackwell AD, Bamber DJ, Goodyer I et al. The subjective and cognitive effects of acute phenylalanine and tyrosine depletion in patients recovered from depression. Neuropsychopharmacology 2005;30:775-85.
- 13. Harmer CJ, McTavish SF, Clark L, Goodwin GM, Cowen PJ. Tyrosine depletion attenuates dopamine function in healthy volunteers. Psychopharmacology (Berl) 2001;154:105-11.
- Oli MW, Petschow BW, Buddington RK. Evaluation of fructooligosaccharide supplementation of oral electrolyte solutions for treatment of diarrhea: recovery of the intestinal bacteria. Digestive Diseases & Sciences. 1998;43(1):138-47.
- 15. Allen A, Cunliffe WJ, Pearson JP, et.al. Studies on gastrointestinal mucous. Scand J Gastroenterol 1984; s93:101-13.
- 16. van der Wiel-Korstanje JA, Winkler KC. The faecal flora in ulcerative colitis. J-Med-Microbiol. 1975;8:491-501.
- 17. Edwards CA.Duerden Bl.Read NW. The effects of pH on colonic bacteria grown in continuous culture. Journal of Medical Microbiology.19(2):169-80, 1985.
- 18. Caldarini MI, Pons S, D'Agostino D et al. Abnormal fecal flora in a patient with short bowel syndrome. An in vitro study on effect of pH on D-lactic acid production. Dig Dis Sci. 1996;41:1649-1652
- 19. Hove H, Mortensen PB. Colonic lactate metabolism and D-lactic acidosis. Dig Dis Sci 1995:40.
- 20. Shah M, Beuerlein M, Danayan K. An approach to the patient with a life-threatening acid-base disturbance: the acidemias. . University of Toronto Medical Journal 2001;78:122-28.
- 21. Uribarri J, Oh MS, Carroll HJ. D-lactic acidosis. A review of clinical presentation, biochemical features, and pathophysiologic mechanisms. Medicine (Baltimore) 30 1998;77:73-82.
- 22. Salvioli G, Salati R, Bondi M, et al. Bile acid transformation by the intestinal flora and cholesterol saturation in bile. Effects of Streptococcus faecium administration. Digestion. 1982;23:80-88.
- 23. Jacob SE, James WD. From Road Rash to Top Allergen in a Flash: Bacitracin. American Society for Dermatologic Surgery 2004;30:521-24.
- 24. Hirauchi K, Sakano T, Notsumoto S, Nagaoka T, Morimoto A. Measurement of K vitamins in food by high-performance liquid chromatography with fluorometric detection. Vitamins 1989;63:147-51.
- 25. E.A.Smith and G.T.Macfarlane. Studies on Amine Production in the Human Colon: Enumeration of Amine forming Bacteria and Physiological Effects of Carbohydrate and pH . Anaerobe 1996;2:285-97.
- 26. Haan E.Brown G.Bankier A.Mitchell D.Hunt S.Blakey J.Barnes G. Severe illness caused by the products of bacterial metabolism in a child with a short gut. European Journal of Pediatrics. 144(1):63-5, 1985.
- 27. Zhang X, Rimpiläinen M, Simelyte E, Toivanen P. Characterisation of Eubacterium cell wall: peptidoglycan structure determines arthritogenicity. Ann Rheum Dis. 2001 Mar;60(3):269-74.