

Supporting Information

Rapid and sensitive quantification of cellular associated multi-walled carbon nanotubes

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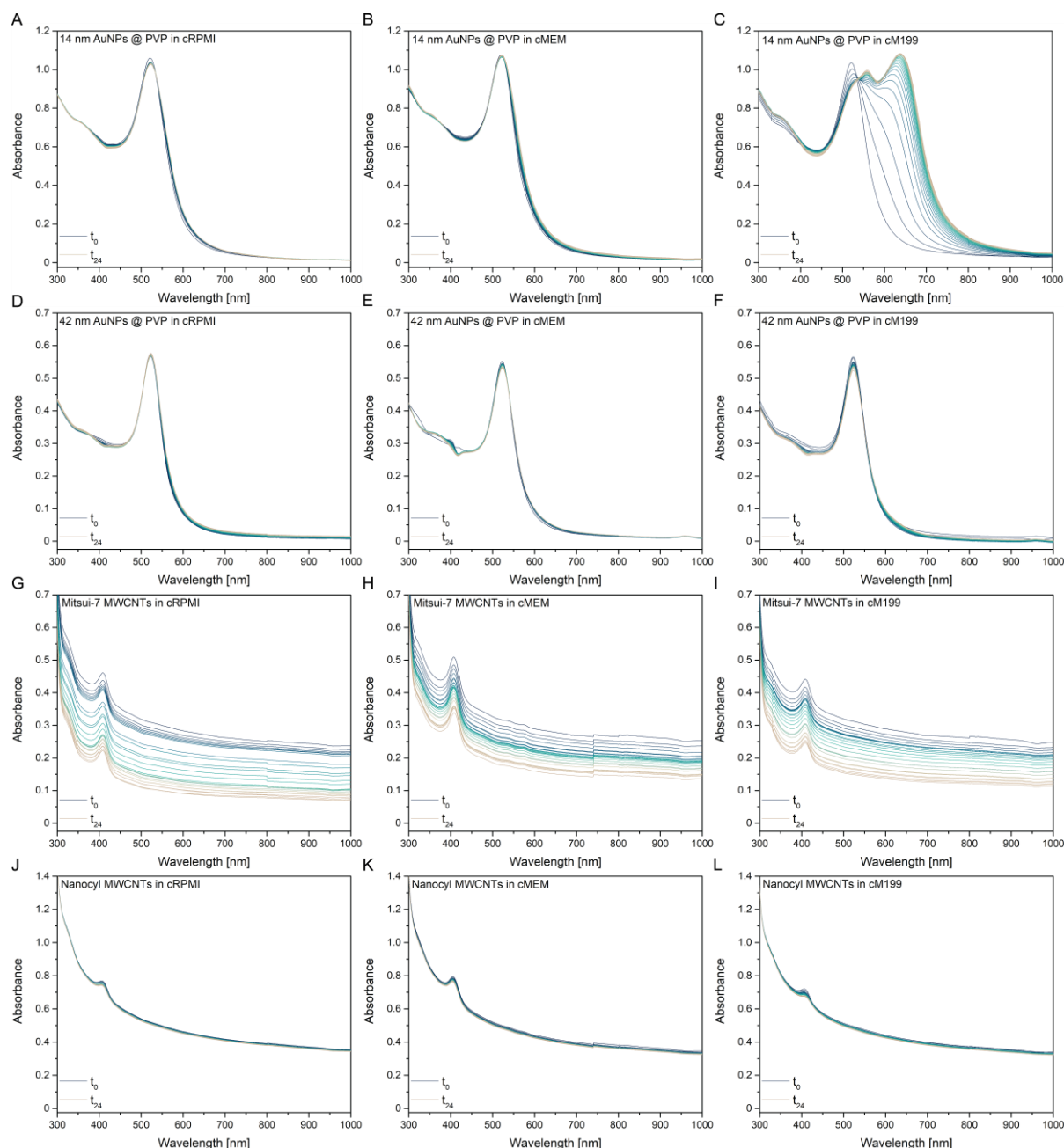


Figure S1. UV-Vis absorbance spectra of 14 & 42 nm AuNPs and Mitsui-7 & Nanocyl MWCNTs in complete cell culture media. Spectra were recorded over 24 h in one-hour steps. Extended signs of aggregation were only observed for 14 nm AuNPs in cM199. The reduction of absorbance over time for Mitsui-7 MWCNTs can be related to their sedimentation in the cuvette, which is caused by their relatively large size.

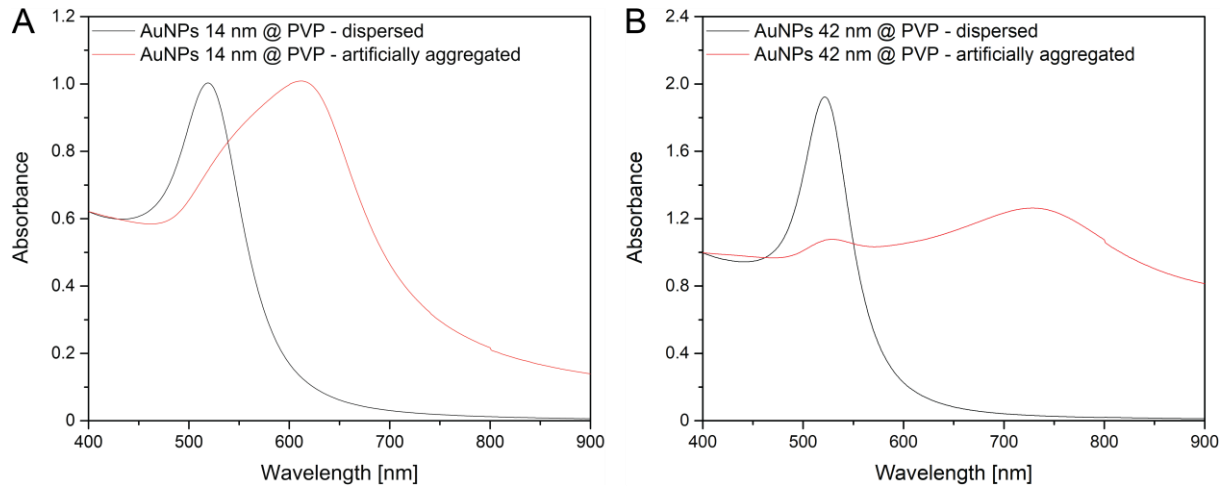


Figure S2. UV-Vis absorbance spectra of 14 nm (A) & 42 nm (B) AuNPs @ PVP. Dispersed and aggregated NPs are compared. Aggregation of NPs was achieved by the addition of 0.1 M HCl.

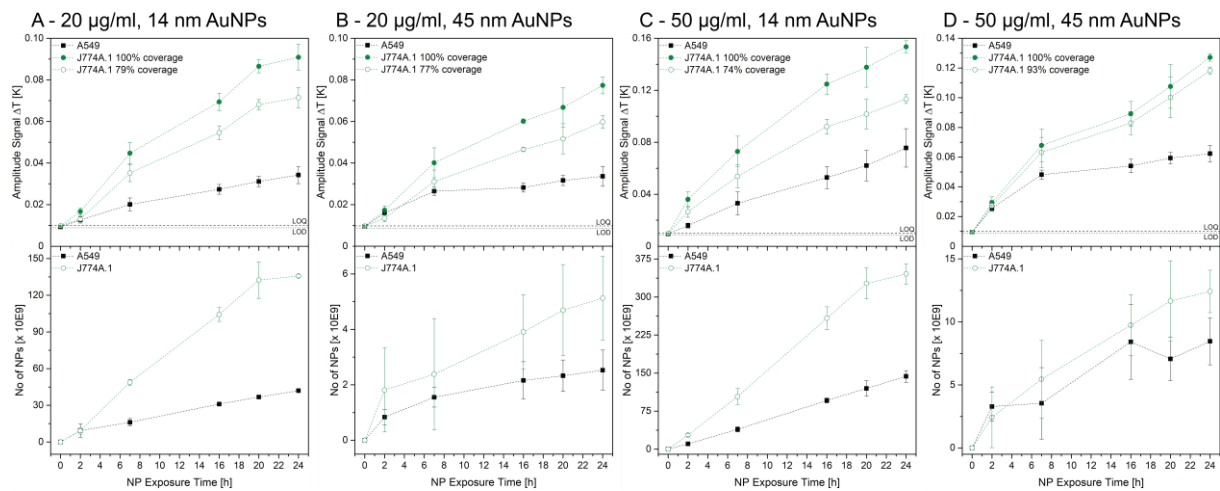


Figure S3. LIT AuNP-cell association trends (top row) and ICP-OES analysis (bottom row) for J774A.1 cells (macrophages). Due to their non-confluent growth pattern macrophages can not be directly compared to the investigated epithelial and mesothelial cells. Therefore, the cell coverage was determined (79 %, 77 %, 74 % and 93 %, for A-D) and normalized to a theoretical 100 % coverage.

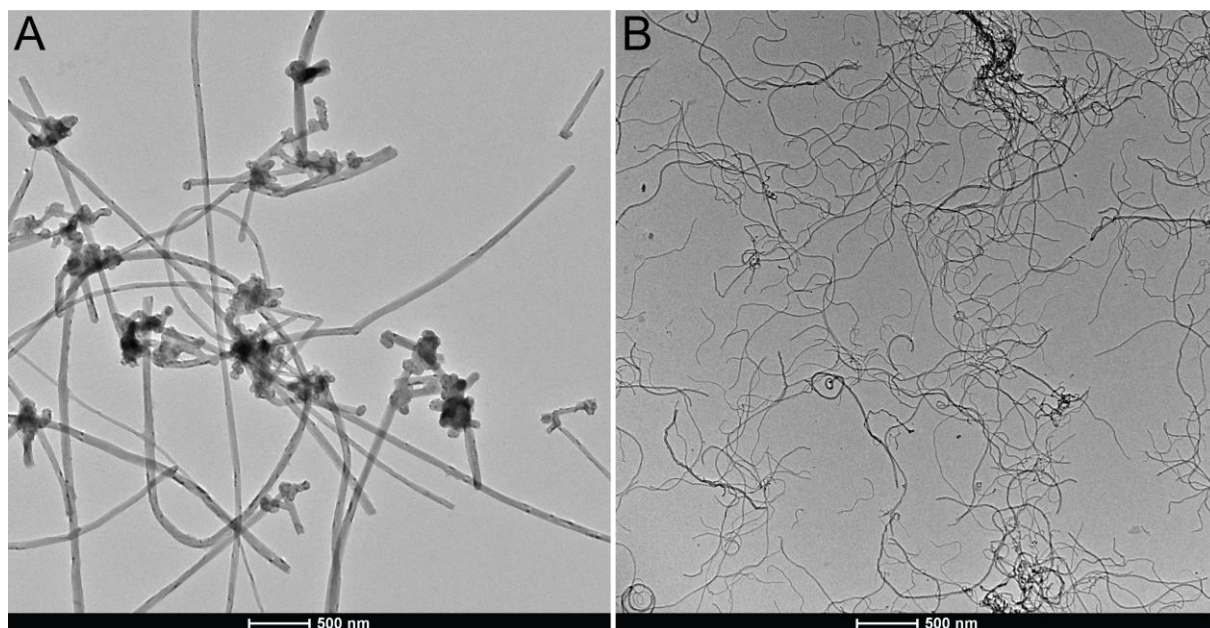


Figure S4. TEM micrographs of Mitsui-7 (A) & Nanocyl (B) MWCNTs in water, highlighting the difference in dimensions and stiffness.

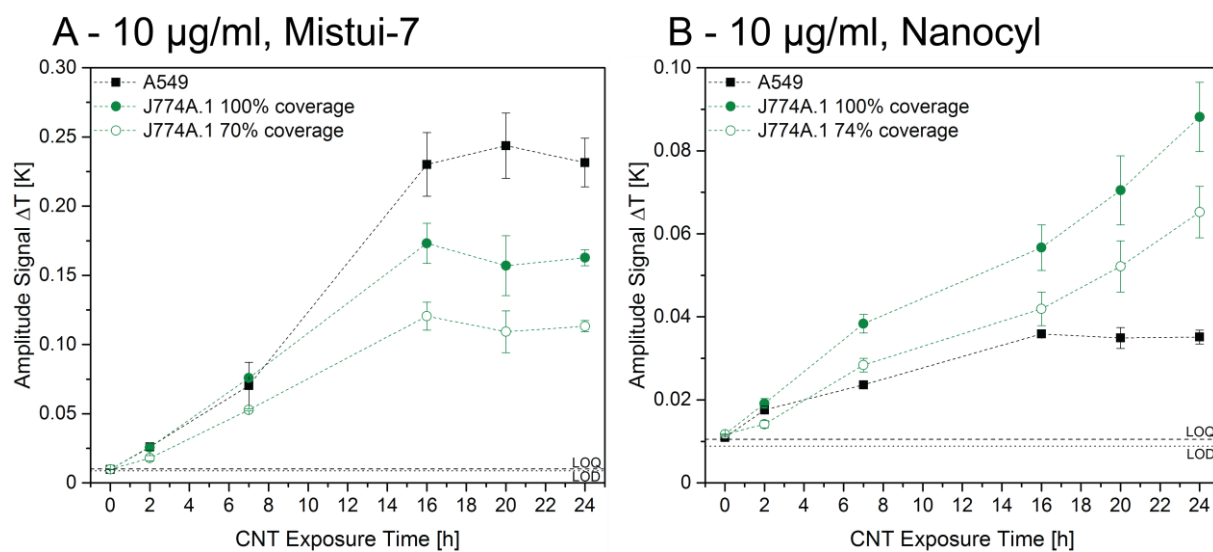


Figure S5. LIT MWCNT-association trends for J774A.1 cells (macrophages) at an exposure concentration of 10 $\mu\text{g/ml}$. Cell coverage was determined (70 % A & 74 % B) and normalized to a theoretical 100 % coverage.

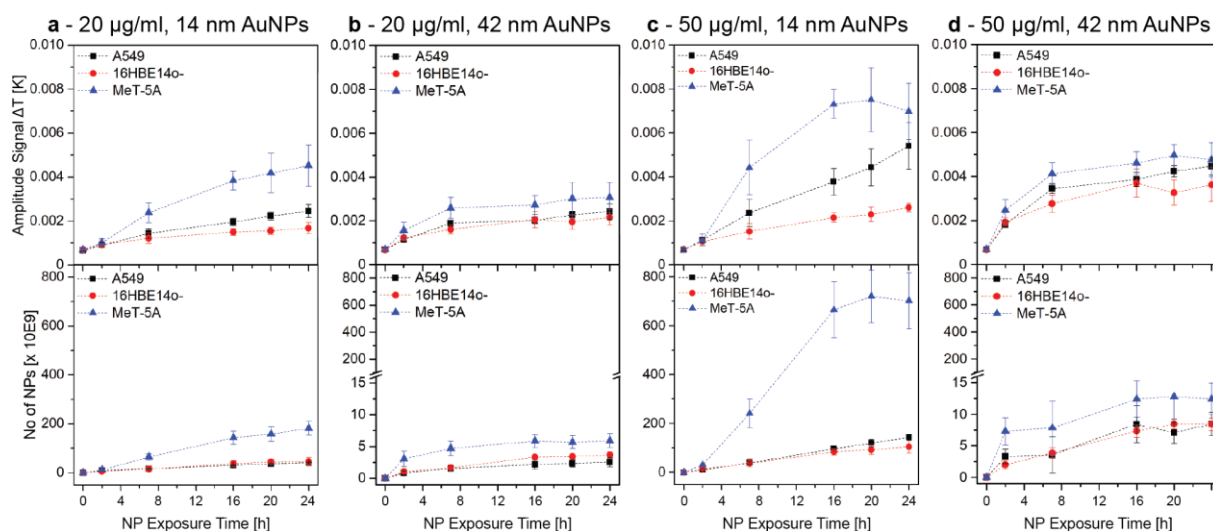


Figure S6. Re-printed Figure 3 with identical y-axes. AuNP-cell association trends over 24 h, obtained by LIT (top row, 525 nm excitation wavelength) and ICP-OES (bottom row). The association trends of A549, 16HBE14o-, and MeT-5A cells exposed to 20 and 50 $\mu\text{g/mL}$ of 14 nm (A, C) and 42 nm AuNPs (B, D) were investigated. Hence, it was possible to determine that, independent of the NP size and concentration, the AuNP association is higher for MeT-5A. The plateau reached under some conditions indicated a NP-cell association saturation.

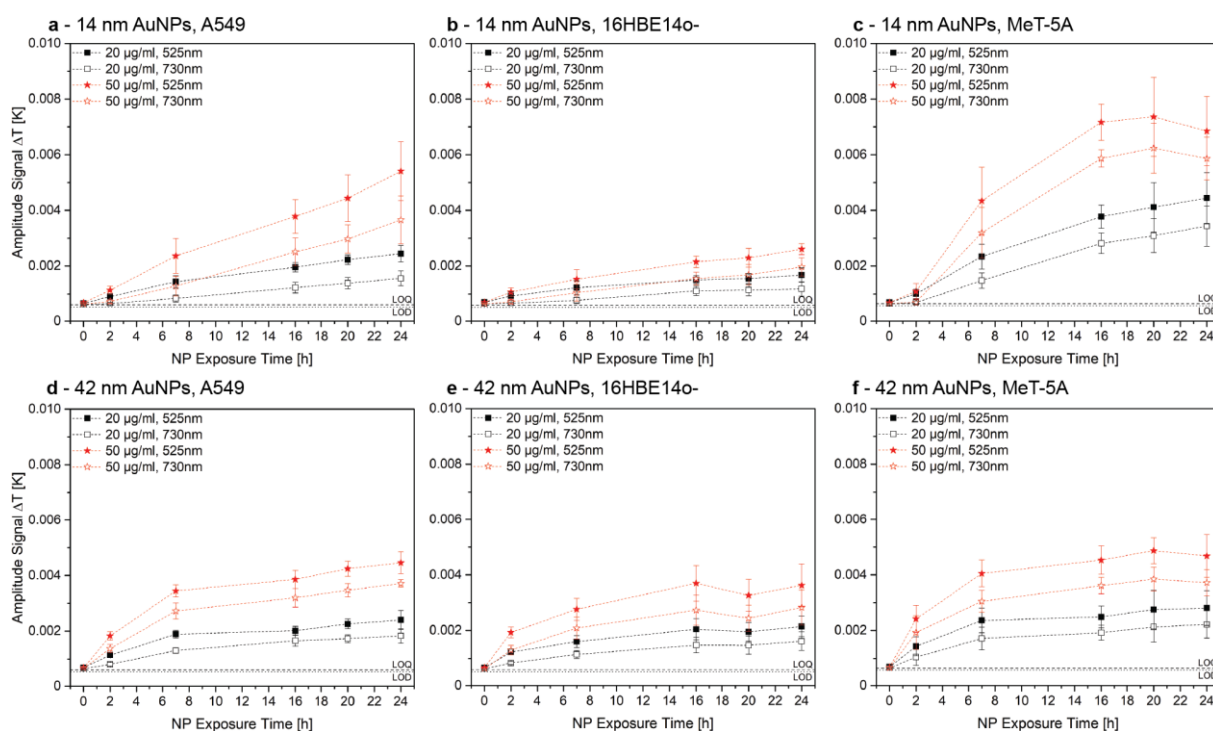


Figure S7. Re-printed Figure 4 with identical y-axes. Comparison of LIT measurements for cell-associated AuNPs at an excitation wavelength of 525 nm (closed symbols) and 730 nm (open symbols). The generation of heat at an excitation wavelength of 730 nm is a clear indication for NP aggregation. However, association trends for 20 $\mu\text{g/mL}$ (black symbols) and 50 $\mu\text{g/mL}$ (red symbols) evolve in an almost identical manner over time. Therefore, either all AuNPs aggregate due to the association or single NPs and aggregates associate to a similar extent.

| | 14 nm AuNPs | | | 42 nm AuNPs | | |
|------------------------|--------------------|---------|----------|--------------------|----------|----------|
| <i>[nm]</i> | cRPMI | cMEM | cM199 | cRPMI | cMEM | cM199 |
| <i>t₀</i> | 44 ± 23 | 36 ± 19 | 59 ± 15 | 132 ± 8 | 108 ± 15 | 118 ± 48 |
| <i>t_{1h}</i> | 59 ± 10 | 40 ± 5 | 98 ± 22 | 117 ± 7 | 108 ± 15 | 116 ± 13 |
| <i>t_{5h}</i> | 73 ± 3 | 36 ± 10 | 135 ± 13 | 111 ± 11 | 127 ± 20 | 124 ± 66 |
| <i>t_{24h}</i> | 70 ± 4 | 43 ± 1 | 144 ± 4 | 108 ± 6 | 119 ± 20 | 121 ± 1 |

Table S1. DDLS measurements of AuNPs in complete cell culture media, recorded over 24 h at 37°C.