

**Supplementary Table 1.** Impact of Diet and *Ex Vivo* HMO Exposure on  $^3\text{H}$ -thymidine Incorporation (CPM) into PBMC as a Measure of Cellular Proliferation.

Treatment	FF	SR
Unstimulated	5099.0 ± 1003.6	1724.2 ± 1171.5
TriGal	6695.4 ± 2342.9	2059.2 ± 1179.5
sLeX	5176.6 ± 2391.3	1641.2 ± 1264.8
LNFPIII	6951.4 ± 3116.8	2074.7 ± 1146.4
6'-SL	5806.2 ± 3439.2	2249.8 ± 1793.6
3'-SL	5324.3 ± 2802.3	2053.6 ± 1763.4
SA	4296.3 ± 2395.4	2240.2 ± 1625.6
2'-FL	2090.9 ± 1542.0	1537.9 ± 1144.6
3'-FL	2813.8 ± 1156.2	1423.7 ± 830.7
LNnT	4085.4 ± 904.1	1542.4 ± 922.6
FL mix	3109.0 ± 1731.1	1218.9 ± 842.4
SL mix	3374.4 ± 3033.8	1732.6 ± 901.8
iHMO	2724.6 ± 1216.2	1801.1 ± 1049.7
LPS	4363.9 ± 2861.1	1911.1 ± 2353.5
LNnT + LPS	3747.7 ± 1828.3	2656.7 ± 875.7
FL mix + LPS	4220.9 ± 3096.9	3326.0 ± 819.3
SL mix + LPS	7428.9 ± 3916.8	4416.5 ± 850.5
iHMO + LPS	4791.1 ± 1962.5	2526.5 ± 722.2
PHA	33296.3 ± 25838.0	37066.6 ± 19181.4
LNnT + PHA	65456.9 ± 42244.3	65057.6 ± 12965.9
FL mix + PHA	56967.9 ± 41640.6	46354.3 ± 26199.7
SL mix + PHA	81476.0 ± 30898.6	55457.9 ± 17412.3
iHMO + PHA	75079.6 ± 27027.9	63107.2 ± 16206.6

Data are expressed as mean ± SD

Proc GLM: model p<0.0001, stimulant p=0.05, diet p<0.0001, and stimulation \* diet p=0.63.

When analyzing upon exclusion of cells stimulated with LPS or PHA, PBMC from SR pigs proliferated less ( $1485.1 \pm 1165.0$  CPM) than those isolated from FF pigs ( $3452.8 \pm 2525.9$  CPM).

**Supplementary Table 2.** Impact of Diet and 72h *Ex Vivo* HMO Exposure on Serum T Cell Populations (data expressed as a percent of total CD3+ events).

Treatment	T helper cells (CD3+CD4+CD8-) <sup>1</sup>		Cytotoxic T cells (CD3+CD4-CD8+) <sup>2</sup>		T helper to Cytotoxic T cells <sup>3</sup> Ratio		Double Positive T cells (CD3+CD4+CD8+) <sup>4</sup>	
	FF	SR	FF	SR	FF	SR	FF	SR
Unstimulated	65.8 ± 5.8	54.5 ± 9.0	3.9 ± 1.4	5.6 ± 1.9	18.3 ± 5.4	10.5 ± 3.1	1.7 ± 0.8	1.7 ± 0.5
TriGal	70.0 ± 6.0	59.8 ± 9.6	3.5 ± 1.8	6.2 ± 2.6	24.1 ± 10.6	11.0 ± 4.3	2.4 ± 2.1	2.2 ± 1.3
sLeX	69.2 ± 5.0	60.0 ± 8.4	3.9 ± 1.5	6.2 ± 2.2	19.9 ± 6.3	10.4 ± 2.8	2.0 ± 1.3	2.7 ± 1.9
LNnT	66.8 ± 7.6	60.8 ± 5.3	4.2 ± 1.8	4.7 ± 1.5	18.1 ± 6.0	13.9 ± 4.1	1.9 ± 0.9	2.5 ± 1.4
LNFPIII	66.6 ± 8.9	60.8 ± 11.0	4.5 ± 1.3	6.4 ± 2.6	15.9 ± 5.2	10.4 ± 3.0	1.8 ± 1.3	3.0 ± 2.0
6'-SL	65.0 ± 6.8	57.2 ± 7.7	3.8 ± 1.6	5.8 ± 2.6	19.9 ± 7.8	11.2 ± 3.9	1.9 ± 1.1	1.9 ± 0.7
3'-SL	69.0 ± 4.7	57.0 ± 12.1	3.7 ± 1.7	6.3 ± 2.3	23.0 ± 12.6	9.7 ± 2.7	1.9 ± 1.1	2.1 ± 1.1
SA	65.3 ± 7.6	53.7 ± 7.2	3.9 ± 1.1	5.8 ± 1.6	17.7 ± 4.6	9.6 ± 2.1	1.8 ± 1.1	1.6 ± 0.7
2'-FL	63.0 ± 6.2	46.8 ± 10.5	3.8 ± 1.5	5.8 ± 2.1	18.6 ± 6.9	8.6 ± 2.7	1.7 ± 1.2	1.2 ± 0.4
3'-FL	65.0 ± 6.5	54.8 ± 6.3	3.8 ± 1.4	5.2 ± 1.9	18.7 ± 6.4	11.5 ± 3.0	1.5 ± 1.0	1.3 ± 0.5
FL mix	65.7 ± 7.5	54.8 ± 11.3	4.2 ± 1.3	4.1 ± 2.8	17.0 ± 5.2	11.1 ± 1.8	1.9 ± 0.9	1.3 ± 0.7
SL mix	64.8 ± 6.2	52.5 ± 7.0	3.6 ± 1.7	5.4 ± 1.9	21.4 ± 9.7	10.6 ± 3.1	1.7 ± 0.9	1.3 ± 0.4
iHMO	66.9 ± 6.5	54.4 ± 4.4	3.7 ± 1.5	6.5 ± 1.3	21.1 ± 10.8	8.6 ± 1.4	1.9 ± 1.3	1.5 ± 0.4
LPS	76.4 ± 1.6	71.1 ± 4.6	3.6 ± 1.4	4.2 ± 2.8	24.0 ± 8.8	15.7 ± 8.5	3.9 ± 0.8	2.3 ± 0.1
LNnT + LPS	74.6 ± 4.8	66.4 ± 6.5	3.4 ± 0.8	3.0 ± 3.0	22.9 ± 4.8	28.9 ± 28.0	3.1 ± 1.2	2.0 ± 1.3
FL mix + LPS	72.5 ± 5.4	64.7 ± 5.5	3.7 ± 1.1	3.1 ± 3.1	21.2 ± 6.6	14.7 ± 8.1	3.3 ± 1.3	2.5 ± 1.9
SL mix + LPS	74.9 ± 3.2	66.2 ± 5.3	3.2 ± 1.1	4.8 ± 1.4	25.1 ± 7.2	15.1 ± 6.2	2.6 ± 1.3	1.9 ± 0.8
iHMO + LPS	72.4 ± 4.7	65.8 ± 6.1	3.8 ± 1.3	5.2 ± 1.4	21.1 ± 6.9	13.6 ± 5.2	2.5 ± 1.6	1.7 ± 0.4
PHA	79.8 ± 5.1	73.4 ± 4.7	2.3 ± 0.9	2.4 ± 0.6	40.4 ± 17.2	32.5 ± 9.1	9.0 ± 3.7	12.3 ± 1.9
LNnT + PHA	82.0 ± 3.9	68.9 ± 18.5	2.1 ± 0.9	1.6 ± 1.2	45.2 ± 14.8	34.8 ± 19.1	8.4 ± 2.2	8.2 ± 5.6
FL mix + PHA	81.5 ± 3.0	64.9 ± 16.9	2.0 ± 1.2	1.4 ± 1.3	52.7 ± 22.9	33.9 ± 10.3	8.9 ± 2.6	9.6 ± 6.0
SL mix + PHA	82.5 ± 4.1	72.7 ± 5.1	2.1 ± 0.9	2.6 ± 0.5	44.9 ± 19.0	28.8 ± 7.6	6.9 ± 2.3	8.9 ± 1.7
iHMO + PHA	81.9 ± 4.3	75.0 ± 5.2	2.1 ± 1.1	2.4 ± 0.6	50.1 ± 27.4	30.9 ± 11.9	6.8 ± 2.2	8.6 ± 1.9

Data are expressed as mean  $\pm$  SD

<sup>1</sup>Proc GLM: model p<0.0001, stimulant p<0.0001, diet p<0.0001, stimulant\*diet p=0.999.

<sup>2</sup>Proc GLM: model p<0.0001, stimulant p<0.0001, diet p<0.0001, stimulant\*diet p=0.87.

<sup>3</sup>Proc GLM: model p<0.0001, stimulant p<0.0001, diet p<0.0001, stimulant\*diet p=0.96.

<sup>4</sup>Proc GLM: model p=0.88, stimulant p=0.58, diet p=0.88, and stimulation \* diet p=0.90. When analyzing upon exclusion of cells stimulated with LPS or PHA, PBMC from SR pigs had similar CD4+CD8+ T cell populations ( $1.9 \pm 1.1$ ) to that isolated from FF pigs ( $1.9 \pm 1.1$ ).

**Supplementary Table 3.** Impact of Diet and 72h *Ex Vivo* HMO Exposure on Cytokine Production (pg/ml)

<b>Treatment</b>	<b>IL-10<sup>1</sup></b>		<b>TNF-α<sup>2</sup></b>		<b>IL-4<sup>3</sup></b>		<b>IFN-γ<sup>4</sup></b>	
	<b>FF</b>	<b>SR</b>	<b>FF</b>	<b>SR</b>	<b>FF</b>	<b>SR</b>	<b>FF</b>	<b>SR</b>
Unstimulated	9.3 ± 4.5	8.3 ± 4.7	7.3 ± 2.7	5.7 ± 1.8	2.2 ± 1.3	2.2 ± 0.7	14.6 ± 7.8	15.4 ± 11.5
TriGal	12.0 ± 7.7	36.3 ± 58.6	8.5 ± 3.4	8.1 ± 8.1	4.5 ± 5.1	3.6 ± 0.01	29.0 ± 36.2	57.5 ± 66.0
sLeX	13.2 ± 4.0	16.7 ± 17.0	7.4 ± 4.0	10.5 ± 13.1	4.5 ± 4.9	3.5 ± 3.0	43.5 ± 42.5	25.4 ± 35.8
LNnT	16.1 ± 12.2	5.8 ± 3.3	7.4 ± 4.7	4.8 ± 2.4	0.4 ± 0.01	2.7 ± 1.3	13.7 ± 10.3	25.2 ± 15.6
LNFPIII	11.2 ± 2.1	15.2 ± 13.7	9.7 ± 5.2	5.9 ± 5.0	4.1 ± 0.3	2.2 ± 1.3	20.7 ± 15.5	18.1 ± 10.8
6'-SL	10.4 ± 2.4	9.0 ± 5.4	6.0 ± 1.1	5.3 ± 2.9	2.0 ± 0.5	1.7 ± 0.7	18.4 ± 17.4	36.1 ± 42.2
3'-SL	13.0 ± 3.1	8.5 ± 4.6	12.3 ± 10.9	5.4 ± 2.6	4.1 ± 1.7	2.7 ± 1.5	39.9 ± 27.9	39.8 ± 32.2
SA	8.3 ± 3.5	7.3 ± 3.8	7.1 ± 2.3	4.7 ± 1.8	2.7 ± 1.6	3.9 ± 3.0	24.1 ± 15.5	18.8 ± 13.2
2'-FL	9.3 ± 7.2	14.9 ± 8.6	6.0 ± 4.9	5.4 ± 2.2	2.8 ± 2.0	2.4 ± 0.7	10.0 ± 5.0	21.2 ± 21.8
3'-FL	8.8 ± 4.4	11.9 ± 12.6	7.2 ± 3.3	5.3 ± 3.6	3.2 ± 1.7	1.4 ± 0.3	20.1 ± 15.1	32.3 ± 30.9
FL mix	9.8 ± 5.5	9.9 ± 8.3	5.7 ± 3.9	5.8 ± 2.8	0.9 ± 0.6	1.5 ± 0.9	11.4 ± 4.2	12.8 ± 8.8
SL mix	7.7 ± 2.6	18.4 ± 16.4	3.8 ± 2.3	9.6 ± 1.4	0.5 ± 0.3	1.3 ± 0.8	9.6 ± 2.8	10.7 ± 7.4
iHMO	17.3 ± 6.7	17.6 ± 10.8	7.9 ± 4.2	6.6 ± 3.4	1.9 ± 0.6	2.4 ± 1.3	17.6 ± 15.9	11.1 ± 4.1
LPS	234.6 ± 80.4	225.0 ± 160.7	45.2 ± 31.8	77.5 ± 47.3	3.3 ± 0.4	9.8 ± 6.5	165.9 ± 117.1	70.1 ± 66.5
LNnT + LPS	298.7 ± 137.5	281.7 ± 106.9	38.4 ± 15.3	65.6 ± 11.6	0.7 ± 0.3	2.7 ± 2.2	71.9 ± 53.6	36.7 ± 23.7
FL mix + LPS	261.2 ± 141.7	283.5 ± 111.8	32.2 ± 13.4	65.8 ± 13.3	1.6 ± 0.6	3.6 ± 2.9	49.5 ± 21.5	46.7 ± 29.5
SL mix + LPS	316.7 ± 205.8	301.2 ± 118.1	35.8 ± 16.8	96.2 ± 25.9	1.6 ± 1.3	4.3 ± 2.5	68.5 ± 71.2	79.9 ± 71.3
iHMO + LPS	341.3 ± 172.8	258.5 ± 103.7	46.1 ± 13.6	72.9 ± 37.5	2.1 ± 1.6	1.4 ± 0.6	79.3 ± 58.1	62.1 ± 53.1
PHA	175.4 ± 85.0	111.7 ± 42.1	56.0 ± 21.5	69.5 ± 45.1	296.2 ± 227.6	451.9 ± 269.9	401.0 ± 285.4	563.6 ± 518.6
LNnT + PHA	390.9 ± 484.3	231.6 ± 179.2	85.5 ± 39.5	98.3 ± 39.6	296.1 ± 266.7	408.7 ± 323.9	363.8 ± 202.9	433.4 ± 392.5
FL mix + PHA	213.6 ± 201.3	128.1 ± 64.4	55.0 ± 27.2	81.0 ± 21.3	304.5 ± 319.2	464.4 ± 290.7	326.5 ± 173.9	625.9 ± 778.8
SL mix + PHA	203.8 ± 126.3	115.9 ± 32.7	61.4 ± 26.1	76.1 ± 21.7	272.3 ± 191.3	455.3 ± 292.2	246.3 ± 111.2	477.4 ± 475.5
iHMO + PHA	315.5 ± 325.9	161.9 ± 74.3	63.0 ± 23.8	95.4 ± 50.7	312.8 ± 281.0	365.7 ± 266.5	369.9 ± 310.9	535.4 ± 521.9

Data are expressed as mean ± SD

<sup>1</sup> Proc GLM: model p<0.0001, stimulant p<0.0001, diet p=0.02, stimulant\*diet p=0.97.

<sup>2</sup>. Proc GLM: model p<0.0001, stimulant p<0.0001, diet p=0.10, stimulant\*diet p=0.01. The interaction between diet and stimulant was driven by the fact that diet had a strong effect on TNF- $\alpha$  production in response to PHA or LPS stimulation (p<0.0001) but no effect when PBMC were unstimulated or stimulated with HMO alone (p=0.13). When stimulated by PHA or LPS, PBMC from SR pigs produced more TNF- $\alpha$  ( $81.7 \pm 34.0$  pg/ml) than PBMC from FF pigs ( $52.1 \pm 26.8$  pg/ml).

<sup>3</sup>. Upon analysis of the PHA-stimulated samples, the full model was not significant (p=0.59). When the PHA-stimulated samples were analyzed by one-way ANOVA with diet as the factor, p=0.01. PBMC from SR pigs produced more IL-4 upon PHA stimulation ( $444.6 \pm 271.0$  pg/ml) than PBMC from FF pigs ( $296.4 \pm 238.6$  pg/ml).

<sup>4</sup>. Proc GLM: model p<0.0001, stimulant p<0.0001, diet p=0.83, stimulant\*diet p=0.998.PHA stimulation significantly increased IFN- $\gamma$  production by PBMC from 10 day-old pigs.