Electronic Supplementary Material (ESI) for Environmental Science: Nano.

Fate, uptake and gut toxicity of two colloidal silver products in mice: how micro X-ray fluorescence, micro X-ray absorption spectroscopy and near-infrared spectroscopy provide new insights in food nanotoxicology

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Supplementary Information

Table

Table S1. Oligonucleotide sequences for real-time PCR.

Gene	Forward primer (5'-3')	Reverse primer (5'-3')
Ocln	GCTGGATGACTACAGAGAGGAGA	TTTGCTCTTCAACTGCTTGC
Zo1	AATTATCCCACAAGGAGCCATT	ACACATCACTAGGGGGCTCAG
Cln1	GCTGTCATTGGGGGCATAATA	GGGGTCAAGGGGTCATAGAAT
Cln2	GGGCTGTTAGGCACATCCAT	GTCGCACACTCCATCCAGAG
Cln5	TTTCTTCTATGCGCAGTTGG	GCAGTTTGGTGCCTACTTCA
JamA	TCTTGATTTTTGGCGTCTGG	ACTGGGCTGGCTGTAAATGA
Lyz	CTGGCTGACTGGGTGTGTTTAG	GGATCTCTCACCACCCTCTTTG
Camp	AAGGAACAGGGGGTGGTG	CCGGGAAATTTTCTTGAACC
Muc1	TTGGTTGCTTTGGCTATCGT	TTACCTGCCGAAACCTCCT
Muc2	ATTATTCTGAAGCCTGGGGAGA	TGGAGCCTGAAACACAATCACT
Muc3	GTGATCCTCGTGATCCTCCTC	GTTTAGGGAAGGCTGGAAGGT
Muc4	GGCTCTGAACCTAAGTATGCTG	CCGTAGAATGCCCCAAGTTT
Tff3	CTGGTGCTTCAAACCTCTGC	GCCTGGACAGCTTCAAAATG
Tnfα	CCACGCTCTTCTGTCTACTGAAC	GGTCTGGGCCATAGAACTGATG
Il1β	GCCCATCCTCTGTGACTCAT	AGGCCACAGGTATTTTGTCG
Il6	TGATGGATGCTACCAAACTGG	AAGAGATAAGCTGGAGTCACAGAA
hprt	TCAGTCAACGGGGGACATAAA	GGGGCTGTACTGCTTAACCAG

Figures

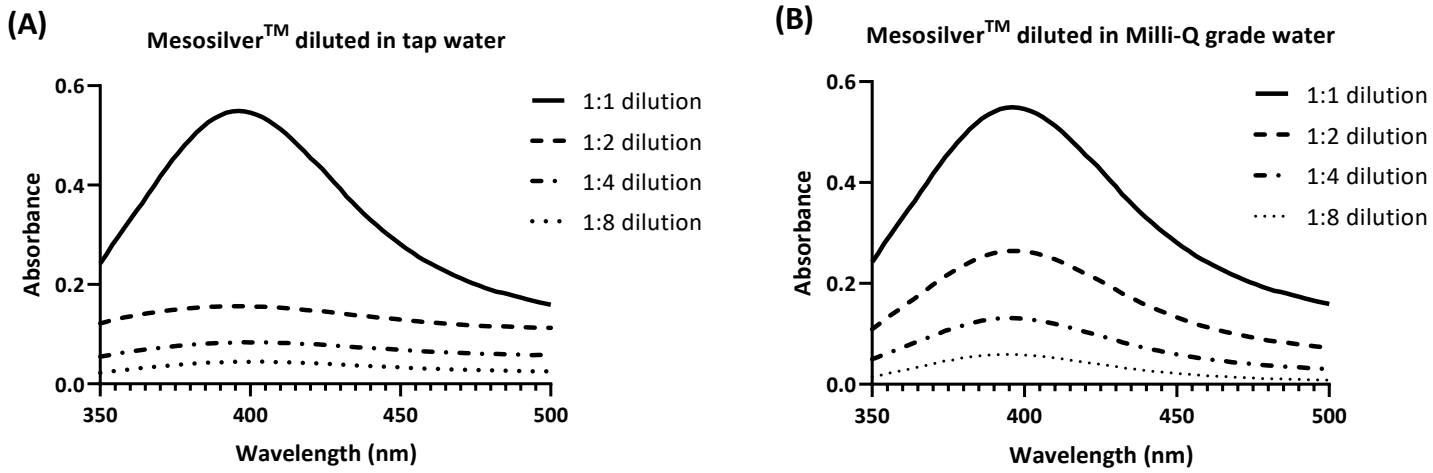

Figure S1. Changes in UV–vis absorbance of Mesosilver™ in its pure form and after dilution (1/2, 1/4 or 1/8) in (A) tap water and (B) Milli-Q grade water. The strong absorption peak at a maximum wavelength around 400 nm, as a result of plasmon resonance, is characteristic of AgNPs.

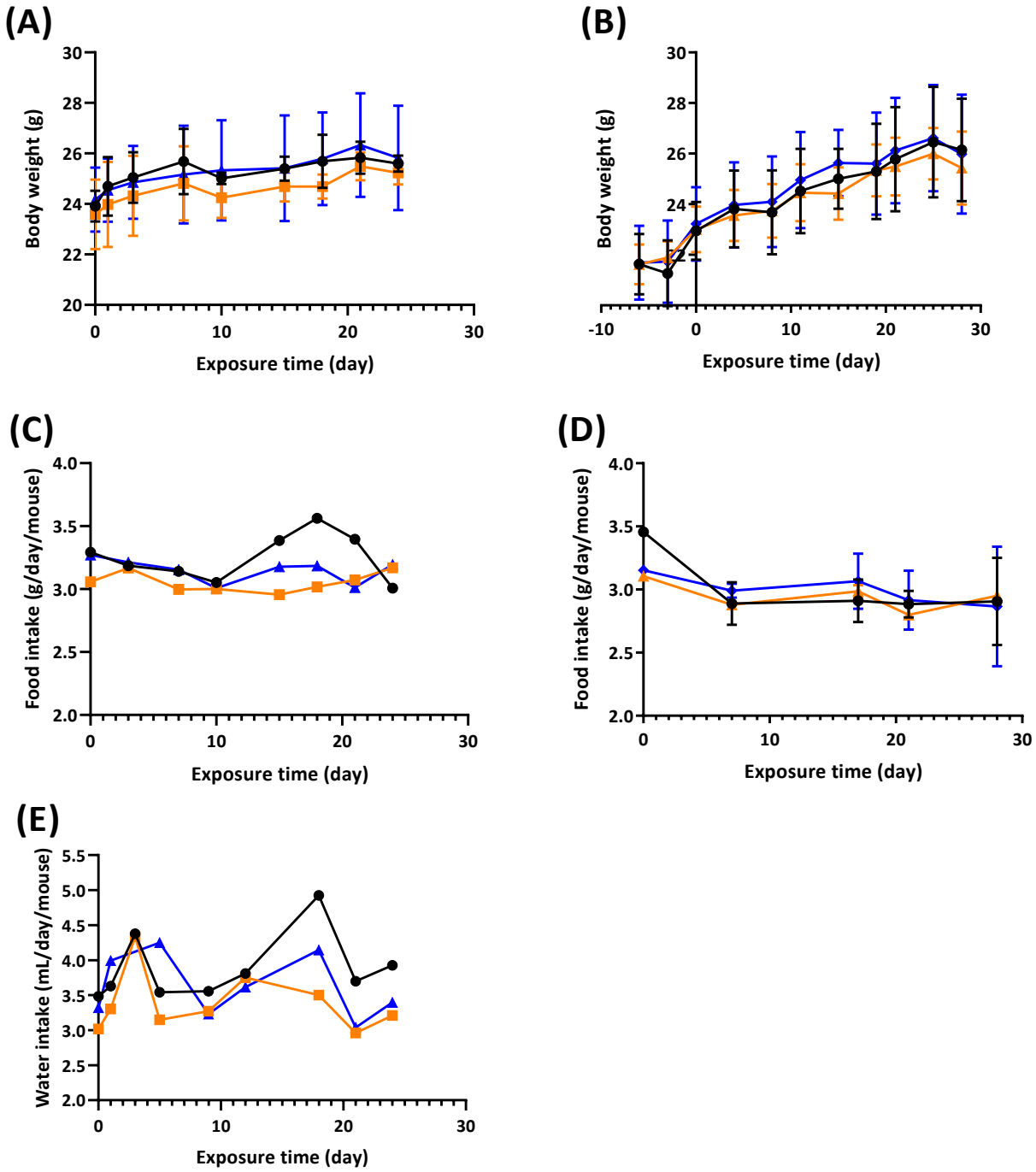

Figure S2. Monitoring of body weight (A,B), food intake (C,D) and water intake (E) of animals exposed to Mesosilver™ (in orange), AgC (in blue) or water (control; in black) for 28 days of administration via drinking (high dose of 3 mg/kg bw/day; n=3-4 animals per group) (A,C,E) or oral gavage (low dose of 150 µg/kg bw/day; n=12 animals per group) (B,D). For panels A, B and D, data are expressed as mean ± SD. For panels C and E, values correspond to the average intake for one cage housing the animals.

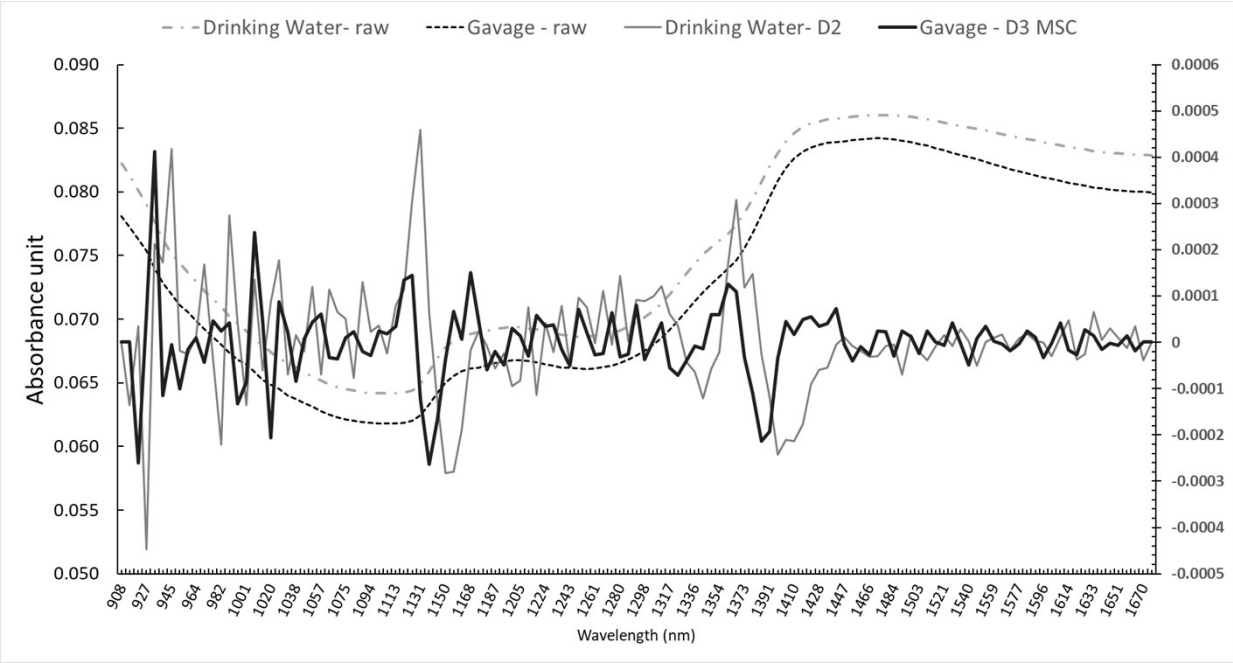

Figure S3. Average infrared absorbance spectra of contents and tissues of drinking water and oral gavage trials, before and after preprocessing.

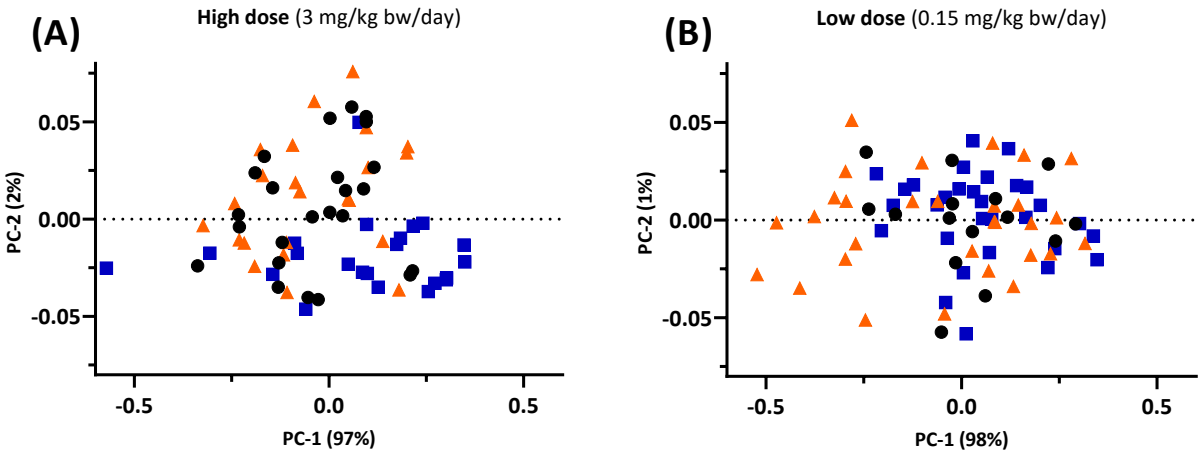

Figure S4. Score plot considering all NIRS spectra (combining liver, intestinal content and tissue conditions) after oral exposure to Mesosilver™ (orange triangles) or AgC (blue squares) vs control (black circles) for 28 days at high dose (3 mg/kg bw/day; 3 animals/group) (A) or low dose (150 µg/kg bw/day; 3 control, 5 Mesosilver™-treated and 5 AgC-treated mice) (B).

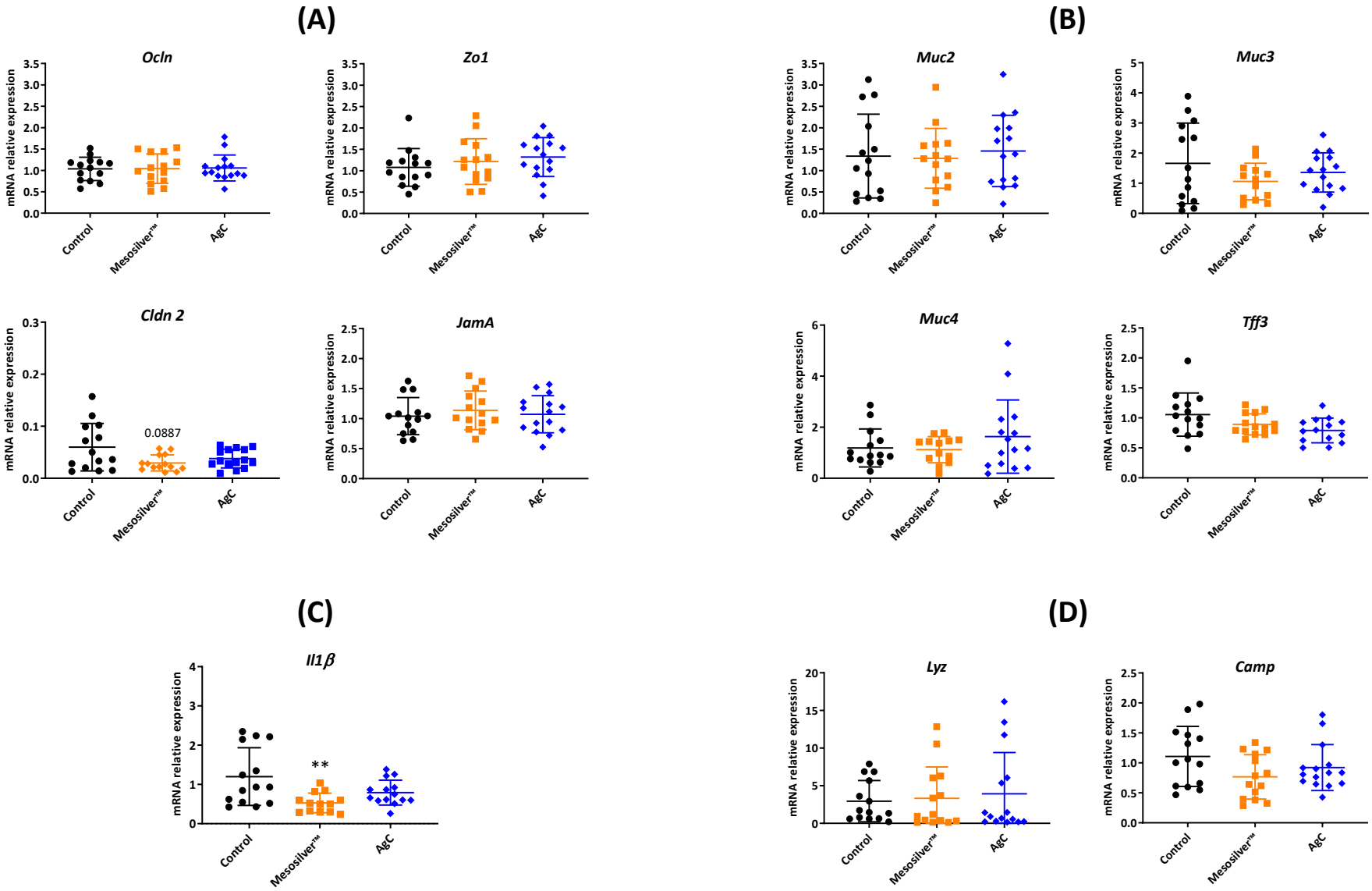

Figure S5. Effect of a 28-day sub-chronic exposure to Mesosilver™ (in orange) or AgC (in blue) at 150 µg/kg bw/day vs water (control; in black) on mRNA expression in the jejunum. The expression level of genes involved in (A) tight junction, (B) mucus pathways, (C) inflammation and (D) antimicrobial peptide was analysed. Expression was normalised to the reference gene *Hprt* and expressed as $2^{\Delta\Delta Ct}$. Data are expressed as mean ± SD; n=12 animals per group. Significant differences were assessed by one-way ANOVA with ** p-value < 0.01, compared to the control condition.

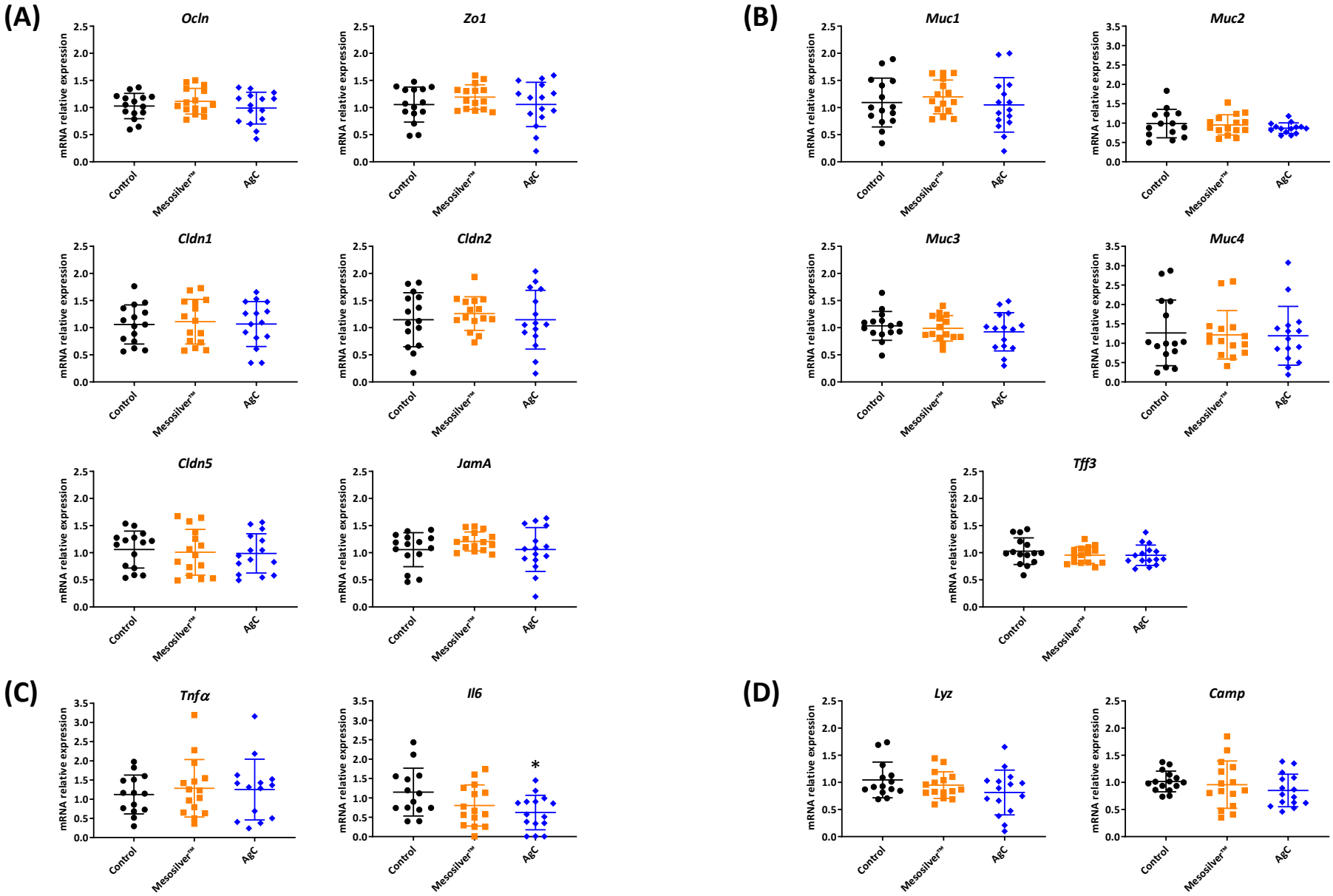

Figure S6. Effect of a 28-day sub-chronic exposure to Mesosilver™ (in orange) or AgC (in blue) at 150 µg/kg bw/day vs water (control; in black) on mRNA expression in the colon. The expression level of genes involved in (A) tight junction, (B) mucus, (C) inflammation and (D) antimicrobial peptide was analysed. Expression was normalised to the reference gene *Hprt* and expressed as $2^{\Delta\Delta Ct}$. Data are expressed as mean ± SD; n=12 animals per group. Significant differences were assessed by one-way ANOVA with * p-value < 0.05 compared to the control condition.