ESI Supporting Information

Quantification of silver nanoparticles up taken by single cells using Inductively Coupled Plasma Mass Spectrometry in the single cell mode

Ana Lopez-Serrano Oliver[1], Sabine Baumgart[2], Wolfram Bremser[1], Sabine Flemig[1], Doreen Wittke[3], Andreas Grützkau [2], Andrea Haase[3], Andreas Luch[3], Norbert Jakubowski[1]

[1] Bundesanstalt für Materialforschung und –prüfung (BAM), Richard-Willstatter-Str. 11, 12489, Berlin Germany

[2] DRFZ, German Rheumatism Research Centre Berlin, Charitéplatz 1, 10117 Berlin, An Institute of Leibniz Asscociation

[3] The Federal Institute for Risk Assessment Department of Product Safety (BfR), Max-Dohrn-Str. 8-10

D - 10589 Berlin.

Table S-1. Typical operating parameters for the ICP-MS (Element XR/2).

Detector voltage	1580-1650 V (for AgNPs and cell suspensions) 1900 V (for Ag standard solutions and digested samples
Rf power/ W	1550
Ar cooling gas flow rate/ L min^{-1}	15
Ar auxiliary gas flow rate/ L min^{-1}	1.0
Sample and skimmer cone	Ni
Micro nebulizer	Micromist 200 µL
Data acquisition mode	Time Resolved Analysis (TRA) in counting mode
Isotopes	^{107}Ag; ^{115}In
Sample uptake rate (mL min^{-1})	0.25
Dwell time (ms)	0.1, 1, 10 (parameter optimized)
Total acquisition time (s)	52

Table S-2. Determination of nebulization efficiency by using AgNPs aqueous suspension. The transport efficiency, **ε**, was determined by using AgNPs aqueous suspensions with known concentrations of particles mL^{-1} at a given flow rate

AgNPs ng mL^{-1}	**ng/particle**	**AgNPs particles mL^{-1}**	**Events s^{-1}**	**flow rate mL s^{-1}**	**ε**	**ε_{mean}**
$3x10^{-2}$	$6.86x10^{-7}$	$3.64x10^{+4}$	3	0.004	0.02	0.02
$5x10^{-2}$		$7.29x10^{+4}$	5.09		0.02	
$1x10^{-1}$		$1.46x10^{+5}$	9.72		0.02	

Table S-3. Quantitative analysis of AgNPs in single cells by SC-ICP-MS. The mode of the distribution and a 67% inter-quantile was selected in order to mimic the one-sigma of a normal distribution. An uncertainty u- and an uncertainty u+ are used due to the lower and upper bound of the heavily asymmetric quantile. Note that broken numbers (fractions) for the mode like 2.5 are a consequence of a distributed mode, and broken numbers for the uncertainties result from the very short left-tail with a zero zone as described above.

		THP-1 Monocytes				THP-1 partially differentiated macrophages			
Exposure time	Exposure dose	mode	u_down	u_up	67% inter-quantile	mode	u_down	u_up	67% inter-quantile
(h)	(µg Ag mL^{-1})	AgNP/cell				AgNP/cell			
4	-	0	0	0	0	0	0	0	0
24									
4	0.1	1	0,5	1	1,5	1	0,2	7	7,2
24		1	0,5	1	1,5	3	1	3	4
4	1	2,5	0,3	1,5	1,8	2	0,2	40	40,2
24		4	1	4	5	4,5	2	54	56

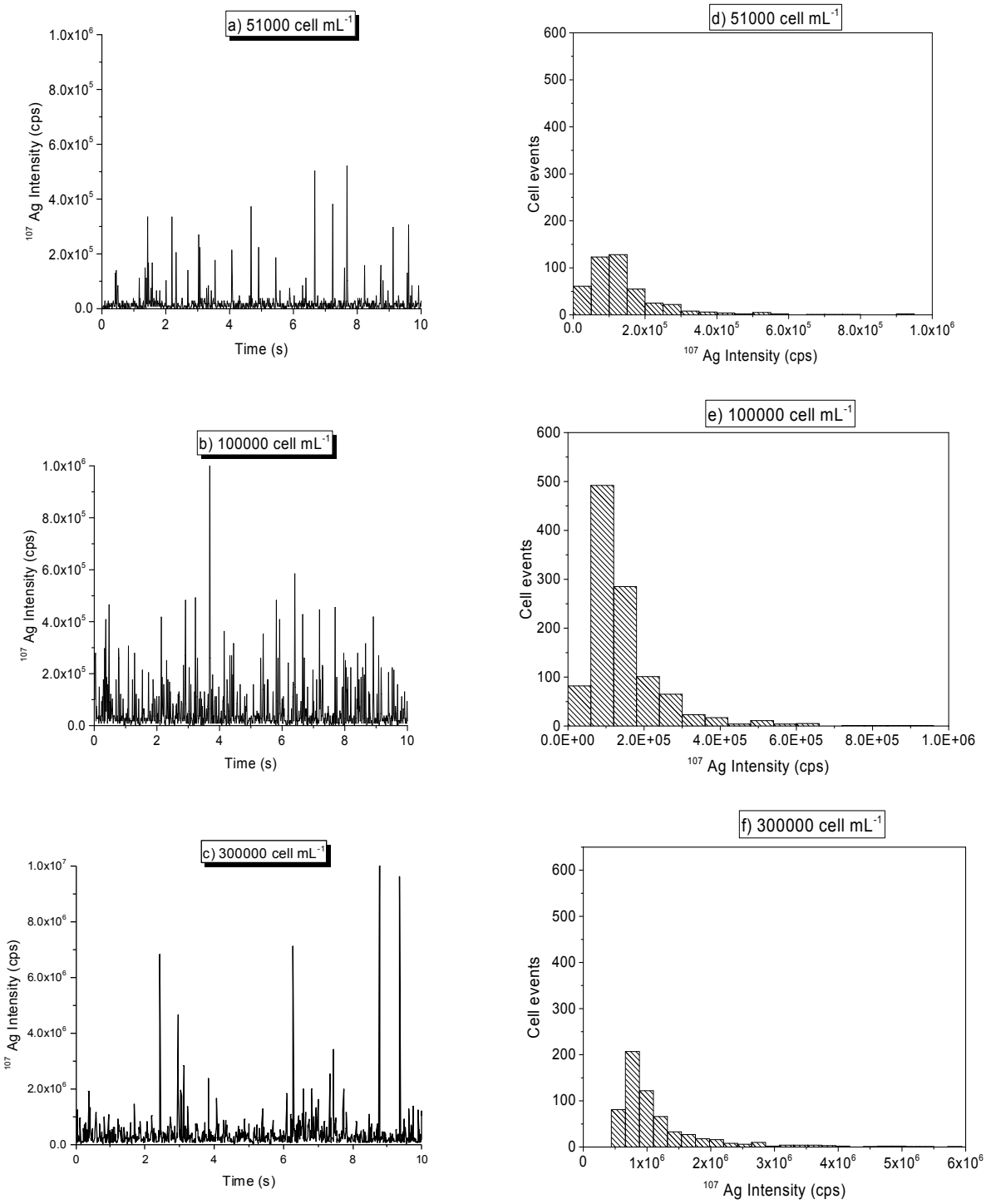

Figure S-1. Effects of different cell concentrations on the signal profile of single cells. ^{107}Ag chromatograms (a-c) and histograms (e-f) at different concentrations of cell suspensions THP-1 monocytes incubated with 1 µg AgNPs mL^{-1} for 24h.

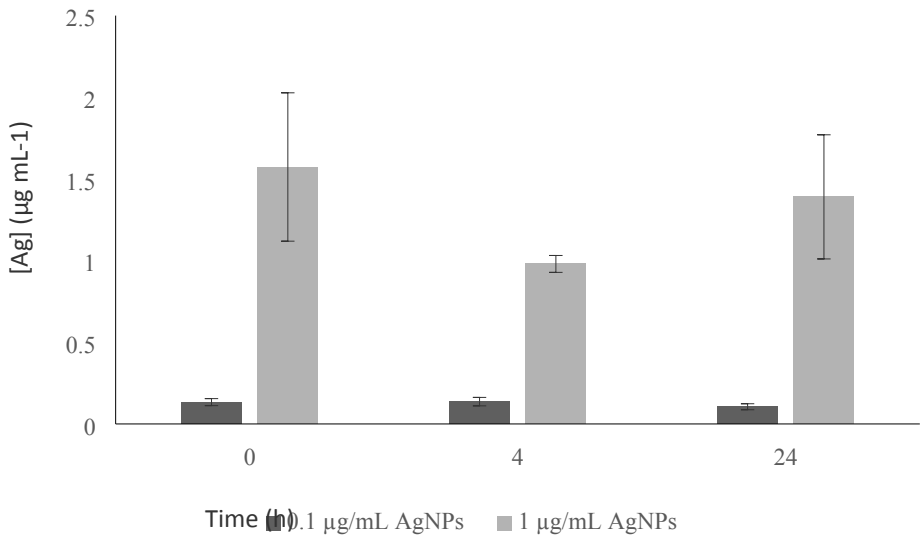

Figure S-2. Concentration of AgNPs , calculated as the absolute mass of Ag (µg mL^{-1}) in the supernatant of the exposure/incubation medium after 4 and 24 h AgNPs exposure at 0.1 (black bars) and 1µg mL^{-1} (gray bars) to THP-1 monocytes.

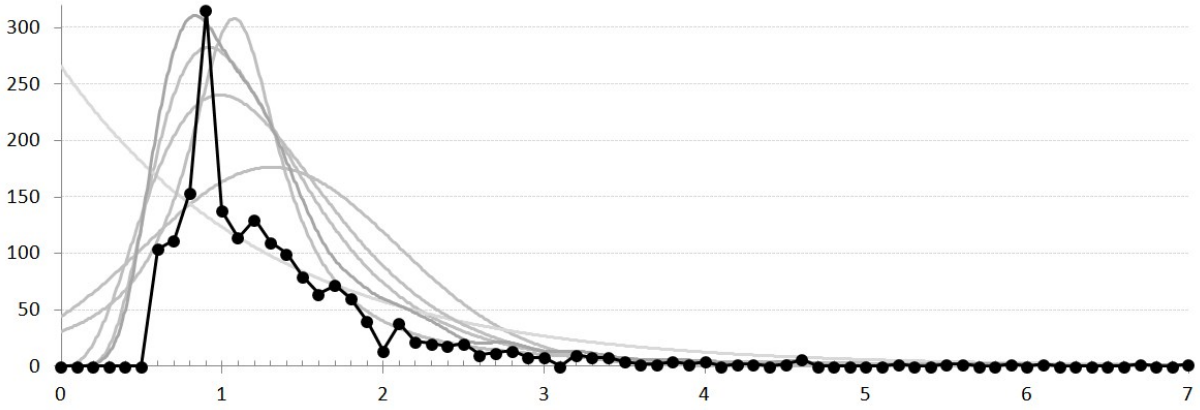

Figure S-3. Experimental event distribution (black dots) and fitted common distributions (grey lines): Normal, log-normal, exponential, Gamma, Lorentz distributions, and a Kernel smoothing.

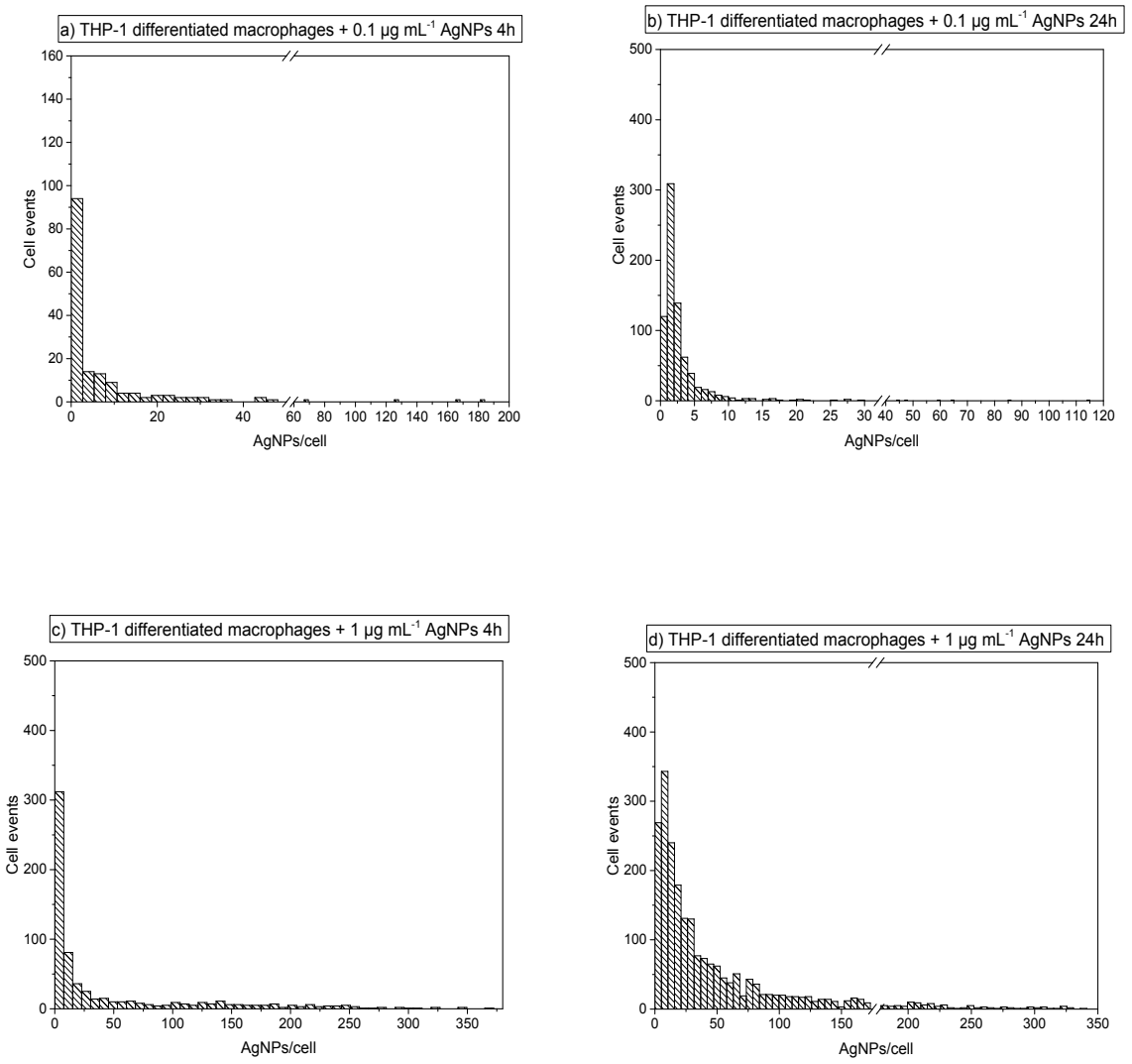

Figure S-4. SC-ICP-MS of THP-1 partially differentiated macrophages as single cell suspensions at a concentration of $5x10^4$-$1x10^5$ cell mL^{-1}. Histograms obtained of THP-1 partially differentiated macrophages incubated with 50 nm AgNPs at 0.1 (a-b) and 1 µg mL^{-1} (c-d) for 4 and 24 hours. Results correspond to one biological experiment measured in two replicates.