

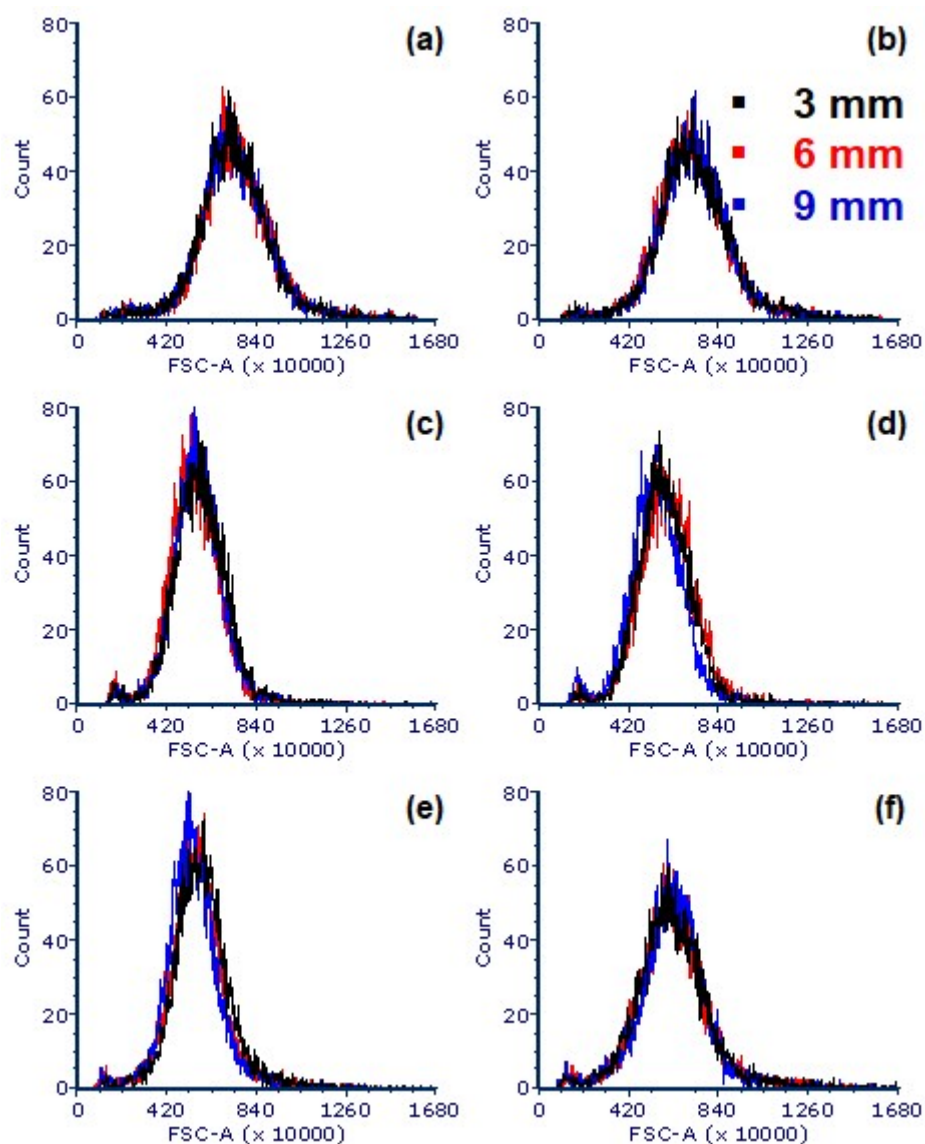
Effects of Agglomeration on *In Vitro* Dosimetry and Cellular Association of Silver Nanoparticles

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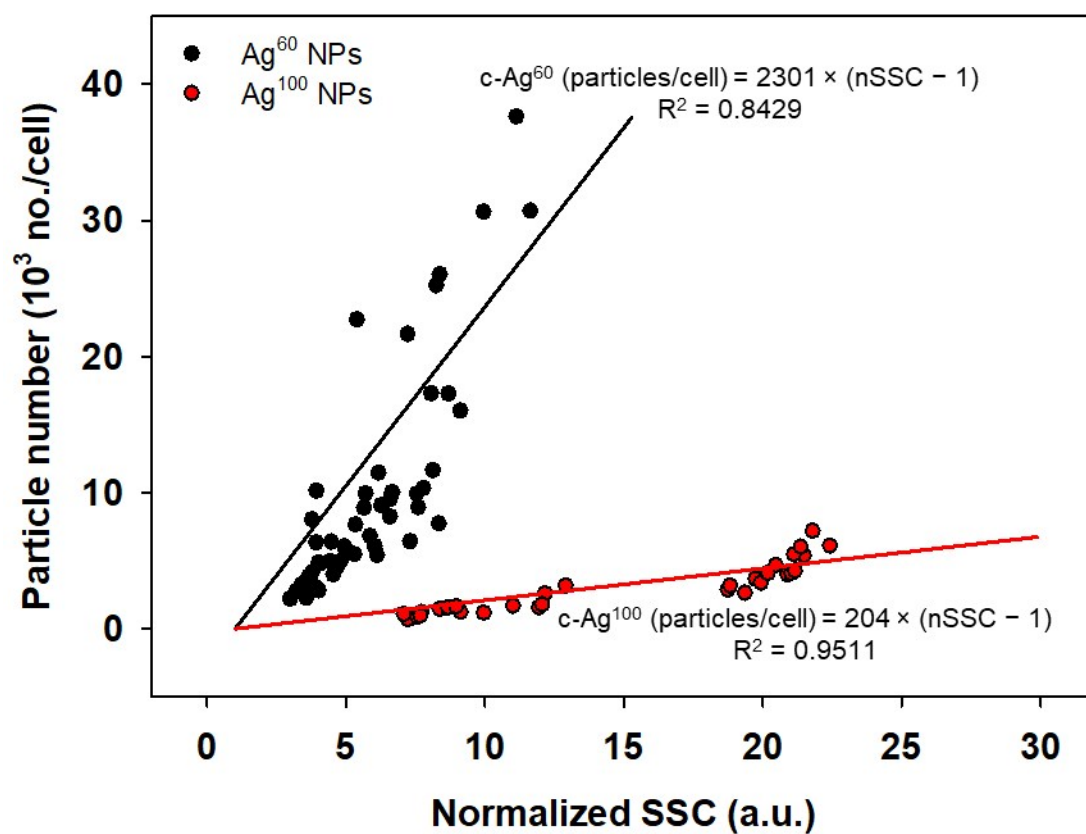
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SUPPLEMENTARY FIGURES



Supplementary Figure S1. Histograms of forward scattering intensity of (a, b) control cells and cells exposed to (c, d) Ag⁶⁰ NPs and (e, f) Ag¹⁰⁰ NPs in (a, c, e) upright and (b, d, f) inverted configurations. Exposure concentration was 10 µg/mL and exposure time was 24 h. Black histogram indicates 3 mm media height, red histogram 6 mm and blue histogram 9 mm.



Supplementary Figure S2. Linear regression between cellular dose and normalized SSC for Ag⁶⁰ NPs and Ag¹⁰⁰ NPs.

SUPPLEMENTARY TABLES

Supplementary Table S1. Effective doses for Ag⁶⁰ NPs and Ag¹⁰⁰ NPs simulated by DG model using DLS and NTA size distributions in upright/inverted configurations and different media heights. Simulated exposure time was 24 h.

Size distribution	Media height (mm)	Effective dose for Ag ⁶⁰ NPs (10 ⁻⁴ µg/cm ²)		Effective dose for Ag ¹⁰⁰ NPs (10 ⁻⁴ µg/cm ²)	
		Upright	Inverted	Upright	Inverted
DLS	3	7,161	65.9	6,383	5.49
	6	8,011	66.7	6,512	11.0
	9	8,673	66.8	6,556	16.5
NTA	3	6,070	0.675	4,266	0.405
	6	11,018	0.676	7,695	0.406
	9	14,682	0.677	10,335	0.410

Supplementary Table S2. Training data for linear regression between cellular dose and normalized SSC for Ag⁶⁰ NPs and Ag¹⁰⁰ NPs.

NPs	nSSC	Cellular dose (no./cell)	NPs	nSSC	Cellular dose (no./cell)
Ag ⁶⁰	9.14	15964	Ag ⁶⁰	3.01	2141
Ag ⁶⁰	8.71	17215	Ag ⁶⁰	3.24	2722
Ag ⁶⁰	9.98	30570	Ag ⁶⁰	3.43	3190
Ag ⁶⁰	5.37	7602	Ag ⁶⁰	5.89	6763
Ag ⁶⁰	5.73	9892	Ag ⁶⁰	6.61	9477
Ag ⁶⁰	8.27	25174	Ag ⁶⁰	6.69	9977
Ag ⁶⁰	8.09	17232	Ag ⁶⁰	3.69	3801
Ag ⁶⁰	11.66	30642	Ag ⁶⁰	3.95	6295
Ag ⁶⁰	11.16	37566	Ag ⁶⁰	4.50	6338
Ag ⁶⁰	3.80	4074	Ag ¹⁰⁰	20.92	3948
Ag ⁶⁰	4.06	4736	Ag ¹⁰⁰	21.52	5339
Ag ⁶⁰	4.96	5986	Ag ¹⁰⁰	22.45	6047
Ag ⁶⁰	6.29	9012	Ag ¹⁰⁰	11.98	1511
Ag ⁶⁰	7.57	9858	Ag ¹⁰⁰	12.19	2538
Ag ⁶⁰	8.15	11582	Ag ¹⁰⁰	12.93	3101
Ag ⁶⁰	4.04	4806	Ag ¹⁰⁰	19.39	2581
Ag ⁶⁰	4.47	4901	Ag ¹⁰⁰	21.05	4037
Ag ⁶⁰	5.67	8834	Ag ¹⁰⁰	21.19	4205
Ag ⁶⁰	6.06	6018	Ag ¹⁰⁰	7.25	649
Ag ⁶⁰	7.81	10279	Ag ¹⁰⁰	8.40	1400
Ag ⁶⁰	7.63	8861	Ag ¹⁰⁰	12.09	1734
Ag ⁶⁰	3.94	2999	Ag ¹⁰⁰	18.76	2817
Ag ⁶⁰	4.70	4484	Ag ¹⁰⁰	19.77	3614
Ag ⁶⁰	4.83	4893	Ag ¹⁰⁰	20.50	4613
Ag ⁶⁰	6.15	5362	Ag ¹⁰⁰	7.74	1156
Ag ⁶⁰	7.33	6371	Ag ¹⁰⁰	7.10	1026
Ag ⁶⁰	8.36	7674	Ag ¹⁰⁰	9.15	1173
Ag ⁶⁰	3.59	2213	Ag ¹⁰⁰	18.86	3109
Ag ⁶⁰	4.04	2763	Ag ¹⁰⁰	19.96	3332
Ag ⁶⁰	4.57	3938	Ag ¹⁰⁰	20.22	4040
Ag ⁶⁰	6.19	11410	Ag ¹⁰⁰	7.60	825
Ag ⁶⁰	7.25	21603	Ag ¹⁰⁰	7.70	900
Ag ⁶⁰	8.40	25980	Ag ¹⁰⁰	9.98	1117
Ag ⁶⁰	3.80	7976	Ag ¹⁰⁰	21.15	5413
Ag ⁶⁰	3.96	10075	Ag ¹⁰⁰	21.40	5962
Ag ⁶⁰	5.42	22666	Ag ¹⁰⁰	21.82	7138
Ag ⁶⁰	5.32	5426	Ag ¹⁰⁰	8.68	1510
Ag ⁶⁰	6.61	8193	Ag ¹⁰⁰	8.99	1578
Ag ⁶⁰	6.32	8992	Ag ¹⁰⁰	11.04	1640

Supplementary Table S3. Cellular dose and effective dose for Ag⁶⁰ NPs and Ag¹⁰⁰ NPs in upright and inverted configurations. Exposure time was 24 h.

Dose metrics	Media height (mm)	Dose for Ag ⁶⁰ NPs (10 ³ no./cell)		Dose for Ag ¹⁰⁰ NPs (10 ³ no./cell)	
		Upright	Inverted	Upright	Inverted
Cellular dose by ICPMS	3	5.9 ± 0.6	2.8 ± 0.8	2.8 ± 0.3	0.9 ± 0.3
	6	8.6 ± 1.7	4.1 ± 1.7	3.7 ± 0.4	1.1 ± 0.3
	9	8.9 ± 0.9	4.6 ± 1.4	4.3 ± 0.3	1.3 ± 0.3
Cellular dose by FCM-SSC	3	11.9 ± 0.2	6.5 ± 0.4	3.7 ± 0.1	1.3 ± 0.1
	6	14.9 ± 0.6	7.6 ± 0.8	3.9 ± 0.1	1.4 ± 0.1
	9	16.4 ± 0.8	9.5 ± 1.2	4.0 ± 0.1	1.9 ± 0.3
Effective dose by DLS size	3	9.7	2.0	5.0	0.1
	6	13.0	2.1	5.3	0.3
	9	13.8	2.4	5.9	0.4
Effective dose by NTA size	3	8.2	1.3	6.2	0.1
	6	17.8	1.4	6.9	0.1
	9	23.4	1.6	7.8	0.1