

## **SUPPLEMENTARY MATERIAL**

### **‘Pera’ Orange and ‘Moro’ Blood Orange Juice Improves Oxidative Stress and Inflammatory Response Biomarkers and Modulates the Gut Microbiota of Individuals with Insulin Resistance and Different Obesity Classes**

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## Material and Methods

### *Quantification of Soluble Sugar, Organic Acids, and Chemical Composition*

The soluble sugars were analyzed by high performance liquid chromatography (HPLC) coupled to a pulse amperometric detector according to Shiga *et al.* (2011). The organic acid contents were analyzed by HPLC in a HP1100 system (Hewlett-Packard Company, Palo Alto, CA, USA) coupled with a diode-array detector, equipped with a  $\mu$ Bondpack C18 (300 mm  $\times$  3.6 mm i.d., Waters, Milford, MA, USA) and elution (flow rate of 0.5 mL  $\cdot$  min<sup>-1</sup>) was carried out in isocratic conditions with 0.1% H<sub>3</sub>PO<sub>4</sub>, monitored at 210 nm. The content of total dietary fiber and fractions were measured according to the method described by Association of Official Analytical Chemists (AOAC 991.43) (AOAC, 1995).

### *Analysis of Total Phenolic Content*

The total phenolic content of samples was determined using the Folin–Ciocalteu colorimetric method described by Singleton and Rossi (1965) with some modifications. A previously shaken sample of orange juice (2 mL) was added to 10 mL of methanol/water (80:20, v/v). The sample was vortexed for 1 min and then placed in an ultrasonic bath (15 min) according to the procedure described by Stella *et al.* (2011). Afterward, the samples were centrifuged at 10000  $\times g$  during 15 min at 20 °C, and supernatants were filtered by a Whatman filter and analyzed. The absorbance was measured spectrophotometrically at 763 nm. Measurements were recorded on an UV–vis spectrophotometer Helios Epsilon (Thermo Scientific, Madison, WI, USA). The results were expressed as milligrams of gallic acid per 100 mL of orange juice.

### *Quantification of Flavonoids in Orange Juice*

Orange juices (10 mL) were centrifuged at  $10,000 \times g$  for 15 min at 4 °C. The supernatant was eluted in a column of 1 g of polyamide (CC 6, Macherey–Nagel), previously preconditioned by passing methanol followed by deionized water. The phenolic compounds were eluted with methanol acidified with 2.5% acetic acid. The eluates were completely dried by rotary evaporation (Rotavapor, RE 120, Büchi, Flawil, Switzerland) under a vacuum at 40 °C, resuspended with methanol acidified with 5% acetic acid, and filtered through a 0.45 µm PVDF Millex filter (Millipore Ltd.a., Bedford, MA, USA) before HPLC analysis. The pellet was added to 20 mL of dimethyl sulfoxide, homogenized overnight at room temperature, centrifuged, and filtered through a 0.45 µm PVDF filter.

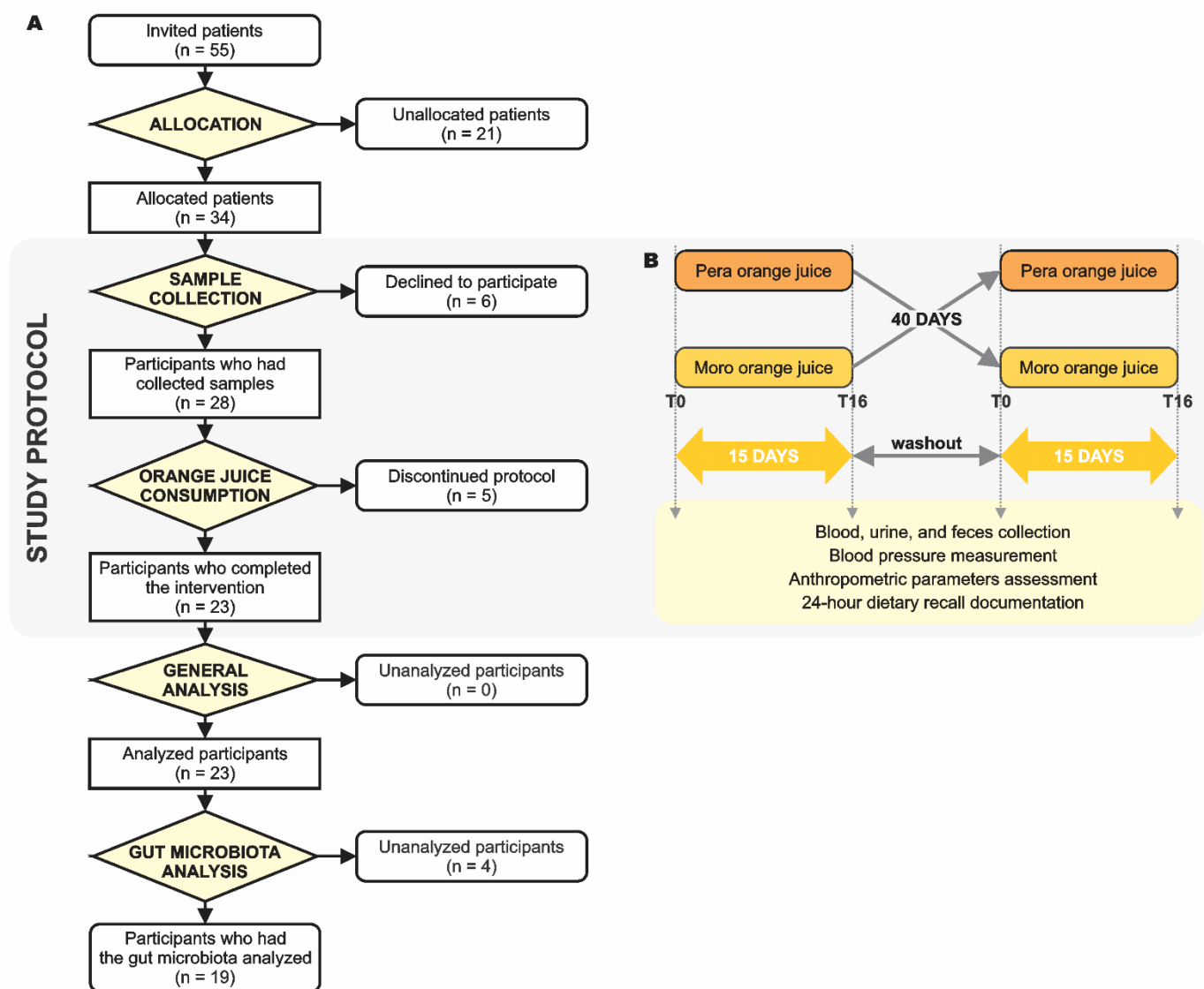
Samples were analyzed by HPLC on Agilent 2100 equipment coupled to a diode array detector (DAD) using a Prodigy 5 µm ODS3 column (250 × 4.60 mm) (Phenomenex Ltd., Cheshire, UK) with a flow rate of  $1 \text{ mL} \cdot \text{min}^{-1}$  at 25 °C. Elution was carried out with a solvent gradient constituted of 0.5% formic acid in water (A) and 0.5% formic acid in acetonitrile (B). The solvent concentration gradient applied was 8% B at the beginning, 10% for 5 min, 17% for 10 min, 25% for 15 min, 50% for 25 min, 90% for 30 min, 50% for 32, and 8% for 35 min. The eluates were monitored at 280 and 525 nm (Brasili *et al.*, 2017). Quantification was performed using a calibration curve of cyanidin-3-O-glucoside (at 525 nm) and hesperidin, narirutin, and didymin at 280 nm.

Peak identification was carried out by Prominence liquid chromatography (Shimadzu, Japan) coupled to an ion trap mass spectrometer (Esquire HCT model, Bruker Daltonics, Billerica, MA, USA). The separation conditions were the same as those used for HPLC/DAD, and the flow rate was changed to 0.2

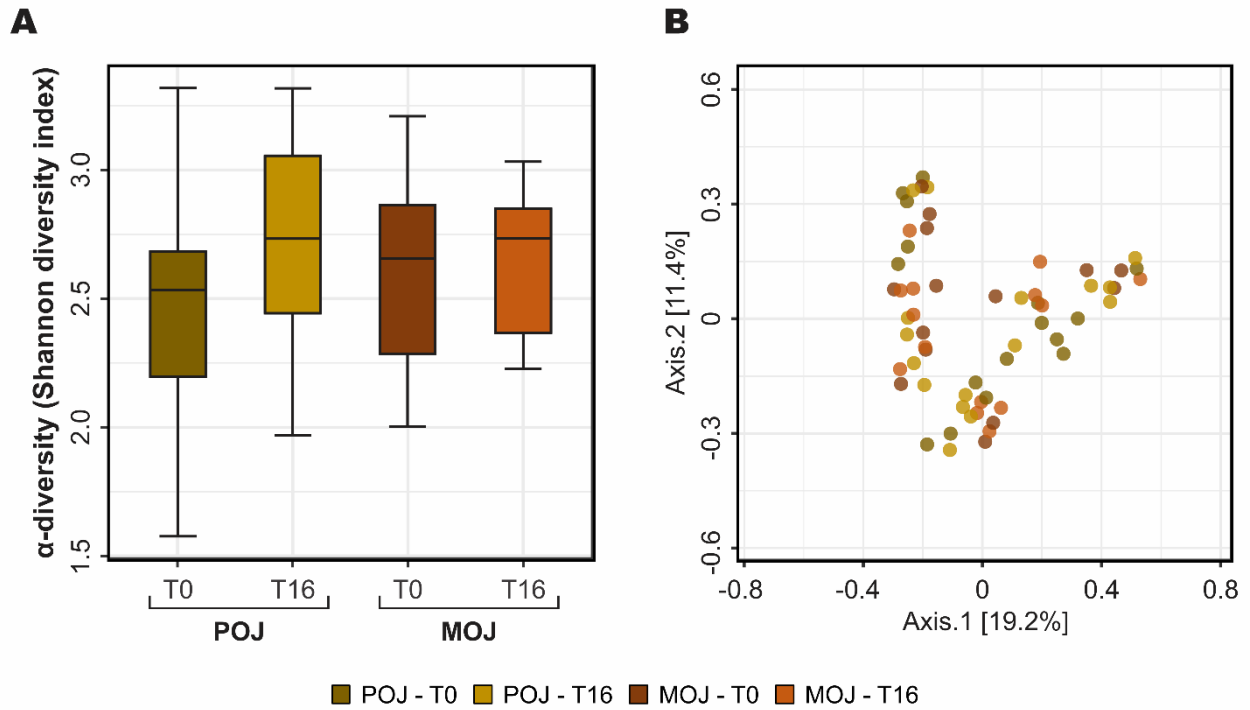
mL·min<sup>-1</sup> to allow the eluate to pass through the mass spectrometer. The ESI was maintained in positive and negative modes for anthocyanins and other flavonoid classes, respectively. The mass operating conditions were programmed to perform a full scan (m/z 100–1000), with a collision energy of 3000–3500 V, and a capillary temperature of 275 °C. Peak identification was carried out by the combined information provided by mass spectra, retention time, and literature data (Hillebrand *et al.*, 2004). The identity was confirmed by co-elution with authentic standards.

## References

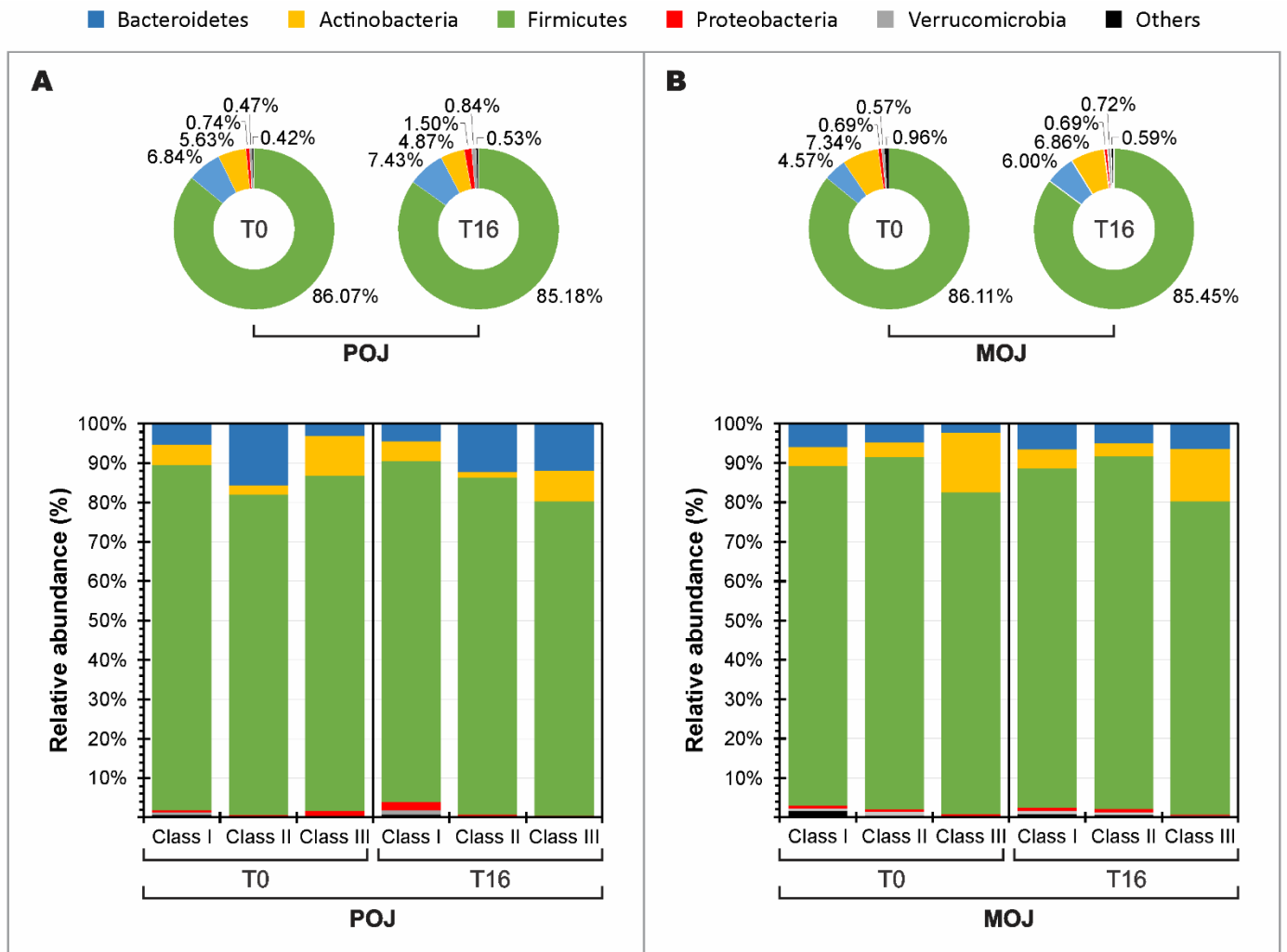
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**Figure S1.** Flowchart of the clinical study **(A)** and schematic representation of the clinical study design **(B)**.



**Figure S2.** Gut microbiota diversity analysis. **(A)** Shannon diversity index as a measurement of alpha-diversity. **(B)** beta-diversity indicated by principal coordinates analyses (PCoA) plot. POJ: Pera orange juice. MOJ: Moro orange juice.



**Figure S3.** Effect of Pera (POJ) and Moro (MOJ) orange juice intake on the gut microbiota OTUs. Mean relative abundance **(A)** and relative abundance by obesity class **(B)** of the most abundant bacteria phyla in the gut microbiota of the patients before (T0) and after (T16) each intervention. OTU: Operational taxonomic unit.

**Table S1.** Quality parameters and chemical composition of pasteurized Pera and Moro orange juices

	POJ	MOJ
Quality Parameters		
pH	3.52 ± 0.00	3.48 ± 0.00
TSS (°Brix)	9.50 ± 0.00	10.00 ± 0.00
Soluble sugars (mg.100 mL <sup>-1</sup> )		
Sucrose	3.10 ± 0.25	2.80 ± 0.25
Fructose	1.56 ± 0.10	1.90 ± 0.15
Glucose	1.50 ± 0.08	1.69 ± 0.18
Total	6.16	6.39
Organic acids (mg.100 mL <sup>-1</sup> )		
Citric acid	1092.36 ± 3.09	1161.94 ± 81.19
Malic acid	301.77 ± 3.34	273.97 ± 18.50
Ascorbic acid	16.00 ± 1.50	32.00 ± 2.60
Tartaric acid	N.D.	26.25 ± 0.95
Total	1410.13	1494.16
Total phenolic compounds (mg.100 mL <sup>-1</sup> )	50.31 ± 0.00	60.75 ± 0.01
Dietary fiber (g.100 mL <sup>-1</sup> )		
Soluble dietary fiber	0.04 ± 0.01	0.08 ± 0.01
Insoluble dietary fiber	0.25 ± 0.05	0.26 ± 0.02
Total dietary fiber	0.29	0.34

POJ: Pera orange juice. MOJ: Moro orange juice. TSS: Total soluble solids. N.D.: Not detected. Results are expressed as the mean ± standard error ( $n = 3$ ).



**Table S2.** Flavonoid content in pasteurized Pera and Moro orange juices

	POJ			MOJ		
	Supernatant	Pellet	Total	Supernatant	Pellet	Total
Narirutin	0.25 ± 0.03 <sup>a</sup>	1.79 ± 0.08 <sup>a</sup>	2.04	0.14 ± 0.02 <sup>b</sup>	0.40 ± 0.07 <sup>b</sup>	0.54
Hesperidin	1.26 ± 0.44	28.40 ± 1.52 <sup>a</sup>	29.66	1.48 ± 0.20	22.29 ± 2.11 <sup>b</sup>	23.77
Didymin	N.D.	N.D.	N.D.	0.42 ± 0.02	7.59 ± 0.42	8.01
Total flavanones	1.51	30.19	31.70	2.04	30.28	32.32
Cyn-3-O-glu	N.D.	N.D.	N.D.	2.73 ± 0.08	N.D.	2.73
Cyn-3-O-(malonyl)glu **	N.D.	N.D.	N.D.	0.08 ± 0.03	N.D.	0.08
Total anthocyanins	N.D.	N.D.	N.D.	2.81	N.D.	2.81
Total flavonoids	1.51	30.19	31.70	4.85	30.28	35.13

POJ: Pera orange juice. MOJ: Moro orange juice. Cyn-3-O-glu: cyanidin-3-O-glucoside. Cyn-3-O-(malonyl)glu: cyanidin-3-O-(6"-malonyl)glucoside (quantified as cyanidin-3-O-glucoside equivalent). N.D.: Not detected. Results are expressed as mg.100 mL<sup>-1</sup> (mean ± standard error, *n* = 3). Different superscript letters indicate statistical significance (*p* < 0.05) between POJ and MOJ at each fraction.

**Table S3.** Creatinine and flavanone metabolites excreted by the patients before (T0) and after (T16) the consumption of Pera and Moro orange juices

	POJ			MOJ		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
Creatinine (mg.dL <sup>-1</sup> )	136.01 ± 23.29	113.68 ± 12.35	0.733	130.82 ± 14.46	120.42 ± 14.82	0.354
Narirutin metabolites (mg equivalents)	<b>0.11 ± 0.03<sup>b</sup></b>	<b>0.47 ± 0.14<sup>a</sup></b>	<b>0.021</b>	<b>0.35 ± 0.09<sup>b</sup></b>	<b>0.71 ± 0.19<sup>a</sup></b>	<b>0.013</b>
Hesperidin metabolites (mg equivalents)	0.26 ± 0.17	0.64 ± 0.22	0.110	<b>0.72 ± 0.26<sup>b</sup></b>	<b>1.46 ± 0.49<sup>a</sup></b>	<b>0.028</b>
Phenolic acids (mg equivalents)	<b>32.26 ± 13.17<sup>b</sup></b>	<b>72.38 ± 19.34<sup>a</sup></b>	<b>0.043</b>	<b>37.17 ± 6.39<sup>b</sup></b>	<b>77.15 ± 19.83<sup>a</sup></b>	<b>0.028</b>

POJ: Pera orange juice. MOJ: Moro orange juice. Results are expressed as the mean ± standard error (*n* = 23). The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16.

**Table S4.** Energy and macronutrient consumption before (T0) and after (T16) the intervention with daily consumption of 500 mL of orange juice

	POJ			MOJ		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
Energy (kcal)	1280.10 ± 127.74	1408.28 ± 112.34	0.287	1306.99 ± 103.72	1457.55 ± 109.76	0.191
Carbohydrates (g.100 g <sup>-1</sup> )	172.27 ± 18.25	204.61 ± 17.31	0.162	180.21 ± 13.83	211.55 ± 20.02	0.144
Proteins (g.100 g <sup>-1</sup> )	54.25 ± 5.52	63.48 ± 7.04	0.429	65.85 ± 9.67	60.72 ± 6.75	1.000
Lipids (g.100 g <sup>-1</sup> )	41.54 ± 6.05	36.56 ± 4.46	0.484	37.80 ± 4.64	39.96 ± 4.07	0.648
Cholesterol (mg.100 g <sup>-1</sup> )	173.30 ± 27.93	227.01 ± 43.90	0.227	232.25 ± 39.62	194.67 ± 32.77	0.661
SFA (g.100 g <sup>-1</sup> )	9.88 ± 1.91	9.80 ± 1.83	0.833	11.07 ± 1.63	9.91 ± 1.54	0.456
MUFA (g.100 g <sup>-1</sup> )	10.45 ± 2.13	9.22 ± 1.67	0.948	9.88 ± 1.53	8.65 ± 1.27	0.494
PUFA (g.100 g <sup>-1</sup> )	7.85 ± 2.25	5.80 ± 1.03	0.820	6.77 ± 1.44	5.72 ± 0.80	0.637
Dietary fiber (g.100 g <sup>-1</sup> )	11.41 ± 1.75	10.83 ± 1.36	0.976	11.13 ± 1.09	9.35 ± 1.00	0.224

POJ: Pera orange juice. MOJ: Moro orange juice. SFA: Saturated fatty acids. MUFA: Monounsaturated fatty acids. PUFA: polyunsaturated fatty acids. Results are expressed as the mean ± standard error (*n* = 23). The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16.

**Table S5.** Anthropometric variables and biochemical parameters of patients with different classes of obesity before (T0) and after (T16) the consumption of Pera orange juice

	Obese I class (n = 12)			Obese II class (n = 5)			Obese III class (n = 6)		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
Body weight (kg)	87.81 ± 3.44	88.06 ± 3.59	0.610	99.96 ± 2.63	98.94 ± 3.41	0.686	103.25 ± 5.20	103.42 ± 4.85	0.293
Abdominal circumference (cm)	108.75 ± 1.81	109.38 ± 1.71	0.498	118.96 ± 2.08	117.40 ± 2.29	0.461	125.58 ± 3.62	125.08 ± 3.12	0.400
BMI (kg.m <sup>-2</sup> )	32.48 ± 0.40	32.54 ± 0.36	0.638	37.35 ± 0.83	36.95 ± 0.98	0.686	44.46 ± 2.33	44.54 ± 2.18	0.249
Blood pressure (mmHg)									
Systolic	130.92 ± 5.26	126.58 ± 4.58	0.444	134.40 ± 4.58	119.40 ± 7.61	0.138	133.83 ± 4.09	129.50 ± 7.18	0.463
Diastolic	81.83 ± 3.52	81.00 ± 3.56	0.052	76.40 ± 4.11	71.60 ± 4.20	1.000	88.17 ± 3.88	84.00 ± 5.27	0.344
Cholesterol (mg.dL <sup>-1</sup> )									
HDL	42.33 ± 2.29	40.33 ± 1.88	0.116	48.60 ± 3.40	47.80 ± 2.33	0.273	50.83 ± 5.02	45.33 ± 2.56	0.345
LDL	105.42 ± 8.69	99.75 ± 7.42	0.071	122.00 ± 13.61	135.00 ± 13.41	0.498	101.00 ± 15.80	91.83 ± 12.46	0.463
VLDL	31.92 ± 2.94	29.83 ± 2.80	0.666	23.00 ± 4.66	20.80 ± 2.29	0.500	32.33 ± 4.22	32.33 ± 4.19	0.344
Total	<b>180.25 ± 10.53<sup>a</sup></b>	<b>170.42 ± 8.17<sup>b</sup></b>	<b>0.004</b>	193.60 ± 15.80	203.60 ± 12.27	0.500	184.17 ± 14.56	169.50 ± 12.15	0.917
Triglycerides (mg.dL <sup>-1</sup> )	161.25 ± 14.43	148.42 ± 14.17	0.272	115.20 ± 23.21	104.60 ± 12.03	0.500	161.33 ± 20.97	161.83 ± 20.85	0.344
Blood glucose (mg.dL <sup>-1</sup> )	95.42 ± 2.65	93.08 ± 1.76	0.929	92.40 ± 2.44	94.60 ± 3.31	0.715	94.83 ± 4.71	97.33 ± 4.12	0.917

BMI: Body mass index. HDL: High-density lipoprotein. LDL: Low-density lipoprotein. VLDL: Very low-density lipoprotein. The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16 for each class of obesity (mean ± standard error).

**Table S6.** Anthropometric variables and biochemical parameters of patients with different classes of obesity before (T0) and after (T16) the consumption of Moro orange juice

	Obese I class (n = 12)			Obese II class (n = 5)			Obese III class (n = 6)		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
Body weight (kg)	87.35 ± 3.51	87.20 ± 3.60	0.307	97.73 ± 3.39	97.98 ± 3.66	0.273	103.20 ± 4.46	102.80 ± 4.54	0.462
Abdominal circumference (cm)	108.71 ± 1.84	109.29 ± 2.14	0.836	116.40 ± 1.33	117.00 ± 2.26	0.336	124.33 ± 2.87	118.48 ± 4.88	0.680
BMI (kg.m <sup>-2</sup> )	32.28 ± 0.33	32.21 ± 0.34	0.367	36.51 ± 1.11	36.61 ± 1.26	0.273	44.41 ± 1.84	44.21 ± 1.73	0.600
Blood pressure (mmHg)									
Systolic	129.17 ± 4.12	126.25 ± 4.68	0.272	130.20 ± 3.92	120.80 ± 4.07	0.225	128.33 ± 3.87	131.00 ± 4.58	0.463
Diastolic	83.92 ± 3.18	80.42 ± 3.80	0.784	73.00 ± 0.84	73.40 ± 2.86	0.138	83.17 ± 3.03	82.50 ± 3.97	0.207
Cholesterol (mg.dL <sup>-1</sup> )									
HDL	45.25 ± 2.82	43.00 ± 2.94	0.134	50.60 ± 3.89	49.00 ± 2.90	0.715	50.50 ± 2.92	48.33 ± 3.61	0.138
LDL	123.25 ± 14.82	113.67 ± 13.53	0.146	121.20 ± 9.32	114.80 ± 13.50	0.500	96.50 ± 7.52	98.67 ± 11.50	0.207
VLDL	34.33 ± 3.62	31.67 ± 4.15	0.533	23.60 ± 2.16	22.00 ± 3.58	0.686	36.50 ± 3.99	34.00 ± 4.49	1.000
Total	206.83 ± 14.49	185.25 ± 13.18	0.100	195.40 ± 10.52	185.80 ± 13.68	0.686	181.00 ± 9.16	182.17 ± 15.01	0.173
Triglycerides (mg.dL <sup>-1</sup> )	193.50 ± 26.94	158.00 ± 20.76	0.480	117.60 ± 10.72	111.00 ± 17.56	0.686	182.17 ± 19.66	170.33 ± 22.11	0.893
Blood glucose (mg.dL <sup>-1</sup> )	96.50 ± 2.63	97.08 ± 1.95	0.287	94.80 ± 2.11	95.60 ± 2.68	0.500	<b>99.17 ± 4.14<sup>a</sup></b>	<b>98.50 ± 2.36<sup>b</sup></b>	<b>0.041</b>

BMI: Body mass index. HDL: High-density lipoprotein. LDL: Low-density lipoprotein. VLDL: Very low-density lipoprotein. The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16 for each class of obesity (mean ± standard error).

**Table S7.** Inflammatory biomarkers of patients with different classes of obesity before (T0) and after (T16) the consumption of Pera and Moro orange juices

	Obese I class (n = 12)			Obese II class (n = 5)			Obese III class (n = 6)		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
<b>Pera orange juice</b>									
IL-6 (pg.mL <sup>-1</sup> )	5.26 ± 1.06	5.53 ± 0.92	0.424	4.45 ± 1.33	2.68 ± 0.26	0.465	6.74 ± 2.30	6.36 ± 1.97	0.345
IL-10 (pg.mL <sup>-1</sup> )	1.05 ± 0.16	1.46 ± 0.43	0.221	0.93 ± 0.26	1.15 ± 0.29	0.893	1.06 ± 0.26	1.00 ± 0.24	0.893
TNF-α (pg.mL <sup>-1</sup> )	97.33 ± 29.80	178.67 ± 34.98	0.499	78.25 ± 15.15	200.00 ± 55.45	0.686	<b>110.00 ± 51.70<sup>a</sup></b>	<b>105.50 ± 48.95<sup>b</sup></b>	<b>0.043</b>
IL-8 (pg.mL <sup>-1</sup> )	<b>623.60 ± 82.01<sup>b</sup></b>	<b>639.00 ± 138.67<sup>a</sup></b>	<b>0.026</b>	689.00 ± 162.17	606.40 ± 154.78	0.345	678.00 ± 143.83	736.00 ± 79.38	0.500
IL-1β (pg.mL <sup>-1</sup> )	196.14 ± 54.67	206.00 ± 89.18	0.314	377.50 ± 283.86	370.75 ± 250.19	0.080	230.25 ± 93.21	152.00 ± 99.88	0.715
MCP-1 (pg.mL <sup>-1</sup> )	6983.89 ± 2652.45	4120.78 ± 468.29	0.333	4908.20 ± 1593.20	3407.20 ± 302.87	0.893	3223.75 ± 1122.10	2686.00 ± 490.26	0.893
<b>Moro orange juice</b>									
IL-6 (pg.mL <sup>-1</sup> )	5.47 ± 1.57	3.80 ± 0.67	0.799	6.48 ± 4.00	2.79 ± 0.47	0.080	5.68 ± 0.86	4.77 ± 1.06	0.285
IL-10 (pg.mL <sup>-1</sup> )	1.24 ± 0.36	1.37 ± 0.15	0.515	1.43 ± 0.24	1.40 ± 0.34	0.138	1.12 ± 0.31	0.97 ± 0.17	0.854
TNF-α (pg.mL <sup>-1</sup> )	136.75 ± 33.45	184.38 ± 41.50	0.249	144.00 ± 34.81	137.40 ± 66.52	0.345	173.00 ± 49.50	94.00 ± 51.93	0.713
IL-8 (pg.mL <sup>-1</sup> )	666.00 ± 106.42	519.45 ± 124.49	0.721	811.40 ± 162.54	593.60 ± 84.02	0.068	805.20 ± 142.95	803.00 ± 157.51	0.465
IL-1β (pg.mL <sup>-1</sup> )	213.90 ± 70.82	205.40 ± 78.41	0.917	343.20 ± 159.96	195.40 ± 101.76	1.000	234.40 ± 72.34	230.20 ± 81.56	0.068
MCP-1 (pg.mL <sup>-1</sup> )	12245.00 ± 4702.55	4048.20 ± 513.73	0.374	4218.60 ± 615.44	4049.40 ± 869.92	0.500	6432.80 ± 3066.14	3897.60 ± 638.89	0.715

IL: interleukin. TNF-α: tumor necrosis factor alpha. MCP-1: monocyte chemoattractant protein-1. The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16 for each class of obesity (mean ± standard error).

**Table S8.** Oxidative stress biomarkers of patients with different classes of obesity before (T0) and after (T16) the consumption of Pera and Moro orange juices

	Obese I class (n = 12)			Obese II class (n = 5)			Obese III class (n = 6)		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
<b>Pera orange juice</b>									
Plasma									
ORAC ( $\mu\text{M}$ eq Trolox.mL <sup>-1</sup> )	2.51 $\pm$ 0.23	2.81 $\pm$ 0.23	0.480	2.35 $\pm$ 0.35	2.86 $\pm$ 0.45	0.068	2.41 $\pm$ 0.19	2.62 $\pm$ 0.29	0.344
GPx (U.mg <sup>-1</sup> )	0.72 $\pm$ 0.14	0.59 $\pm$ 0.10	0.610	1.49 $\pm$ 0.31	0.68 $\pm$ 0.23	0.080	<b>0.91 <math>\pm</math> 0.16<sup>b</sup></b>	<b>1.01 <math>\pm</math> 0.06<sup>a</sup></b>	<b>0.046</b>
SOD (U.mg <sup>-1</sup> )	2.98 $\pm$ 0.46	3.66 $\pm$ 0.41	0.262	2.23 $\pm$ 0.94	2.34 $\pm$ 0.74	0.068	1.76 $\pm$ 0.67	2.68 $\pm$ 0.91	0.144
Urine									
8-isoprostane (pg.mg <sup>-1</sup> )	2.29 $\pm$ 0.35	2.53 $\pm$ 0.22	0.695	3.87 $\pm$ 2.21	1.78 $\pm$ 0.40	0.893	2.02 $\pm$ 0.35	1.37 $\pm$ 0.15	0.600
8-OHdG (ng.mg <sup>-1</sup> )	<b>1.53 <math>\pm</math> 0.30<sup>a</sup></b>	<b>0.58 <math>\pm</math> 0.15<sup>b</sup></b>	<b>0.002</b>	1.70 $\pm$ 0.78	0.32 $\pm$ 0.07	0.500	<b>1.73 <math>\pm</math> 0.59<sup>a</sup></b>	<b>0.38 <math>\pm</math> 0.08<sup>b</sup></b>	<b>0.028</b>
<b>Moro orange juice</b>									
Plasma									
ORAC ( $\mu\text{M}$ eq Trolox.mL <sup>-1</sup> )	2.79 $\pm$ 0.23	2.89 $\pm$ 0.25	0.099	<b>3.59 <math>\pm</math> 0.16<sup>a</sup></b>	<b>3.26 <math>\pm</math> 0.09<sup>b</sup></b>	<b>0.043</b>	3.01 $\pm$ 0.30	3.10 $\pm$ 0.28	0.463
GPx (U.mg <sup>-1</sup> )	0.93 $\pm$ 0.14	0.83 $\pm$ 0.11	0.444	1.19 $\pm$ 0.26	0.72 $\pm$ 0.16	0.144	1.37 $\pm$ 0.22	0.62 $\pm$ 0.08	0.674
SOD (U.mg <sup>-1</sup> )	3.98 $\pm$ 0.40	3.36 $\pm$ 0.51	0.386	3.39 $\pm$ 0.77	2.58 $\pm$ 0.57	1.000	3.33 $\pm$ 0.33	4.12 $\pm$ 0.42	0.465
Urine									
8-isoprostane (pg.mg <sup>-1</sup> )	2.51 $\pm$ 0.33	2.60 $\pm$ 0.46	0.508	1.82 $\pm$ 0.26	2.49 $\pm$ 1.19	0.686	2.91 $\pm$ 0.33	2.66 $\pm$ 0.45	0.116
8-OHdG (ng.mg <sup>-1</sup> )	<b>1.36 <math>\pm</math> 0.25<sup>a</sup></b>	<b>0.43 <math>\pm</math> 0.06<sup>b</sup></b>	<b>0.011</b>	<b>0.66 <math>\pm</math> 0.08<sup>a</sup></b>	<b>0.57 <math>\pm</math> 0.24<sup>b</sup></b>	<b>0.043</b>	<b>1.36 <math>\pm</math> 0.19<sup>a</sup></b>	<b>0.78 <math>\pm</math> 0.22<sup>b</sup></b>	<b>0.046</b>

8-OHdG: 8-hydroxy-2'-deoxyguanosine. GPx: glutathione Peroxidase. ORAC: oxygen radical absorbance capacity. SOD: superoxide dismutase.

The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16 for each class of obesity (mean  $\pm$  standard error).

**Table S9.** Relative abundance of significant gut microbiota phyla and Firmicutes/Bacteroidetes ratio in patients before (T0) and after (T16) Pera and Moro orange juice intake

	POJ			MOJ		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
Firmicutes	86.07 ± 2.55	85.18 ± 1.97	0.256	86.11 ± 1.62	85.45 ± 1.86	0.500
Bacteroidetes	6.84 ± 2.76	7.43 ± 1.97	0.394	4.57 ± 0.77	6.00 ± 1.04	0.122
F/B ratio	<b>60.66 ± 19.06<sup>a</sup></b>	<b>25.16 ± 7.96<sup>b</sup></b>	<b>0.041</b>	51.56 ± 23.70	40.00 ± 13.22	0.231
Actinobacteria	5.63 ± 1.20	4.87 ± 1.17	0.691	7.34 ± 1.80	6.86 ± 1.75	0.811
Proteobacteria	0.74 ± 0.27	1.50 ± 0.63	0.233	0.73 ± 0.23	0.69 ± 0.20	0.723
Verrucomicrobia	0.52 ± 0.21	0.84 ± 0.36	0.484	0.57 ± 0.25	0.76 ± 0.38	0.445
Others	0.42 ± 0.11	0.53 ± 0.14	0.173	0.96 ± 0.36	0.59 ± 0.14	0.528

POJ: Pera orange juice. MOJ: Moro orange juice. F/B: Firmicutes/Bacteroidetes ratio.

Results are expressed as the mean ± standard error (*n* = 16). The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16.



**Table S10.** Relative abundance of significant gut microbiota phyla and Firmicutes/Bacteroidetes ratio in patients with different classes of obesity before (T0) and after (T16) Pera and Moro orange juice intake

	Obese I class			Obese II class			Obese III class		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
<b>Pera orange juice</b>									
Firmicutes	87.96 ± 1.72	86.86 ± 1.91	0.260	81.45 ± 12.70	85.47 ± 7.17	0.593	84.99 ± 3.88	79.86 ± 4.72	0.285
Bacteroidetes	5.20 ± 1.68	4.39 ± 0.76	0.953	15.55 ± 13.45	12.17 ± 7.82	1.000	3.03 ± 1.53	11.82 ± 5.69	0.109
F/B ratio	61.98 ± 27.65	33.25 ± 12.61	0.374	49.43 ± 36.22	15.17 ± 6.44	0.285	67.93 ± 46.14	10.88 ± 4.54	0.109
Actinobacteria	5.19 ± 0.92	5.03 ± 1.18	0.953	2.39 ± 0.99	1.43 ± 0.67	0.109	10.19 ± 4.87	7.8 ± 4.55	1.000
Proteobacteria	0.54 ± 0.14	2.18 ± 0.99	0.139	0.44 ± 0.22	0.69 ± 0.48	1.000	1.64 ± 1.36	0.30 ± 0.14	1.000
Verrucomicrobia	0.67 ± 0.26	1.12 ± 0.43	0.345	0.09 ± 0.00	0.02 ± 0.00	-	0.08 ± 0.00	0.02 ± 0.00	-
Others	0.66 ± 0.16	0.80 ± 0.19	0.260	0.11 ± 0.06	0.16 ± 0.10	0.285	0.12 ± 0.09	0.22 ± 0.21	1.000
<b>Moro orange juice</b>									
Firmicutes	86.47 ± 1.89	86.41 ± 2.48	0.889	89.94 ± 1.85	89.77 ± 1.36	0.893	81.72 ± 4.24	79.61 ± 4.53	0.345
Bacteroidetes	5.93 ± 0.87	6.45 ± 1.78	0.575	4.68 ± 1.92	4.96 ± 1.27	0.686	<b>2.30 ± 1.08<sup>b</sup></b>	<b>6.33 ± 2.38<sup>a</sup></b>	<b>0.043</b>
F/B ratio	17.77 ± 3.45	38.69 ± 16.36	0.575	28.65 ± 6.17	25.04 ± 7.25	0.686	<b>128.53 ± 80.33<sup>a</sup></b>	<b>57.06 ± 41.74<sup>b</sup></b>	<b>0.043</b>
Actinobacteria	4.70 ± 1.77	4.93 ± 2.00	0.484	3.71 ± 1.63	3.36 ± 0.96	0.893	15.19 ± 4.06	13.47 ± 4.41	0.686
Proteobacteria	0.87 ± 0.39	0.78 ± 0.29	0.612	0.63 ± 0.40	0.89 ± 0.54	0.138	0.63 ± 0.46	0.35 ± 0.12	0.893
Verrucomicrobia	0.47 ± 0.25	0.83 ± 0.65	0.917	0.93 ± 0.70	0.75 ± 0.15	1.000	0.09 ± 0.00	0.37 ± 0.00	-
Others	1.78 ± 0.71	0.90 ± 0.24	0.779	0.48 ± 0.19	0.57 ± 0.26	0.225	0.14 ± 0.06	0.12 ± 0.06	0.500

F/B: Firmicutes/Bacteroidetes ratio. Results are expressed as the mean ± standard error. The *p*-values were calculated using the Wilcoxon test.

Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16 for each class of obesity.

**Table S11.** Relative abundance of significant gut microbiota OTUs (bacterial genera) in patients before (T0) and after (T16) Pera and Moro orange juices intake

ID	OTU	POJ			MOJ		
		T0	T16	p-value	T0	T16	p-value
01	[Eubacterium]_coprostanoligenes_group	2.77 ± 0.63	2.84 ± 0.60	0.820	<b>2.11 ± 0.53<sup>b</sup></b>	<b>2.97 ± 0.64<sup>a</sup></b>	<b>0.044</b>
02	[Eubacterium]_ruminantium_group	0.43 ± 0.12	1.10 ± 0.54	0.345	0.60 ± 0.16	0.48 ± 0.23	0.463
03	[Ruminococcus]_gauvreauui_group	0.86 ± 0.30	0.82 ± 0.32	0.173	0.88 ± 0.33	1.11 ± 0.37	0.570
04	[Ruminococcus]_gnavus_group	1.10 ± 0.71	0.23 ± 0.09	0.225	0.73 ± 0.51	1.36 ± 0.94	0.600
05	[Ruminococcus]_torques_group	<b>1.70 ± 0.50<sup>a</sup></b>	<b>1.01 ± 0.20<sup>b</sup></b>	<b>0.046</b>	0.84 ± 0.17	1.10 ± 0.30	0.653
06	Agathobacter	10.94 ± 3.65	6.04 ± 1.44	0.191	13.05 ± 1.92	9.85 ± 1.80	0.078
07	Akkermansia	0.52 ± 0.21	0.84 ± 0.36	0.484	0.57 ± 0.25	0.76 ± 0.37	0.445
08	Alistipes	0.91 ± 0.49	0.99 ± 0.41	0.125	0.50 ± 0.12	0.51 ± 0.10	0.811
09	Anaerostipes	1.40 ± 1.36	3.11 ± 2.30	0.138	0.29 ± 0.19	0.33 ± 0.19	0.441
10	Bacteroides	4.20 ± 1.88	4.64 ± 1.40	0.532	2.52 ± 0.50	3.74 ± 0.82	0.267
11	Bifidobacterium	4.31 ± 1.32	3.33 ± 1.14	0.433	4.62 ± 1.48	3.65 ± 1.15	0.352
12	Blautia	<b>6.91 ± 1.28<sup>a</sup></b>	<b>5.13 ± 1.07<sup>b</sup></b>	<b>0.011</b>	6.49 ± 1.20	6.02 ± 1.38	0.446
13	Butyricicoccus	0.69 ± 0.15	0.70 ± 0.14	0.955	0.97 ± 0.20	1.02 ± 0.14	0.586
14	Butyrivibrio	3.48 ± 1.98	1.68 ± 0.60	0.144	1.07 ± 0.97	1.14 ± 0.99	0.345
15	Catenibacterium	4.11 ± 1.70	4.04 ± 1.52	0.575	4.25 ± 1.32	4.77 ± 1.56	0.248
16	Christensenellaceae_R-7_group	3.69 ± 0.87	3.07 ± 0.75	0.609	2.83 ± 0.41	4.20 ± 0.99	0.679
17	Collinsella	1.24 ± 0.20	1.39 ± 0.27	0.861	2.48 ± 0.86	3.09 ± 1.42	0.535
18	Coprococcus_2	1.46 ± 0.58	1.93 ± 0.9	0.310	2.29 ± 0.59	2.42 ± 0.86	0.799
19	Coprococcus_3	1.37 ± 0.68	1.19 ± 0.42	0.778	1.18 ± 0.19	1.53 ± 0.41	0.918
20	Dialister	<b>1.52 ± 0.42<sup>b</sup></b>	<b>2.83 ± 0.80<sup>a</sup></b>	<b>0.050</b>	3.22 ± 0.51	3.11 ± 0.5	0.767
21	Dorea	2.34 ± 0.51	1.98 ± 0.41	0.496	2.54 ± 0.35	1.93 ± 0.32	0.094
22	Erysipelotrichaceae_UCG-003	2.07 ± 0.61	1.39 ± 0.54	0.071	1.84 ± 0.69	1.44 ± 0.34	0.701
23	Escherichia-Shigella	0.58 ± 0.42	1.35 ± 0.87	0.203	0.49 ± 0.24	0.27 ± 0.19	0.799
24	Faecalibacterium	5.49 ± 1.11	7.22 ± 1.07	0.233	6.54 ± 1.19	8.05 ± 1.34	0.078
25	Fusicatenibacter	2.22 ± 0.67	2.12 ± 0.62	0.551	1.61 ± 0.39	1.98 ± 0.42	0.349
26	Holdemanella	4.64 ± 1.31	3.80 ± 1.08	0.213	5.18 ± 1.76	4.17 ± 1.25	0.609
27	Lachnospiraceae_ND3007_group	0.52 ± 0.08	0.74 ± 0.17	0.169	0.53 ± 0.11	0.67 ± 0.10	0.363
28	Lachnospiraceae_NK4A136_group	0.60 ± 0.24	1.53 ± 0.87	0.776	0.61 ± 0.18	0.56 ± 0.11	0.943
29	Lactobacillus	0.15 ± 0.05	1.09 ± 0.60	0.139	0.70 ± 0.23	0.90 ± 0.41	0.878
30	Megamonas	0.52 ± 0.00	1.23 ± 0.00	-	2.67 ± 0.87	0.91 ± 0.65	0.180
31	Megasphaera	0.61 ± 0.24	0.49 ± 0.28	1.000	0.49 ± 0.10	0.54 ± 0.25	1.000
32	Prevotella_9	0.63 ± 0.44	0.44 ± 0.14	0.674	1.06 ± 0.30	1.13 ± 0.48	0.678
33	Roseburia	1.57 ± 0.34	2.27 ± 0.66	0.394	1.49 ± 0.35	1.34 ± 0.25	0.267
34	Ruminiclostridium_5	0.97 ± 0.34	0.53 ± 0.08	1.000	0.54 ± 0.09	0.53 ± 0.09	0.948
35	Ruminococcaceae_NK4A214_group	0.79 ± 0.19	1.22 ± 0.24	0.096	0.99 ± 0.24	0.85 ± 0.20	0.463
36	Ruminococcaceae_UCG-002	2.16 ± 0.54	3.07 ± 0.57	0.078	2.18 ± 0.54	2.28 ± 0.39	0.569

**Table S11. Cont.**

ID	OTU	POJ			MOJ		
		T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
37	Ruminococcaceae_UCG-005	1.15 ± 0.30	1.41 ± 0.31	0.109	1.14 ± 0.25	1.27 ± 0.22	0.523
38	Ruminococcaceae_UCG-013	1.24 ± 0.37	0.85 ± 0.18	0.460	1.17 ± 0.42	0.96 ± 0.19	0.943
39	Ruminococcaceae_UCG-014	2.84 ± 0.86	2.87 ± 1.09	0.878	2.07 ± 0.58	1.78 ± 0.43	0.424
40	Ruminococcus_1	1.39 ± 0.34	1.76 ± 0.38	0.397	1.25 ± 0.25	1.48 ± 0.24	0.246
41	Ruminococcus_2	6.99 ± 1.42	6.81 ± 1.73	0.730	6.67 ± 1.83	6.64 ± 1.95	0.831
42	Streptococcus	1.57 ± 0.58	2.41 ± 1.40	0.683	2.14 ± 0.84	2.59 ± 1.48	0.687
43	Subdoligranulum	7.50 ± 1.43	7.94 ± 1.20	0.826	8.39 ± 1.38	7.50 ± 1.22	0.407
44	Succinivibrio	0.20 ± 0.11	1.22 ± 0.56	0.285	0.81 ± 0.79	0.43 ± 0.20	1.000
45	Tyzzarella_3	0.46 ± 0.26	0.64 ± 0.43	1.000	0.51 ± 0.30	0.49 ± 0.24	0.655
	Uncultured	1.21 ± 0.28	1.62 ± 0.33	0.173	0.96 ± 0.20	1.03 ± 0.18	0.500
	Others	11.66 ± 1.39	12.91 ± 0.92	0.173	10.11 ± 0.74	10.02 ± 0.65	0.711

OTU: Operational taxonomic unit. POJ: Pera orange juice. MOJ: Moro orange juice.

Results are expressed as the mean ± standard error (*n* = 16). The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16.

**Table S12.** Relative abundance of significant gut microbiota OTUs (bacterial genera) in patients with different classes of obesity before (T0) and after (T16) Pera and Moro orange juice intake

ID	OTU	Obese I class			Obese II class			Obese III class		
		T0	T16	p-value	T0	T16	p-value	T0	T16	p-value
Pera Orange juice										
01	[Eubacterium]_coprostanoligenes_group	4.09 ± 0.74	3.90 ± 0.66	0.767	1.28 ± 0.60	2.08 ± 1.51	0.593	0.30 ± 0.08	0.42 ± 0.30	1.000
02	[Eubacterium]_ruminantium_group	0.39 ± 0.15	0.63 ± 0.33	0.715	0.58 ± 0.00	2.98 ± 0.00	-	N.D.	N.D.	-
03	[Ruminococcus]_gauvreauii_group	0.73 ± 0.20	0.70 ± 0.36	0.110	0.23 ± 0.16	0.44 ± 0.27	0.285	3.91 ± 0.00	3.09 ± 0.00	-
04	[Ruminococcus]_gnavus_group	0.17 ± 0.07	0.15 ± 0.09	0.180	0.11 ± 0.00	0.55 ± 0.00	-	2.52 ± 1.31	0.15 ± 0.03	0.180
05	[Ruminococcus]_torques_group	0.81 ± 0.23	0.72 ± 0.20	0.110	3.07 ± 1.51	1.31 ± 0.52	0.285	3.63 ± 2.10	1.89 ± 0.33	0.655
06	Agathobacter	5.04 ± 1.99	4.87 ± 1.49	0.767	16.23 ± 8.33	7.65 ± 5.43	0.285	23.38 ± 14.33	7.92 ± 3.40	0.109
07	Akkermansia	0.67 ± 0.26	1.12 ± 0.43	0.345	0.09 ± 0.00	0.02 ± 0.00	-	0.08 ± 0.00	0.02 ± 0.00	-
08	Alistipes	0.58 ± 0.20	0.67 ± 0.28	0.260	2.74 ± 2.40	2.38 ± 1.91	1.000	0.08 ± 0.04	0.53 ± 0.16	0.109
09	Anaerostipes	0.05 ± 0.02	1.21 ± 1.15	0.285	0.02 ± 0.00	0.02 ± 0.00	-	6.82 ± 0.00	11.91 ± 0.00	-
10	Bacteroides	2.51 ± 0.68	2.22 ± 0.55	0.859	10.75 ± 9.43	6.54 ± 4.40	1.000	2.74 ± 1.54	10 ± 4.60	0.109
11	Bifidobacterium	2.99 ± 0.92	2.60 ± 0.91	0.401	2.13 ± 0.00	0.73 ± 0.00	-	8.53 ± 4.29	6.15 ± 3.88	1.000
12	Blautia	6.77 ± 1.81	5.37 ± 1.52	0.110	3.46 ± 0.86	1.84 ± 0.54	0.109	10.80 ± 1.91	7.71 ± 1.73	0.109
13	Butyricicoccus	0.48 ± 0.05	0.65 ± 0.18	0.515	0.46 ± 0.23	0.41 ± 0.14	0.593	1.57 ± 0.51	1.12 ± 0.39	0.593
14	Butyrivibrio	4.63 ± 2.27	2.18 ± 0.48	0.109	0.01 ± 0.00	0.17 ± 0.00	-	N.D.	N.D.	-
15	Catenibacterium	3.64 ± 2.75	1.91 ± 0.76	0.893	3.80 ± 0.86	4.75 ± 2.10	0.655	7.08 ± 0.00	13.28 ± 0.00	-
16	Christensenellaceae_R-7_group	5.33 ± 1.11	4.31 ± 0.99	0.594	2.35 ± 0.52	2.27 ± 1.16	1.000	0.14 ± 0.10	0.16 ± 0.07	0.593
17	Collinsella	1.20 ± 0.27	1.46 ± 0.37	0.441	1.05 ± 0.46	0.81 ± 0.51	0.180	1.65 ± 0.05	1.62 ± 0.36	0.655
18	Coprococcus_2	1.27 ± 0.65	1.82 ± 1.06	0.249	2.58 ± 0.00	2.58 ± 0.00	-	N.D.	N.D.	-
19	Coprococcus_3	1.69 ± 1.06	1.26 ± 0.64	0.515	0.67 ± 0.35	0.96 ± 0.52	0.285	0.94 ± 0.50	1.20 ± 0.25	0.180

**Table S12. Cont.**

ID	OTU	Obese I class			Obese II class			Obese III class		
		T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
20	Dialister	1.40 ± 0.52	2.41 ± 1.01	0.173	N.D.	N.D.	-	1.88 ± 0.88	4.10 ± 0.68	0.180
21	Dorea	2.48 ± 0.69	2.20 ± 0.65	0.314	2.51 ± 1.78	1.71 ± 0.60	1.000	1.78 ± 0.29	1.60 ± 0.40	0.285
22	Erysipelotrichaceae_UCG-003	1.77 ± 0.75	1.30 ± 0.64	0.263	0.73 ± 0.35	0.19 ± 0.13	0.180	4.63 ± 0.07	2.98 ± 1.93	0.655
23	Escherichia-Shigella	0.25 ± 0.12	2.57 ± 1.64	0.080	0.01 ± 0.00	0.25 ± 0.18	0.180	1.49 ± 1.42	0.07 ± 0.04	0.285
24	Faecalibacterium	6.87 ± 1.52	7.09 ± 1.31	0.953	3.42 ± 1.18	7.63 ± 3.10	0.109	3.41 ± 2.54	7.22 ± 3.08	0.285
25	Fusicatenibacter	1.58 ± 0.25	1.54 ± 0.30	0.767	5.94 ± 4.63	4.80 ± 4.49	0.180	1.67 ± 0.45	2.04 ± 1.07	0.593
26	Holdemanella	4.04 ± 1.69	3.04 ± 1.07	0.249	5.76 ± 3.47	3.88 ± 3.38	0.109	4.77 ± 3.64	5.97 ± 2.51	0.180
27	Lachnospiraceae_ND3007_group	0.58 ± 0.10	0.84 ± 0.22	0.176	0.50 ± 0.10	0.45 ± 0.30	0.655	0.13 ± 0.00	0.59 ± 0.00	-
28	Lachnospiraceae_NK4A136_group	0.91 ± 0.37	0.62 ± 0.22	0.374	0.16 ± 0.12	5.70 ± 3.89	0.285	0.11 ± 0.03	0.11 ± 0.04	1.000
29	Lactobacillus	0.14 ± 0.05	1.13 ± 0.91	0.249	0.43 ± 0.00	0.29 ± 0.00	-	0.02 ± 0.02	1.36 ± 0.38	0.180
30	Megamonas	N.D.	N.D.	-	N.D.	N.D.	-	0.52 ± 0.00	1.23 ± 0.00	-
31	Megasphaera	0.80 ± 0.19	0.49 ± 0.39	0.593	0.03 ± 0.00	0.48 ± 0.00	-	N.D.	N.D.	-
32	Prevotella_9	0.70 ± 0.50	0.40 ± 0.15	1.000	0.11 ± 0.00	0.73 ± 0.00	-	N.D.	N.D.	-
33	Roseburia	1.34 ± 0.23	2.67 ± 1.03	0.139	0.67 ± 0.42	0.62 ± 0.35	0.593	3.17 ± 1.19	2.72 ± 0.98	0.593
34	Ruminiclostridium_5	1.00 ± 0.53	0.45 ± 0.08	0.594	1.05 ± 0.64	0.63 ± 0.16	1.000	0.81 ± 0.43	0.69 ± 0.35	0.285
35	Ruminococcaceae_NK4A214_group	0.91 ± 0.26	1.38 ± 0.30	0.173	0.83 ± 0.38	1.42 ± 0.50	0.593	0.17 ± 0.16	0.21 ± 0.17	0.180
36	Ruminococcaceae_UCG-002	3.04 ± 0.74	3.36 ± 0.46	0.260	1.51 ± 0.44	4.96 ± 1.73	0.285	0.20 ± 0.17	0.32 ± 0.14	0.593
37	Ruminococcaceae_UCG-005	1.62 ± 0.38	1.59 ± 0.38	0.767	0.44 ± 0.15	1.69 ± 0.74	0.109	0.09 ± 0.07	0.21 ± 0.01	0.180
38	Ruminococcaceae_UCG-013	0.69 ± 0.19	0.62 ± 0.20	0.859	1.07 ± 0.46	0.87 ± 0.19	1.000	3.07 ± 1.39	1.54 ± 0.51	0.285
39	Ruminococcaceae_UCG-014	3.40 ± 0.98	3.58 ± 1.24	0.674	0.09 ± 0.00	0.01 ± 0.00	-	1.12 ± 0.00	0.06 ± 0.00	-
40	Ruminococcus_1	1.69 ± 0.48	2.01 ± 0.48	0.374	1.21 ± 0.43	1.66 ± 1.03	1.000	0.29 ± 0.29	0.78 ± 0.77	0.180

Table S12. Cont.

ID	OTU	Obese I class			Obese II class			Obese III class		
		T0	T16	p-value	T0	T16	p-value	T0	T16	p-value
41	Ruminococcus_2	6.35 ± 2.01	5.05 ± 1.82	0.767	10.65 ± 0.29	14.94 ± 2.60	0.109	4.36 ± 3.13	2.51 ± 1.10	0.655
42	Streptococcus	0.78 ± 0.29	1.56 ± 0.52	0.093	2.89 ± 2.64	0.16 ± 0.03	0.285	2.33 ± 0.72	6.94 ± 6.59	1.000
43	Subdoligranulum	9.67 ± 1.78	8.75 ± 1.41	0.515	3.34 ± 2.09	3.84 ± 1.52	0.593	4.03 ± 0.04	10.50 ± 4.15	0.180
44	Succinivibrio	0.11 ± 0.07	1.69 ± 0.53	0.180	0.40 ± 0.00	0.28 ± 0.00	-	N.D.	N.D.	-
45	Tyzzerella_3	0.60 ± 0.39	0.22 ± 0.20	0.180	N.D.	N.D.	-	0.18 ± 0.00	1.47 ± 0.00	-
	Uncultured	1.51 ± 0.37	1.92 ± 0.46	0.314	1.11 ± 0.77	1.94 ± 0.66	0.109	0.44 ± 0.19	0.42 ± 0.15	1.000
	Others	12.76 ± 1.61	14.52 ± 1.01	0.173	11.94 ± 5.15	11.76 ± 2.18	1.000	8.06 ± 1.42	9.21 ± 1.27	0.285
Moro orange juice										
01	[Eubacterium]_coprostanoligenes_group	2.42 ± 0.57	3.11 ± 1.03	0.208	3.04 ± 1.42	3.24 ± 1.13	0.345	0.31 ± 0.14	2.35 ± 1.42	0.068
02	[Eubacterium]_ruminantium_group	0.69 ± 0.21	0.61 ± 0.35	0.715	0.41 ± 0.23	0.23 ± 0.05	0.655	N.D.	N.D.	-
03	[Ruminococcus]_gauvreauii_group	0.78 ± 0.27	1.15 ± 0.56	0.866	0.37 ± 0.10	0.62 ± 0.21	0.138	1.97 ± 1.54	1.81 ± 1.37	0.593
04	[Ruminococcus]_gnavus_group	0.23 ± 0.09	0.72 ± 0.57	0.593	0.19 ± 0.00	0.06 ± 0.00	-	1.74 ± 1.51	2.98 ± 2.85	0.655
05	[Ruminococcus]_torques_group	0.79 ± 0.28	1.11 ± 0.48	0.866	0.81 ± 0.16	0.99 ± 0.27	0.686	0.93 ± 0.47	1.20 ± 0.83	0.893
06	Agathobacter	8.31 ± 2.05	9.65 ± 2.67	0.401	<b>17.29 ± 2.97<sup>a</sup></b>	<b>9.03 ± 2.74<sup>b</sup></b>	<b>0.043</b>	16.38 ± 4.44	11.01 ± 4.66	0.138
07	Akkermansia	0.47 ± 0.24	0.83 ± 0.64	0.917	0.93 ± 0.70	0.75 ± 0.15	1.000	0.09 ± 0.00	0.37 ± 0.00	-
08	Alistipes	0.56 ± 0.15	0.46 ± 0.14	0.208	0.75 ± 0.34	0.72 ± 0.26	0.893	<b>0.14 ± 0.08<sup>b</sup></b>	<b>0.38 ± 0.15<sup>a</sup></b>	<b>0.043</b>
09	Anaerostipes	0.14 ± 0.07	0.52 ± 0.41	0.273	0.05 ± 0.00	0.12 ± 0.00		0.50 ± 0.44	0.20 ± 0.14	0.465
10	Bacteroides	2.94 ± 0.67	3.65 ± 1.15	1.000	2.54 ± 1.26	2.50 ± 1.07	0.893	<b>1.82 ± 0.92<sup>b</sup></b>	<b>5.14 ± 2.15<sup>a</sup></b>	<b>0.043</b>
11	Bifidobacterium	1.57 ± 0.42	1.93 ± 0.83	0.866	2.83 ± 1.80	2.10 ± 0.89	0.715	10.30 ± 3.45	7.31 ± 3.03	0.138
12	Blautia	5.93 ± 1.65	5.66 ± 2.47	0.575	3.07 ± 1.23	3.47 ± 1.08	0.686	10.79 ± 2.33	9.15 ± 2.60	0.225
13	Butyricicoccus	1.03 ± 0.28	0.76 ± 0.15	0.401	0.37 ± 0.08 <sup>b</sup>	1.19 ± 0.37 <sup>a</sup>	0.043	1.49 ± 0.51	1.25 ± 0.24	0.893

**Table S12. Cont.**

ID	OTU	Obese I class			Obese II class			Obese III class		
		T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
14	Butyrivibrio	1.54 ± 1.46	1.53 ± 1.52	1.000	0.14 ± 0.12	0.36 ± 0.31	0.180	N.D.	N.D.	-
15	Catenibacterium	1.78 ± 1.20	3.92 ± 2.94	0.225	5.05 ± 2.04	6.14 ± 0.86	0.285	7.57 ± 3.47	4.83 ± 3.64	1.000
16	Christensenellaceae_R-7_group	3.59 ± 0.62	4.61 ± 1.32	0.889	2.04 ± 0.27	4.24 ± 2.12	0.345	2.10 ± 1.17	3.03 ± 2.61	1.000
17	Collinsella	2.33 ± 1.52	2.54 ± 1.59	0.161	0.69 ± 0.12	0.65 ± 0.09	0.715	4.55 ± 1.21	6.63 ± 4.71	1.000
18	Coprococcus_2	2.15 ± 0.91	1.50 ± 0.48	0.463	2.22 ± 0.92	1.94 ± 0.96	0.593	3.29 ± 0.00	9.33 ± 0.00	-
19	Coprococcus_3	1.06 ± 0.26	1.58 ± 0.82	0.484	1.23 ± 0.39	1.43 ± 0.32	0.686	1.41 ± 0.51	1.58 ± 0.56	0.285
20	Dialister	2.58 ± 0.29	2.09 ± 0.62	0.715	2.95 ± 1.00	3.55 ± 0.61	0.593	4.91 ± 1.40	4.46 ± 1.18	0.180
21	Dorea	2.91 ± 0.67	2.25 ± 0.64	0.208	1.72 ± 0.37	1.78 ± 0.34	0.686	2.76 ± 0.44	1.56 ± 0.46	0.138
22	Erysipelotrichaceae_UCG-003	2.45 ± 1.41	1.45 ± 0.55	0.600	1.10 ± 0.79	1.65 ± 0.88	0.109	1.49 ± 0.72	1.27 ± 0.57	0.715
23	Escherichia-Shigella	0.26 ± 0.15	0.02 ± 0.01	0.225	0.40 ± 0.37	0.75 ± 0.61	0.109	1.17 ± 1.17	0.18 ± 0.18	0.655
24	Faecalibacterium	8.05 ± 1.94	9.78 ± 2.42	0.208	7.66 ± 2.33	8.09 ± 1.58	0.500	3.00 ± 1.23	5.22 ± 2.30	0.138
25	Fusicatenibacter	1.19 ± 0.25	2.23 ± 0.76	0.401	2.18 ± 1.33	1.44 ± 0.58	0.686	1.71 ± 0.53	2.12 ± 0.76	0.345
26	Holdemanella	3.47 ± 1.38	4.51 ± 2.74	0.753	6.45 ± 4.66	3.77 ± 1.21	0.686	6.14 ± 3.31	4.15 ± 2.41	0.068
27	Lachnospiraceae_ND3007_group	0.62 ± 0.17	0.82 ± 0.11	0.600	0.62 ± 0.21	0.62 ± 0.20	0.893	0.20 ± 0.09	0.45 ± 0.27	0.109
28	Lachnospiraceae_NK4A136_group	0.79 ± 0.25	0.63 ± 0.23	0.398	0.79 ± 0.46	0.64 ± 0.17	0.893	0.17 ± 0.08	0.36 ± 0.14	0.345
29	Lactobacillus	0.74 ± 0.42	0.33 ± 0.15	0.686	0.53 ± 0.16	0.55 ± 0.52	0.655	0.75 ± 0.41	2.08 ± 1.17	0.593
30	Megamonas	N.D.	N.D.	-	3.54 ± 0.00	0.26 ± 0.00	-	1.81 ± 0.00	1.57 ± 0.00	-
31	Megasphaera	0.50 ± 0.19	0.66 ± 0.55	0.655	0.48 ± 0.17	0.43 ± 0.23	0.655	N.D.	N.D.	-
32	Prevotella_9	1.42 ± 0.35	1.43 ± 0.68	0.917	0.85 ± 0.00	1.38 ± 0.00	-	0.08 ± 0.07	0.09 ± 0.02	0.655
33	Roseburia	1.43 ± 0.54	1.09 ± 0.25	0.674	1.93 ± 0.93	1.84 ± 0.70	0.500	1.14 ± 0.32	1.23 ± 0.40	0.500
34	Ruminiclostridium_5	0.40 ± 0.12	0.54 ± 0.20	0.401	0.69 ± 0.10	0.50 ± 0.09	0.225	0.60 ± 0.25	0.54 ± 0.12	0.893

**Table S12. Cont.**

ID	OTU	Obese I class			Obese II class			Obese III class		
		T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
35	Ruminococcaceae_NK4A214_group	1.13 ± 0.42	0.77 ± 0.24	0.123	1.27 ± 0.40	0.80 ± 0.32	0.225	0.37 ± 0.28	1.05 ± 0.68	0.068
36	Ruminococcaceae_UCG-002	2.36 ± 0.54	2.05 ± 0.62	0.575	3.04 ± 1.37	2.72 ± 0.63	0.893	0.29 ± 0.22	2.16 ± 0.92	0.109
37	Ruminococcaceae_UCG-005	1.50 ± 0.41	1.35 ± 0.31	0.779	1.16 ± 0.44	1.20 ± 0.36	0.893	0.38 ± 0.20	1.20 ± 0.68	0.068
38	Ruminococcaceae_UCG-013	1.36 ± 0.76	0.93 ± 0.30	0.575	0.71 ± 0.22	1.11 ± 0.38	0.138	1.38 ± 1.04	0.83 ± 0.36	0.465
39	Ruminococcaceae_UCG-014	<b>3.21 ± 0.71<sup>a</sup></b>	<b>1.75 ± 0.58<sup>b</sup></b>	<b>0.028</b>	0.99 ± 0.77	2.38 ± 1.09	0.109	0.25 ± 0.21	0.96 ± 0.72	0.180
40	Ruminococcus_1	1.75 ± 0.38	1.38 ± 0.29	0.484	1.26 ± 0.32	1.90 ± 0.52	0.225	0.24 ± 0.13	1.12 ± 0.59	0.068
41	Ruminococcus_2	9.25 ± 3.36	9.04 ± 3.62	0.779	6.48 ± 2.35	7.67 ± 2.23	0.500	1.77 ± 1.39	0.53 ± 0.30	0.715
42	Streptococcus	0.74 ± 0.27	0.25 ± 0.11	0.128	3.14 ± 2.80	5.24 ± 4.81	0.225	3.09 ± 0.71	3.21 ± 1.69	0.893
43	Subdoligranulum	6.72 ± 1.37	5.03 ± 1.08	0.093	6.22 ± 1.35	7.85 ± 2.49	0.345	14.46 ± 3.93	11.99 ± 2.63	0.715
44	Succinivibrio	1.21 ± 1.18	0.60 ± 0.19	0.655	0.01 ± 0.00	0.09 ± 0.00	-	N.D.	N.D.	-
45	Tyzzereella_3	0.80 ± 0.00	0.73 ± 0.00	-	0.21 ± 0.00	0.26 ± 0.00	-	N.D.	N.D.	-
	Uncultured	1.14 ± 0.30	0.98 ± 0.26	0.401	1.27 ± 0.46	1.51 ± 0.38	0.500	<b>0.37 ± 0.13<sup>b</sup></b>	<b>0.63 ± 0.20<sup>a</sup></b>	<b>0.043</b>
	Others	12.59 ± 0.51	11.14 ± 0.98	0.123	9.19 ± 1.36	10.19 ± 1.20	0.500	7.04 ± 0.85	8.06 ± 0.96	0.138

OTU: Operational taxonomic unit. Results are expressed as the mean ± standard error. The *p*-values were calculated using the Wilcoxon test.

Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16 for each class of obesity.



**Table S13.** Effect of Pera (POJ) and Moro orange juice (MOJ) intake on the fecal short-chain fatty acids

	POJ			MOJ		
	T0	T16	<i>p</i> -value	T0	T16	<i>p</i> -value
Acetate	<b>1.68 ± 0.12<sup>a</sup></b>	<b>1.05 ± 0.22<sup>b</sup></b>	<b>0.022</b>	0.44 ± 0.05	0.63 ± 0.14	0.157
Propionate	1.36 ± 0.06	1.13 ± 0.11	0.287	0.59 ± 0.07	0.45 ± 0.08	0.995
Butyrate	1.65 ± 0.07	1.51 ± 0.09	1.000	0.73 ± 0.01	0.70 ± 0.08	0.815
Isobutyrate	1.40 ± 0.05	1.14 ± 0.10	0.169	0.67 ± 0.01	0.49 ± 0.26	0.833

Results are expressed as the mean ± standard error. The *p*-values were calculated using the Wilcoxon test. Values in bold and different superscript letters indicate statistical significance (*p* < 0.05) between T0 and T16.