

Supplemental Table 1. Demographics, Bowel Diary, Fasting Serum C4 and Fecal BA Measured from Random, Single and 48-Hour Stool Samples

	Healthy	IBS-D without BAD	IBS-D with BAD	Terminal ileum resection	Kruskal-Wallis p
N	25	34	25	4	
Age (years)	39.5 ± 10.7	41.8 ± 10.7	46 ± 12.9	50.75 ± 22.6	0.2
Sex (F/M)	18/7	26/8	19/6	0/4	0.01
BMI (kg/m ²)	24.8 ± 3.1 ^{^#}	30.6 ± 8.0 [#]	33.5 ± 7.0 [^]	28 ± 6.8	0.0001
Stool frequency (BM/day)	1.3 ± 0.7 ^{*^#}	2.2 ± 1.1 ^{*^#}	3.3 ± 2.0 [^]	5.4 ± 1.7	<0.001
Stool consistency (BSFS)	4.1 ± 0.5 ^{*^}	4.4 ± 1.1 ^{*^}	5.5 ± 0.8 [^]	6.2 ± 0.2	<0.001
C4 (ng/mL)	13.6 ± 9.0 [^]	19.8 ± 14.2 ^{^#}	50.5 ± 31.5 ^{^#}		<0.001
Single stool					
Primary BA (%)	2.5 ± 4.2 [*]	2.2 ± 4.1 ^{*^}	19.5 ± 27.3 ^{*^}	73.4 ± 18.2	0.002
Primary BA (µmol/L)	2.8 ± 8.4 ^{*^}	2.9 ± 8.4 ^{*^}	45.1 ± 85.2 ^{*^}	345.2 ± 188.5	<0.001
CDCA (%)	1.0 ± 1.1 [*]	1.2 ± 1.8 ^{*^}	8.6 ± 12.3 ^{*^}	24.0 ± 3.2	0.002
CDCA (µmol/L)	0.94 ± 1.9 ^{*^}	1.45 ± 3.3 ^{*^}	19.25 ± 36.3 ^{*^}	129.5 ± 98.4	0.0003
CA (%)	1.4 ± 3.2 [*]	1.0 ± 2.4 ^{*^}	10.9 ± 15.6 ^{*^}	49.4 ± 18.1	0.0007
CA (µmol/L)	1.8 ± 6.6 ^{*^}	1.5 ± 5.3 ^{*^}	25.6 ± 49.7 ^{*^}	215.7 ± 96.4	0.0002
DCA (%)	50.4 ± 13.4 [*]	55.5 ± 12.1 [*]	47.2 ± 16.9 [*]	16.1 ± 16.3	<0.01
DCA (µmol/L)	37.7 ± 35.7	61.5 ± 53.8	67.8 ± 47.6	114.9 ± 142.5	0.14
LCA (%)	46.4 ± 15.6 ^{*^}	41.3 ± 13.4 ^{*^}	30.9 ± 15.9 ^{*^}	3.8 ± 4.3	<0.001
LCA (µmol/L)	25.0 ± 14.5	37.5 ± 24.7	37.3 ± 22.8	30.0 ± 43.9	0.1
Total fecal BA (µmol)	212 ± 249 [^]	260 ± 290.4	534.2 ± 561.8 [^]	973.8 ± 1201	0.04
Stool weight (g)	106.8 ± 99.8	88.1 ± 67.7	127 ± 100.9	97 ± 82.8	0.26
48 hour stool sample					
Primary BA (%)	2.94 ± 5.4 ^{*^}	2.1 ± 2.6 ^{*^}	27.2 ± 21.3 ^{*^}	82.4 ± 15.3	<0.0001
Primary BA (µmol/L)	1.6 ± 2.9 ^{*^}	2.2 ± 3.0 ^{*^}	56.9 ± 69.8 ^{*^}	258.5 ± 86.4	<0.0001
CDCA (%)	1.7 ± 3.2 ^{*^}	1.2 ± 1.4 ^{*^}	10.9 ± 7.1 ^{*^}	31.4 ± 4.6	<0.0001
CDCA (µmol/L)	0.92 ± 1.8 ^{*^}	1.23 ± 1.7 ^{*^}	22.9 ± 26.4 ^{*^}	100.3 ± 41	<0.0001
CA (%)	1.23 ± 2.3 ^{*^}	0.9 ± 1.3 ^{*^}	16.4 ± 14.5 ^{*^}	51 ± 11.9	<0.0001
CA (µmol/L)	0.67 ± 1.2 ^{*^}	0.9 ± 1.4 ^{*^}	34.0 ± 44.1 ^{*^}	158.2 ± 48	<0.0001
DCA (%)	56.6 ± 11.9 ^{*^}	56.9 ± 9.4 ^{*^}	47.2 ± 14.9 ^{*^}	9.5 ± 13.3	0.0013
DCA (µmol/L)	39.1 ± 28.7 ^{^#}	61.6 ± 46.8 ^{^#}	97.2 ± 71.7 [^]	47.5 ± 75.5	0.006

LCA (%)	39.5 ± 12.5*^	40.0 ± 10.1*^	23.3 ± 10.9*^	2.7 ± 4.5	<0.0001
LCA (μmol/L)	24.7 ± 17^#	38.9 ± 24.4#	41.6 ± 34.2^	14.3 ± 24.5	0.03
Total fecal BA (μmol)	585.8 ± 465*^	718 ± 598.2*^	1891.6 ± 848^	7204 ± 8500	<0.0001
Stool weight (g/48h)	318.9 ± 165^	287.3 ± 123^	527.3 ± 257^	993 ± 950	0.0001

* represents significant difference (p<0.05) to terminal ileum resection patients

^ represents significant difference (p< 0.05) to IBS-D with BAD; # represents significant difference (p< 0.05) to IBS-D without BAD

BA=bile acid; BAD=bile acid diarrhea; BM=bowel movement; BSFS=Bristol Stool Form Scale; C4=7a-hydroxy-4-cholesten-3-one;

CA=cholic acid; CDCA=chenodeoxycholic acid; DCA=deoxycholic acid; IBS-D=diarrhea-predominant irritable bowel syndrome;

LCA=lithocholic acid; ⁷⁵SeHCAT=⁷⁵selenium homocholeic acid taurine

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Supplemental Table 2. BAD diagnostic accuracy of fasting serum C4 >52.5ng/mL and various cut-offs for single stool primary BA (%)

All participants with measurements of serum C4 and single stool primary BA % using 10% cut-off			
Fasting serum C4 and primary BA % in single stool sample	Diagnosis based on 48-hour stool sample		Predictive value
	BAD (+)	BAD (-)	
C4 >52.5 ng/mL or primary BA >10%	19	3	PPV = 86%
C4 <52.5 ng/mL and primary BA <10%	10	56	NPV = 85%
	Sensitivity = 66%	Specificity = 95%	
All participants with measurements of serum C4 and single stool primary BA % using 15% cut-off			
C4 >52.5 ng/mL or primary BA >15%	19	2	PPV = 90%
C4 <52.5 ng/mL and primary BA <15%	10	57	NPV = 85%
	Sensitivity = 66%	Specificity = 97%	
All participants with measurements of serum C4 and single stool primary BA % using 20% cut-off			
C4 >52.5 ng/mL or primary BA >20%	19	2	PPV = 90%
C4 <52.5 ng/mL and primary BA <20%	10	57	NPV = 85%
	Sensitivity = 66%	Specificity = 97%	
Only IBS-D participants with measurements of serum C4 and single stool primary BA % using 10% cut-off			
C4 >52.5 ng/mL or primary BA >10%	15	2	PPV = 88%
C4 <52.5 ng/mL and primary BA <10%	10	32	NPV = 76%
	Sensitivity = 60%	Specificity = 94%	

BA=bile acid; BAD=bile acid diarrhea; C4=7a-hydroxy-4-cholesten-3-one; IBS-D=diarrhea-predominant irritable bowel syndrome