

Table 1 Standard Polystyrene and Commercial AgNP Hydrodynamic Diameter

Particle & Source	Hydrodynamic Diameter						
	Nominal Size (nm)	TEM Nominal Size (nm)	NTA Mean (nm)	NTA Mode (nm)	DLS_Z-Avg	(PDI)	Nominal Z-Avg (nm)
Polystyrene, NIST	100	100 ± 3	101 ± 0.3	94 ± 0.7	109 ± 0.7	0.03 ± 0.01	nr
Polystyrene, NIST	50	46 ± 2	43 ± 2.3	20 ± 0.9	61 ± 0.6	0.21 ± 0.02	57
Polystyrene, NIST	40	41 ± 4	45 ± 0.5	26 ± 1.7	42 ± 0.2	0.03 ± 0.01	nr
AgNP, NCX	5	4.6 ± 0.6	45 ± 2.1	26 ± 1.6	16 ± 0.49	0.3 ± 0.02	nr
AgNP, NCX	10	9.7 ± 1.8	76 ± 1.2	54 ± 3.3	86 ± 7.5	0.4 ± 0.06	19
AgNP, NCX	20	20.6 ± 3.1	50 ± 0.4	40 ± 0.6	88 ± 2.4	0.4 ± 0.06	41
AgNP, NCX	50	47 ± 4	84 ± 0.4	75 ± 1.1	80 ± 0.7	0.12 ± 0.01	62
AgNP, NCX	75	74.3 ± 3.9	96 ± 1.1	75 ± 0.8	105 ± 0.46	0.04 ± 0.01	98

Silver nanoparticles (AgNP)

Nanoparticle Tracking Analysis (NTA)

National Institute of Standards and Technology (NIST) secondary standard

NanoComposix (NCX)

nr not reported

Nominal, TEM Nominal and Nominal Z-Avg sizes were reported by the manufacturer

Table 2 Consumer product silver concentration claimed, measured, form (soluble, particulate)

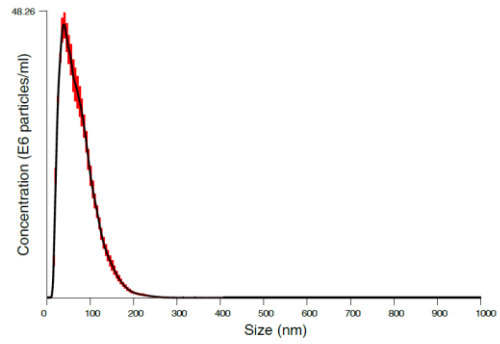
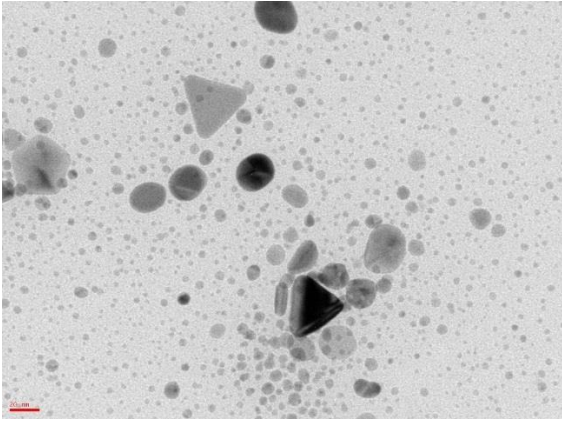
Product	Description	Color	Claimed Ag (mg/L)	Total Ag (mg/L)	Filtrate Ag (mg/L)
1	Colloidal/ ionic Ag, dietary supplement	colorless	120	17.9	18.6
2	Colloidal Ag, dietary supplement	colorless	100	1.6	nd
3	Colloidal Ag, dietary supplement	yellow-brown	15	1.6	nd
4	Colloidal Ag, immune support	dark brown	500	466	4.9
5	"silver-sol", ionic Ag, immune support	colorless	10	10.3	8.5
6	Colloidal/ ionic Ag, dietary supplement	dark brown	200	194.5	2.1
7	Colloidal Ag, foot spray	turbid yellow	nr	6.4	nd
8	Colloidal Ag, sanitizing spray	turbid yellow	nr	6.1	nd
9	"Silver-hydrosol" Ag, immune support	colorless	10	12.9	11.8
10	Colloidal/ ionic Ag, anti-infection	colorless	240	157.5	164
11	Colloidal/ ionic Ag, anti-infection	colorless	10	5.4	4.7
12	"Silver-sol" Ag, dietary supplement	colorless	10	9.7	9.2
13	Colloidal Ag, throat spray	orange	30	32.6	nd
14	Colloidal Ag, anti-microbial	colorless	20	12.6	10.1
15	Colloidal Ag, immune support	colorless	30	54.1	31.3
16	Colloidal Ag, dietary supplement	yellow-brown	20	23.7	10.0
17	Colloidal Ag	colorless	25	30.7	25.8
18	Ionic Ag, anti-microbial, immune support	colorless	10	10.3	10.8
19	Ionic Ag, first aid spray	colorless	20	15.1	11.5
20	Colloidal Ag, sanitizing spray, H <sub>2</sub> O <sub>2</sub>	yellow	nr	6.3	nd
21	Colloidal Ag, skin spray	colorless	30	50.5	31.6
22	Ionic Ag, dietary supplement	light yellow	20	19.6	14.6

nr not reported

nd not determined

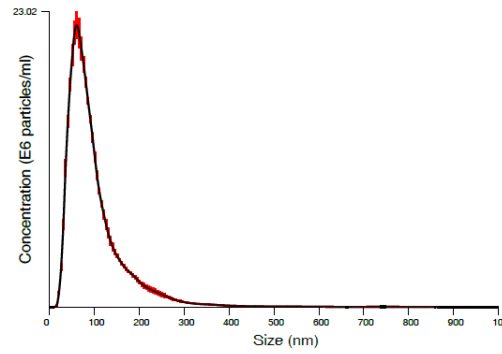
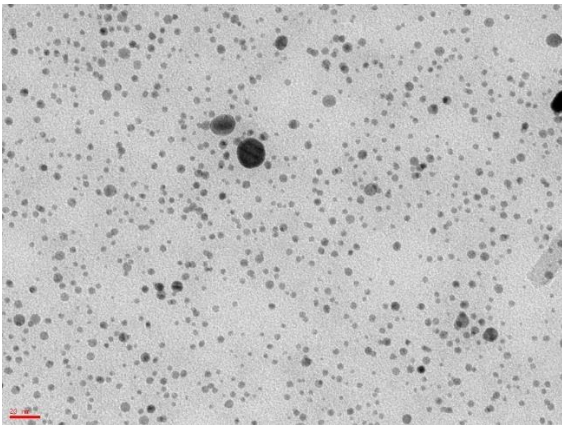
Table 3 Particle size; hydrodynamic diameter, primary particle size.

Product	Hydrodynamic Diameter				TEM Particle Diameter	
	NTA Mean (nm)	NTA Mode (nm)	DLS_Z-AVG (nm)	PDI	P-1 (nm)	P-2 (nm)
1	82 ± 1.8	52 ± 2.1	166.9 ± 31.47	0.43	2.3	30-200
2	na	na	284.7 ± 29.53	0.51	nd	200
3	39 ± 2	25 ± 0.4	264.1 ± 17.26	0.45	6.6	nd
4	71 ± 1.4	39 ± 1.3	57.46 ± 0.90	0.46	4.5	20-40
5	96 ± 4.8	66 ± 2	82.99 ± 5.59	0.69	9.1	20-60
6	70 ± 0.7	64 ± 3	55.91 ± 15.69	0.42	3.6	15-40
7	168 ± 23.2	63 ± 13.1	245.5 ± 11.62	0.29	15.5	15-30
8	39 ± 29.7	20 ± 13.4	220.2 ± 5.86	0.34	5.2	15-30
9	105 ± 8.1	64 ± 2.51	68.4 ± 100.2	0.47	10.1	15-100
10	220 ± 28.1	52 ± 6.2	484.4 ± 224.8	0.72	5.1	20
11	72 ± 0.4	64 ± 2.6	149.5 ± 84.55	0.58	2.7	20-40
12	75 ± 8.7	52 ± 4.2	181.5 ± 57.56	0.57	4.2	20-30
13	71 ± 0.7	56 ± 2.0	144.6 ± 58.51	0.52	9.1	20-50
14	93 ± 1.1	72 ± 1.7	119.3 ± 36.53	0.45	4	60
15	nd	nd	221.2 ± 9.53	0.43	4.2	20-40
16	105 ± 2.4	74 ± 7.7	58.8 ± 1.88	0.60	4.6	15-40
17	86 ± 1.6	45 ± 3.1	159.6 ± 28.04	0.40	2.8	20-40
18	127 ± 2.6	110 ± 10.9	479.6 ± 374.8	0.73	3.6	20-100
19	89 ± 1.7	64 ± 3.2	299.7 175.9	0.67	6.2	20-100
20	225 ± 142.5	171 ± 115.1	421 ± 40.41	0.43	nd	nd
21	nd	nd	126.8 ± 245		2.7	aggregated
22	126 ± 4.9	100 ± 9.5	234 ± 29.08	0.50	5.7	20-60

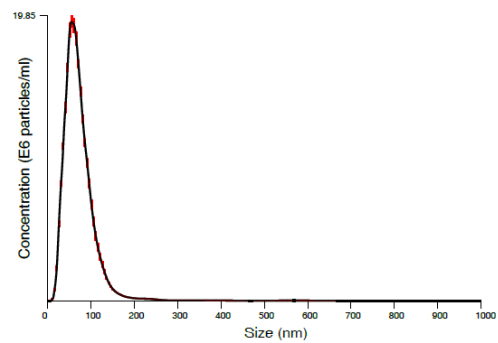
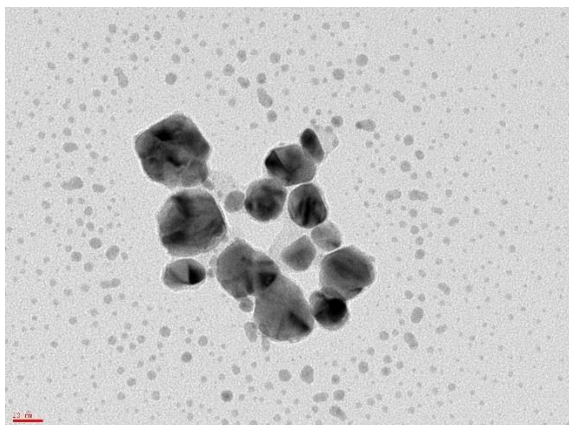


Averaged Size / Concentration  
Red error bars indicate +/- 1 standard error of the mean

Product4

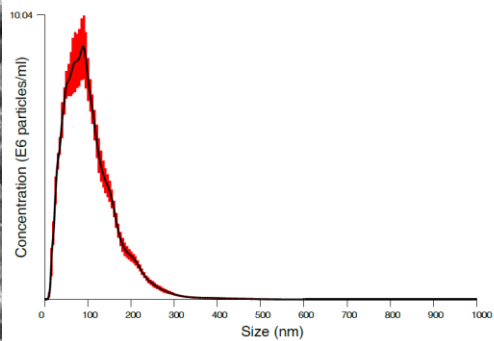
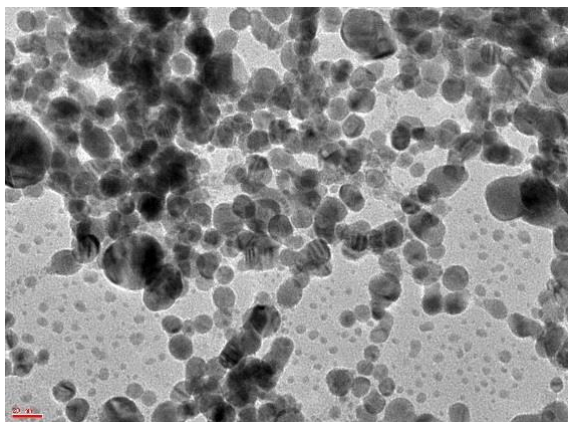


Product 6



Averaged Size / Concentration  
Red error bars indicate +/- 1 standard error of the mean

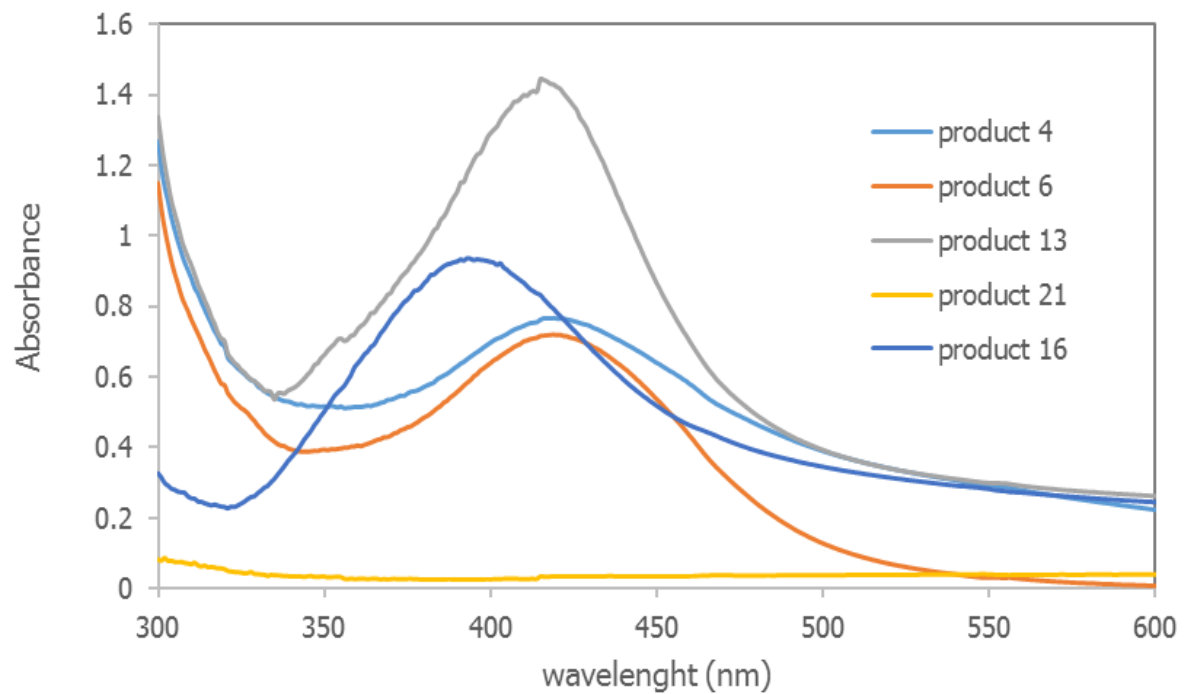
Product 13



Averaged Size / Concentration  
Red error bars indicate +/- 1 standard error of the mean

Product 16

Representative TEM Micrographs and NTA Size Distributions for Products 4, 6, 13 and 16.



**UV-Vis Plasmon Resonance Signatures:** Spectra were recorded for products 4, 6, 13, 16, & 21. Absorbance peaks were observed at 420nm, 420nm, 414nm, 398nm & no peak, respectively.

## **2.0 Methods**

### *2.1 Materials*

Twenty-two commercially available colloidal silver spray products were purchased from the internet. These products were advertised as health products intended for internal consumption or as surface sanitizers intended for external use. Products were either packaged in pump spray devices or advertised for spray applications. Secondary standard polystyrene beads (100, 50, 40 nm, traceable to National Institute of Standards) were obtained from Thermo Scientific, Fremont, CA. Nanosilver particles (pvp-stabilized) (5, 10, 20, 50, 75 nm) were obtained from Nanocompositix, San Diego, CA. All other chemicals used were of reagent grade.

### *2.2 Total silver concentration by ICP-OES*

For the determination of total silver in the consumer products 1mL of the product was acid digested (in triplicate) with 1mL of concentrated nitric acid (67–70% Optima™, Fisher Scientific, Inc., Pittsburgh, PA) in a hot block (DigiPrep, SCP Science, Quebec, Canada) at 60 °C for 12 hours. Prior to ICP-OES analysis, all samples were diluted 10 fold or greater (depending on the initial concentration) using DI water (DI, 18 MΩ × cm, Millipore, Bedford, MA). The analysis of the digested samples was performed using an iCAP 6500 Duo ICP-OES instrument (Thermo Scientific, Waltham, MA, USA). For quantification of silver, the spectral line 328.0 nm was used. Scandium (spectral line 361.3 nm) was used as an internal standard. Commercially available certified reference standards (SCP Science) were used for the preparation of calibration standards.

### *2.3 Centrifugal Ultrafiltration*

To estimate the dissolved silver fraction in the consumer products centrifugal ultrafiltration was used; 5ml of the product was transferred to a 10kDa centrifuge filter unit (Amicon Ultra-15, 10 K, Millipore, Bedford, MA) and centrifuged at 5911 x *g* for 20 min. After centrifugation, an aliquot of 1 ml of the filtrate was acid digested (in triplicate) and analyzed as described above.

#### *2.4 Dynamic Light Scattering (DLS)*

DLS measurements were performed with a Malvern Zetasizer Nano ZS (Malvern, UK) equipped with a 633 nm He-Ne laser and operating at an angle of 173°. 1 mL of each product was measured in single-use acrylic cuvettes (Sarstedt, Germany). The measurements were performed at a controlled temperature of 25°C with an automatic attenuation. For each sample, 3 measurements were performed. Polystyrene beads 40, 50, 100 nm (NIST beads; Thermo Scientific, Fremont, California) and pvp-stabilized silver nanoparticles 5, 10, 20, 50, 75 nm (NanoComposix) were used for a quality control. The intensity size distribution, the Z-average diameter (Z-ave) and the polydispersity index (PDI) were obtained from the autocorrelation function using the “general purpose model”.

#### *2.5 Nanoparticle Tracking Analysis (NTA)*

NTA measurements of the consumer products were performed with a NanoSight NS500 instrument (NanoSight, UK), equipped with a 640 nm laser. Measurements were performed at 25°C. All samples were measured for 60 s with manual shutter and gain adjustments. The measurement settings were optimized using 100 nm polystyrene beads. The software used to capture and analyzing the data was the NTA 2.3 (NanoSight Ltd.). Each video was analyzed to give a particle size distribution and an estimate of the particle concentration.

#### *2.6 UV-Visible Spectrophotometry*

The presence of laboratory synthesized or commercial AgNPs in aqueous suspension is often indicated by a light yellow to brown color and absorbance peaks at or around 400 nm due to local surface plasmon resonance (LSPR). UV-visible spectra were collected using a Hewlett Packard single beam instrument (Agilent, Santa Clara, CA) at 25 °C. Concentrated AgNP suspensions were diluted with DDI water to an initial absorbance of about 1.0. Spectra (from 300 to 600 nm) were recorded in the second to minute timeframe.



## *2.7 Transmission Electron Microscopy-Energy Dispersive Spectroscopy (TEM-EDS)*

TEM-EDS was used to verify the presence of nanoparticles in the consumer products and to measure their size and shape. Two types of grids and two TEM instruments were used. Samples were prepared by depositing a drop of the consumer product onto carbon-coated nickel grids and allowing it to dry in air overnight at room temperature. A JEOL-1200 EX (JEOL Ltd.) operating at 120 kV was used to acquire images at magnifications between 25kx and 350kx. The image processing program Image J was used to determine the nanoparticle size distribution from TEM micrographs. For the second TEM experiment, AgNPs were applied to amine functionalized silicon dioxide grids (Dune Scientific, Eugene OR) by leaning the chemically modified side of the grid against a sample drop (placed on para-film) for 10 min. The grids were then rinsed by placement in deionized (DI) water for 15 s and allowed to dry. Micrograph images were obtained using an FEI Titan 80–300 probe aberration corrected scanning Transmission Electron Microscope with a monochromator operating at 200 kV. A Bruker 4 SDD Energy Dispersive Spectroscopy (EDS) instrument was used to perform elemental mapping. Images were acquired at 300 kV. All images were representative of at least 3 grids. Image J size calculations were conducted using particle counts of 100 to 1000 particles. For the larger particles, size range values were typically determined from observations of between 10 and 20 particles.