Supplementary Materials

Table S1. Forward and reverse primer sequences and final cDNA concentrations of all target genes, as determined in sigmoid biopsies.

Name	Gene description	Primer sequences	Final cDNA concentration
CAMP	Cathelicidin antimicrobial peptide	5′-AGGATTGTGACTTCAAGAAGGACG-3′ 5′-GTTTATTTCTCAGAGCCCAGAAGC-3′	80 ng/µl
CDH1	Cadherin 1	5′-CACCTGGAGAGAGGCCGCGT -3′ 5′-AACGGAGGCCTGATGGGGCG -3′	20 ng/µl
CLDN2	Claudin 2	5′-AACTACTACGATGCCTACC-3′ 5′-GAACTCACTCTTGACTTTGG -3′	20 ng/µl
CLDN3	Claudin 3	5′-TTCATCGGCAGCAACATCATC-3′ 5′-CGCCTGAAGGTCCTGTGG-3′	20 ng/µl
CLDN4	Claudin 4	5′-ACAGACAAGCCTTACTCC-3′ 5′-GGAAGAACAAAGCAGAG-3′	20 ng/µl
CTNNB1	Catenin beta 1	5′-GTGCTATCTGTCTGCTCTAGTA -3′ 5′-CTTCCTGTTTAGTTGCAGCATC -3′	20 ng/µl
DEFB1	Defensin beta 1	5′-CTCTGTCAGCTCAGCCTC-3′ 5′-CTTGCAGCACTTGGCCTTCCC-3′	20 ng/µl
GAPDH	Glyceraldehyde-3-phosphate dehydrogenase	5′-TGCACCACCAACTGCTTAGC-3′ 5′-GGCATGGACTGTGGTCATGAG-3′	20 ng/µl
IL1B	Interleukin 1 beta	5′-AAACAGATGAAGTGCTCCTTCCAGG-3′ 5′-TGGAGAACACCACTTGTTGCTCCA-3′	40 ng/µl
IL10	Interleukin 10	5′-TCAGGGTGGCGACTCTAT-3′ 5′-TGGGCTTCTTTCTAAATCGTTC-3′	80 ng/µl
MUC2	Mucin 2	5′-GTCAACCCTGCCGACACCTG-3′ 5′-ACTCACACCAGTAGAAAGGACAGC-3′	20 ng/µl
MLCK	Myosin light chain kinase	5-GCCTGACCACGAATATAAGTT-3′ 5′-GCTCCTTCTCATCATCATCTG-3′	20 ng/µl
OCLN	Occludin	5′-TCAGGGAATATCCACCTATCACTTCAG-3′ 5′-CATCAGCAGCAGCCATGTACTCTTCAC-3′	20 ng/µl
TFF3	Trefoil factor 3	5′-CTTGCTGTCCTCCAGCTCT-3′ 5′-CCGGTTGTTGCACTCCTT-3′	20 ng/µl
TJP1 (ZO-1)	Tight junction protein 1	5′-AGGGGCAGTGGTGGTTTTCTGTTCTTTC-3′ 5′-GCAGAGGTCAAAGTTCAAGGCTCAAGAGG-3′	20 ng/µl
TLR1	Toll like receptor 1	5′-CAGTGTCTGGTACACGCATGGT-3′ 5′-TTTCAAAAACCGTGTCTGTTAAGAGA-3′	80 ng/µl
TLR2	Toll like receptor 2	5′-GCCAAAGTCTTGATTGATTGG-3′	20 ng/µl

		5'-TATACCACAGGCCATGGAAAC-3'	
TLR4	Toll like receptor 4	5'-CCTGCGTGAGACCAGAAAGC-3'	80 ng/μl
		5'-TCAGCTCCATGCATTGATAAGTAATA-3'	
TLR6	Toll like receptor 6	5'-GAAGAAGAACAACCCTTTAGGATAGC-3'	20 ng/μl
		5'-AGGCAAACAAAATGGAAGCTT-3'	
TNF	Tumor Necrosis Factor	5'-CCGAGTGACAAGCCTGTAGC-3'	40 ng/μl
		5'-GAGGACCTGGGAGTAGATGAG-3'	
18S RNA	18S ribosomal RNA	5'-GTAACCCGTTGAACCCCATT-3'	20 ng/μl
		5'-CCATCCAATCGGTAGTAGCG-3'	

Table S2. Baseline characteristics of the subgroups of young adults (n=22) and elderly (n=22), undergoing sigmoidoscopy after the pectin or placebo intervention

	Young adults (n=22)			Elderly (n=22)		
	Pectin (n=12)	Placebo (n=10)	*P*-value	Pectin (n=12)	Placebo (n=10)	*P*-value
Age (yrs, mean ± SD)	23.9 ± 4.6	24.2 ± 5.4	0.893	70.4 ± 3.0	70.7 ± 2.8	0.784
Sex (% female)	58.3	30.0	0.184	41.7	20.0	0.277
BMI (kg/m², mean ± SD)	22.3 ± 2.8	23.7 ± 3.2	0.299	25.6 ± 2.6	27.3 ± 1.7	0.083
Serum CRP (mg/L, mean ± SD)	1.8 ± 1.8	0.6 ± 0.7	0.061	0.7 ± 0.6	1.8 ± 1.6	0.032
Medication (%)						
PPI				8.3	20.0	0.427
Statins	N.A.	N.A.	N.A.	8.3	0	0.350
Antihypertensives				0	10	0.262
Alcohol consumption (units/week, mean ± SD)	3.0 ± 2.7	6.3 ± 6.3	0.156	5.5 ± 5.0	10.8 ± 6.8	0.049

BMI: body mass index, CRP: C-reactive protein, N.A: not applicable, PPI: proton-pump inhibitors. Age, BMI, CRP and alcohol consumption were compared between intervention groups with the use of an independent samples t-test. Sex and medication were compared between intervention groups with the use of a Pearson Chi-square test.
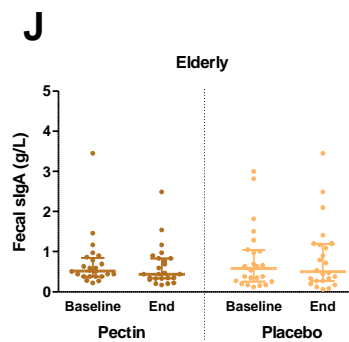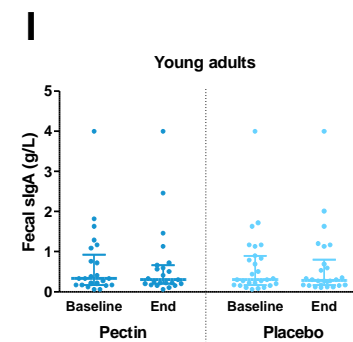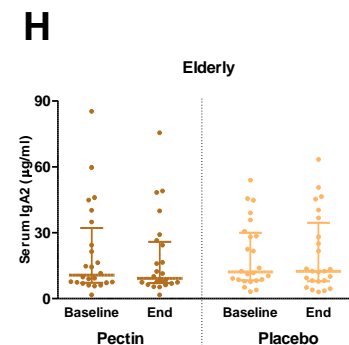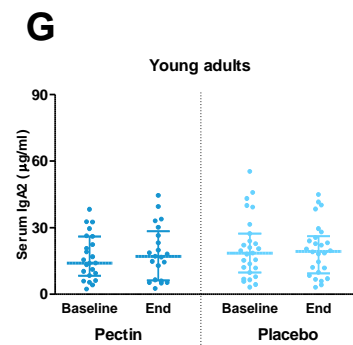
A Young adults

Salivary sIgA1 (μg/ml)

B Elderly

Salivary sIgA1 (μg/ml)

C Young adults

Salivary sIgA2 (μg/ml)

D Elderly

Salivary sIgA2 (μg/ml)

E Young adults

Serum IgA1 (μg/ml)

F Elderly

Serum IgA1 (μg/ml)

G Young adults

Serum IgA2 (μg/ml)

H Elderly

Serum IgA2 (μg/ml)

I Young adults

Fecal sIgA (g/L)

J Elderly

Fecal sIgA (g/L)

Baseline   End   Baseline   End
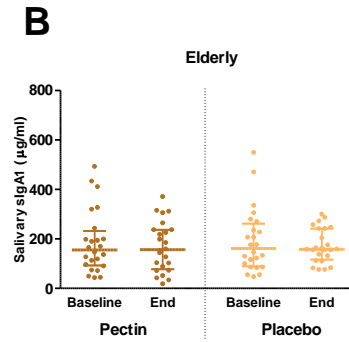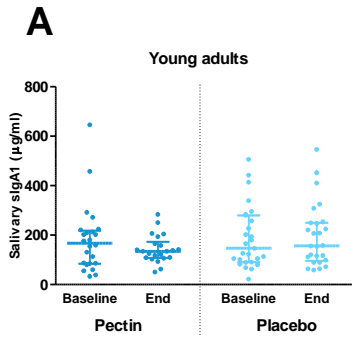Pectin   Placebo
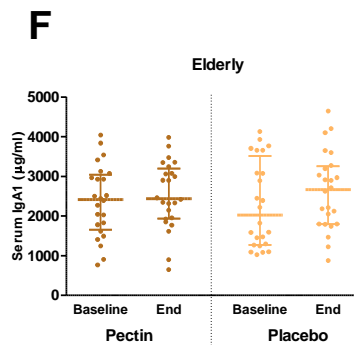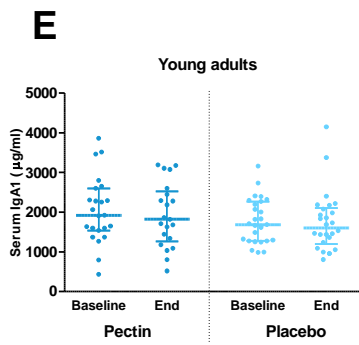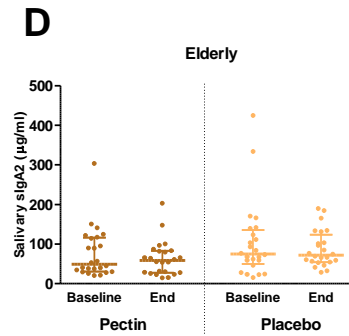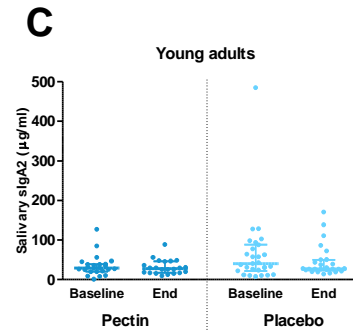
Figure S1. Mucosal defense parameters at baseline and after four weeks of pectin (circles) or placebo (triangles) intervention in young adults and elderly. A: Salivary sIgA1 (g/ml) in young adults. B: Salivary sIgA1 (g/ml) in elderly. C: Salivary sIgA2 (g/ml) in young adults. D: Salivary sIgA2 (g/ml) in elderly. E: Serum IgA1 (g/ml) in young adults. F: Serum IgA1 (g/ml) in elderly. G: Serum IgA2 (g/ml) in young adults. H: Serum IgA2 (g/ml) in elderly. I: Fecal sIgA (g/L) in young adults. J: Fecal sIgA (g/L) in elderly. Values are presented in scatter plots with median line and IQR (25-75th interquartile range). Sample sizes vary due to drop-outs and technical reasons. Within age groups, mucosal defense parameters were compared between intervention groups with unstructured linear mixed models and correction for baseline values. IgA, Immunoglobulin A; sIgA, secretory Immunoglobulin A.
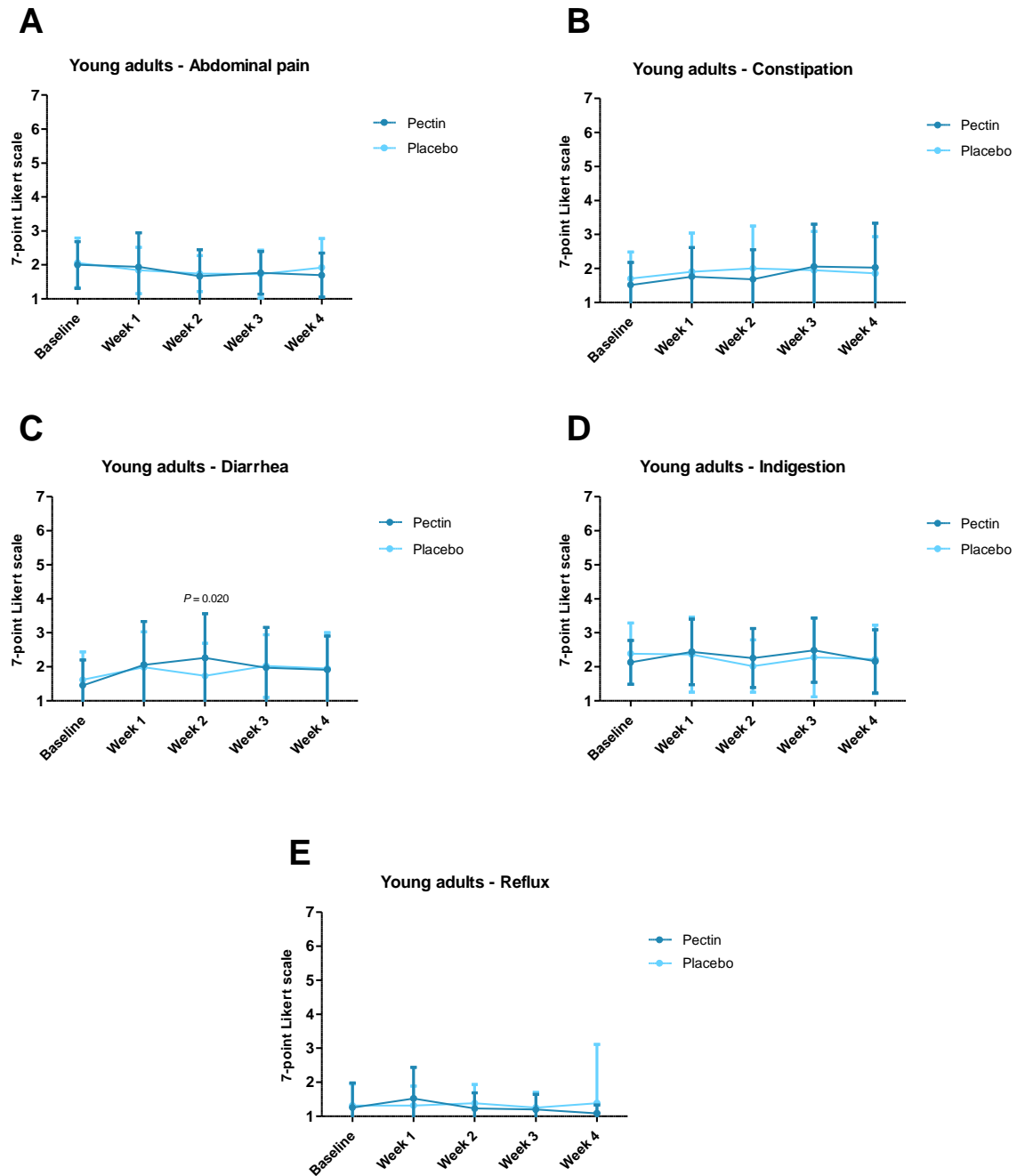

Figure S2. Gastrointestinal symptoms at baseline and every week of pectin (fixed lines) and placebo (dashed lines) intervention in young adults. A: Abdominal pain scores. B: Constipation scores. C:

Diarrhea scores. D: Indigestion scores. E: Reflux scores. Means and standard deviations are shown. Missing values at specific weeks were due to drop-outs. Gastrointestinal symptom scores were compared between intervention groups with random intercept linear mixed models and correction for baseline values. *P*-values per time point were corrected for multiple testing by calculating the false-discovery-rate (FDR) of Benjamini-Hochberg.
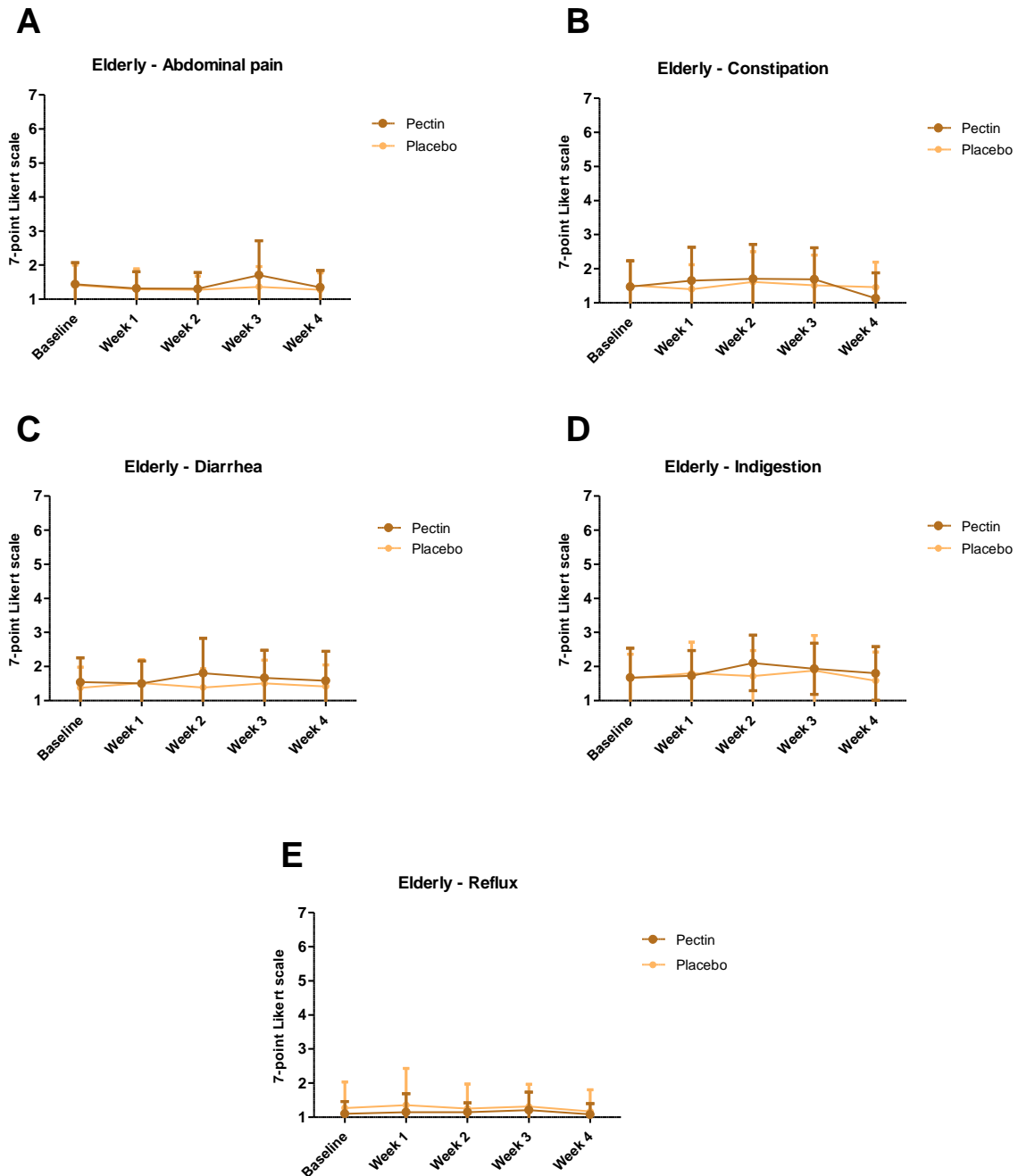

Figure S3. Gastrointestinal symptoms at baseline and every week of pectin (fixed lines) and placebo (dashed lines) intervention in elderly. A: Abdominal pain scores. B: Constipation scores. C: Diarrhea scores. D: Indigestion scores. E: Reflux scores. Means and standard deviations are shown. Gastrointestinal symptom scores were compared between intervention groups with random intercept linear mixed models and correction for baseline values. *P*-values per time point were corrected for multiple testing by calculating the false-discovery-rate (FDR) of Benjamini-Hochberg.
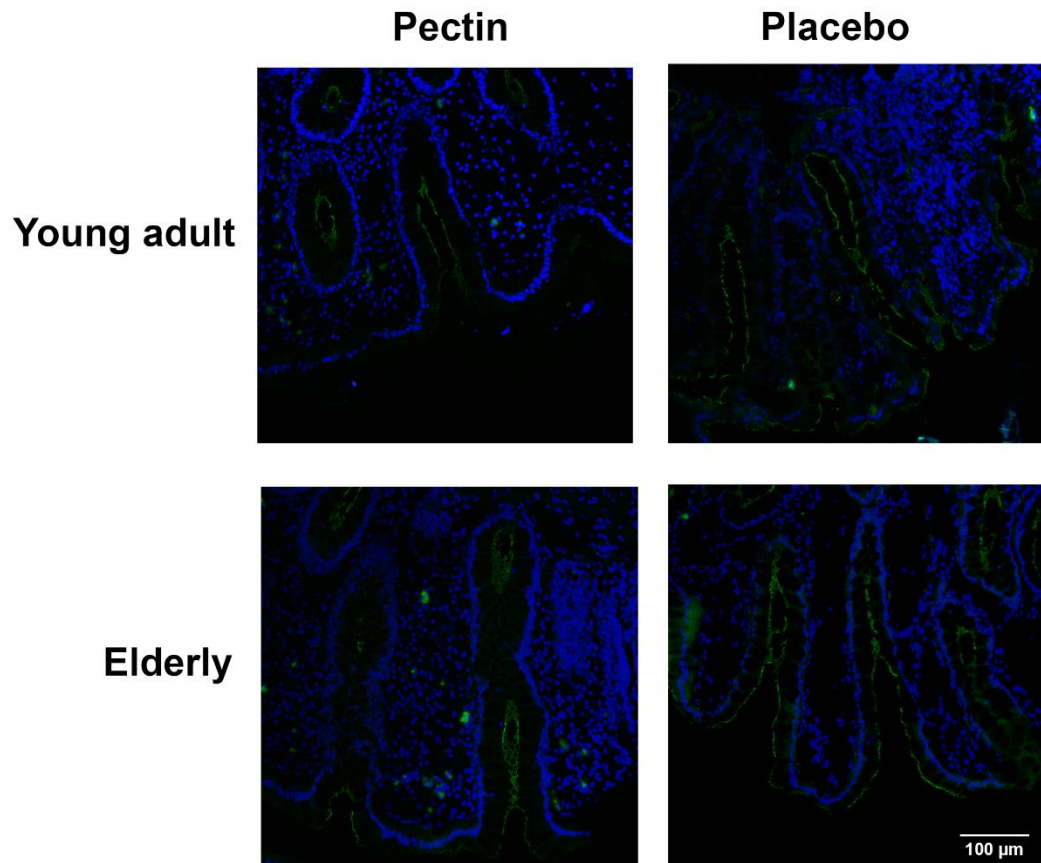
Figure S4. Representative images of tight junction protein TJP1 (green) immunofluorescence staining in sigmoid biopsy sections of a healthy young adult and healthy elderly after four weeks pectin or placebo intervention. Scale bar represents 100 μm. Blue counterstaining (DAPI) shows nuclei. TJP1: Tight junction protein 1.
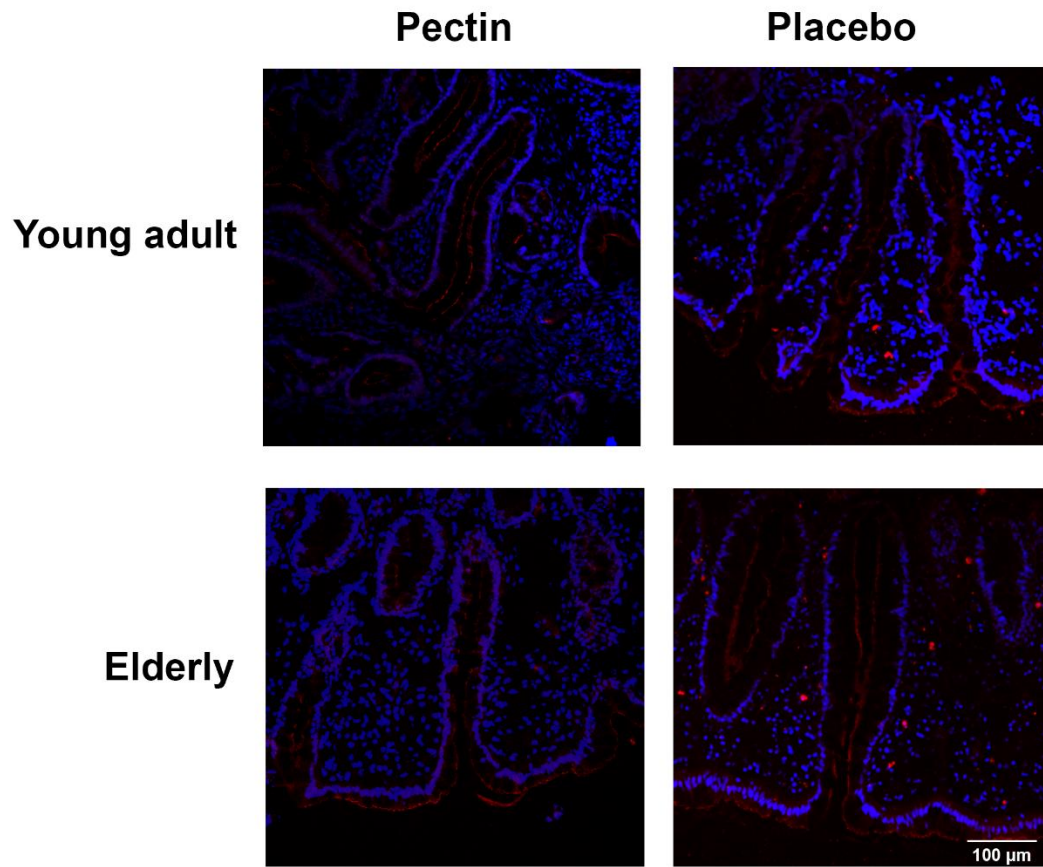
Figure S5. Representative images of tight junction protein occludin (red) immunofluorescence staining in sigmoid biopsy sections of a healthy young adult and healthy elderly after four weeks pectin or placebo intervention. Scale bar represents 100 μm. Blue counterstaining (DAPI) shows nuclei.