Microbial Overgrowth Syndromes in 2026

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Disclosures

- Consultant for Ardelyx. Equity in Salvo Health and Cylinder Health. Equity in Gemelli Biotech. Cedars-Sinai has a licensing agreement with Gemelli Biotech and Hobbs medical.

Opportunistic Microbial Overgrowth Syndromes

Organism	Present in Patients without Disease	Higher density increases disease likelihood	Disease Outcome
Helicobacter pylori	Yes	Yes	Peptic Ulcer
Clostridioides difficile	Yes	Yes	Pseudomembranous colitis
Candida albicans	Yes	Yes	Thrush/esophageal candidiasis
E. coli/Klebsiella	Yes	Yes	IBS/Other
Methanobrevibacter smithii	Yes	Yes	Constipation
H_2S producer	Yes	Yes	Diarrhea/GERD
Rothia mucilaginosa	Yes	Yes	PCOS

Causes of SIBO

Table 4. Conditions associated with small intestinal bacterial overgrowth

Category	Specific condition
Mechanical causes	Small bowel tumor Volvulus Intussusception Postsurgical causes
Systemic disease	Diabetes Scleroderma Amyloidosis
Motility	IBS Pseudo-obstruction Visceral myopathies Mitochondrial diseases
Medications	Opiates Potent antisecretory agents
Malabsorptive conditions	Pancreatic insufficiency Cirrhosis (altered bile acid composition) Other malabsorptive conditions
Immune-related	Human immunodeficiency virus Combined variable immunodeficiency IgA deficiency
Other	Aging (the elderly) Small bowel diverticulosis

IBS, irritable bowel syndrome; IgA, immunoglobulin A.

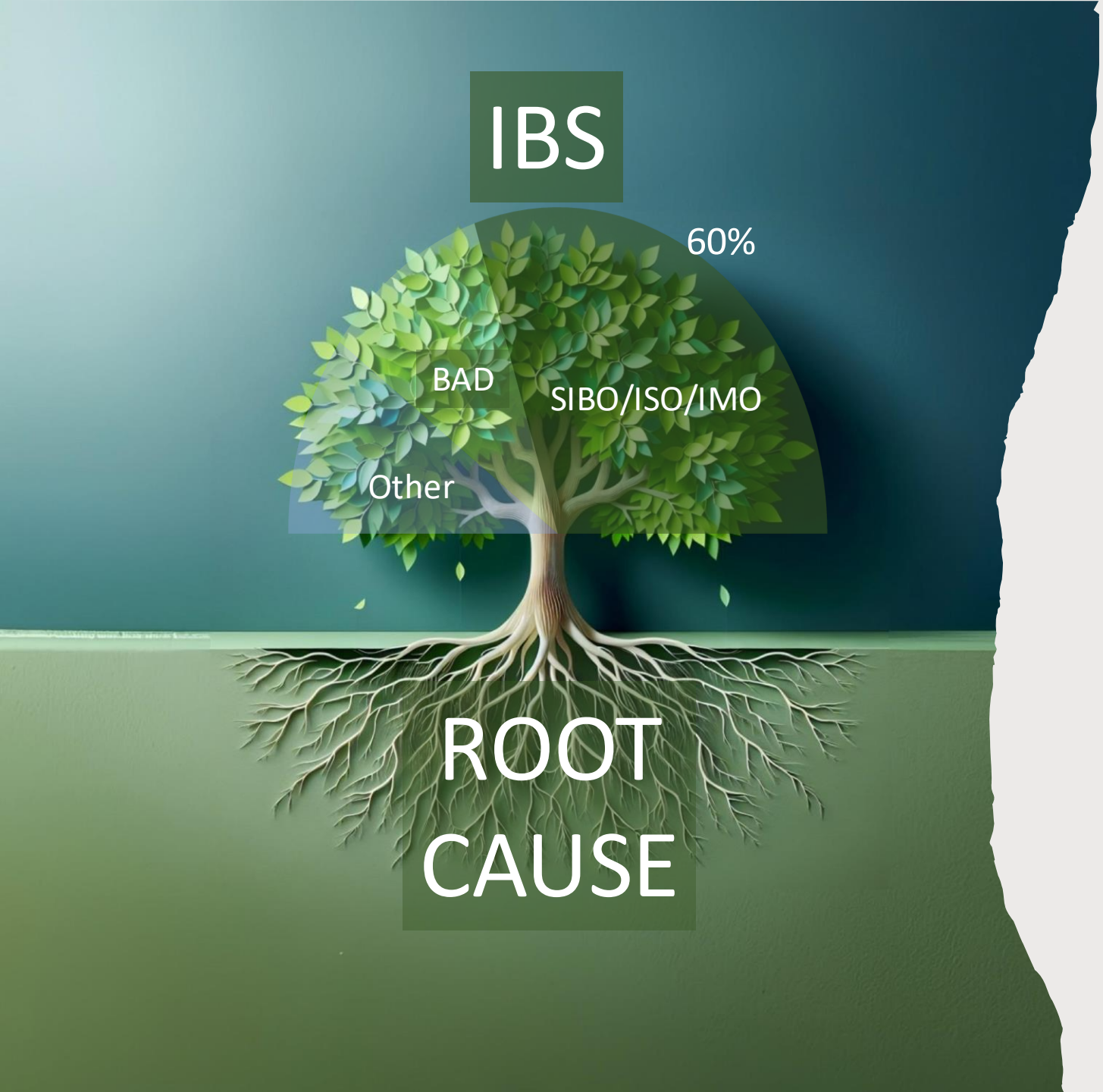
OFFICIAL JOURNAL OF THE AMERICAN COLLEGE OF GASTROENTEROLOGY | ACG

Pimentel, et al. Am J Gastroenterol 2020

Myths in SIBO- Not your Grandparents SIBO

- Old school – malabsorption, B12 deficiency
- Methanogens are only in the colon
- SIBO is the movement of colon bacteria into the small intestine
- The lactulose breath test measures intestinal transit
- Focused on iatrogenic SIBO (blind loops, Bilroth II, gastrectomy, adhesions…)

Bushyhead and Quigley. Gastroenterol 2022



Roots of IBS

- Bile acid diarrhea – but what is the root cause of this?
- SIBO/ISO/IMO – Root cause identified

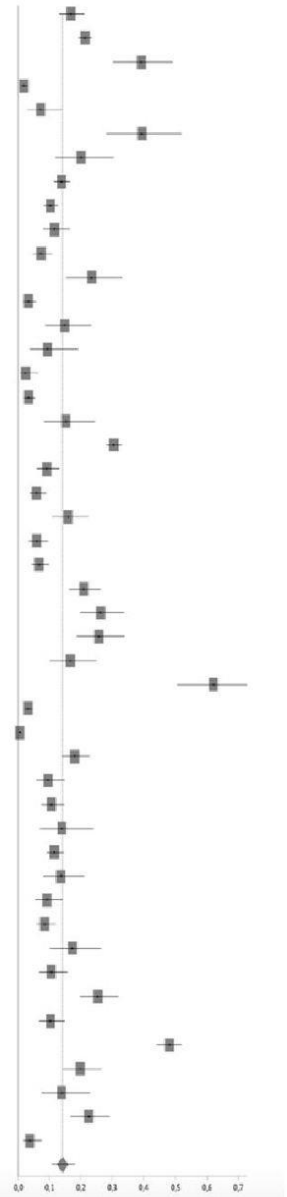
Acute Gastroenteritis Causes IBS

49 Outbreaks Studied

Pooled prevalence = 14.5% (CI=11.2-18.1)

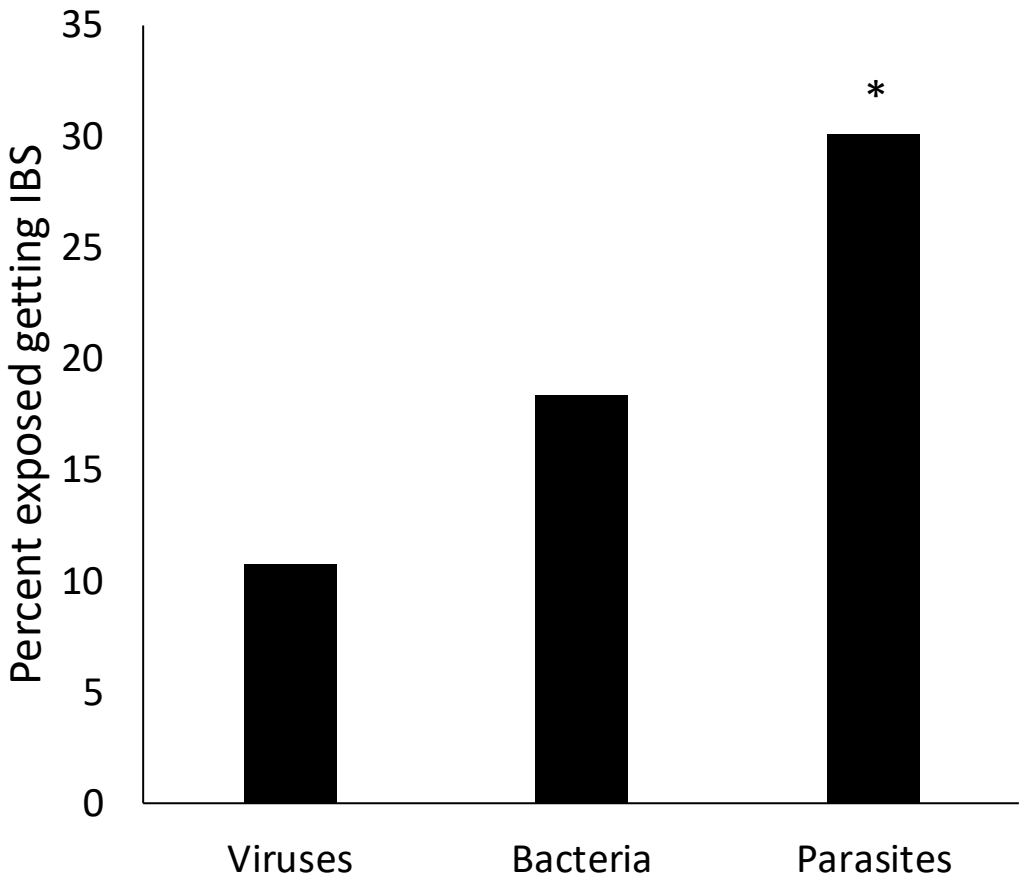
That's 1 in every 6.9 infections!

Study		Event	Total	Event Rate (95% CI)
Andresen	2015	59	351	0,17 (0,13, 0,21)
Berumen	2020	301	1418	0,21 (0,19, 0,23)
Blackett	2022a	44	112	0,39 (0,30, 0,49)
Blackett	2022b	13	749	0,02 (9,3E-3, 0,03)
Borgaonkar	2006	7	99	0,07 (0,03, 0,14)
Cremon	2014	28	71	0,39 (0,28, 0,52)
Dormond	2016	16	80	0,20 (0,12, 0,30)
Dunlop	2003	103	747	0,20 (0,12, 0,30)
Espana Cueto	2023	68	669	0,10 (0,08, 0,13)
Farsi	2021	27	233	0,12 (0,08, 0,16)
Ghoshal	2021	20	280	0,07 (0,04, 0,11)
Gwee	1999	22	94	0,23 (0,15, 0,33)
Golla	2022	10	320	0,03 (0,02, 0,06)
JI	2005	15	101	0,15 (0,09, 0,23)
Koh	2012	6	65	0,09 (0,03, 0,19)
Kowalcyk	2014	3	134	0,02 (4,6E-3, 0,06)
Marasco	2022	14	435	0,03 (0,02, 0,05)
Marshall	2007	13	86	0,15 (0,08, 0,24)
Marshall	2006	417	1368	0,30 (0,28, 0,33)
Mearin	2005	24	266	0,09 (0,06, 0,13)
Nair	2014	20	348	0,06 (0,04, 0,09)
Nakhli	2021	26	164	0,16 (0,11, 0,22)
Nazarewska	2022	15	257	0,06 (0,03, 0,09)
Neal	1997	23	347	0,07 (0,04, 0,10)
Nielsen	2014	56	268	0,21 (0,16, 0,26)
Noviello	2021	43	164	0,26 (0,20, 0,34)
Parida	2019	35	136	0,26 (0,19, 0,34)
Parry	2003	18	108	0,17 (0,10, 0,25)
Peters	2021	49	79	0,62 (0,50, 0,73)
Pitzurra	2011	26	852	0,03 (0,02, 0,04)
Porter	2012	7	1718	4,1E-3 (1,6E-3, 8,4E-3)
Rahman	2018	57	318	0,18 (0,14, 0,23)
Sharifi	2022	17	179	0,09 (0,06, 0,15)
Siyal	2023	32	303	0,11 (0,07, 0,15)
Soyturk	2007	10	72	0,14 (0,07, 0,24)
Spence	2007	63	547	0,12 (0,09, 0,14)
Stermer	2006	16	118	0,14 (0,08, 0,21)
Thornley	2001	17	188	0,09 (0,06, 0,15)
Törnblom	2007	28	333	0,08 (0,06, 0,12)
Trivedi	2011	16	93	0,17 (0,10, 0,26)
Velez	2022	21	200	0,11 (0,07, 0,16)
Wadhwa	2016	52	205	0,25 (0,20, 0,32)
Wang	2004	24	235	0,10 (0,07, 0,15)
Wensaas	2016	339	707	0,48 (0,44, 0,52)
Wouters	2015	34	172	0,20 (0,14, 0,27)
Youn	2016	12	87	0,14 (0,07, 0,23)
Zanini	2012	40	178	0,22 (0,17, 0,29)
Zhang	2023	7	190	0,04 (0,01, 0,07)
Total (95% CI)		2313	16244	0,14 (0,11, 0,19)

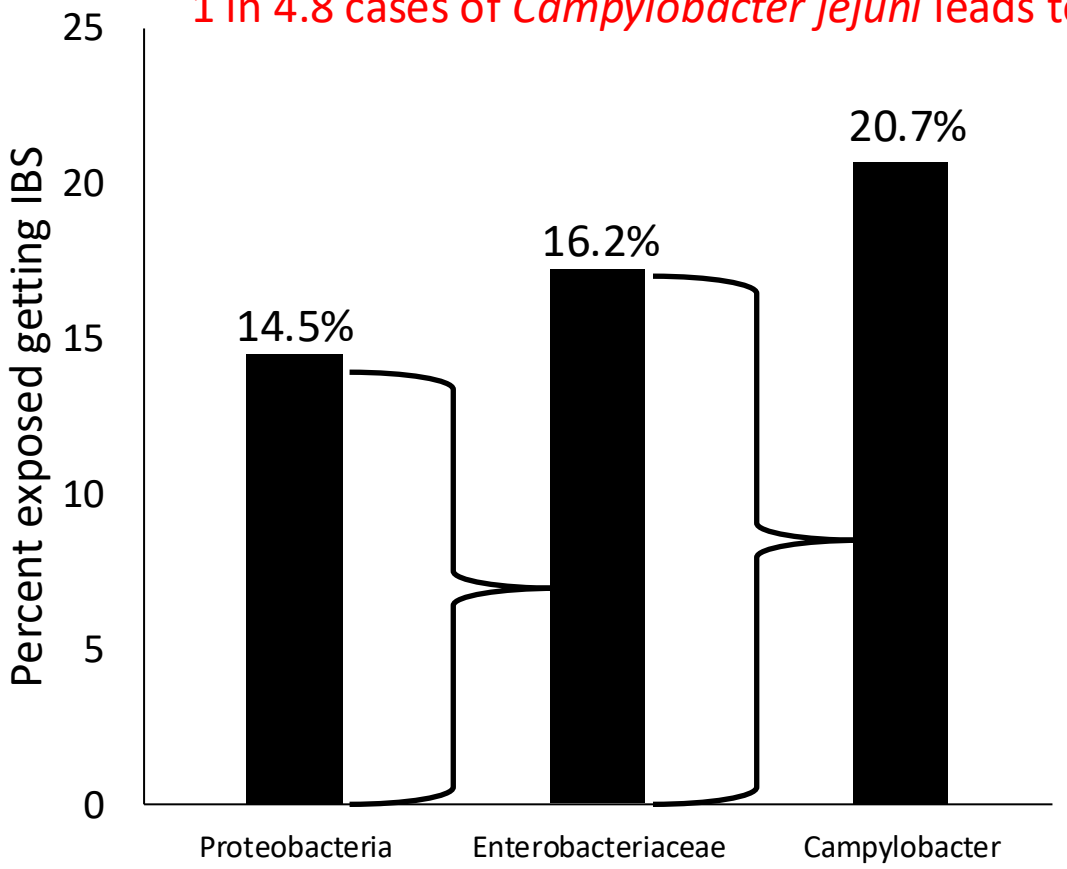


Porcari, et al. Gut 2024.

Campylobacter jejuni is the biggest culprit



1 in 4.8 cases of *Campylobacter jejuni* leads to IBS



*Only 2 studies

Porcari, et al. Gut 2024

Gastroenteritis (not stress) linked to IBS

Variable	Any constipation	Any IBS	Any dyspepsia	Any FGD
Race = white	1.43 (0.85, 2.42)	1.55 (0.70, 3.39)	1.32 (0.46, 3.83)	1.43 (0.92, 2.22)
Married	1.32 (0.79, 2.31)	0.72 (0.32, 1.59)	0.92 (0.36, 2.35)	1.04 (0.67, 1.61)
Junior enlisted (E1–E4)	2.43 (1.18, 4.97)*	3.70 (1.28, 10.71)*	1.78 (0.50, 6.33)	2.08 (1.18, 3.65)*
Any diarrhea	2.27 (1.24, 4.15)*	5.27 (2.28, 12.21)**	1.62 (0.53, 4.98)	2.54 (1.59, 4.07)**
Sought care for diarrhea	2.26 (1.23, 4.15)*	5.24 (2.26, 12.18)**	1.83 (0.58, 5.85)	2.72 (1.68, 4.39)**
Lost duty for diarrhea	2.88 (1.19, 6.97)*	4.76 (1.55, 14.55)*	2.41 (0.64, 9.06)	2.95 (1.48, 5.90)*
Any vomiting	4.14 (1.89, 9.08)**	7.00 (2.70, 18.14)**	4.35 (1.41, 13.44)*	4.73 (2.65, 8.45)**
Sought care for vomiting	4.43 (1.95, 10.04)**	7.67 (2.75, 21.40)**	5.42 (1.61, 18.26)*	5.23 (2.98, 10.26)**
Lost duty for vomiting	2.51 (1.00, 6.28)*	7.13 (1.82, 28.00)*	2.65 (0.64, 10.96)	3.30 (1.57, 6.90)*
Feelings of being in danger	0.88 (0.49, 1.57)	1.73 (0.82, 3.67)	1.06 (0.36, 3.16)	1.15 (0.73, 1.83)
Seeing killed persons	1.05 (0.58, 1.89)	2.00 (0.89, 4.46)	0.66 (0.19, 2.23)	1.18 (0.73, 1.89)
Firing one's weapon	0.73 (0.22, 2.43)	1.27 (0.34, 4.75)	1.00 (0.18, 5.58)	1.07 (0.47, 2.41)
Any war stressor	1.29 (0.78, 2.14)	2.30 (1.06, 4.96)*	1.06 (0.39, 2.89)	1.51 (0.99, 2.31)
Feeling detached	1.70 (0.76, 3.80)	1.44 (0.43, 4.77)	1.27 (0.24, 6.59)	1.57 (0.80, 3.06)
Excessive alcohol use	2.04 (0.64, 6.57)	0.67 (0.73, 6.10)	N/A	2.30 (0.84, 6.26)
Binge drinking	0.60 (0.31, 1.19)	1.37 (0.59, 3.17)	1.75 (0.53, 5.76)	0.97 (0.58, 1.62)
Wanting to cut back on drinking	1.20 (0.30, 4.72)	1.88 (0.30, 11.71)	N/A	1.66 (0.48, 5.78)
Feeling down in past month	1.40 (0.83, 2.34)	1.86 (0.88, 3.92)	1.83 (0.63, 5.30)	1.35 (0.88, 2.08)
Little interest in doing things	1.36 (0.65, 2.85)	1.16 (0.69, 1.95)	1.76 (0.60, 5.18)	1.18 (0.77, 1.82)
Having nightmares	1.12 (0.49, 2.52)	0.99 (0.33, 2.95)	1.53 (0.37, 6.31)	1.10 (0.58, 2.12)
Being "on guard"	0.82 (0.38, 1.77)	0.99 (0.29, 3.46)	2.15 (0.60, 7.66)	1.13 (0.60, 2.10)

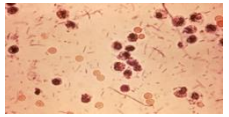
*P<0.05
**P<0.001

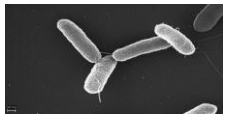
Porter, et al
Dig Dis Sci 2011

Molecular Mimicry

- "Molecular mimicry is one of the leading mechanisms by which infectious or chemical agents may induce autoimmunity."
- "Molecular mimicry is a process that occurs when the immune system mistakes a foreign substance for a part of the body."

Causes of Post-infection IBS


Shigella


Salmonella

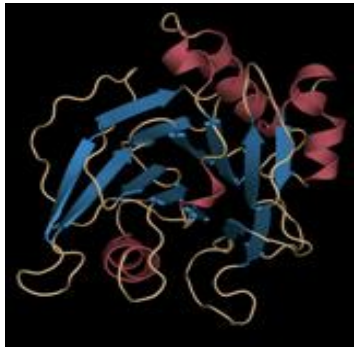

Campylobacter

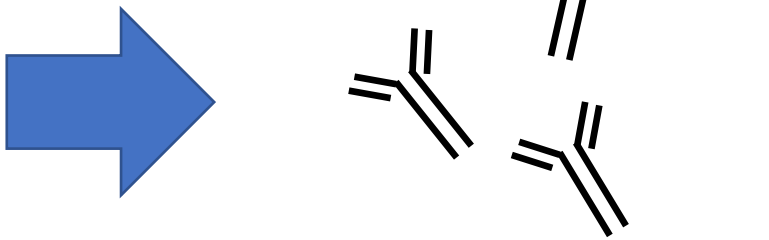

E. coli

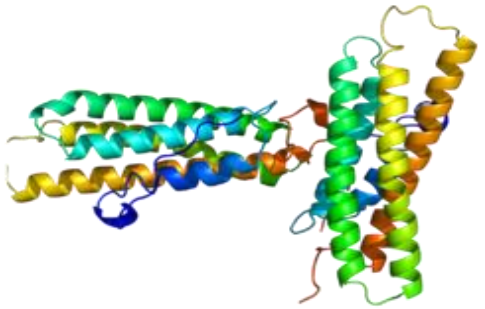
Cytolethal Distending Toxin B (CdtB)

Rojas, et al. J Autoimmun 2018

Molecular Mimicry/Autoimmunity



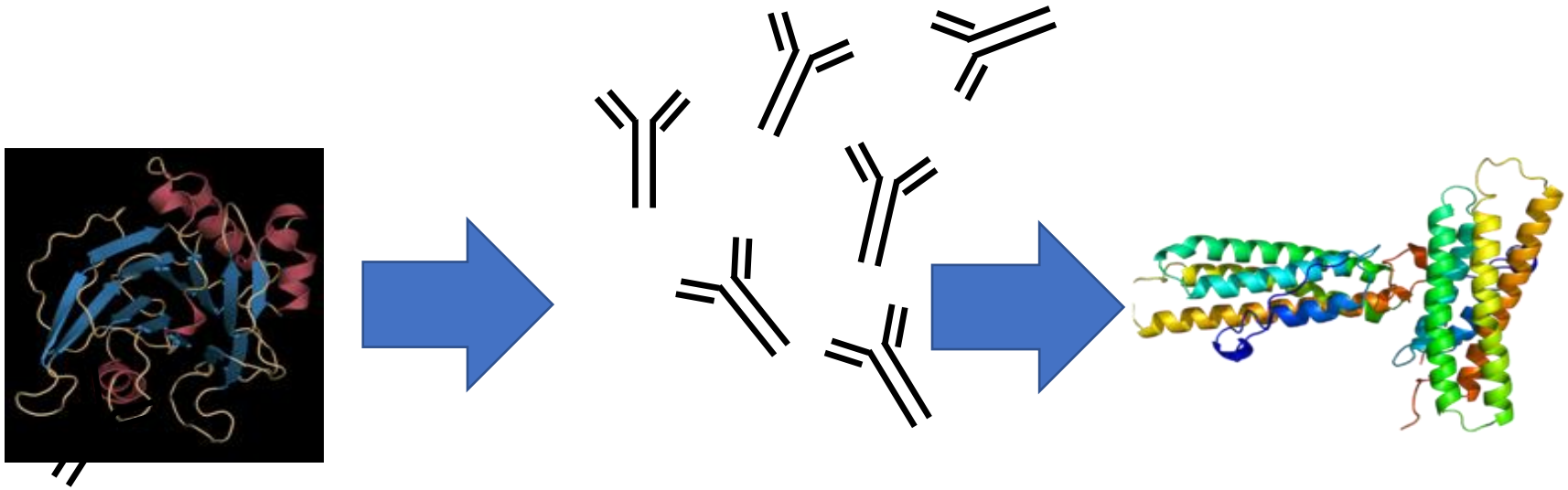




Cytolethal Distending Toxin B

Human Vinculin

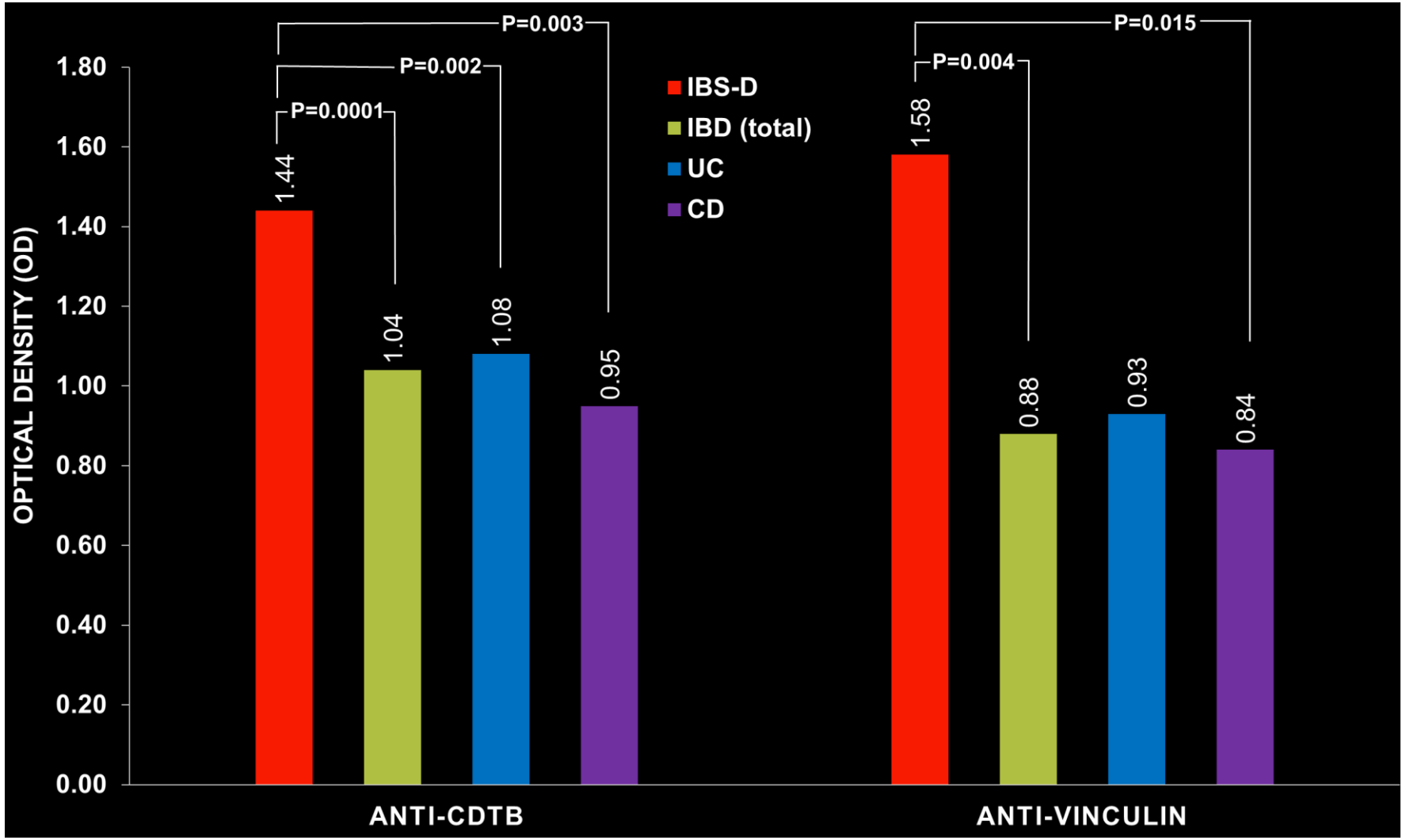
Molecular Mimicry/Autoimmunity



Cytolethal Distending Toxin B

Human Vinculin

Generation 2 Blood Test for Diagnosing IBS-D



Morales, et al. Dig Dis Sci 2019

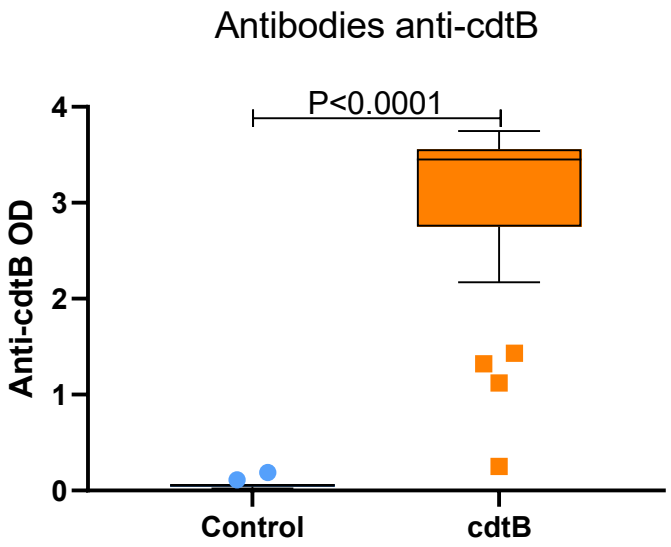
Antibodies (Generation 2) test dynamics

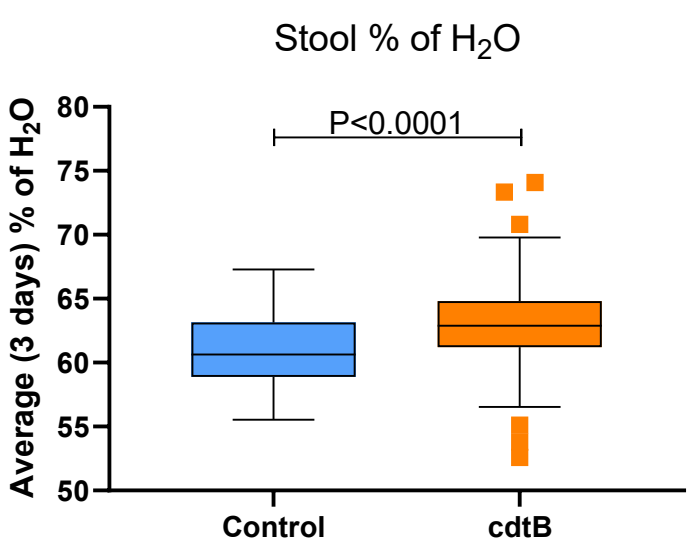
Anti-CdtB	Anti-vinculin	Post-Test Probability of IBS
+	-	89%
-	+	88%
+	+	98%

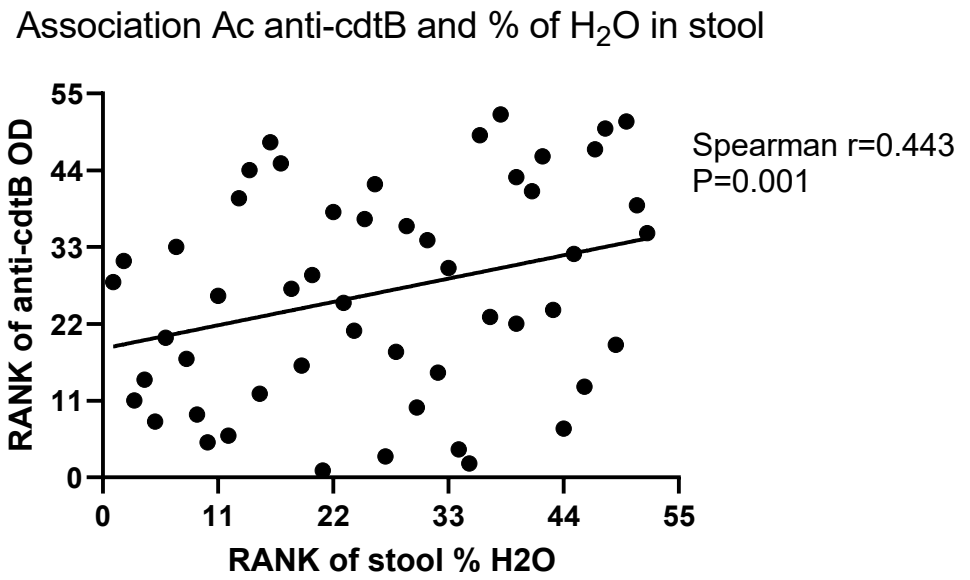
	Sensitivity	Specificity	PPV	NPV	LR+	The old Assay
Anti-CdtB	43%	93.5%	95.6	33.7	6.3	5.2
Anti-vinculin	52%	90.9%	96.3	29.4	5.3	2.0
And/or	56%					

Morales, et al. Dig Dis Sci 2019

Rats get IBS-D phenotype after CdtB injection

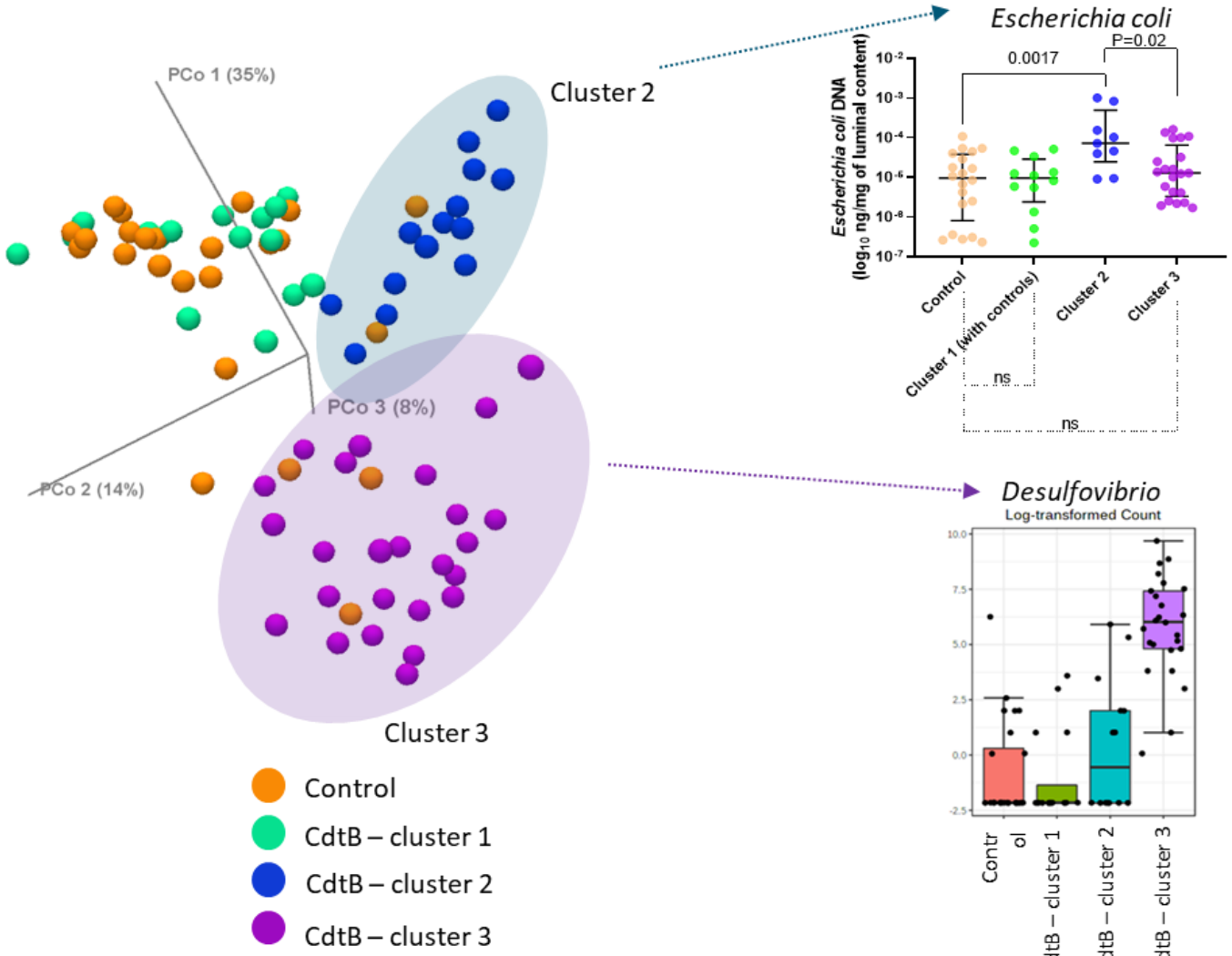






Leite, et al Gut Microbes, 2024.

Animal Model of Post-Infectious IBS-D: 3 Microtypes

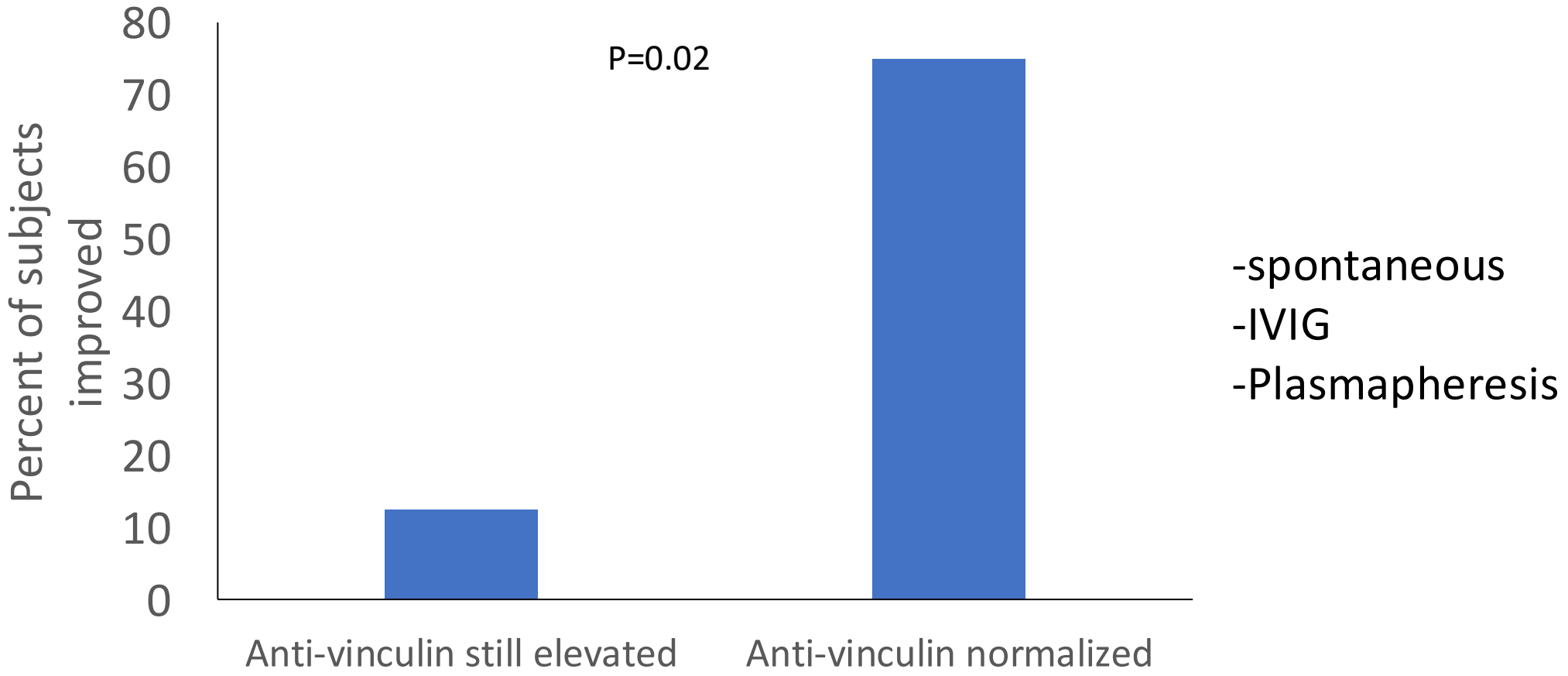


Hydrogen cluster

Hydrogen sulfide cluster

Leite, et al. Gut Microbes, 2024.

Anti-vinculin antibodies and IBS Symptoms



Sharabi, et al. Dig Dis Sci 2025

IBSSmart™

- The first test telling a patient and clinician that you have IBS-D (M)
- The first test telling you what may have caused their IBS-D
- The antibody appear to cause IBS (not just a marker of IBS)
- In my experience higher predict poor response to antibiotics and relapse
- You can guide your patient about travel based on this test
- Reducing anti-vinculin antibodies appears to improve IBS symptoms

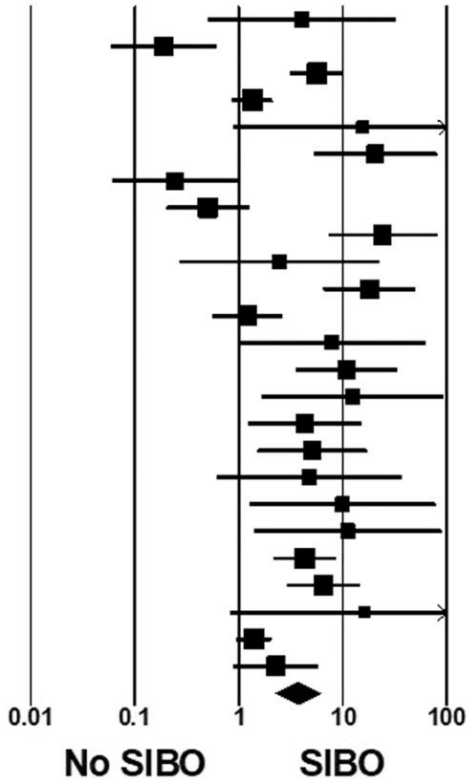
IBS IS AN ORGANIC DISEASE!

Breath Testing Suggests SIBO in IBS

SIBO in IBS patients and controls, all studies included

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value	SIBO / Total IBS	SIBO / Total Controls
Posserud I et al*	4.013	0.503	32.026	1.311	0.190	10 / 162	1 / 62
Choung R et al	0.189	0.058	0.614	-2.771	0.006	3 / 148	52 / 527
Pyleris E et al	5.640	3.098	10.267	5.660	0.000	42 / 112	20 / 208
Giamarellos-Bourboulis E et al	1.345	0.860	2.102	1.299	0.194	33 / 252	65 / 645
Ghoshal U et al 2014*	15.581	0.883	275.075	1.875	0.061	34 / 80	0 / 10
Pimental M et al	20.667	5.293	80.691	4.358	0.000	93 / 111	3 / 15
Walters B et al	0.246	0.060	1.002	-1.958	0.050	4 / 42	6 / 20
Bratten J et al	0.505	0.202	1.265	-1.458	0.145	166 / 224	34 / 40
Scarpellini E et al	24.267	7.347	80.150	5.231	0.000	28 / 43	4 / 56
Park JH et al*	2.484	0.274	22.536	0.809	0.419	7 / 38	1 / 12
Collin B et al	18.041	6.547	49.712	5.593	0.000	68 / 75	14 / 40
Park JS et al*	1.214	0.558	2.642	0.489	0.625	34 / 76	16 / 40
Zhao J et al*	7.778	0.968	62.498	1.929	0.054	35 / 89	1 / 13
Lupascu A et al*	10.889	3.517	33.710	4.141	0.000	20 / 65	4 / 102
Rana S et al 2008*	12.375	1.653	92.658	2.449	0.014	25 / 225	1 / 100
Parodi A et al*	4.303	1.236	14.979	2.293	0.022	21 / 130	3 / 70
Lombardo L et al*	5.084	1.515	17.063	2.632	0.008	49 / 200	3 / 50
Ghoshal U et al 2010*	4.779	0.609	37.485	1.489	0.137	13 / 149	1 / 51
Rana S et al 2011*	9.994	1.275	78.341	2.191	0.028	11 / 175	1 / 150
Sachdeva S et al*	11.200	1.405	89.252	2.281	0.023	14 / 59	1 / 37
Abbasi M et al*	4.317	2.144	8.691	4.096	0.000	40 / 107	13 / 107
Moraru I et al*	6.504	2.920	14.490	4.582	0.000	105 / 331	7 / 105
Galatola G et al	16.280	0.825	321.375	1.833	0.067	5 / 17	0 / 18
Schatz R et al	1.398	0.948	2.061	1.691	0.091	77 / 124	436 / 808
Grover M et al*	2.289	0.892	5.873	1.723	0.085	52 / 158	6 / 34
	3.696	2.261	6.042	5.215	0.000		

25 Studies

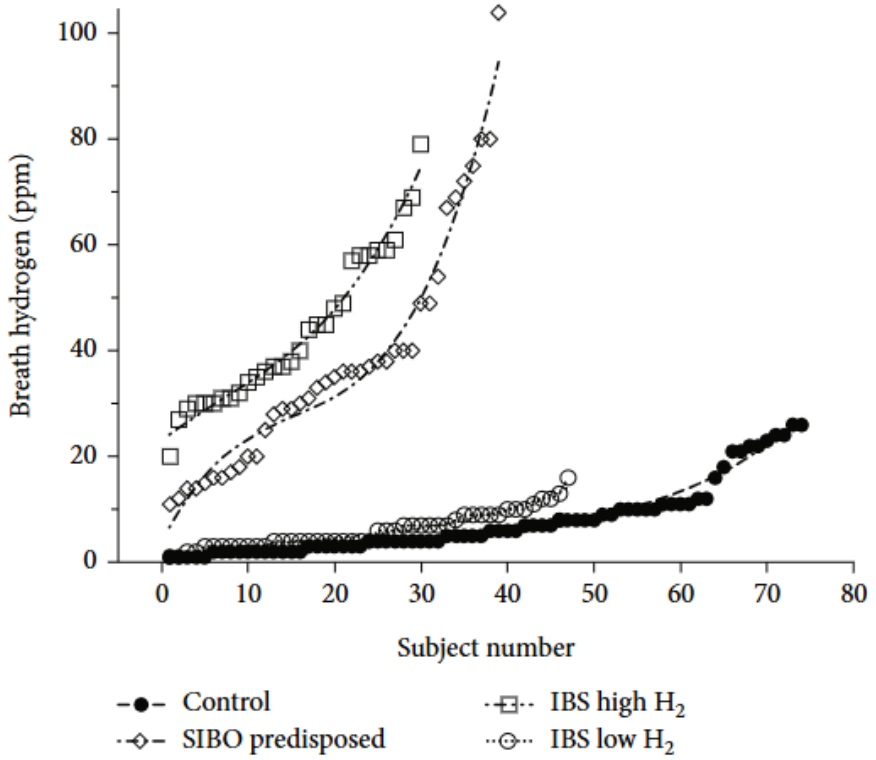


OR = 3.7 [95% CI 2.3–6.0]

*15 are considered high quality studies

Shah, et al. Am J Gastroenterol 2020.

Breath testing and transit- Finally a gold standard study



-Transit did not explain the breath test

-Anything at or before 80 minutes was valid

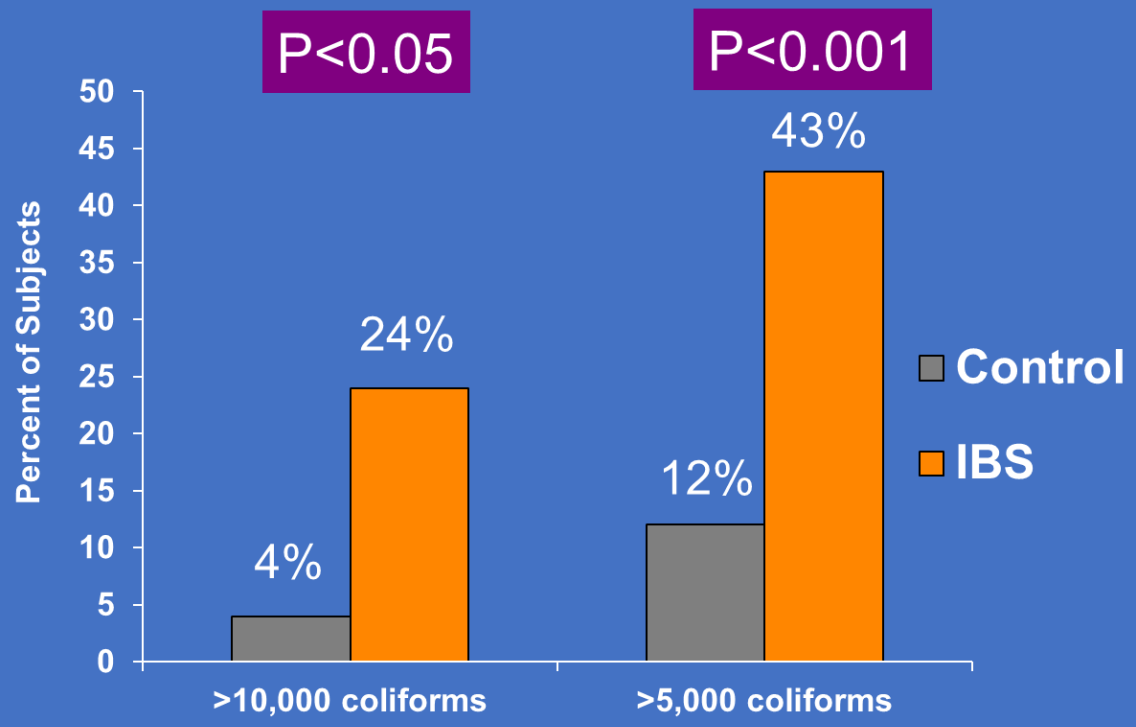
-20ppm by 80 minutes is the cutoff for hydrogen

-69% if IBS patients had high hydrogen

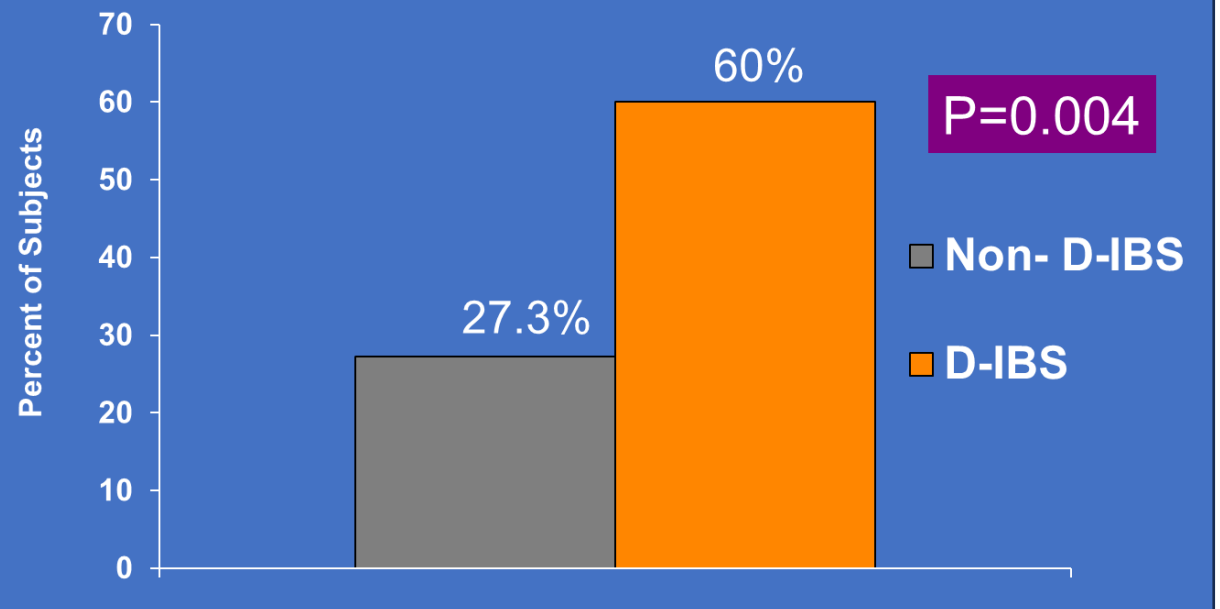
-Those with high hydrogen responded to antibiotics

Dahlgren, et al Gastroenterol Res Pract 2025

Small Bowel Culture Supports SIBO in IBS



N=165 IBS, 26 controls
Posserud, et al, Gut, 2007;56:802-8.



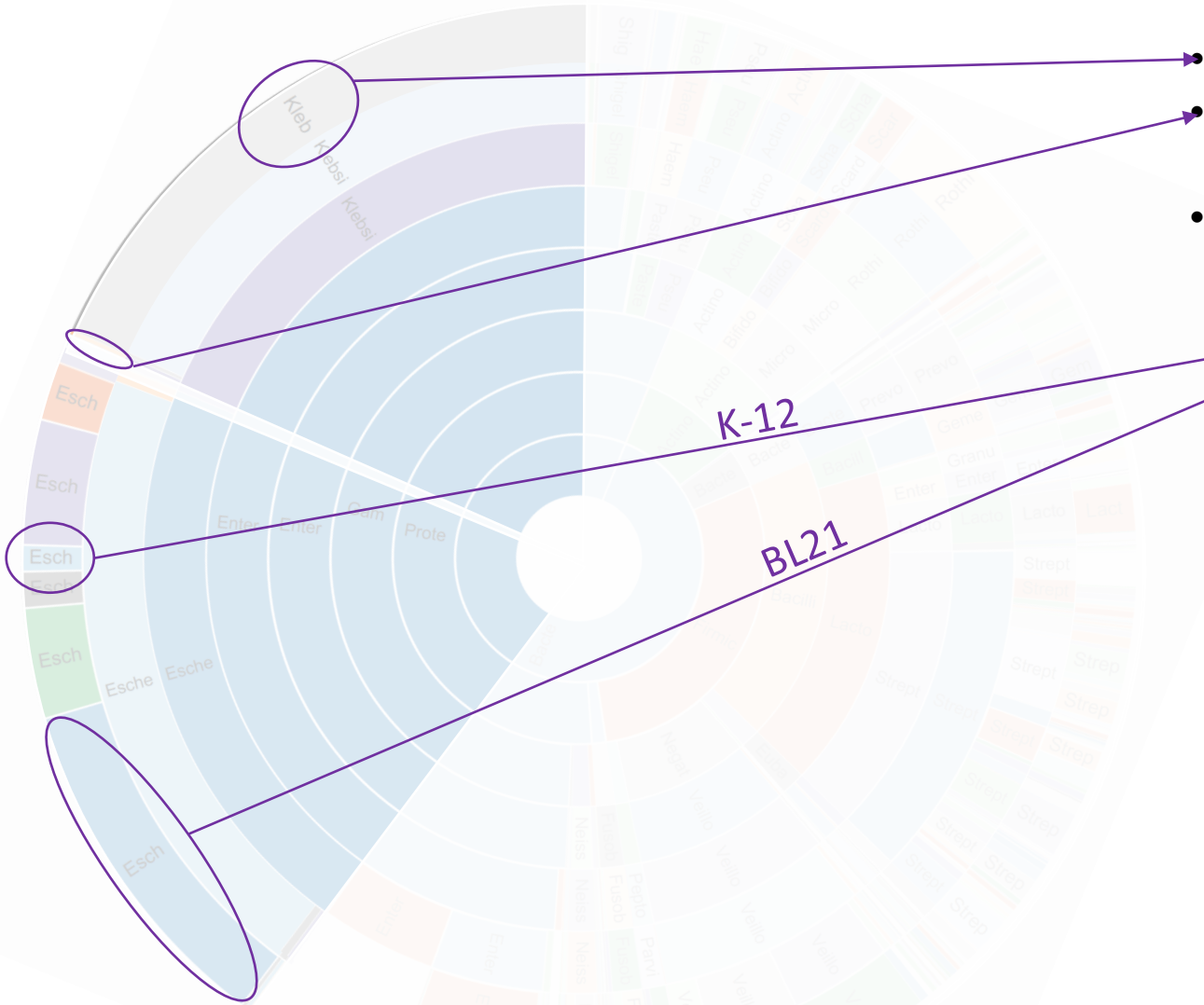
N=77 non-D-IBS, N=35 D-IBS
Pyleris, et al. DDS, 2012

Breath Testing is Validated against SB Culture and Sequencing

Time points for H_2 levels detection in breath after lactulose ingestion (minutes)	SIBO detection Sensitivity (%)	SIBO detection Specificity (%)	Gammaproteobacteria phylum		Firmicutes phylum		Excess of gas		Energy metabolism function (H_2 production)	
			Spearman r	P-value	Spearman r	P-value	Spearman r	P-value	Spearman r	P-value
60	14.28	92.30	0.488	**0.021**	-0.318	0.149	0.355	0.105	0.272	0.221
75	57.14	84.61	0.516	**0.014**	-0.371	0.089	0.390	0.073	0.313	0.156
90	57.14	84.61	0.630	**0.002**	-0.508	**0.016**	0.408	0.059	0.457	**0.033**
105	57.14	69.23	0.689	**3.879E-4**	-0.583	**0.004**	0.444	**0.039**	0.527	**0.012**
120	57.14	61.53	0.781	**1.795E-5**	-0.651	**0.001**	0.368	0.092	0.602	**0.003**

Leite, et al Plos One 2020

SIBO is characterized by *Klebsiella* and *Escherichia coli*



- *Klebsiella pneumoniae* (~18% of the whole microbiome)
- *Klebsiella aerogenes* (less than 1% of the whole microbiome)

- *Escherichia coli* (~28% of the whole microbiome)
  - total of 8 potential strains were detected
  - 2 strains with more genomic similarities to the common strains BL21 and K-12 were more abundant in SIBO and associated with GI symptoms

Bacterial species/strains	SIBO vs. Non-SIBO (Log_2 fold change)	Adj-P-value
Klebsiella pneumoniae	22.39	< 0.0001
Klebsiella aerogenes	12.93	< 0.0001

Leite, et al Clinical Gastro Hepatol 2024

Overgrowth of *Escherichia coli* and *Klebsiella* spp. in the small bowel is associated with enrichment of sugar degradation pathways and biogenic amines metabolism

	Detected pathway	SIBO vs. non-SIBO fold change	Adj. P-value
H_2 production	Glucose degradation	63.07	< 0.0001
	Galactose degradation	1.84	< 0.0001
	Mixed acid fermentation	1.22	< 0.0001

Leite, et al. Clin Gastroenterol Hepatol 2023

Open

Hydrogen and Methane-Based Breath Testing in Gastrointestinal Disorders: The North American Consensus

Rezaie, et al. Am J Gastroenterol, 2018

Ali Rezaie, MD, MSc, FRCP(C)[1], Michelle Buresi, MD[2], Anthony Lembo, MD[3], Henry Lin, MD[4], Richard McCallum, MD[5], Satish Rao, MD[6], Max Schmulson, MD[7], Miguel Valdovinos, MD[8], Salam Zakko, MD[9], Mark Pimentel, MD, FRCP(C)[1] and on behalf of The North American Consensus group on hydrogen and methane-based breath testing

European guideline on indications, performance, and clinical impact of hydrogen and methane breath tests in adult and pediatric patients: European Association for Gastroenterology, Endoscopy and Nutrition, European Society of Neurogastroenterology and Motility, and European Society for Paediatric Gastroenterology Hepatology and Nutrition consensus

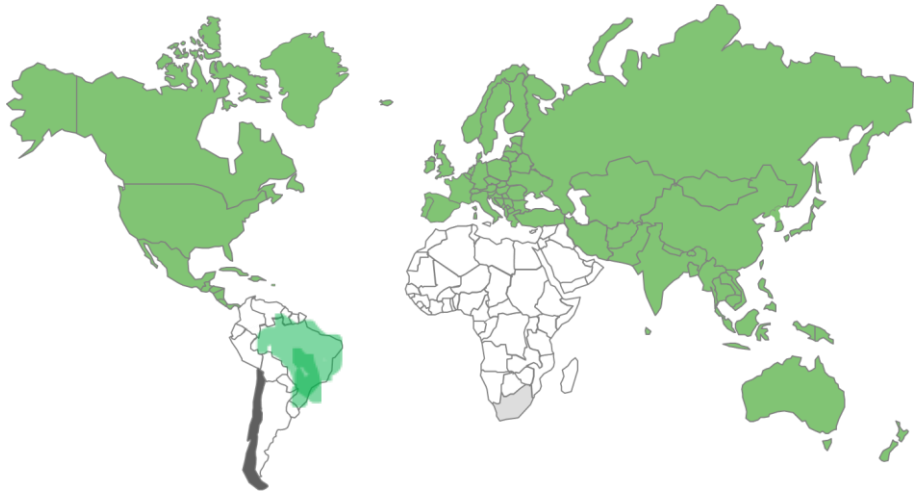
Hammer, et al. United Europ Gastroenterol J, 2021

Heinz F. Hammer[1] | Mark R. Fox[2,3] | Jutta Keller[4] | Silvia Salvatore[5] | Guido Basilisco[6] | Johann Hammer[7] | Loris Lopetuso[8,9] | Marc Benninga[10] | Osvaldo Borrelli[11] | Dan Dumitrascu[12] | Bruno Hauser[13] | Laszlo Herszenyi[14] | Radislav Nakov[15] | Daniel Pohl[3] | Nikhil Thapar[11,16] | Marc Sonyi[1,17] | European H_2-CH_4-breath test group

Asian-Pacific consensus on small intestinal bacterial overgrowth in gastrointestinal disorders: An initiative of the Indian Neurogastroenterology and Motility Association

Ghoshal, et al. Ind J Gastroenterol, 2022

Uday C. Ghoshal[1] · Sanjeev Sachdeva[2] · Ujjala Ghoshal[3] · Asha Misra[1] · Amarender Singh Puri[2] · Nitesh Pratap[4] · Ayesha Shah[5] · M. Masudur Rahman[6] · Kok Ann Gwee[7,8] · Victoria P Y Tan[9] · Tahmeed Ahmed[10] · Yeong Yeh Lee[11,12] · B S Ramakrishna[13] · Rupjyoti Talukdar[14] · S V Rana[15] · Saroj K Sinha[16] · Minhu Chen[17] · Nayoung Kim[18] · Gerald Holtmann[5]



1. Mostly agree about criteria for SIBO
2. Mostly agree about dosing for substrates
3. Mostly agree that SIBO and IBS are interrelated

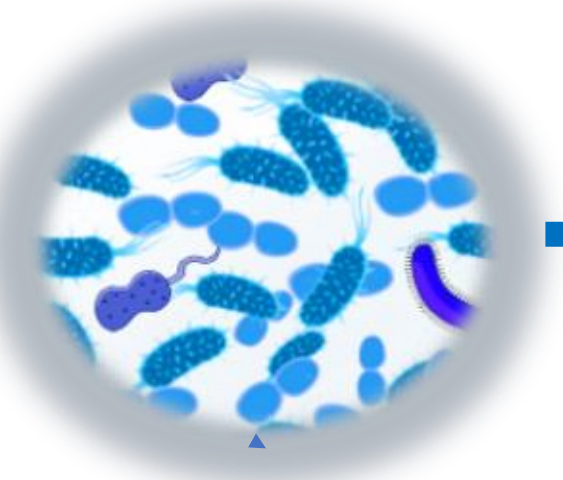
ORIGINAL ARTICLE/GASTROENTEROLOGY • Arq. Gastroenterol. 62 • 2025 • https://doi.org/10.1590/S0004-2803.24612024-107 copy

DIAGNOSIS AND TREATMENT OF SMALL INTESTINAL BACTERIAL OVERGROWTH: AN OFFICIAL POSITION PAPER FROM THE BRAZILIAN FEDERATION OF GASTROENTEROLOGY

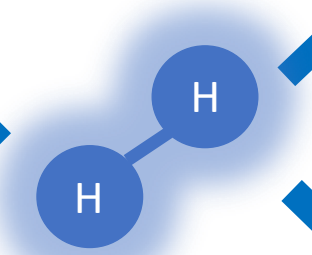
Diagnóstico e tratamento do supercrescimento bacteriano no intestino delgado: um posicionamento oficial da Federação Brasileira de Gastroenterologia

There are 3 Main Gases that must be Measured

Hydrogen Producers

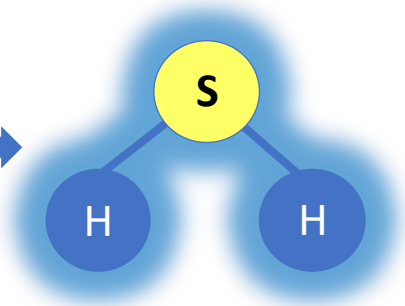


Typical SIBO H_2 is
E. coli and *Klebsiella*[1]
from inside the small intestine



Hydrogen Sulfide Producers

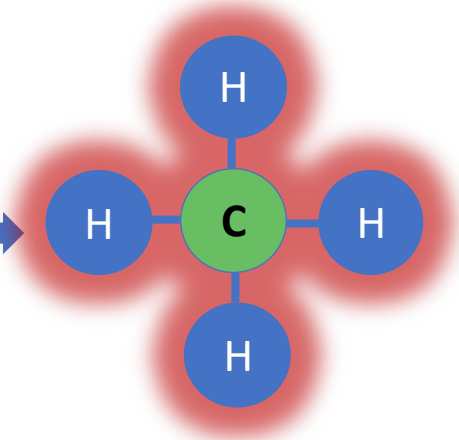
Many H_2S producers



$5H_2 \rightarrow 1H_2S$

Methane Producers

Methanogens such as *M. smithii*[2]
found in colon and small bowel[3]



$4H_2 \rightarrow 1CH_4$

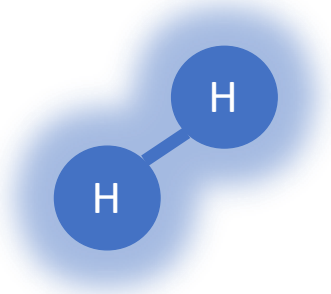
Phenotype

-Diarrhea
-Pain
-Urgency

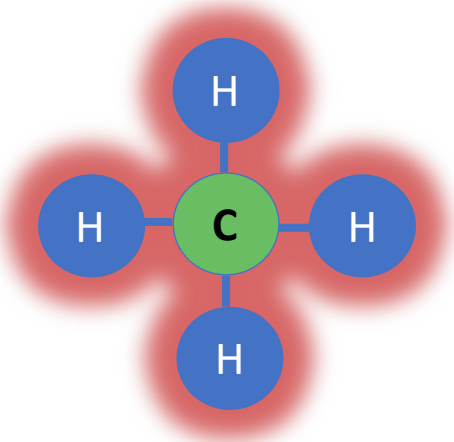
-Constipation
-Discomfort
-Bloating

[1]Leite, et al. PlosOne 2019.
[2]Kunkel, et al. Dig Dis Sci 2011.
[3]Villanueva, et al. DDW 2022.

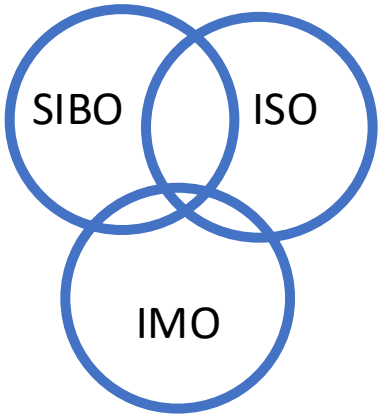
Three Gas Microtypes

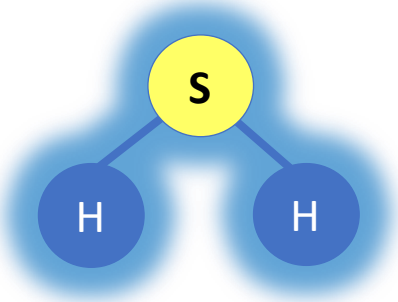


Small Intestinal Bacterial Overgrowth (SIBO)



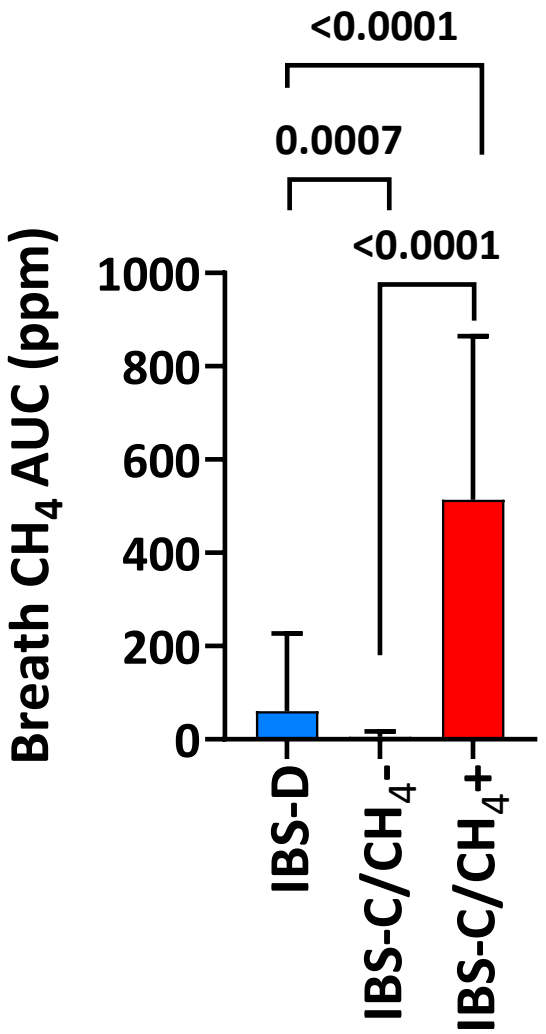
Intestinal Methanogen Overgrowth (IMO)

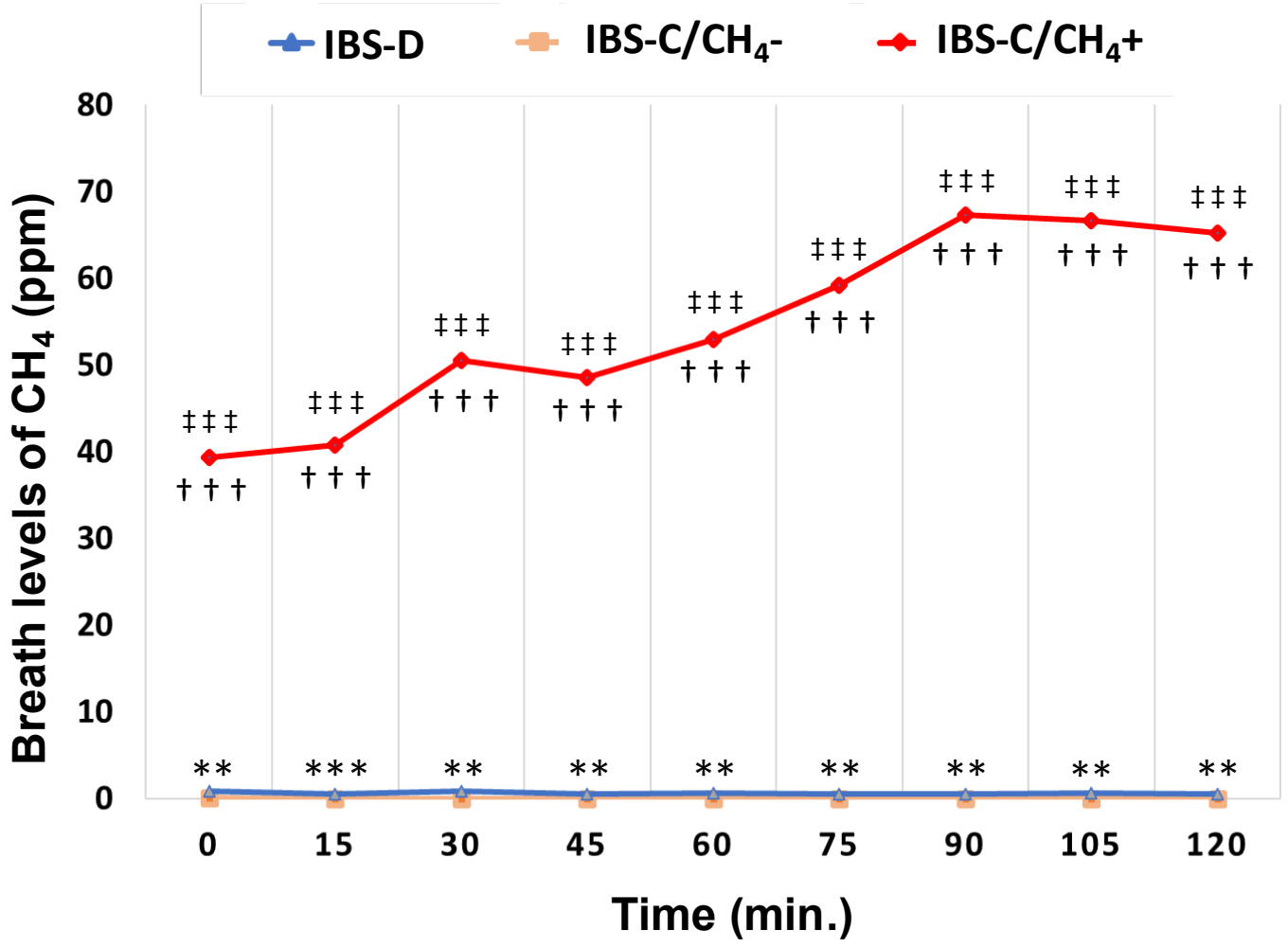




Intestinal Sulfide Overproduction (ISO)

CH_4 Levels Are Not Elevated In IBS-D Subjects

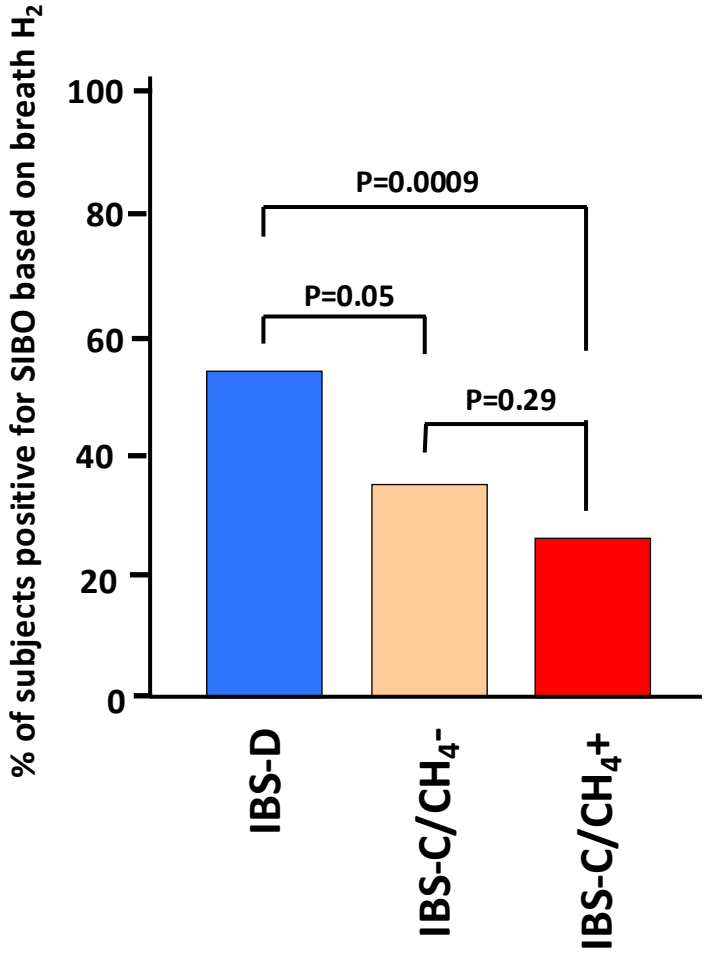


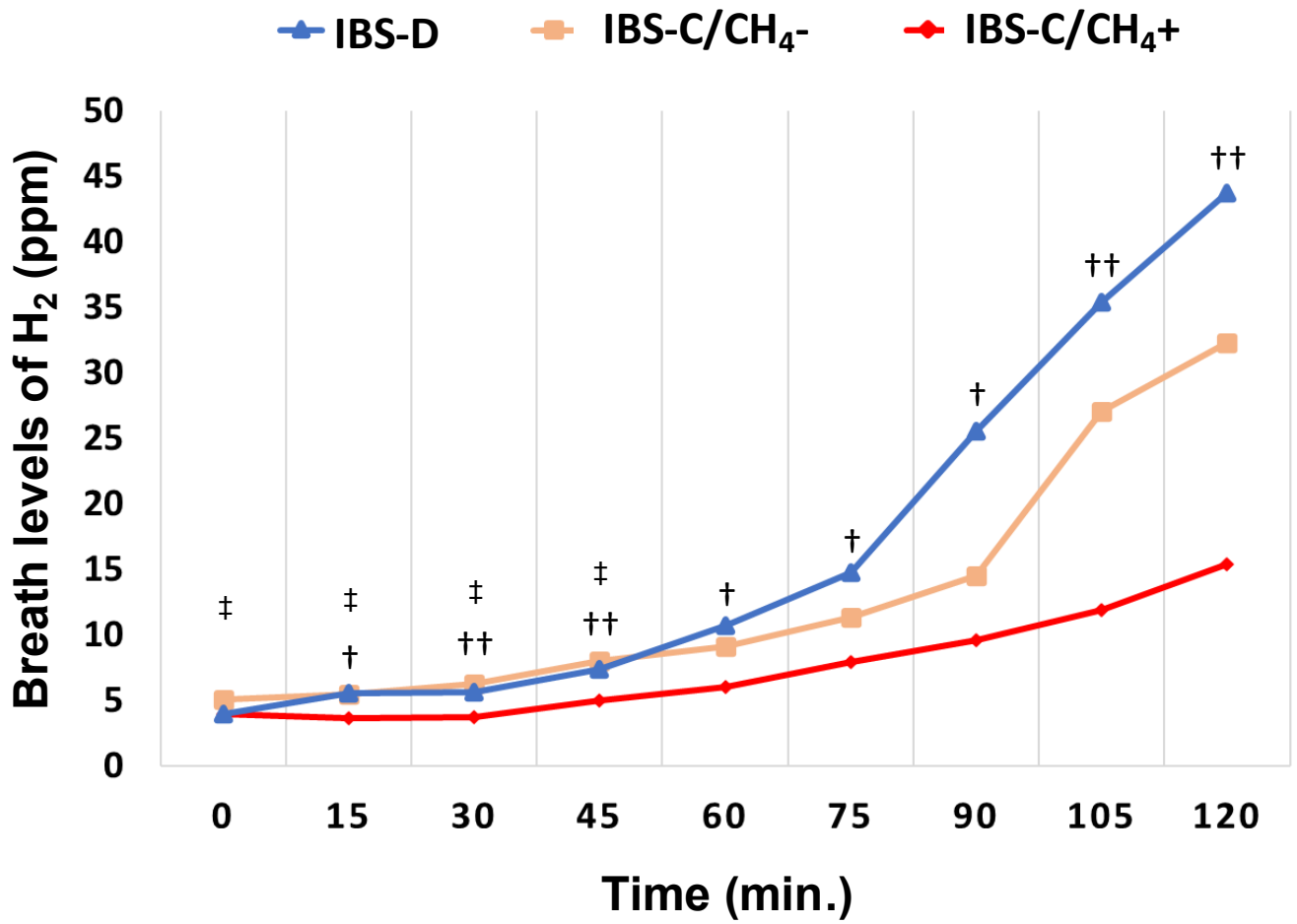


**P < 0.01; ***P < 0.001 IBS-D vs IBS-C/CH_4-
†††P < 0.001 IBS-D vs IBS-C/CH_4+
‡‡‡P < 0.001 IBS-C/CH_4- vs IBS-C/CH_4+

Villanueva, et al. Am J Gastroenterol, 2022

H_2 Levels Are Elevated In IBS-D Subjects



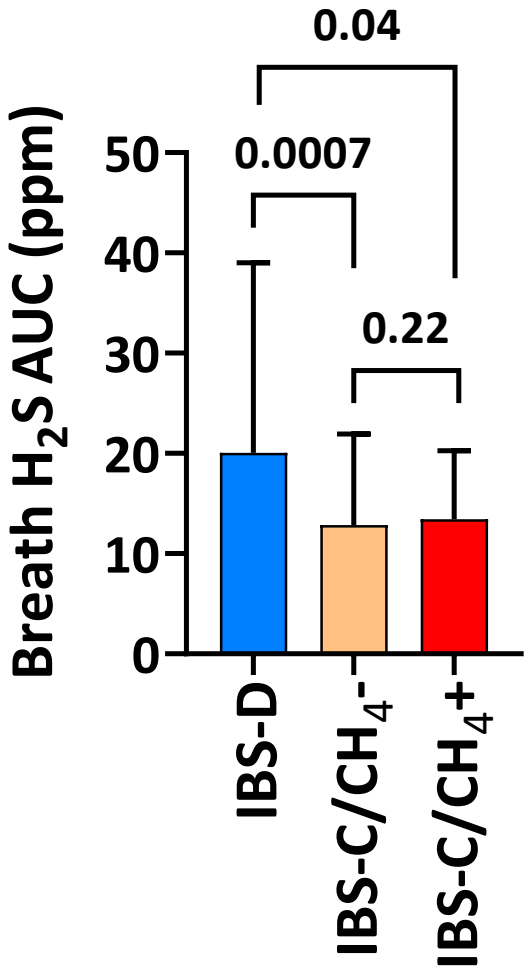


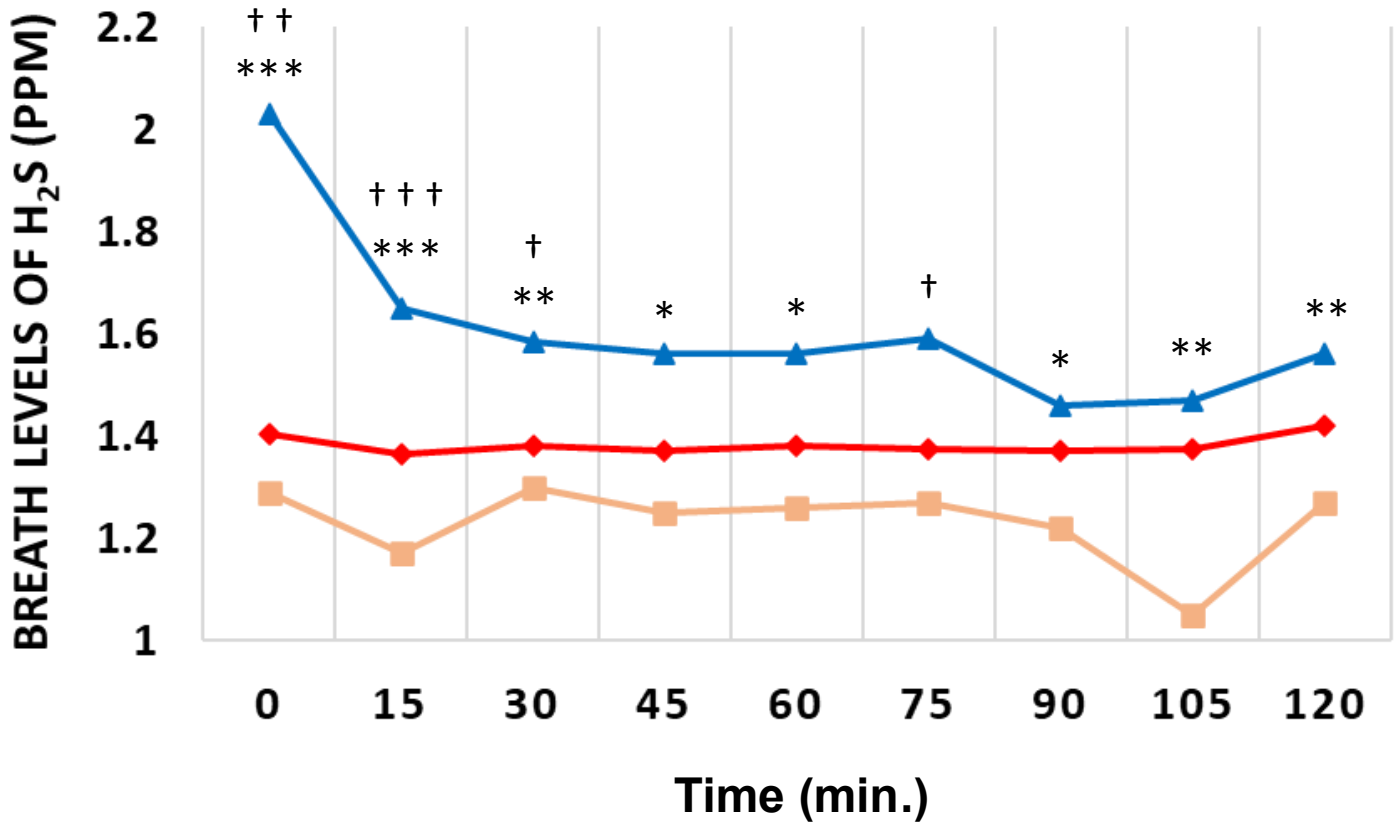
SIBO: ≥ 20ppm rise in H_2 from baseline

†P < 0.05; ††P < 0.01 IBS-D vs IBS-C/CH_4+
‡P < 0.05 IBS-C/CH_4- vs IBS-C/CH_4+

Villanueva, et al. Am J Gastroenterol, 2022

H_2S Levels Are Elevated In IBS-D Subjects





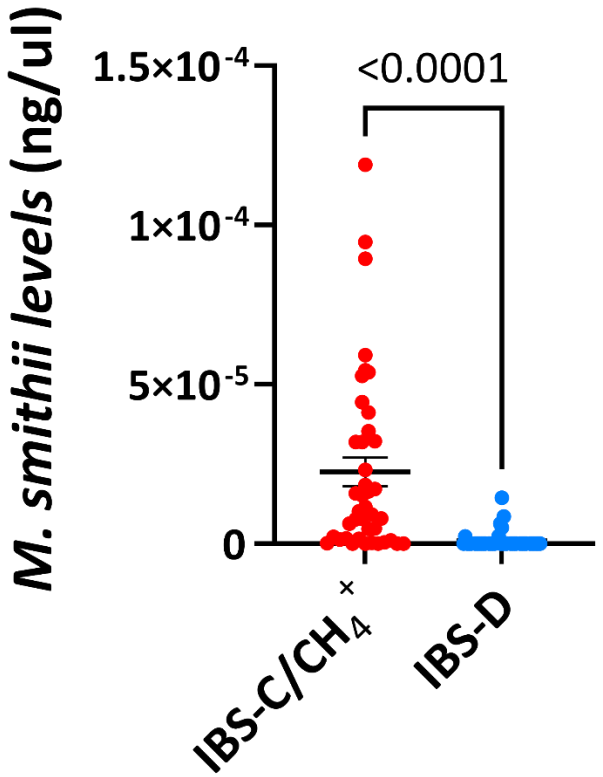
*P < 0.05; **P < 0.01; ***P < 0.001 IBS-D vs IBS-C/CH_4-
†P < 0.05; ††P < 0.01; †††P < 0.001 IBS-D vs IBS-C/CH_4+

Villanueva, et al. Am J Gastroenterol, 2022

Methanogens in IBS-C vs IBS-D

IBS-C subjects had higher abundance of family Methanobacteriaceae and species *Methanobrevibacter smithii*

Methanobrevibacter smithii positively correlated with breath CH_4 and negatively correlated with H_2

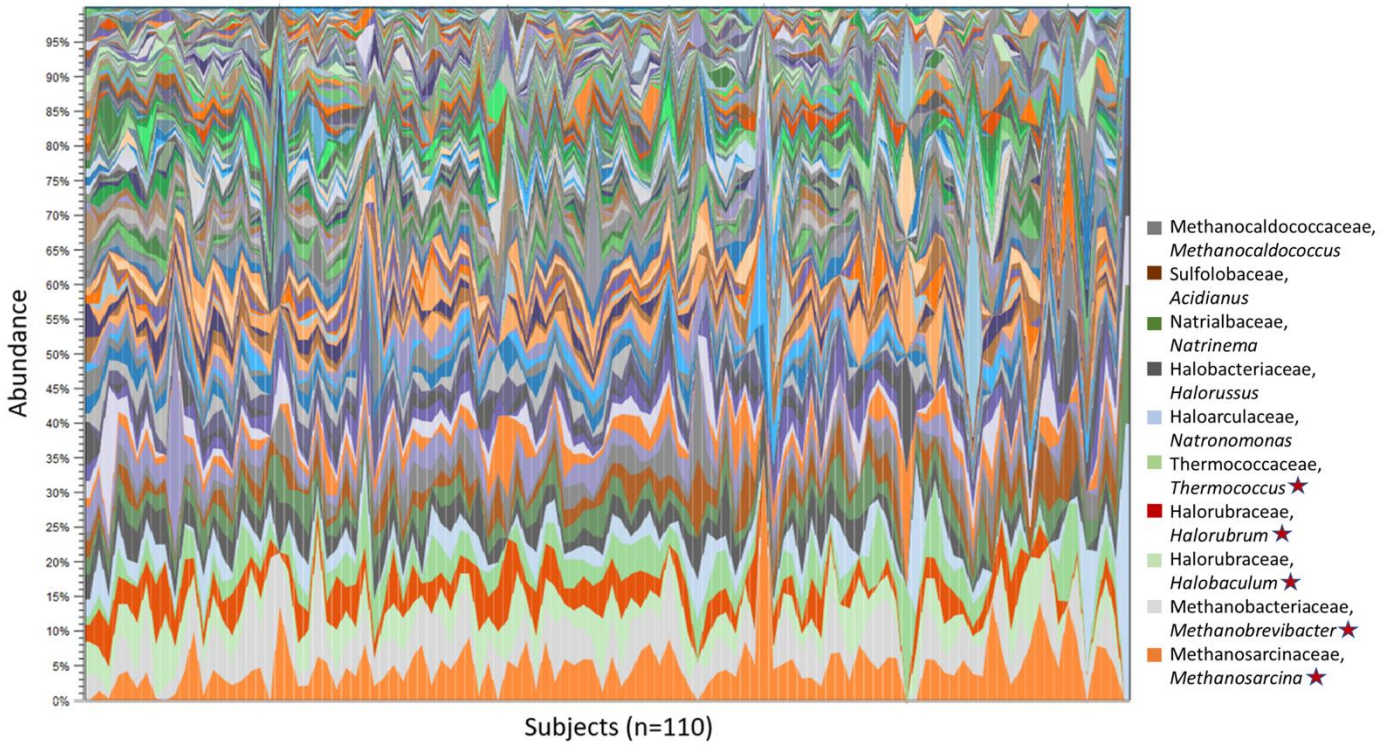


Breath test time point (minutes)	Absolute load of *M. smithii* (qPCR)			
	Methane levels		Hydrogen levels	
	R	P-value	R	P-value
0	0.410	0.003	-0.047	0.750
15	0.478	0.001	0.073	0.621
30	0.480	<0.0001	0.045	0.759
45	0.533	<0.0001	0.100	0.494
60	0.540	<0.0001	-0.019	0.897
75	0.564	<0.0001	-0.179	0.217
90	0.545	<0.0001	-0.265	0.065
105	0.560	<0.0001	-0.375	0.008
120	0.492	<0.0001	-0.332	0.020

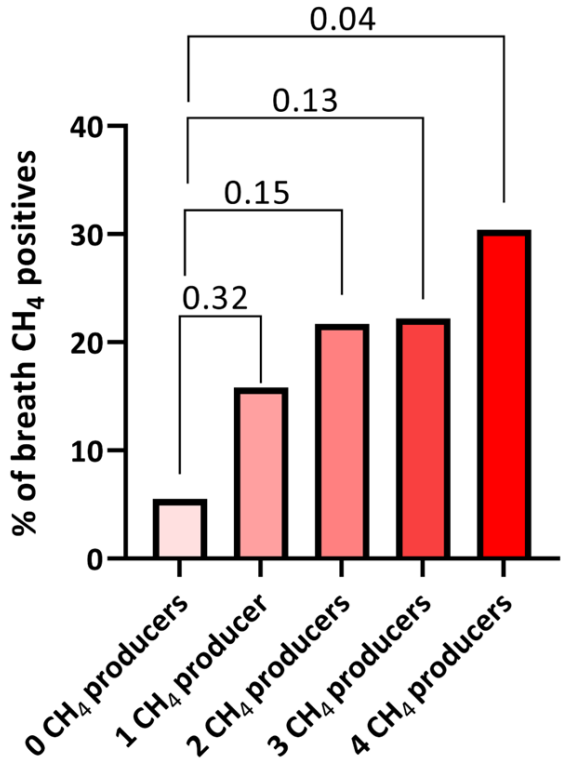
All subjects pooled (n=81)

Villanueva, et al. Am J Gastroenterol, 2022

Methane on Breath Test Reflects the small bowel



The Archaeome in the small intestine



Breath Methane and Methanogens

N=110 subjects with small bowel aspirate and shotgun sequencing
Of the 110 subject, 22 had IMO based on single baseline breath test

Villanueva-Milan, et al. DDW 2024

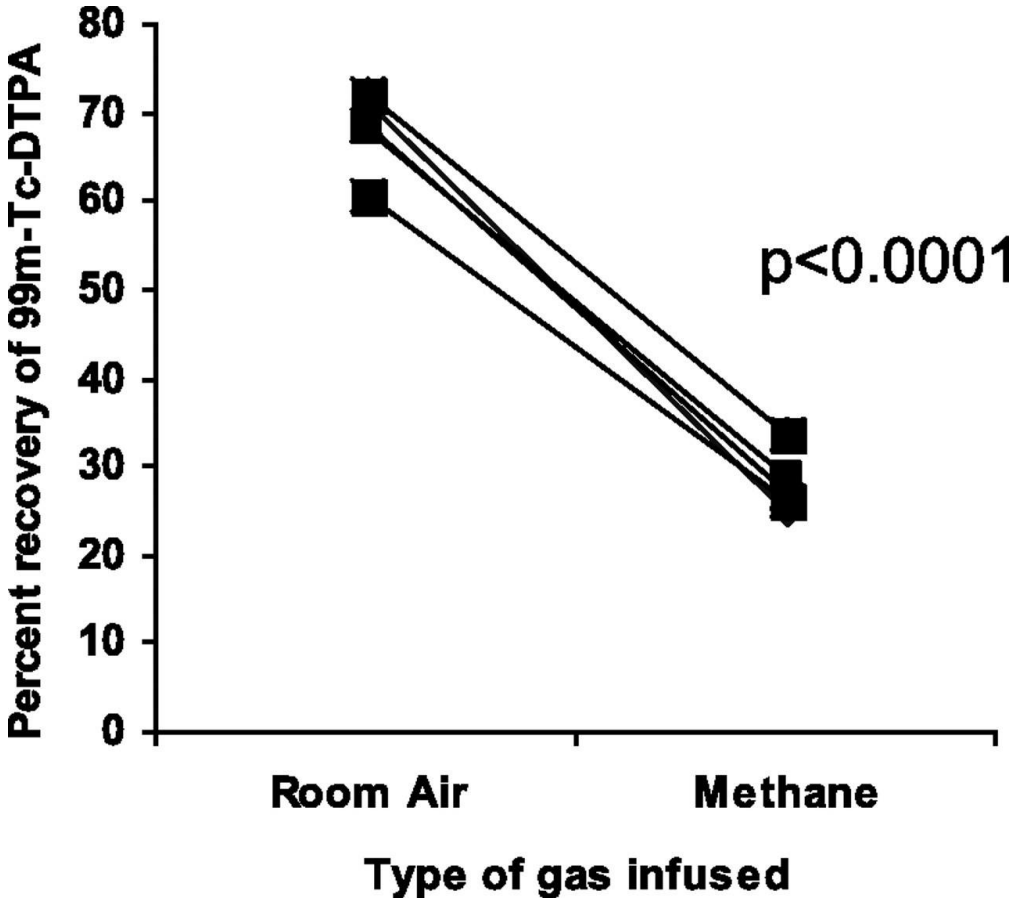
Methanogens in small bowel and CH_4

	Archaeal species	Prevalence (n=110)	Correlation with methane		Correlation with *M. smithii*		Methane production
			R	P value	R	P value	
1	***Methanothermococcus okinawensis***	40%	0.319	0.001	0.282	0.003	Uses H_2 and CO_2 as sole sources of energy and carbon. Formate can be used as an alternative source
2	***Halomicroarcula sp. SHR3***	55.45%	0.291	0.002	0.194	0.042	Not enough information
3	***Halarchaeum* sp. CBA1220**	63.64%	0.277	0.003	No	No	Contains enzymes for methane metabolism
4	***Thermococcus argininiproducens***	29.09%	0.226	0.017	0.277	0.003	Contains enzymes for methane metabolism
5	***Methanobrevibacter smithii***	**63.64%**	**0.221**	**0.02**	**N/A**	**N/A**	**Uses H_2 and CO_2 as sole sources of energy and carbon. Formate can be used as an alternative source**
6	***Candidatus Nitrosotenuis cloacae***	36.36%	0.209	0.028	0.379	<0.0001	Contains enzymes for methane metabolism
7	***Halobacterium litoreum***	57.27%	0.209	0.028	0.312	0.025	Contains enzymes for methane metabolism
8	***Desulfurococcus mucosus***	27.27%	0.19	0.046	No	No	Contains enzymes for methane metabolism
9	***Halobaculum rubrum***	61.82%	0.188	0.049	No	No	Contains enzymes for methane metabolism

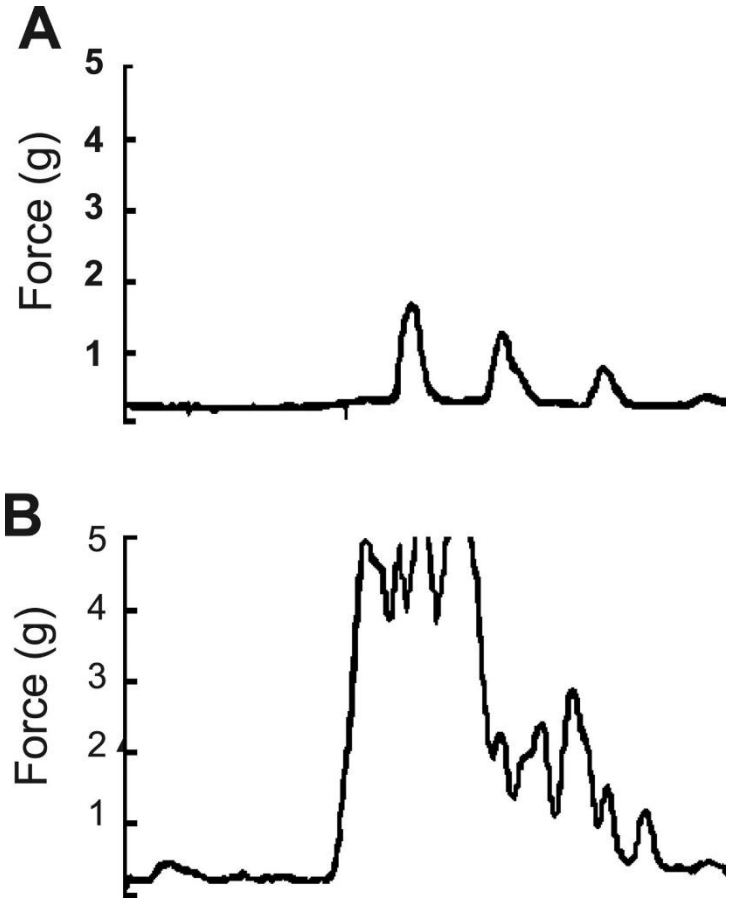
Villanueva-Milan, et al. DDW 2024

Methane and Gut Physiology

Small bowel transit time in animals
-Room air or methane infused in gut



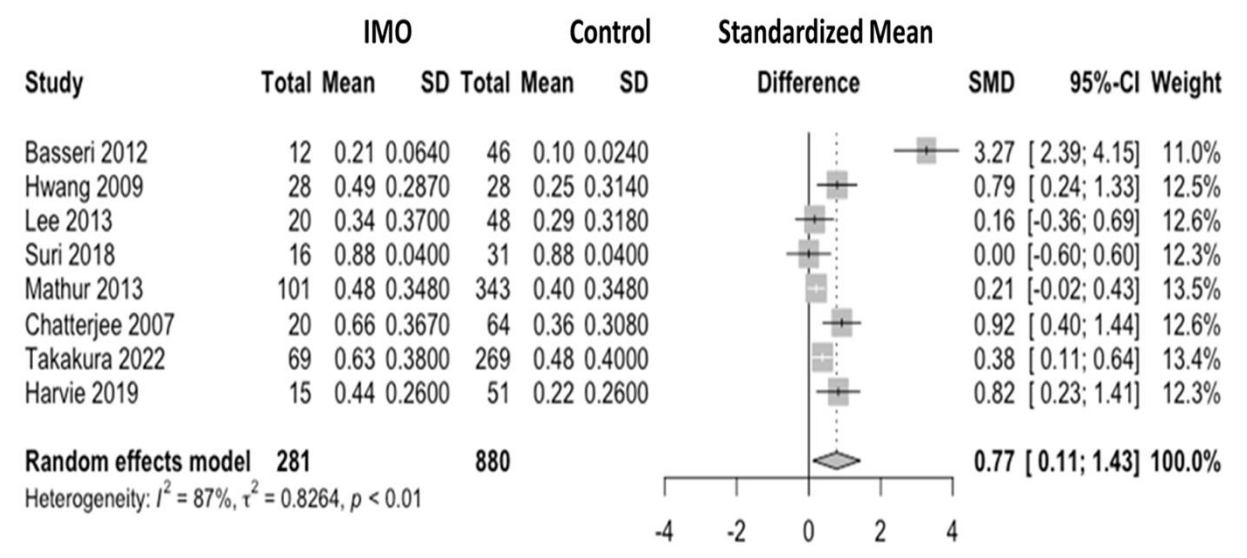
Peristaltic reflex intensity after brush stroke



Pimentel, et al. Am J Phsyiol 2006

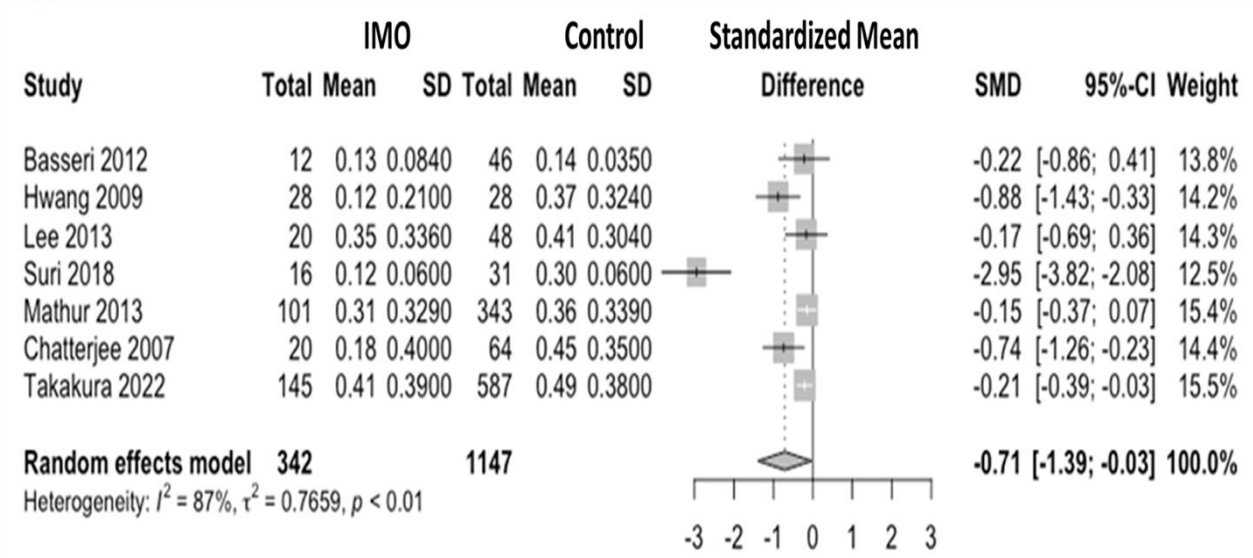
Intestinal Methanogen Overgrowth (IMO) Meta-analysis of CH_4 and Symptoms

A) Constipation (SMD of Severity)



MORE CONSTIPATION WITH CH_4

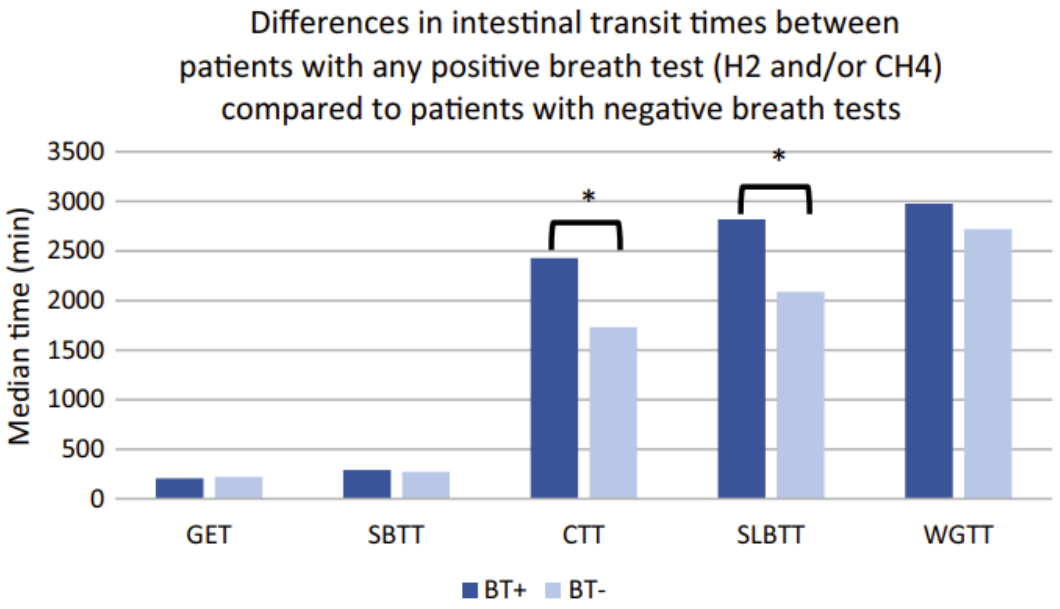
B) Diarrhea (SMD of Severity)

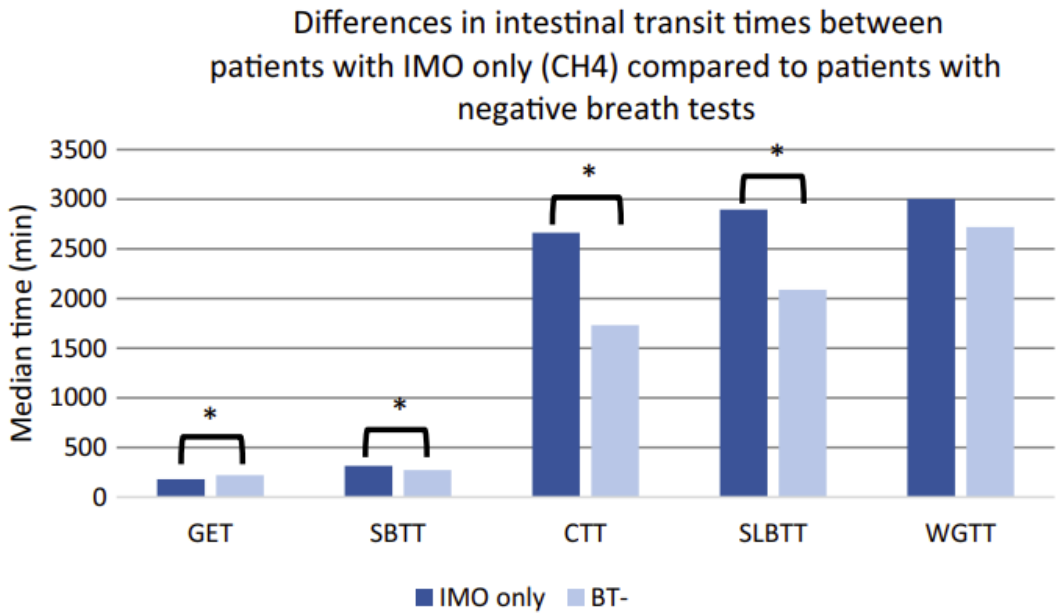


LESS DIARRHEA WITH CH_4

Mehravar, et al. Clin Gastroenterol Hepatol 2024

IMO and Small bowel/Colonic transit



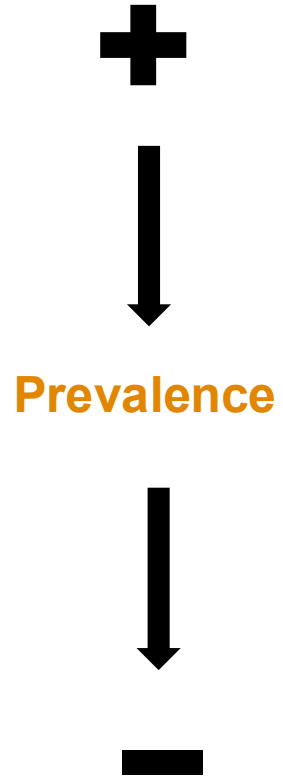


Wireless capsule in SIBO/IMO and non BT-ve groups

Talamantes, et al. Dig Dis Sci 2024

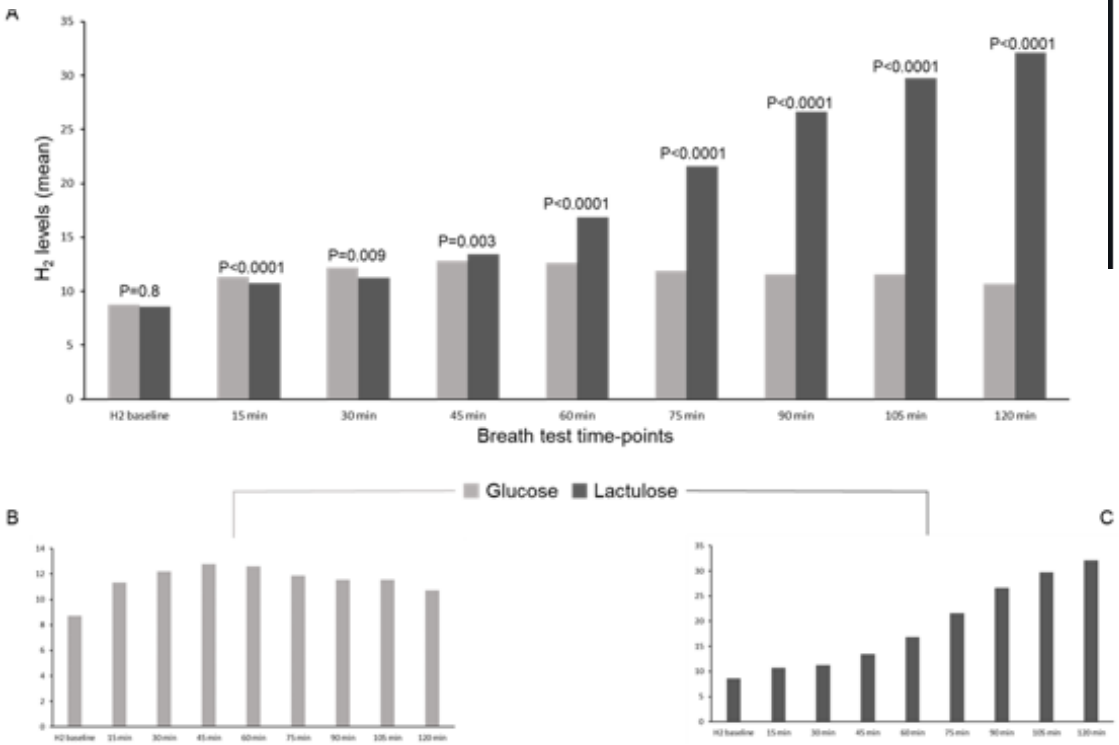
Duodenal H_2S producers were associated with H_2S on Breath

	Bacterial H_2S producers	Prevalence	R	P value
1	*Proteus mirabilis*	77.98%	0.313	0.001
2	*Desulfobulbus oralis*	71.6%	0.194	0.043
3	*Desulfovibrio desulfuricans*	64.2%	0.210	0.028
4	*Desulfuromonas sp.* DDH964	56.9%	0.242	0.011
5	*Desulfosarcina widdelii*	55%	0.243	0.011
6	*Desulfurispirillum indicum*	53.2%	0.229	0.017
7	*Desulfobulbus oligotrophicus*	51.4%	0.202	0.035
8	*Desulfoluna limicola*	41.3%	0.189	0.049



Villanueva-Milan, et al. DDW 2024

Glucose vs Lactulose

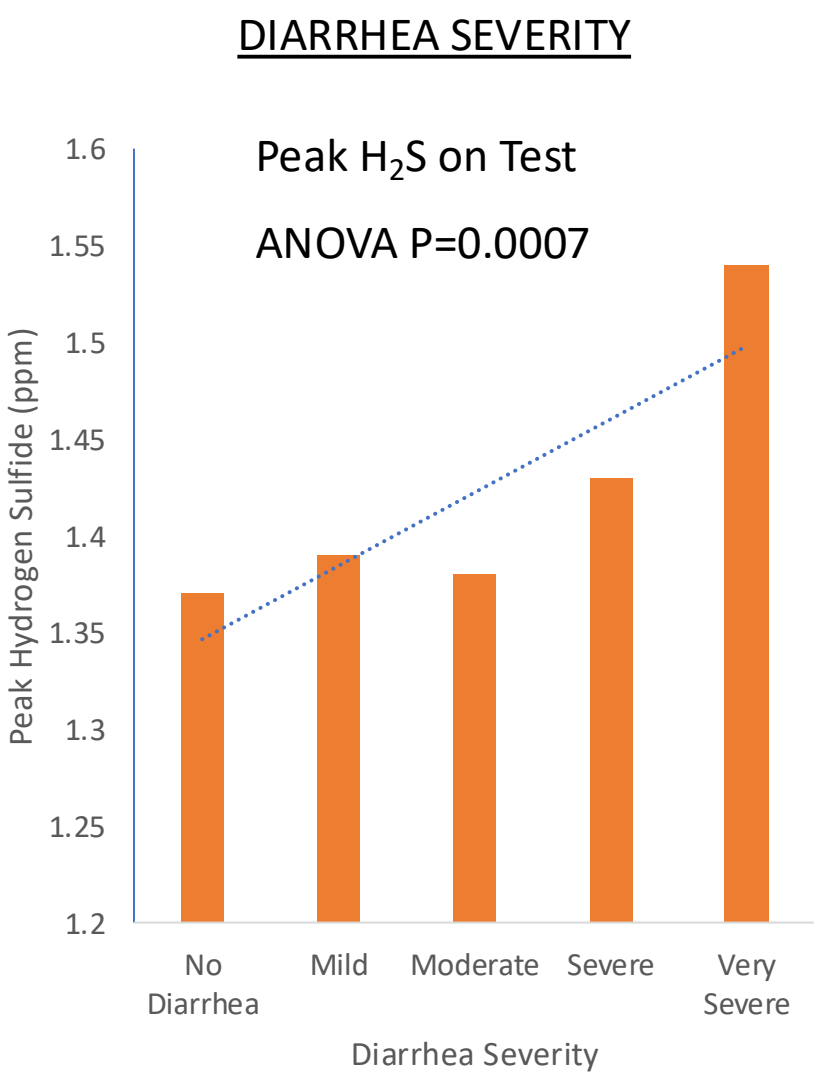


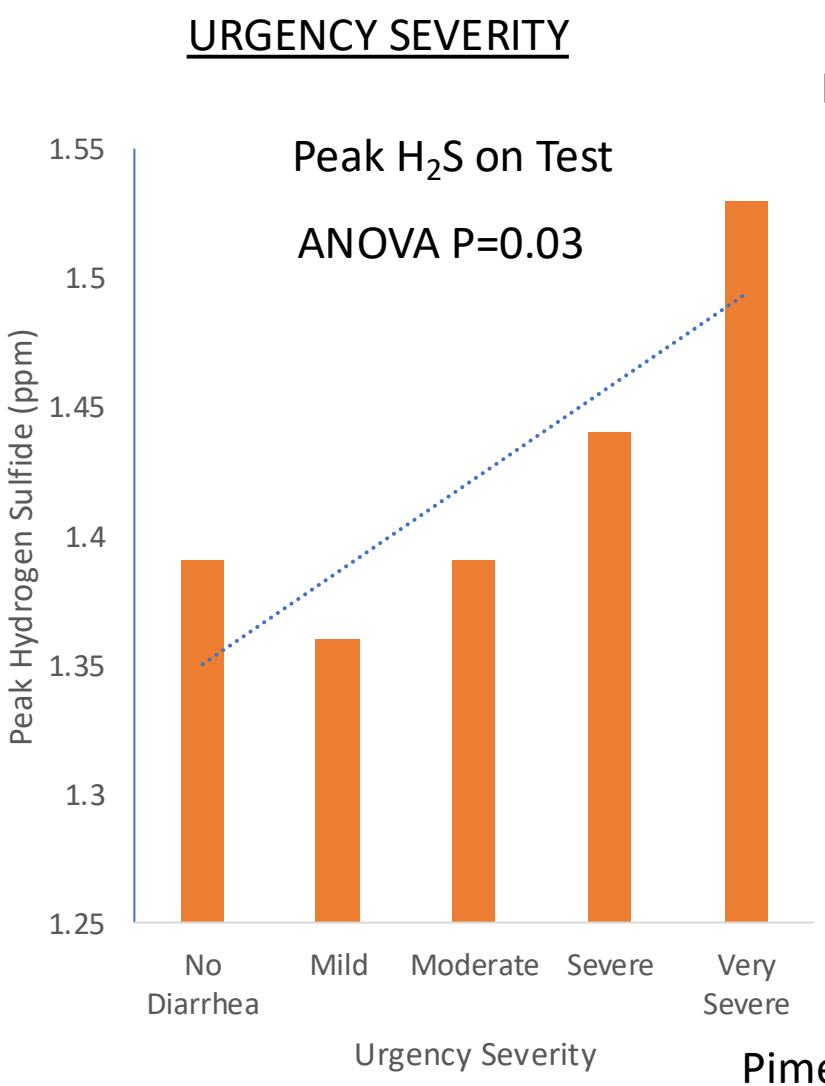
	Glucose	Lactulose	
	Count (%)	Count (%)	P-value*
No-SIBO/No-IMO/No-ISO2.0	707 (62.5%)	800 (42.7%)	
SIBO only	82 (7.3%)	505 (27%)	<0.0001
IMO only	222 (19.6%)	272 (14.5%)	0.44
ISO 2.0 only	60 (5.3%)	89 (4.8%)	0.12
SIBO/IMO (no ISO 2.0)	19 (1.7%)	100 (5.3%)	<0.0001
SIBO/ISO 2.0 (no IMO)	12 (1.1%)	53 (2.8%)	<0.0001
IMO/ISO 2.0 (no SIBO)	26 (2.3%)	40 (2.1%)	0.23
SIBO/IMO/ISO 2.0	3 (0.3%)	14 (0.7%)	0.02

1. Glucose misses people who might benefit
2. Glucose works well for H_2S and CH_4

Pimentel, et al. presented at DDW 2025 now in press

Real World Study – H_2S and Diarrhea/Urgency



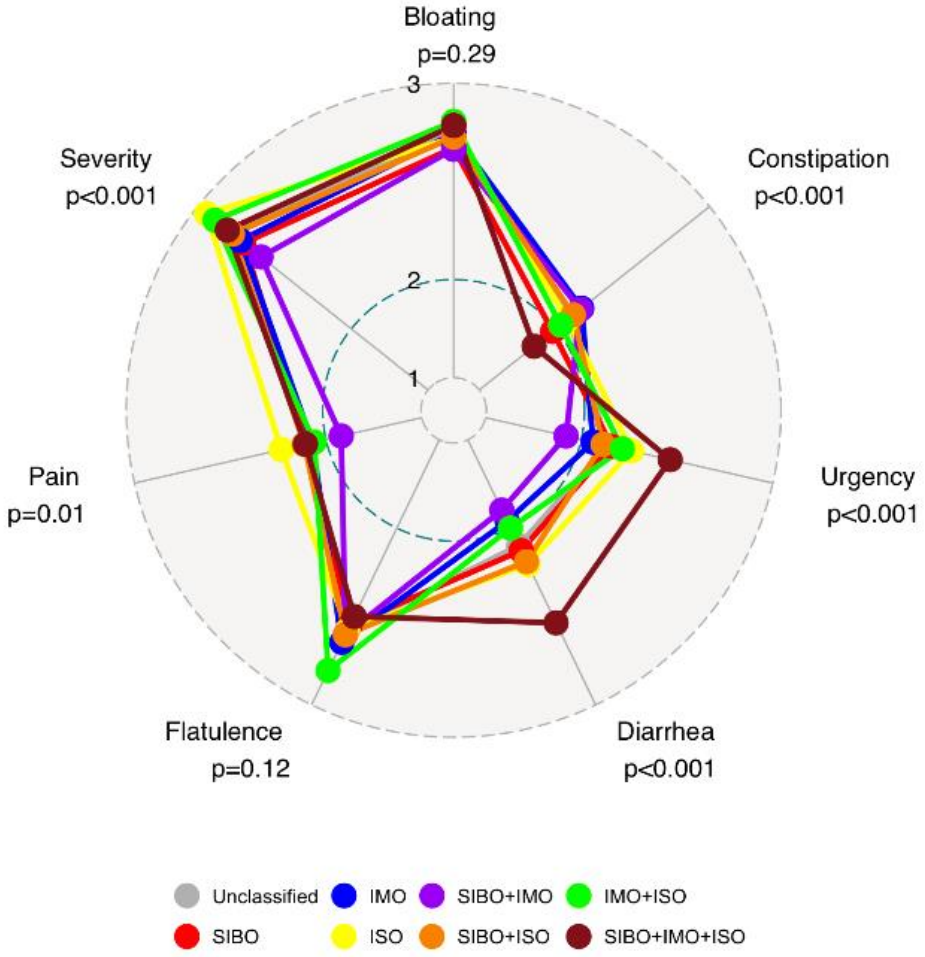


Range of H2S=0.44-4.55

Note:
No correlation with bloating or abdominal pain severity

Pimentel M, DDW 2025 in press

$H_2S \geq 2ppm$ presence or absence is the most important determinant of symptoms



Elevated H_2S

-Diarrhea dominates

-More severe diarrhea

-More severe pain

-More severe urgency

Normal H_2S Positive CH_4

-Constipation dominates

Only H_2 – milder diarrhea

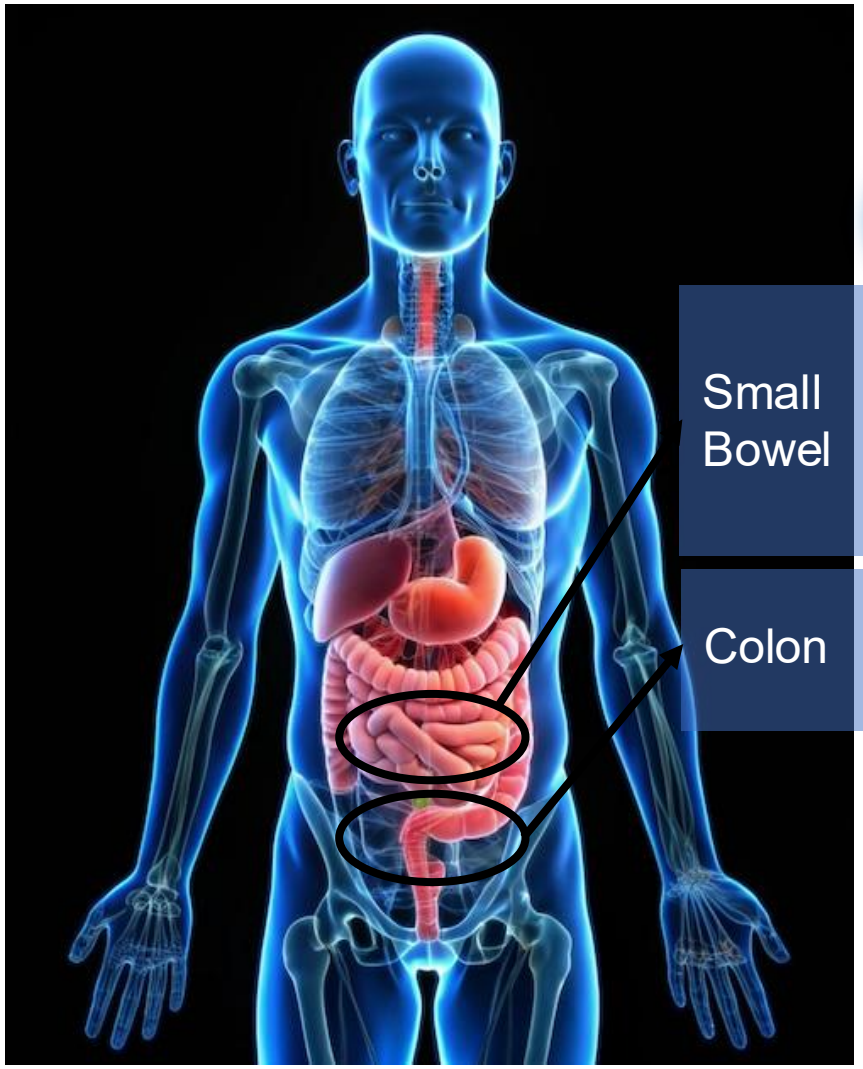
Pimentel, et al. DDW 2025 (in press)

What H_2S does to you….

Functional Change	Finding	Function/Impact
Mitochondrial	↑TXN2	Cells high in TXN2 are more resistant to redox changes
Redox	↓CTH	Reduction in the key enzyme responsible for endogenous H_2S production
	↓SLC7A11	Low levels would produce a pro-oxidative state in the cell
	↑SOD1	Encourages H_2O_2 production (oxidative) Interacts with aquaporin 9 to secrete H_2O_2 Counteracts H_2S
Gut Motor Function	↑HTR3e receptor	Serotonin receptor for sensation
	↓Piezo 2	Detects luminal distension and affects serotonin release and HTR3e function. Increases gut water content.
	↑Serotonin	Binds to HTR3e and other receptors controlling peristalsis and visceral sensation
Aquaporins	↓aquaporin 6	Ion transport
	↑aquaporin 7	Glycerol absorption
	↓aquaporin 9	Reduction in hydrogen peroxide absorption by cells
Defensins	↑DefensinB1	Antimicrobial peptide that is highly sensitive to redox. Highly active when reduced.
Neuroendocrine	↑SST	Expect an increase in somatostatin which affects gut motility

Germano, et al. DDW 2025

IBS DISEASE MICROTYPES



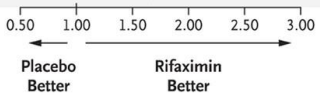
	ISO	SIBO	IMO
Small Bowel	***Proteus mirabilis*** *Desulfosarcina widdelii* *Desulfobulbus oligotrophicus*	***Escherichia coli*** ***Klebsiella pneumoniae*	***Methanobrevibacter smithii*** *Desulfurococcus mucosus* *Halobaculum rubrum* *Halarchaeum sp. CBA1220*
Colon	***Desulfovibrio*** ***Fusobacterium*		***Methabrevibacter smithii*** *Methanosphera stadtmanae* *Methanomasillococcus luminyensis*
	Severe diarrhea	Diarrhea	Constipation

Rifaximin Benefits in IBS

Primary Evaluation Period

A

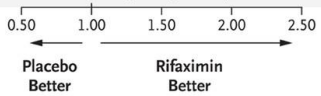
Efficacy Outcome	Proportion of Patients with a Response: Placebo (no./total no.)	Proportion of Patients with a Response: Rifaximin (no./total no.)	Odds Ratio (95% CI)	P Value for Treatment Effect
Primary end point				
Weekly global IBS symptoms				
TARGET 1	98/314	126/309	1.53	0.01
TARGET 2	103/320	128/315	1.45	0.03
Combined	201/634	254/624	1.49	<0.001
Key secondary end point				
Weekly IBS-related bloating				
TARGET 1	90/314	122/309	1.62	0.005
TARGET 2	102/320	129/315	1.49	0.02
Combined	192/634	251/624	1.56	<0.001
Secondary end points				
Daily global IBS symptoms				
TARGET 1	96/314	132/309	1.76	<0.001
TARGET 2	91/320	119/315	1.59	0.007
Combined	187/634	251/624	1.61	<0.001
Daily IBS-related bloating				
TARGET 1	102/314	121/309	1.41	0.05
TARGET 2	99/320	137/315	1.76	<0.001
Combined	201/634	258/624	1.52	<0.001
Daily IBS-related abdominal pain				
TARGET 1	114/314	137/309	1.45	0.03
TARGET 2	110/320	135/315	1.46	0.02
Combined	224/634	272/624	1.42	0.003
Daily abdominal pain and stool consistency				
TARGET 1	121/314	144/309	1.40	0.04
TARGET 2	116/320	147/315	1.55	0.008
Combined	237/634	291/624	1.47	<0.001
Daily abdominal pain				
TARGET 1	132/314	159/309	1.48	0.02
TARGET 2	138/320	165/315	1.46	0.02
Combined	270/634	324/624	1.46	<0.001
Daily stool consistency				
TARGET 1	212/314	244/309	1.80	0.002
TARGET 2	206/320	233/315	1.57	0.01
Combined	418/634	477/624	1.67	<0.001



Full 3 Months Post Treatment

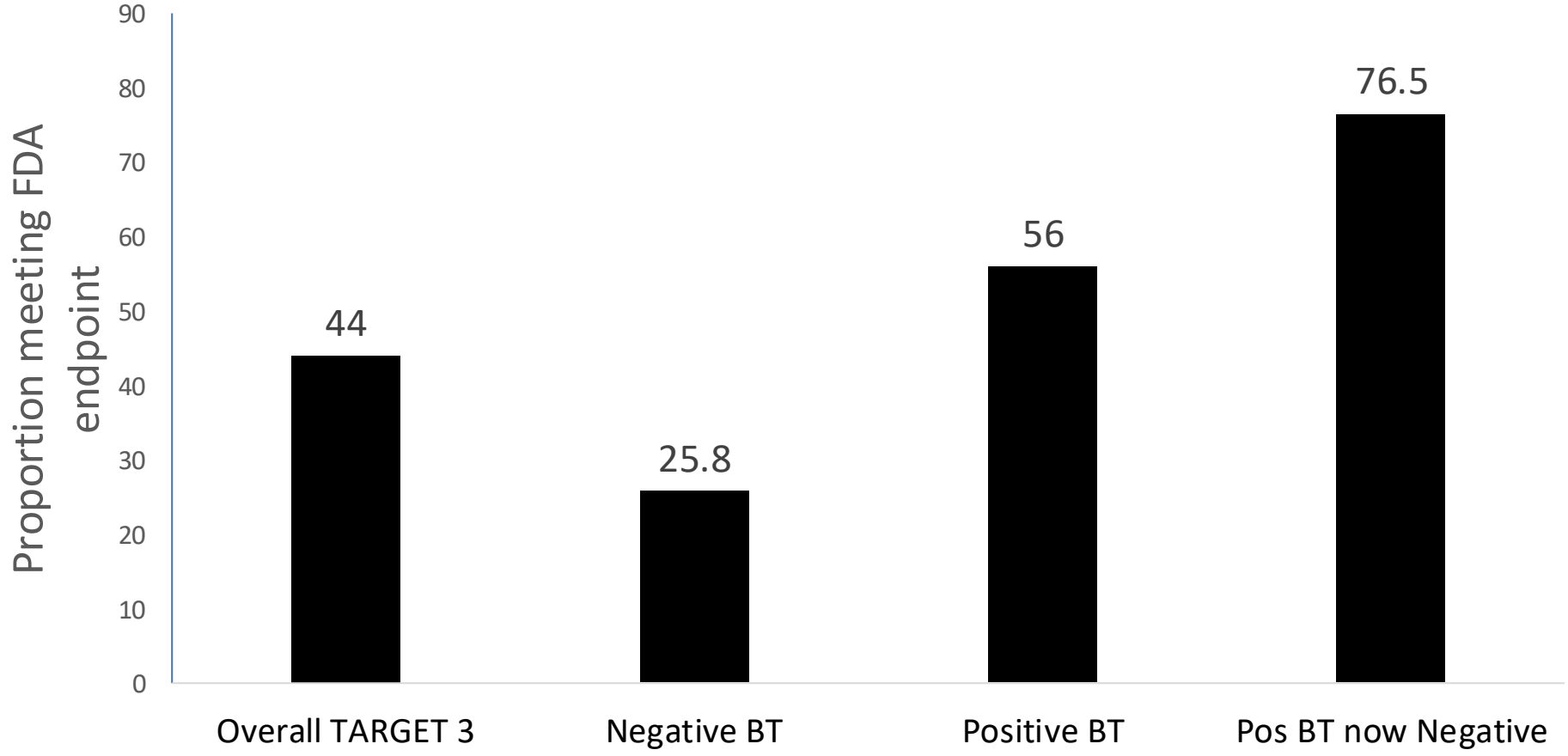
B

Efficacy Outcome	Odds Ratio (95% CI)	P Value for Treatment Effect
Primary end point		
Weekly global IBS symptoms		
TARGET 1	1.35 (1.00–1.82)	0.05
TARGET 2	1.52 (1.13–2.03)	0.005
Combined	1.44 (1.17–1.77)	<0.001
Key secondary end point		
Weekly IBS-related bloating		
TARGET 1	1.28 (0.95–1.73)	0.10
TARGET 2	1.56 (1.16–2.09)	0.003
Combined	1.42 (1.15–1.75)	0.001
Secondary end points		
Daily global IBS symptoms		
TARGET 1	1.60 (1.18–2.18)	0.003
TARGET 2	1.47 (1.09–1.99)	0.01
Combined	1.48 (1.20–1.83)	<0.001
Daily IBS-related bloating		
TARGET 1	1.50 (1.10–2.04)	0.01
TARGET 2	1.67 (1.24–2.25)	<0.001
Combined	1.53 (1.24–1.89)	<0.001
Daily IBS-related abdominal pain		
TARGET 1	1.35 (1.00–1.83)	0.05
TARGET 2	1.35 (1.01–1.81)	0.04
Combined	1.31 (1.06–1.61)	0.01
Daily abdominal pain and stool consistency		
TARGET 1	1.36 (1.01–1.83)	0.04
TARGET 2	1.44 (1.08–1.92)	0.01
Combined	1.40 (1.14–1.72)	0.001
Daily abdominal pain		
TARGET 1	1.31 (0.98–1.75)	0.07
TARGET 2	1.37 (1.03–1.83)	0.03
Combined	1.33 (1.09–1.64)	0.006
Daily stool consistency		
TARGET 1	1.70 (1.24–2.33)	<0.001
TARGET 2	1.48 (1.09–2.00)	0.01
Combined	1.58 (1.27–1.97)	<0.001



Pimentel, et al. N Engl J Med 2011

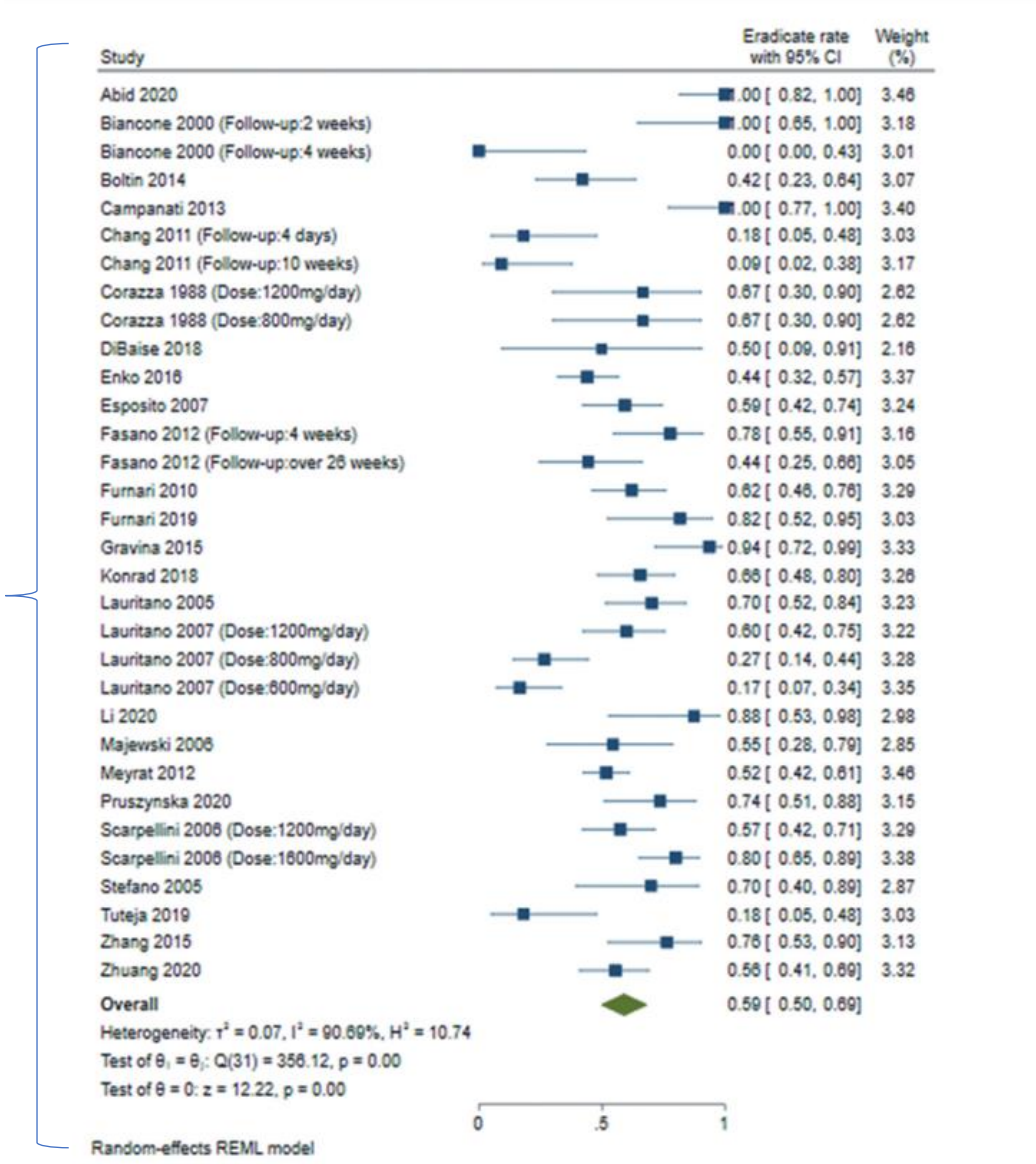
Breath Testing Predicts Rifaximin Response



Rezaie, et al., Am J Gastroenterol 2019

Meta-analysis of Rifaximin for SIBO

32 Studies



Wang, et al. Expert Rev Gastroenterol Hepatol 2021

SIBO Predicts Response to Antibiotics in IBS

Author(s) and year	SIBO Event	SIBO Nonevent	Control Event	Control Nonevent	Weight (%)	Relative ratio [95% CI]
Pimentel, 2003	25	21	6	3	16.35%	1.37 [0.52, 3.64]
Ghoshal, 2016	7	12	18	3	12.82%	4.42 [1.47, 13.31]
Rezaie, 2019	25	37	23	8	39.14%	2.31 [1.23, 4.35]
Zhuang, 2020	27	18	25	8	31.69%	1.65 [0.82, 3.33]
RE model for all studies (Q = 3.03, df = 3, P = 0.390; I^2 = 0.01%, τ^2 = 0.00)					100.00%	2.07 [1.40, 3.08]

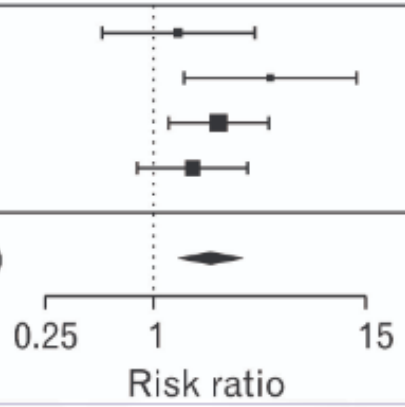


Fig. 3. Forrest plot of studies comparing the efficacy of antibiotics in irritable bowel syndrome (IBS) patients with or without small intestinal bacterial overgrowth (SIBO). The pooled response rate was 51.2% vs 23.4% in the SIBO and no SIBO group, respectively. Relative risk (95% CI) for improvement was 2.07 (1.40-3.08), P = 0.0003. Heterogeneity was I^2 = 0.01%, P = 0.300.

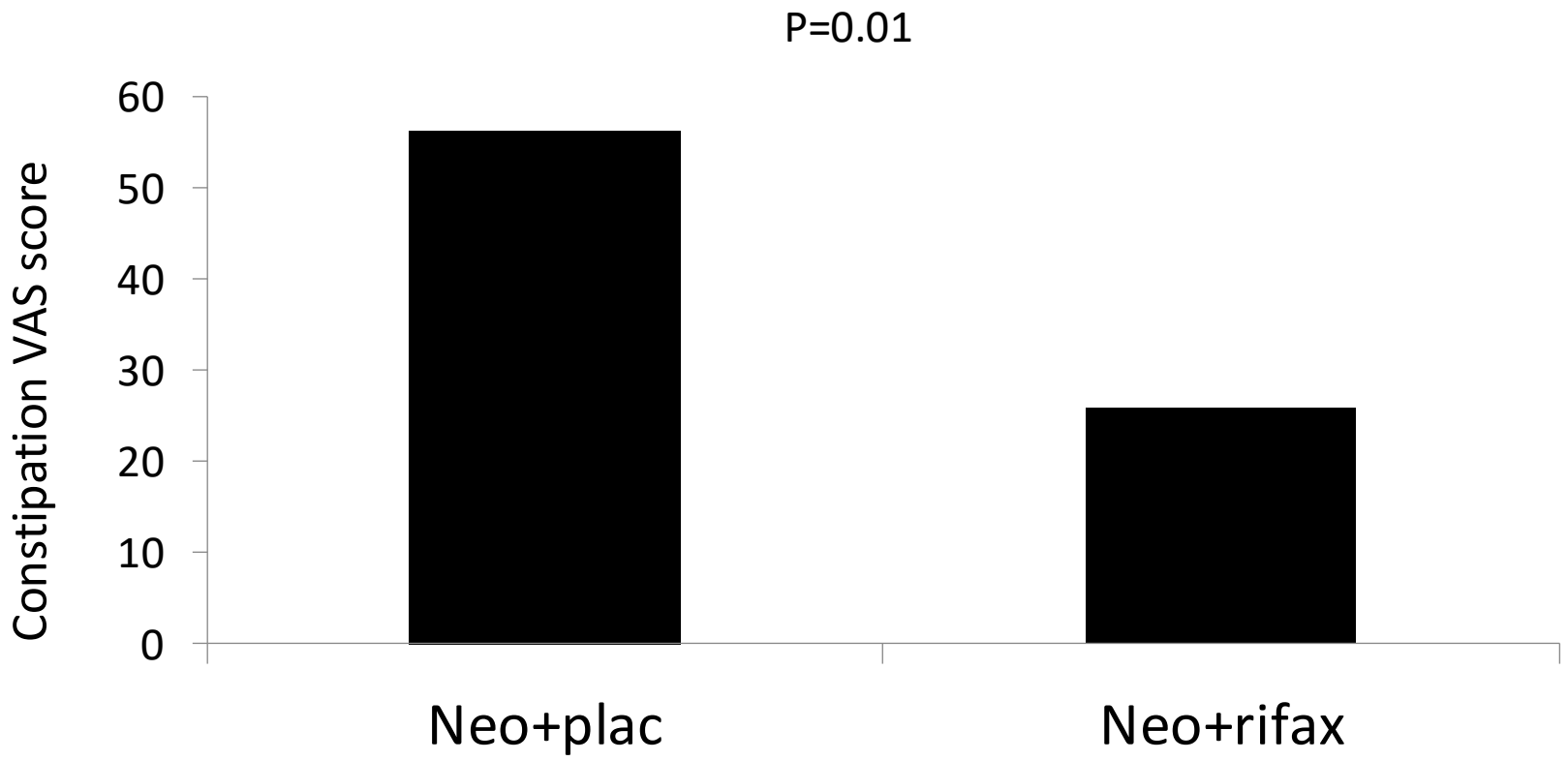
Takakura, et al. J Neurogastroenterol Motil 2024

Harm among IBS Therapies

Therapeutic Agent	NNH	NNT	No. of Subjects Benefiting from Treatment for 1 Patient Harmed
Tricyclic antidepressants *	18.3	8	2.3
Alosetron	19.4	7.5	2.6
Rifaximin	8971	10.6	846

NEW! – Update on this analysis with new drugs expected at DDW 2025. Shah, et al. Am J Med 2012

Methane Positive C-IBS
Double Blind Placebo Controlled Trial



Pimentel, et al. Dig Dis Sci, 2014.

Bismuth Effective for Hydrogen Sulfide

GASTROENTEROLOGY 1998;114:923–929

Bismuth Subsalicylate Markedly Decreases Hydrogen Sulfide Release in the Human Colon

FABRIZIS L. SUAREZ, JULIE K. FURNE, JOHN SPRINGFIELD, and MICHAEL D. LEVITT
Research Department, Minneapolis Veterans Affairs Medical Center, Minneapolis, Minnesota

Background & Aims: Hydrogen sulfide is one of the main malodorous compounds in human flatus. This toxic gas also has been implicated in the pathogenesis of ulcerative colitis. Therefore, a treatment that reduces colonic H_2S levels could be clinically useful in the treatment of flatus odor and of ulcerative colitis. In this study the ability of bismuth subsalicylate, a compound that binds H_2S, to reduce H_2S release in the colon, was tested. *Methods:* Homogenates made from human and rat feces were incubated with and without bismuth subsalicylate, and gas production was measured. Fecal samples from 10 healthy subjects were analyzed before and after ingestion of bismuth subsalicylate (524 mg four times a day) for 3–7 days. *Results:* Fecal homogenates showed a dose-dependent relationship between the concentration of bismuth subsalicylate and H_2S release. Treatment of subjects with bismuth subsalicylate produced a >95% reduction in fecal H_2S release. *Conclusions:* The ability of bismuth subsalicylate to dramatically reduce H_2S could provide a clinically useful means of controlling fecal and/or flatus odor and of decreasing the putative injurious effects of H_2S on the colonic mucosa.

containing compounds to the colon could also limit H_2S production. However, a variety of endogenous compounds (e.g., mucin and taurocholic acid) as well as dietary substances (e.g., amino acids and sulfate) serve as sources of sulfur for the colonic bacteria,[7] and it seems unlikely that delivery of sulfur to the colon can be drastically reduced.

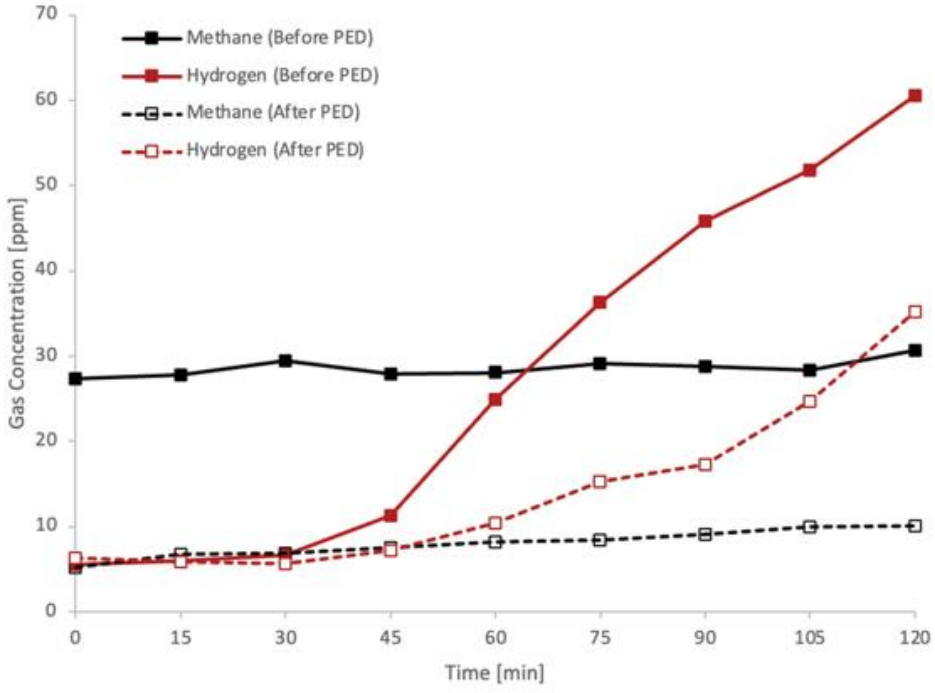
A simple approach to reducing H_2S release in the colon would be the administration of a compound that binds H_2S. In the test tube, bismuth reacts with sulfide to form very insoluble bismuth sulfide. In the present report, we carried out a series of experiments in rats and humans to determine whether bismuth subsalicylate (BSS) decreases the fecal release of H_2S.

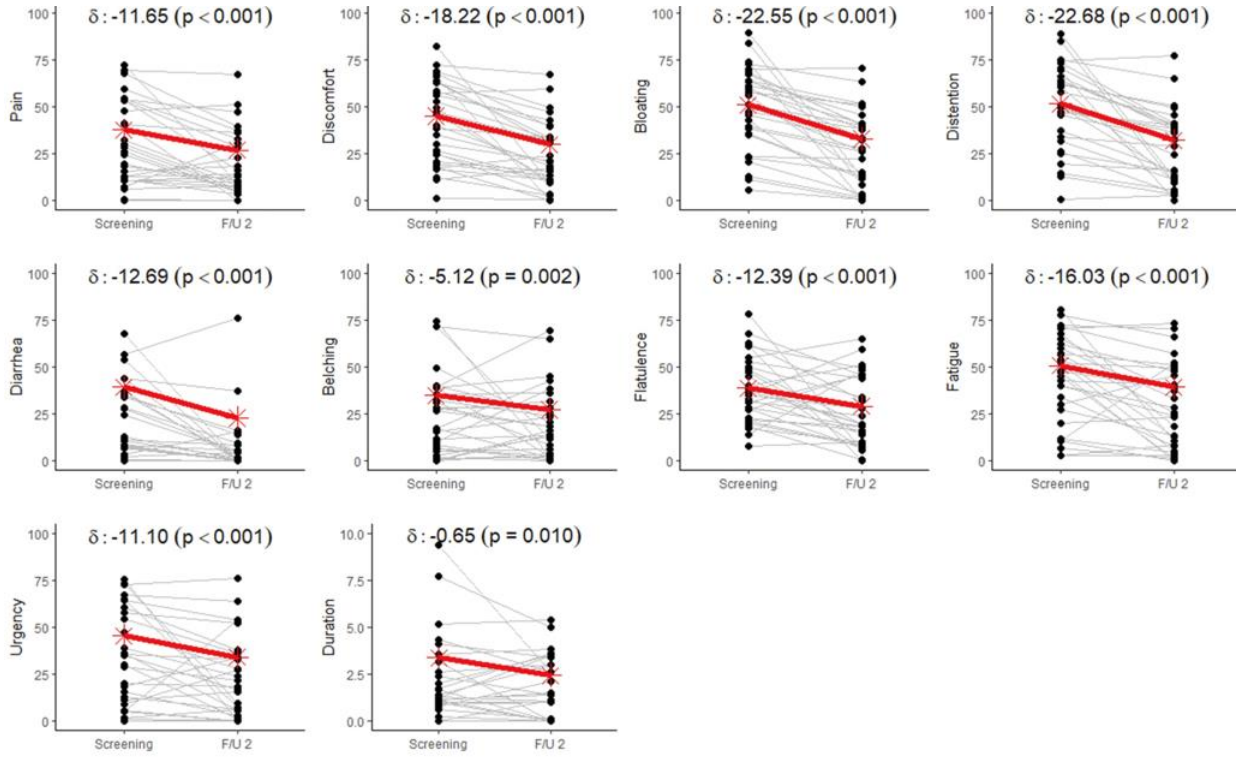
Materials and Methods

In Vitro Studies

The ability of BSS to bind the three major sulfur gases present in human flatus[8] was studied by incubating 1 mL of a 1:10 dilution of BSS (4.84 μmol of bismuth) in the form of Pepto-Bismol (Procter & Gamble, Cincinnati, OH) in buffer (0.1 mol/L sodium phosphate, pH 7.0) with 20 mL of N_2

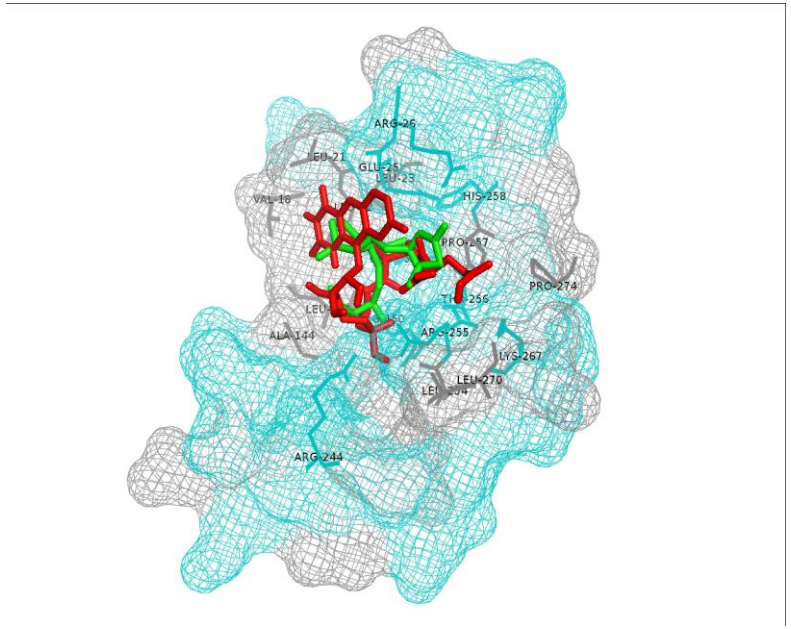
Elemental Diet (mBiota™)



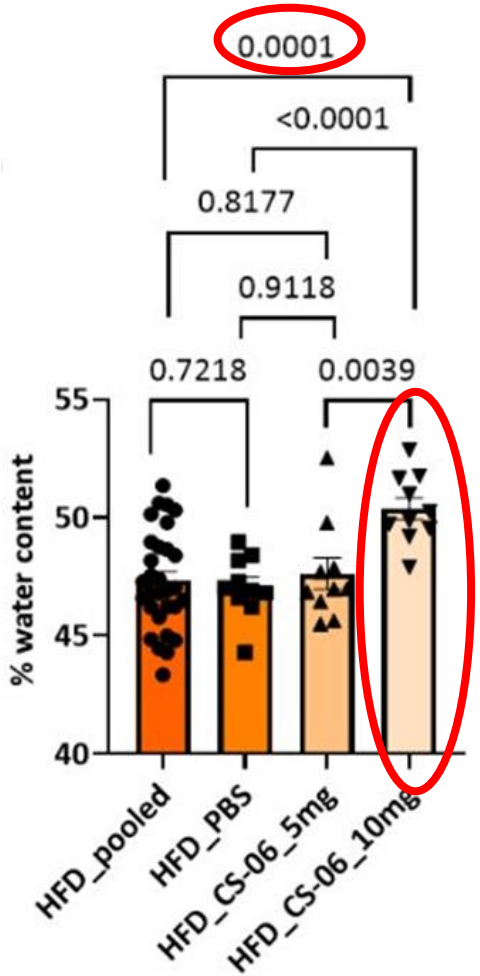


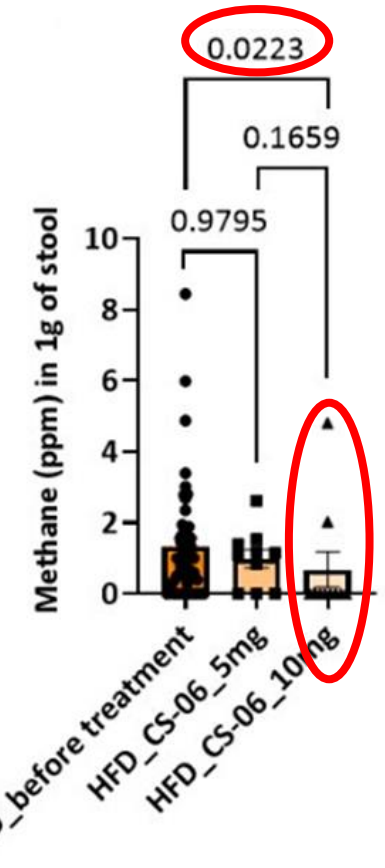
Rezaie, et al. CGH 2025

CS-06 Reduces Methane and Constipation *in vivo*



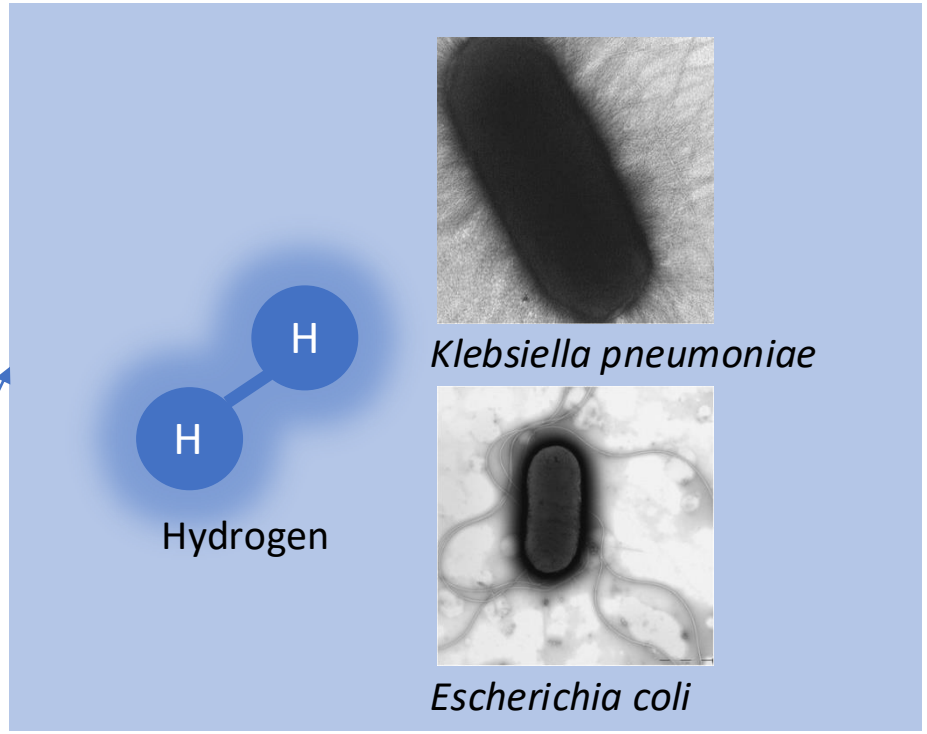
Blocks the binding site for the methylenetetrahydromethanopterin (mtd) enzyme





Villanueva-Millan, et al DDW 2025 Su2010

IBS-D



Many of the main taxa associated with IBS-D partially reside in the mucous of small intestine

Table 1 Differential abundance of the top six phyla in DA-DTT versus DA-U

Taxonomy	DA-DTT (n = 43) versus DA-U (*n* = 112)				
	Average Relative abundance % DA-DTT[a]	Average Relative abundance % DA-U[a]	Fold Change (calculated from the GLM)[b]	*P*-value	FDR *P*-value
Firmicutes	49.3	62.25	1.05	0.65	0.70
Proteobacteria	28.97	14.8	6.22	**1.4E-7**	**7.71E-7**
Actinobacteria	8.91	12.02	−1.23	0.21	0.42
Fusobacteria	5.36	3.93	1.96	**0.01**	**0.03**
Bacteroidetes	6.16	4.63	2.19	**0.01**	**0.03**
TM7	1.17	1.86	−1.34	0.32	0.48

P-value< 0.05 and FDR *P*-value< 0.05 are shown in bold. [a]The relative abundances were calculated from the original counts (number of sequences in the OTU table). [b]Fold changes were calculated from the GLM, which corrects for differences in library size between the samples and the effects of confounding factors. It is therefore not possible to derive these fold changes from the original counts (number of sequences in the OTU table) by simple algebraic calculations

Fernandez-Delgado, et al. Ann Microbiol 2015
Struv, et al. Infect Immunity 2008
Jorgensen, et al. Stand Genom Sci 2018
Leite, et al. BMC Microbiol 2019
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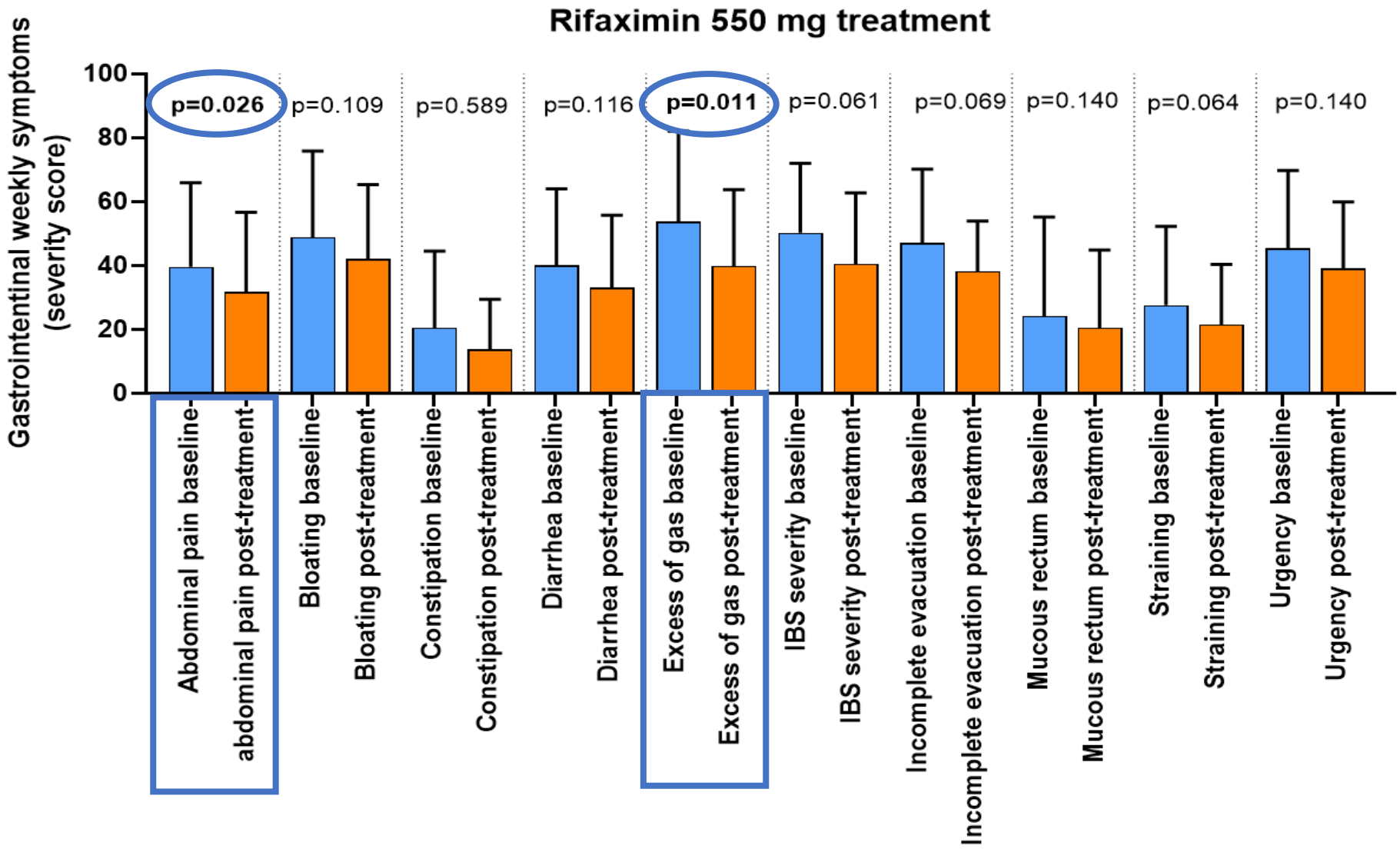
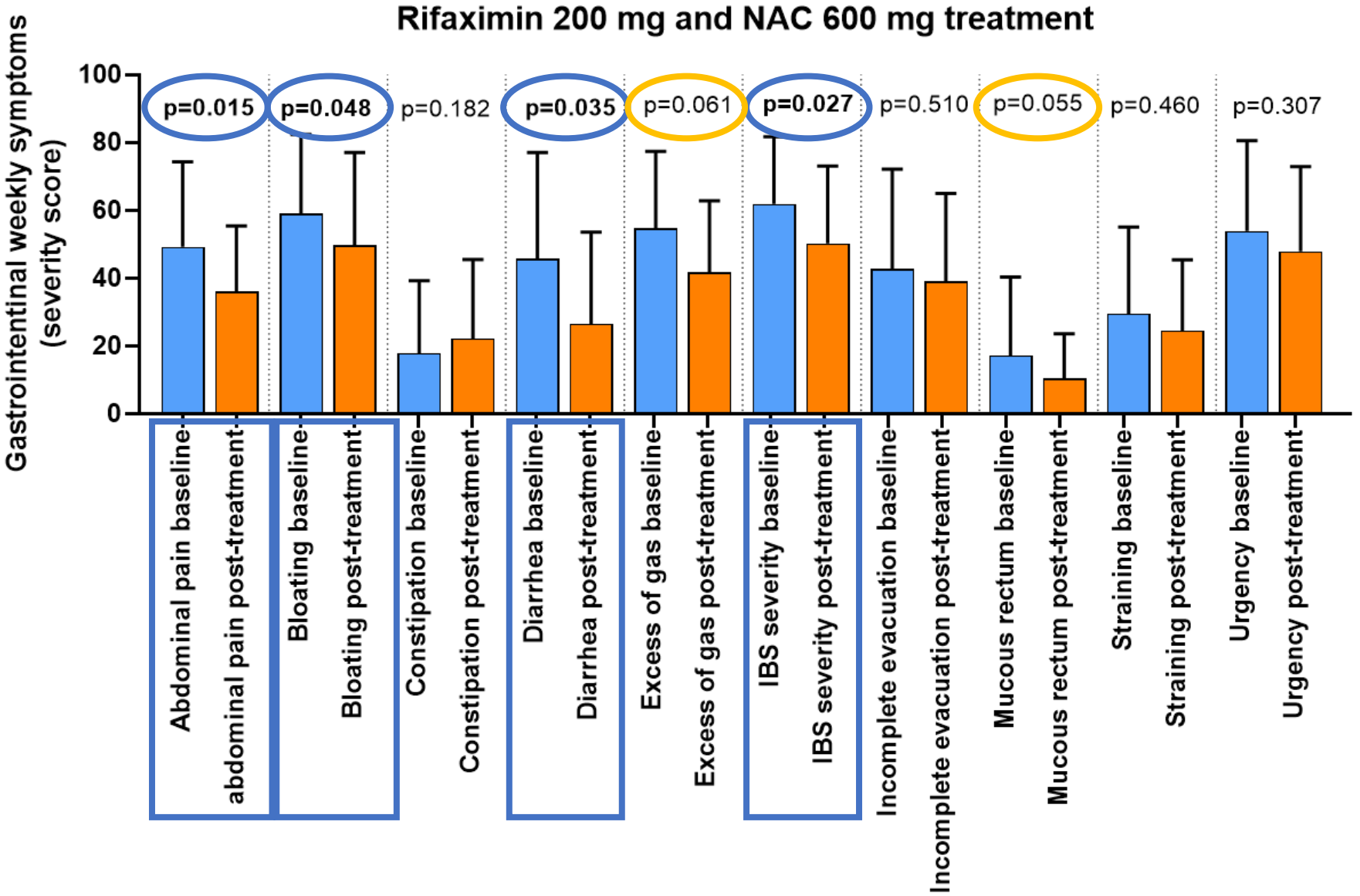
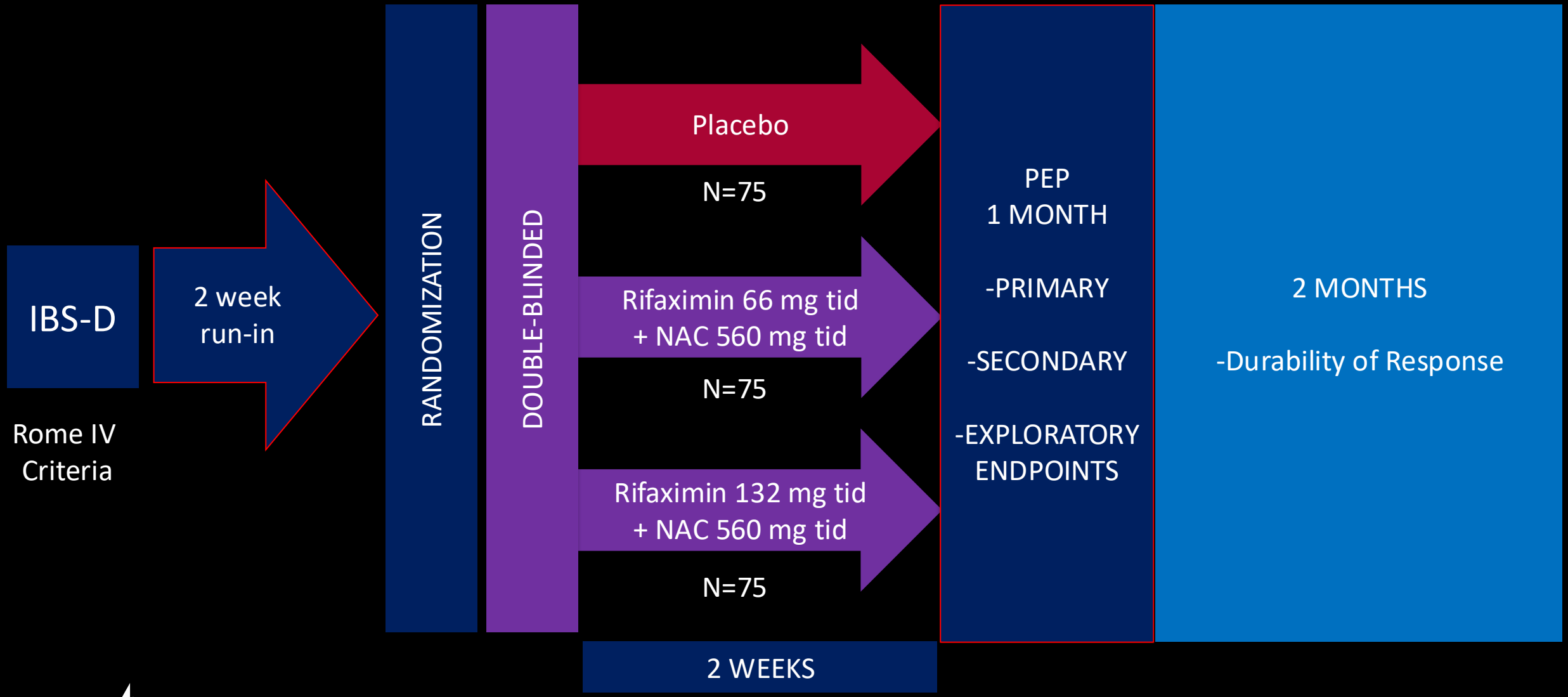
Within group changes (Rifaximin 550mg)



Within group changes (Rifaximin + NAC)



Phase IIB Registered Trial has begun



MAST
MEDICALLY ASSOCIATED SCIENCE AND TECHNOLOGY

NAC = N-acetylcysteine

Indian Journal of Gastroenterology (March–April 2023) 42(2):249–273
https://doi.org/10.1007/s12664-022-01333-5

CONSENSUS

Indian consensus statements on irritable bowel syndrome in adults: A guideline by the Indian Neurogastroenterology and Motility Association and jointly supported by the Indian Society of Gastroenterology

Uday C. Ghoshal[1] · Sanjeev Sachdeva[2] · Nitesh Pratap[3] · Arun Karyampudi[4] · Uzma Mustafa[1] · Philip Abraham[5] · Chetan B. Bhatt[6] · Karmabir Chakravartty[7] · Sujit Chaudhuri[8] · Omesh Goyal[9] · Govind K. Makharia[10] · Manas Kumar Panigrahi[11] · Prasanta Kumar Parida[12] · Sudhanshu Patwari[13] · Rajesh Sainani[14] · Shine Sadasivan[15] · M. Srinivas[16] · Rajesh Upadhyay[17] · Jayanthi Venkataraman[18]

Received: 6 December 2022 / Accepted: 20 December 2022 / Published online: 24 March 2023

Statement 5: Patients with IBS, particularly those with diarrhea-predominant IBS (IBS-D), are more likely to have SIBO and gut dysbiosis

Voting summary: Accepted completely: (72.22%), accepted with minor reservation: (27.78%).
Level of evidence: II-1
Grade of recommendation: A

Statement 6: Excessive methane production slows gut transit and is associated with constipation-predominant IBS (IBS-C)

Voting summary: Accepted completely: (77.78%), accepted with minor reservation: (22.22%).
Level of evidence: I
Grade of recommendation: B

Statement 7: Gastrointestinal infection with varied pathogens may result in post-infection IBS (PI-IBS)

Voting summary: Accepted completely: (100%).
Level of evidence: II-1
Grade of recommendation: A

Digestive Diseases and Sciences
https://doi.org/10.1007/s10620-025-09170-0

INVITED COMMENTARY

Biomarkers in Patients with IBS: Continue the Grail Quest or Give It a Rest?

Mauricio Jin[1] · **Xiao Jing Wang**[2]

Received: 20 May 2025 / Accepted: 5 June 2025

Keywords Enterochromaffin cells · Post-infectious IBS · CdtB · Vinculin · Rome IV criteria · Chromogranin a cell density

Irritable bowel syndrome (IBS) is currently diagnosed based on symptoms and limited diagnostic testing, highlighting the need for noninvasive biomarkers and personalized therapies. Emerging blood-based biomarkers such as cytolethal distending toxin B and antivinculin antibodies, DNA methylation profiles, and intestinal permeability measures, along with stool-based microbiome and metabolite markers, show promise in distinguishing IBS, and its bowel habit subtypes, from other gastrointestinal disorders with overlapping symptoms. Additionally, stool-based biomarkers and breath tests may also predict response to interventions, like the low fermentable oligosaccharides, disaccharides, monosaccharides, and polyols diet, and rifaximin, supporting more personalized treatment strategies.

Review

Mechanisms and pathophysiology leading to development of small intestinal microbial dysbiosis

John A Damianos, Xiao Jing Wang, Michael Camilleri

Small intestinal microbial dysbiosis (SIMD) describes clinical syndromes attributed to overabundance of microbes within the small intestine. Usually attributed to bacteria (but also less commonly to archaea and fungi), SIMD arises as a secondary condition, when there is disruption of one or more key physiological mechanisms. We provide a comprehensive review of the current understanding of SIMD pathophysiology. We highlight the key physiological factors including gastric acid, pancreatic enzymes, bile acids, small intestinal dysmotility and obstruction, colonic motility, and intestinal mucosal immunity. We then describe the disease states associated with failure of these mechanisms, providing SIMD prevalence estimates for each. Finally, we address some of the controversies regarding SIMD including testing modalities, significance of intestinal methanogen overgrowth, and the role of SIMD in irritable bowel syndrome.

Lancet Gastroenterol Hepatol 2026

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January 14, 2026
https://doi.org/10.1016/S2468-1253(25)00295-X

Division of Gastroenterology and Hepatology, Mayo Clinic, Rochester, MN, USA
(J A Damianos MD, X J Wang MD, M Camilleri MD DSc)

Treatments that I Use (present and future)

Gas Type	Current Treatments	Future Treatments
Intestinal Sulfide Overproduction (ISO)	Rifaximin + Pepto Bismol	
	mBiota elemental	
		RNIB21
Small intestinal bacterial overgrowth (SIBO)	Rifaximin	
	mBiota	
		RNIB21
		Novel biologic agent
Intestinal methanogen overgrowth (IMO)	Rifaximin + Neomycin	
	Rifaximin + Metronidazole	
	mBiota	
		CS-06

Conclusions

1. IBS is a disease
2. Food poisoning causes a significant proportion of IBS
3. Blood testing can diagnose and risk-stratify IBS-D
4. Three gas breath testing is the most validated breath test in history
5. H_2S may be the most important gas in how it affects you
6. New therapies are coming soon for SIBO, ISO, and IMO