

Supplementary Information

Quantification of particulate Ag in rainbow trout organs following dietary exposure to silver nitrate, or two forms of engineered silver nanoparticles

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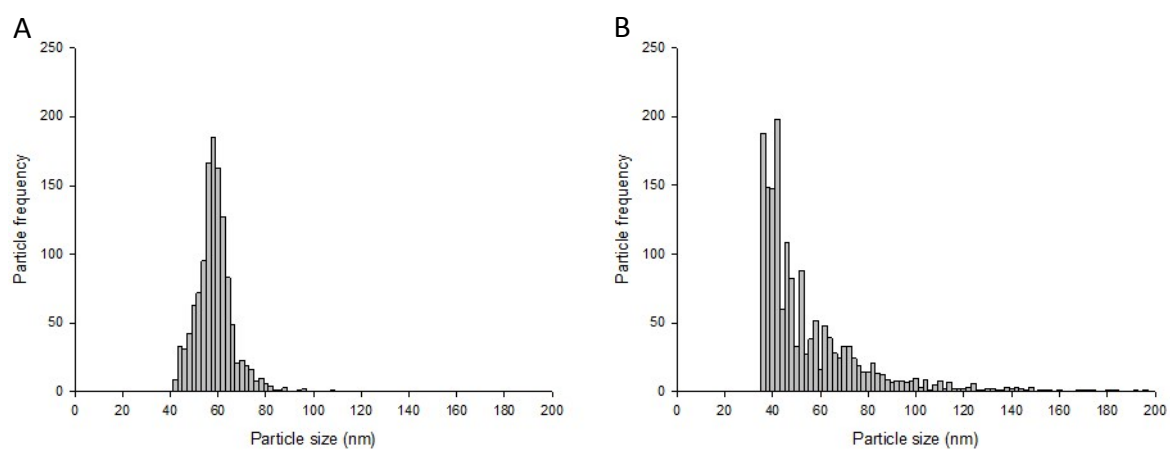


Figure S1. Example size distribution of (A) Ag NPs and (B) Ag₂S NPs in deionised ultrapure water that were subsequently added to the fish diet.

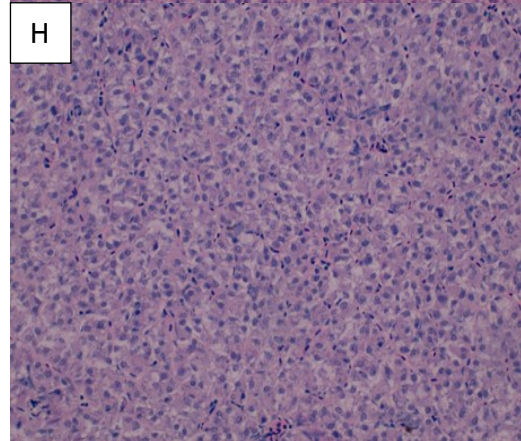
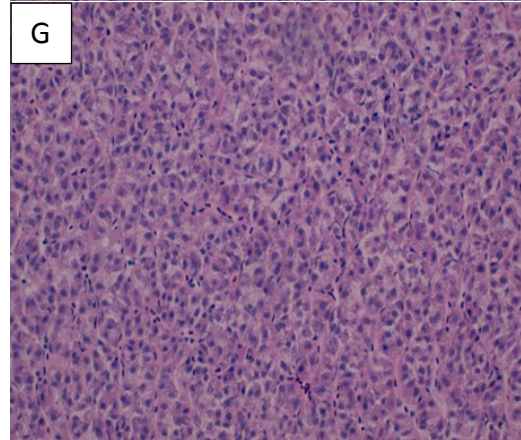
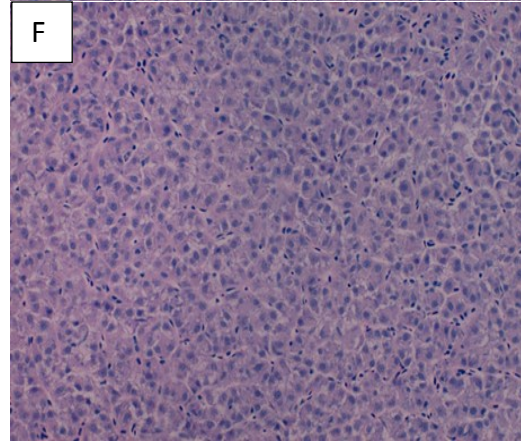
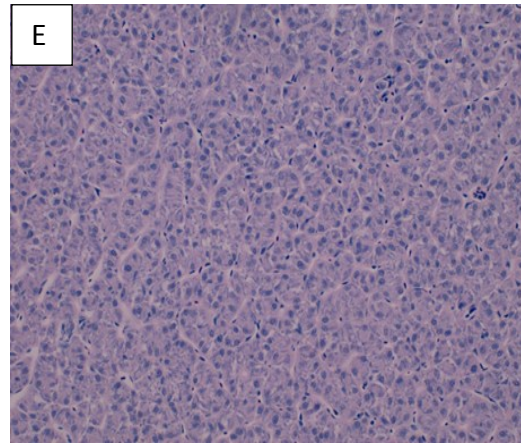
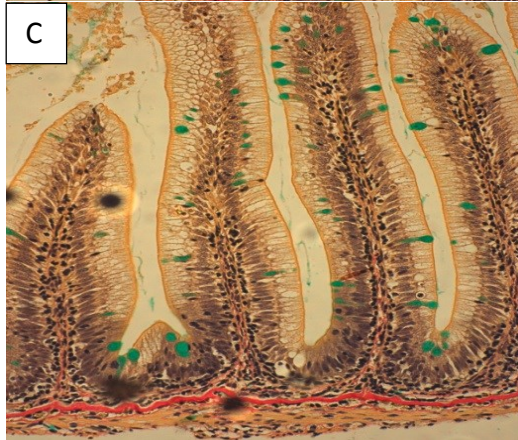
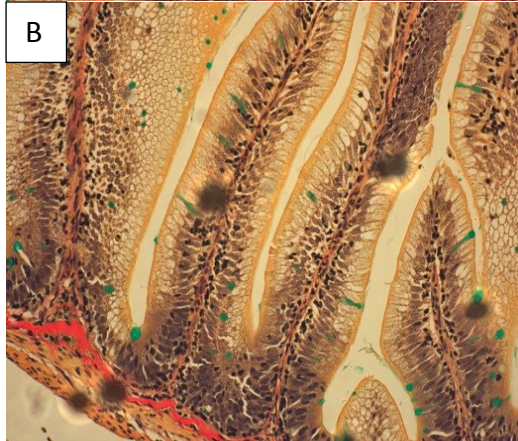
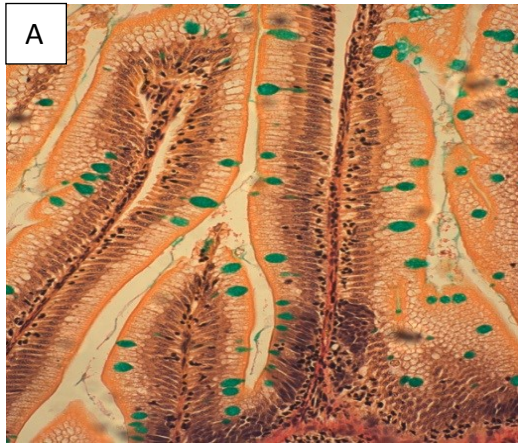


Figure S2. Histological sections of the hind intestine (left) and liver (right) following 4 weeks exposure to control (no added Ag; A, E), or 100 mg/kg of Ag as AgNO₃ (B, F), Ag NPs (C, G) or Ag₂S NPs (D and H). The intestines were stained using haematoxylin/ alcian blue/ van Gieson to highlight collagen (red), mucins (blue/turquoise), muscle and red blood cells (yellow), cytoplasm (pink/yellow) and nuclei (black/brown). The liver was stained with Haematoxylin and Eosin. Note: there was no evidence of pathology in the hind intestine or the liver.

Table S1. The speciation of 0.93 mmol L⁻¹ dissolved silver as percentage (%) of total Ag speciation under different physiological compartments at different concentrations.

Compartment	Percent species (%)
Stomach pH 2	
Ag ⁺	0.045
AgCl (aq)	6.725
AgCl ₂ ⁻	74.719
AgCl ₃ ⁻²	18.512
Upper GIT pH 7.8	
Ag ⁺	0.044
AgCl (aq)	6.741
AgCl ₂ ⁻	74.769
AgCl ₃ ⁻²	18.444
Lower GIT pH 9	
Ag ⁺	0.044
AgCl (aq)	6.741
AgCl ₂ ⁻	74.769
AgCl ₃ ⁻²	18.445
Blood compartment pH 7.8	
Ag ⁺	0.039
AgCl (aq)	6.528
AgCl ₂ ⁻	74.887
AgCl ₃ ⁻²	18.546
Intracellular compartments pH 7.4	
Ag ⁺	0.032
AgCl (aq)	5.865
AgCl ₂ ⁻	73.860
AgCl ₃ ⁻²	20.242

The GIT saline components were (in mmol L⁻¹) NaCl (117.5), KCl, (5.7), CaCl₂, (2.5) and MgSO₄ (1.2; Clark et al. 2019), with the only change between calculations the pH. The blood compartment saline components were (in mmol L⁻¹) NaCl (121.4), KCl (5.1), CaCl₂ (1.4) and MgSO₄ (1.9) at pH 7.8 (Clark et al. 2019). The intracellular components were (in mmol L⁻¹) KCl (140), Na (10), MgSO₄ (1) and CaCl₂ (0.1).