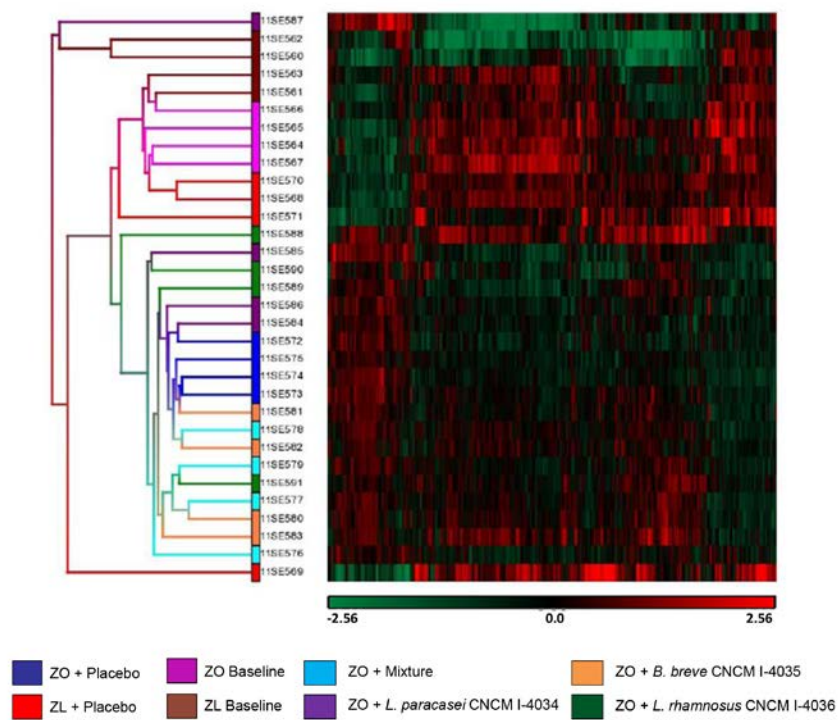


Title: *Adamdec1, Ednrb and Ptgs1/Cox1*, inflammation genes upregulated in the intestinal mucosa of obese rats, are downregulated by three probiotic strains

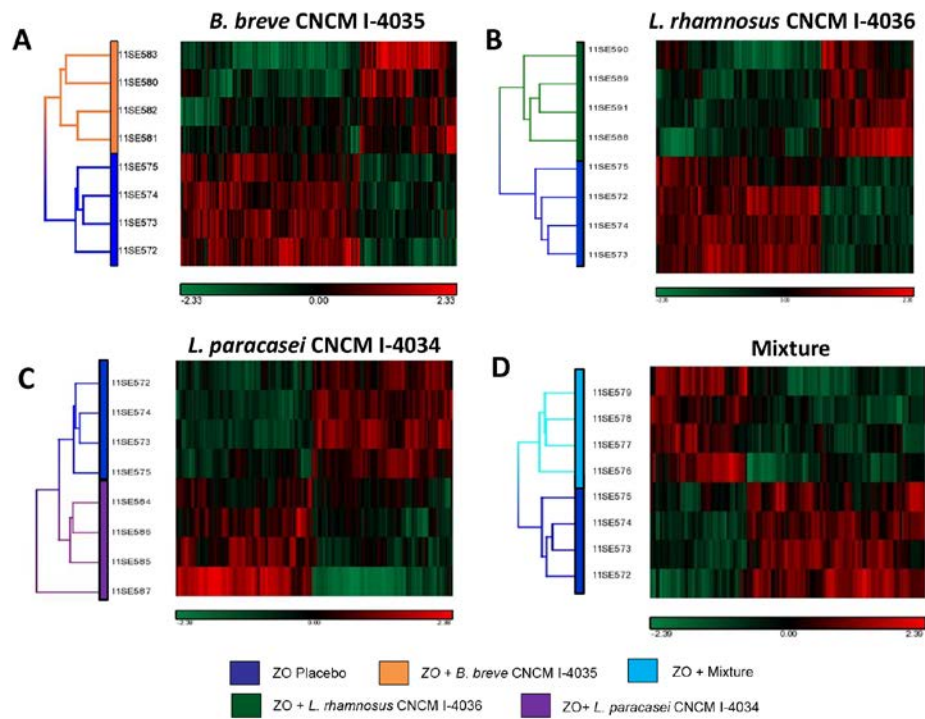
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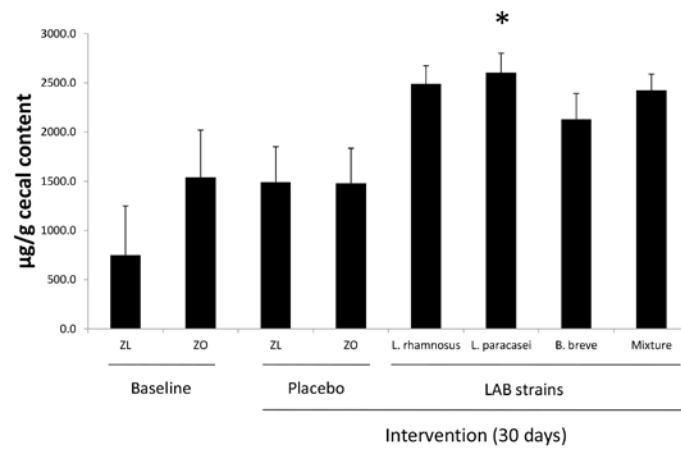
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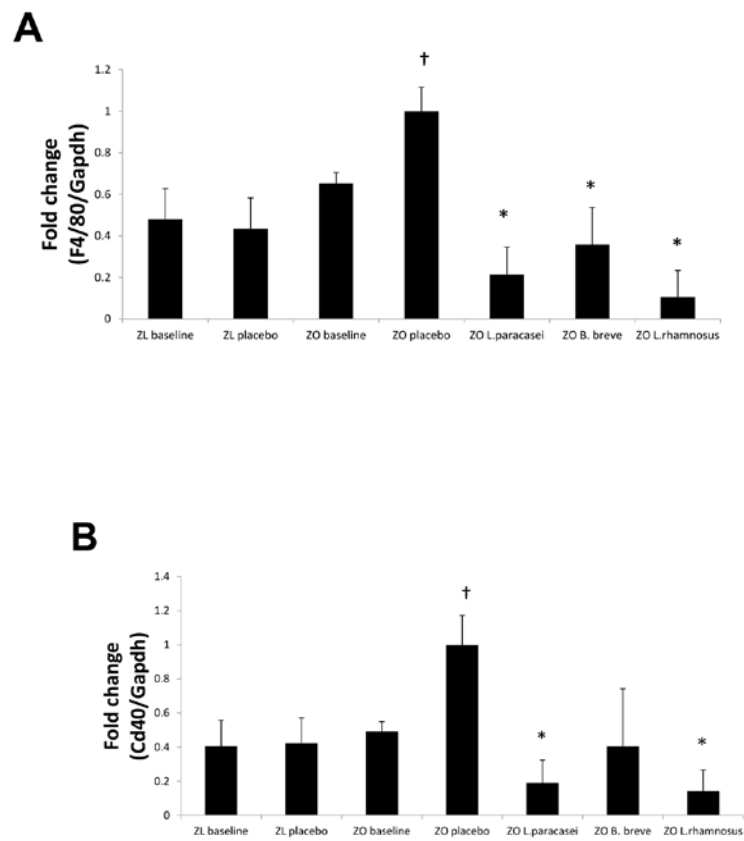
Supplementary Figure 1



Supplementary Figure 2



Supplementary Figure 3



Supplementary Figure 4

Supplementary Figure 1. Microarray results obtained with the Rat Gene 1.1 ST Array Plate (Affymetrix®). These results were uploaded to the Gene Expression Omnibus (GEO) platform as “GSE73848 *Expression data from intestinal mucosa of Zucker rats.*” ZL, Zucker lean $\text{Lepr}^{+/fa}$ rats; ZO, Zucker- $\text{Lepr}^{fa/fa}$ rats; Mixture, *L. paracasei* CNCM I-4034 plus *B. breve* CNCM I-4035 strains. n=4 rats per group.

Supplementary Figure 2. Microarray expression data from the intestinal mucosa of Zucker- $\text{Lepr}^{fa/fa}$ rats treated with *B. breve* CNCM I-4035 **(A)**, *L. rhamnosus* CNCM I-4036 **(B)**, *L. paracasei* CNCM I-4034 **(C)**, or mixture **(D)** compared with Zucker- $\text{Lepr}^{fa/fa}$ rats fed the placebo. ZO, Zucker- $\text{Lepr}^{fa/fa}$ rats; Mixture, *L. paracasei* CNCM I-4034 plus *B. breve* CNCM I-4035 strains. n=4 rats per group.

Supplementary Figure 3. Total content of secretory IgA in the feces of Zucker-lean^{+/fa} rats and Zucker- $\text{Lepr}^{fa/fa}$ rats that were fed either a placebo or probiotic strains for 30 days. Values are expressed as the mean $\pm$ SEM, n=8 rats per group. * $P < 0.05$ versus ZO placebo. ZL, Zucker lean $\text{Lepr}^{+/fa}$ rats. ZO, Zucker- $\text{Lepr}^{fa/fa}$ rats.

Supplementary Figure 4. Obese rats develop intestinal inflammation, and probiotic treatment mitigates it. qRT-PCR showing mRNA levels for (A) F4/80, and (B) Cd40. Values are expressed as the mean $\pm$ SEM, n=8 rats per group. [†] $P < 0.05$ versus ZL baseline, ZL placebo and ZO baseline. * $P < 0.05$ versus ZO placebo. ZL, Zucker lean $\text{Lepr}^{+/fa}$ rats. ZO, Zucker- $\text{Lepr}^{fa/fa}$ rats.

Supplementary Table 1. Validation of microarray results for obese Zucker-Lepr^{fa/fa} rats fed the placebo

Intestinal mucosa genes (qRT-PCR)	ZO baseline versus ZO placebo	<i>P</i> value
Slc7a11	↓-1.548	0.0315
Fkbp5	↓-1.395	0.0362
Ednrb	↑3.682	0.0001
Adamdec1	↑5.941	<0.001
Ptgs1	↑1.46	0.0101
Nfkbia	↓-1.18	0.0269

This table shows the fold change in gene expression as quantitated by qRT-PCR, as well as the statistical significance (*P* value) compared with Zucker-Lepr^{fa/fa} rats at baseline. Inhibited genes appear as ↓, whereas induced genes appear as ↑. n=8 rats per group.

Supplementary Table 2. List of genes (symbol and reference) whose expression was modulated by the probiotic strains

<i>L. rhamnosus</i> CNCM I-4036 group		<i>L. paracasei</i> CNCM I-4034 group			
gene_assignment	Gene Symbol	gene_assignment	Gene Symbol	gene_assignment	Gene Symbol
NM_031345	↓Tsc22d3	NM_017333	↓Ednrb	NM_012521	↑S100g
NM_053551	↓Pdk4	NM_001106046	↓Adamdec1	FQ225348	–
NM_001106611	↓Lsm3	BC091243	↓Igha	BC098746	–
NM_053600	↓Fez2	NM_012802	↓Pdgfra	BC092582	↓LOC314509
ENSRNOT00000065436	↓Grsf1	NM_031048	↓Lifr	ENSRNOT00000040099	↓LOC502822
NM_001108321	↓Rtp4	NM_017272	↓Aldh1a7	<i>L. paracasei</i> CNCM I-4034 and <i>B. breve</i> CNCM I-4035 groups	
NM_001034012	↓Adamtsl4	ENSRNOT00000057971	↓RGD1564318	gene_assignment	Gene Symbol
ENSRNOT00000053807	–	BC088423	↓IgG-2a	NM_013096	↑Hba-a2
ENSRNOT00000054185	–	BC097960	↓Igj	NM_013096	↑Hba-a2
ENSRNOT00000052628	–	NM_022407	↓Aldh1a1	NM_001111269	↑LOC689064
ENSRNOT00000053902	–	NM_001007728	↓Mpzl1	NM_198776	↑Hbb-b1
ENSRNOT00000054104	–	L22652	↓IgG-2a	<i>L. paracasei</i> CNCM I-4034, <i>B. breve</i> CNCM I-4035 and mixture groups	
ENSRNOT00000053447	–	NM_017043	↓Ptgs1	gene_assignment	Gene Symbol
ENSRNOT00000053929	–	NM_031086	↓Pros1	ENSRNOT00000058247	↓RGD1563231
ENSRNOT00000053402	–	NM_001134469	↓Lrrc49	<i>L. rhamnosus</i> CNCM I-4036, <i>B. breve</i> CNCM I-4035 and mixture groups	
<i>L. rhamnosus</i> CNCM I-4036 and <i>L. paracasei</i> CNCM I-4034 groups		ENSRNOT00000041272	↓RGD1564284	gene_assignment	Gene Symbol
gene_assignment	Gene Symbol	NM_001017502	↓Ccdc117	NM_001077589	↓Rgs16
NM_001107673	↓Slc7a11	NM_017260	↓Alox5ap	NM_001034125	↓Per1
NM_001012174	↓Fkbp5	NM_053566	↓Ptch1	<i>L. rhamnosus</i> CNCM I-4036 and <i>B. breve</i> CNCM I-4035 groups	
ENSRNOT00000053268	–	NM_001009920	↓Gsta5	gene_assignment	Gene Symbol
ENSRNOT00000053276	–	NM_133583	↓Ndrp2	ENSRNOT00000053105	–

<i>B. breve</i> CNCM I-4035 group		BC088423	↓IgG-2a	ENSRNOT00000052978	–
gene_assignment	Gene Symbol	NM_001105779	↓Nhp2	FQ233900	–
NM_033234	↑Hbb	NM_022508	↓Mthfd1		
ENSRNOT00000053328	–	NM_031523	↑Klk1b3		

Listed genes were selected from the microarray results based on two criteria: 1) Up- or down-regulation ≥ 1.5 -fold compared with Zucker-Lepr^{fa/fa} rats fed the placebo, and 2) expression regulated by at least two of the probiotic strains. Comparisons were made versus Zucker-Lepr^{fa/fa} rats fed the placebo. Inhibited genes appear as ↓, whereas induced genes appear as ↑. Genes lacking name or symbol are of unknown function (dash). n=4 rats per group.

Supplementary Table 3. Validation of microarray results for obese Zucker-Lepr^{fa/fa} rats that received probiotic strains

Intestinal mucosa genes (qRT-PCR)	Mixture	<i>P</i> value	<i>B. breve</i> CNCM I-4035	<i>P</i> value	<i>L. rhamnosus</i> CNCM I-4036	<i>P</i> value	<i>L. paracasei</i> CNCM I-4034	<i>P</i> value
Rgs16	-1.4475	0.045	-1.1794	0.0169	1.3041	0.0061		0.0229
Per1	-1.1103		-1.0346		-1.1194			
Slc7a11	↓-1.562				-1.5256		-1.4578	
Fkbp5					↓-1.8928		↓-1.4582	
Alox5ap	↓-2.0045	<0.001	↓-1.7617	0.0169	-1.5037	0.0016	↓-1.6926	0.0016
Ednrb	-1.2308		↓-2.2192	0.0063	-2.3345		↓-1.8782	0.0004
Adamdec1	↓-1.6176	0.0429	↓-3.3604	<0.001	↓-3.3349	<0.001	↓-2.7512	<0.001
Ptgs1	-1.2382		↓-1.6275	0.0004	-1.4031		↓-1.3756	0.006
Tlr9	-1.0266		-1.0109		1.8727	0.14	1.4146	0.0611
Naip	1.0523		↓-1.58	0.0409	1.0592		-1.32	
Nfkb1a	1.0191		1.04		1.0348		1.0373	
Nfkb1	-1.003		-1.0792		1.0121		-1.12	

This table shows the fold change in gene expression as quantitated by qRT-PCR, as well as the statistical significance (*P* value) compared with Zucker-Lepr^{fa/fa} rats fed the placebo. Inhibited genes appear as ↓. n=8 rats per group.