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Three probiotic strains exert different effects on plasma bile acid profiles in healthy obese adults: randomised, double-blind placebo-controlled crossover study

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Table S1. The change (final-baseline) in biomarkers of glucose and lipid metabolism in participants with a body mass index <30 or ≥30 kg/m^2 who received placebo and one of the three probiotics.

		Placebo[1]		*B. lactis*[2]		*B. subtilis*		*L. plantarum*	
		BMI <30	BMI ≥30	BMI <30	BMI ≥30	BMI <30	BMI ≥30	BMI <30	BMI ≥30
		(n=40-41)	(n=57-58)	(n=13-15)	(n=17-18)	(n=15-16)	(n=16-18)	(n=11)	(n=24)
Glucose metabolism (plasma)									
Glucose (mmol/l)	*Baseline*	5.49±0.12	6.09±0.24	5.68±0.18	6.07±0.19	5.53±0.21	6.96±0.69	5.42±0.12	5.62±0.15
	Change	0.13±0.09	0.02±0.12	0.09±0.17	-0.06±0.17	0.10±0.11	-0.04±0.13	0.02±0.16	0.03±0.11
Insulin (pmol/l)	*Baseline*	61.5±6.6	85.4±8.1	67.9±14.0	100.0±28.3	47.3±6.7	83.3±12.1	43.6±7.1	77.0±9.3
	Change	-1.15±4.95	-1.79±6.46	0.22±7.18	-7.88±11.62	6.96±7.32	8.04±7.18	8.90±7.25	-0.03±6.17
Blood lipids[4]									
Total cholesterol (mmol/l)	*Baseline*	5.68±0.22	5.64±0.18	5.79±0.34	5.32±0.29	5.61±0.35	5.80±0.36	5.26±0.43	5.60±0.26
	Change	-0.14±0.10	-0.01±0.10	-0.15±0.19	-0.10±0.17	0.09±0.17	-0.35±0.18	0.25±0.21	0.10±0.14
HDL cholesterol (mmol/l)	*Baseline*	15.3±0.6	15.0±0.4	13.6±0.6	14.7±0.8	15.5±1.1	15.0±0.8	15.9±0.8	14.9±0.7
	Change	-0.02±0.03	0±0.03	-0.04±0.05	-0.05±0.04	0.05±0.04	-0.06±0.06	0.12±0.06	0.06±0.04
LDL cholesterol (mmol/l)	*Baseline*	30.5±1.3	30.6±1.1	30.1±1.5	28.7±1.9	29.8±2.1	31.2±2.0	28.9±3.2	30.9±1.6
	Change	-0.10±0.08	0.01±0.05	0.01±0.09	-0.04±0.16	0.10±0.13	-0.27±0.10	0.11±0.15	0.05±0.08
Triglycerides (mmol/l)	*Baseline*	2.38±0.23	2.33±0.17	3.11±0.59	2.14±0.30	2.36±0.39	2.58±0.34	1.70±0.20	2.22±0.26
	Change	-0.05±0.13	-0.07±0.13	-0.27±0.33	0±0.18	-0.15±0.16	-0.06±0.22	0.07±0.12	-0.01±0.13
NEFA (µmol/l)	*Baseline*	404±33	547±35	488±64	513±42	387±40	589±45	541±57	489±46
	Change	47±22	-38±32	15±75	-29±41	73±58	-37±57	57±92	-63±52
ApoA1 (µmol/l)	*Baseline*	63.5±3.4	64.8±2.5	57.6±5.7	70.9±5.4	56.3±3.6	69.5±3.6	58.7±4.3	56.6±2.5
	Change	3.7±1.7	4.0±1.6	4.8±2.7	1.0±1.6	8.1±2.9	1.7±3.1	20.6±11.1	8.9±3.6
ApoB100 (µmol/l)	*Baseline*	1.96±0.10	1.81±0.10	2.03±0.16	1.92±0.25	1.91±0.19	1.79±0.26	1.99±0.30	1.70±0.15
	Change	-0.10±0.07	0.06±0.08	-0.09±0.11	0.39±0.20	0.35±0.15	0.12±0.13	-0.10±0.26	0.18±0.10
HOMA-IR	*Baseline*	2.18±0.30	3.40±0.42	2.51±0.62	3.88±1.14	1.68±0.27	3.71±0.72	1.45±0.23	2.74±0.36
	Change	0.03±0.22	-0.16±0.36	0.14±0.36	-0.22±0.39	0.40±0.32	0.44±0.33	0.35±0.27	0.03±0.26

		Placebo[1]		*B. lactis*[2]		*B. subtilis*		*L. plantarum*	
		BMI <30	BMI ≥30	BMI <30	BMI ≥30	BMI <30	BMI ≥30	BMI <30	BMI ≥30
Bile acids (plasma)[5]		(n=41)	(n=57)	(n=13)	(n=17)	(n=16)	(n=16)	(n=11)	(n=24)
Deconjugated	*Baseline*	996±128	981±120	1360±316	656±127	928±190	640±118	625±112	1335±266
(nmol/l)	*Change*	6±174	98±176	266±319	380±165	-243±211	691±378	30±97	-156±319
Deconjugated /	*Baseline*	1.10±0.13	1.06±0.12	2.01±0.63	0.83±0.14	1.15±0.29	0.86±0.16	1.11±0.15	1.58±0.34
conjugated (ratio)	*Change*	0.464±0.245	0.032±0.170	-0.185±0.504	0.477±0.253	0.138±0.309	0.305±0.381	-0.392±0.149	-0.270±0.240
Secondary (nmol/l)	*Baseline*	932±100	958±99	981±214	641±104	1126±196	818±164	604±68	1075±180
	Change	66±125	-46±87	77±135	337±168	-285±129	466±237	119±66	-19±172
Secondary /	*Baseline*	1.19±0.21	0.96±0.08	0.82±0.14	0.78±0.10	0.99±0.21	1.00±0.10	1.22±0.23	0.91±0.13
primary (ratio)	*Change*	0.102±0.098	-0.120±0.078	-0.148±0.158	0.008±0.094	0.299±0.179	-0.227±0.103	-0.346±0.171	-0.003±0.115
Bile acids (stool)[6]		(n=15)	(n=15)	(n=6)	(n=4)	(n=7)	(n=3)	(n=2)	(n=8)
Deconjugated	*Baseline*	3560±1188	5226±1577	8430±3024	5430±2803	3153±1571	2693±819	127±43	2807±790
(sum)	*Change*	320±910	-1460±1440	-540±3340	1530±3680	-1980±1630	-1440±350	610±710	1210±950
Deconjugated /	*Baseline*	488±107	717±222	734±396	1470±782	746±424	1181±704	54±18	687±339
conjugated (ratio)	*Change*	-83±119	-324±199	74±491	-1101±648	-490±435	-741±807	1±21	-52±431
Secondary (sum)	*Baseline*	1497±428	2363±561	1927±442	1656±641	1887±513	2588±766	105±59	1640±384
	Change	120±430	-1040±540	-120±680	70±550	-810±580	-1390±370	380±480	780±760
Secondary /	*Baseline*	18.8±6.2	14.1±4.7	15.8±11.9	2.2±1.3	24.7±10.6	68.0±50.2	9.7±8.6	16.4±6.1
primary (ratio)	*Change*	3.6±6.0	13.5±6.9	-2.0±10.6	1.3±1.9	-0.9±10.0	-40.1±41.4	-5.1±5.6	-5.5±6.3

[1] Pooled data from the placebo intervention from each of the three crossovers.

[2] Abbreviations: Apo, apolipoprotein; *B. lactis*, *Bifidobacterium animalis* subsp. *lactis* B94; *B. subtilis*, *Bacillus subtilis* R0179; BMI, body mass index (kg/m^2); HDL, high density lipoprotein; HOMA-IR, homeostatic model assessment for insulin resistance; *L. plantarum, Lactobacillus plantarum* HA-119; LDL, low density lipoprotein; NEFA, non-esterified fatty acids.

[3] Values represent means±SEM.

[4] Total, HDL, and LDL cholesterol and NEFA were analysed in the serum and ApoA1 and ApoB100 were analysed in the plasma.

[5] Data represent the sum of individual bile acids.

[6] Data represent arbitrary units.

A

BMI	Intervention	Total n	TUDCA	TCA	TCDCA	TDCA	GCA	GUDCA	TLCA	CA	GCDCA	GDCA	UDCA	CDCA	DCA	GLCA	LCA
<30	Placebo	41	0	3	24	14	54	-2	0	-35	39	30	4	20	18	2	
<30	*B. lactis*	13	1	-3	8	7	-31	14	-1	-4	49	41	-16	226	-7	-2	
<30	*B. subtilis*	16	-1	-64	-95	-40	-358	-18	-1	-2	-395	-103	-67	-120	-54	-2	
<30	*L. plantarum*	11	0	-3	3	6	20	26	1	16	124	75	-18	4	28	2	

B

BMI	Intervention	Total n	TUDCA	TCA	TCDCA	TDCA	GCA	GUDCA	TLCA	CA	GCDCA	GDCA	UDCA	CDCA	DCA	GLCA	LCA
≥30	Placebo	58	0	12	8	-2	51	-7	0	56	9	-26	-19	61	16	-2	
≥30	*B. lactis*	17	1	6	29	18	34	23	1	55	78	83	133	113	80	-1	
≥30	*B. subtilis*	16	1	8	27	8	115	31	1	187	218	93	155	180	99	6	
≥30	*L. plantarum*	24	0	-10	-16	-6	-41	-3	-2	-123	-84	-39	82	-71	-44	-7	

-395	-339	-282	-226	-169	-113	-56	0	32	65	97	129	161	194	226

Figure S1. Heatmap for plasma bile acids. Heatmap showing the degree and direction of the change (final – baseline, nmol/l) in plasma bile acids in nonobese (BMI <30 kg/m^2; Panel A) and obese (BMI ≥30 kg/m^2; Panel B) participants after receiving *Bifidobacterium animalis* subsp. *lactis* B94 (*B. lactis*), *Bacillus subtilis* R0179 (*B. subtilis*), or *Lactobacillus plantarum* HA-119 (*L. plantarum*). Data from the placebo intervention from each of the three crossovers were pooled. Participants who did not have data for baseline and final plasma bile acids were not included. Reasons for missing values included: the participant withdrew from the study (*B. subtilis*, n=5; *B. lactis*, n=4; *L. Plantarum*, n=3) or the participant did not provide a blood sample on one of the appointment days or there was a technical issue with the blood draw study (*B. subtilis*, n=2; *B. lactis*, n=3). Grey coloured boxes denote that all values were below the limit of detection. Abbreviations: body mass index, BMI; tauroursodeoxycholic, TUDCA; taurocholic, TCA; taurochenodeoxycholic acid, TCDCA; taurodeoxycholic, TDCA; glycocholic, GCA; glycoursodeoxycholic, GUDCA; taurolithocholic, TLCA; cholic acid, CA; glycochenodeoxycholic, GCDCA; glycodeoxycholic, GDCA; ursodeoxycholic acid, UDCA; chenodeoxycholic acid, CDCA; deoxycholic acid, DCA; glycolithocolic acid, GLCA.

A

Sample	Intervention	Total n	TUDCA	TCA	TCDCA	TDCA	GCA	GUDCA	TLCA	CA	GCDCA	GDCA	UDCA	CDCA	DCA	GLCA	LCA
Plasma	Placebo	28	3	15	27	7	43	10	-1	-35	17	1	28	-62	-13	2	
Plasma	*B. lactis*	8	-1	-2	3	13	-76	-10	-1	33	-106	30	74	335	75	0	
Plasma	*B. subtilis*	10	1	5	24	-5	36	11	-1	123	100	24	126	104	48	-1	
Plasma	*L. plantarum*	10	1	-4	7	-3	-2	5	-5	-105	40	-45	126	-132	-68	-7	

B

Sample	Intervention	Total n	TUDCA	TCA	TCDCA	TDCA	GCA	GUDCA	TLCA	CA	GCDCA	GDCA	UDCA	CDCA	DCA	GLCA	LCA
Stool	Placebo	30						9		-48	23	0	-31	-52	-432	0	-6
Stool	*B. lactis*	10						-17		352	-2	0	-22	-39	0	0	-5
Stool	*B. subtilis*	10						0		-808	-1	0	-11	-28	-978	-3	9
Stool	*L. plantarum*	10						1		-688	1	0	-57	-39	32	0	3

-978	-838	-699	-559	-419	-279	-140	0	50	101	151	201	251	302	352

Figure S2. Heatmap for plasma and stool bile acids for the subgroup of study participants who provided both stool and plasma samples. Heatmap showing the degree and direction of the change (final – baseline) in bile acids in plasma (nmol/l, panel A) and stool samples (arbitrary units, Panel B) for a subgroup of participants who received *Bifidobacterium animalis* subsp. *lactis* B94 (*B. lactis*), *Bacillus subtilis* R0179 (*B. subtilis*), or *Lactobacillus plantarum* HA-119 (*L. plantarum*). Data from the placebo intervention from each of the three crossovers were pooled. Only those participants who provided a stool sample are included in panel A. Missing data points for plasma bile acids were due to technical error (*B. subtilis* and corresponding placebo, n=2). Grey coloured boxes denote that all values were below the limit of detection. Abbreviations: body mass index, BMI; tauroursodeoxycholic, TUDCA; taurocholic, TCA; taurochenodeoxycholic acid, TCDCA; taurodeoxycholic, TDCA; glycocholic, GCA; glycoursodeoxycholic, GUDCA; taurolithocholic, TLCA; cholic acid, CA; glycochenodeoxycholic, GCDCA; glycodeoxycholic, GDCA; ursodeoxycholic acid, UDCA; chenodeoxycholic acid, CDCA; deoxycholic acid, DCA; glycolithocolic acid, GLCA.