Supporting information for:

Synthesis of Pyridyldisulfide-functionalized Nanoparticles for Conjugating Thiol-containing Small molecules, Peptides and Proteins

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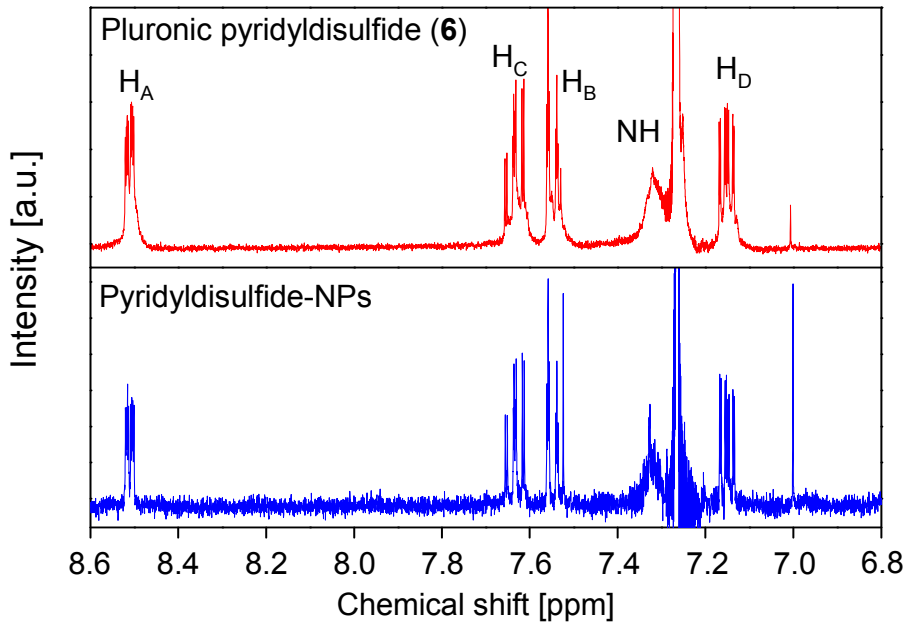
Supporting 1: Calculation NP composition

The following NMR signals were used for calculation: 1.1 ppm (CH_3 Pluronic), 1.4 ppm (CH_3 PPS), 1.8 ppm (CH_2 initiator) and 3.7 ppm (CH_2 PEG). The integral values were normalized by dividing by 3, 3, 8, and 4 respectively giving the molar ratio of -CH_2-$CH(CH_3)$-O-, -CH_2-$CH(CH_3)$-S-, $C(CH_2$-O-CH_2-CH_2-CH_2-$S)_4$- and –CH_2-CH_2-O- units. With this molar ratio and the molecular weights of each fragment a relative molecular weight ($MW_{relative}$) can be calculated. The weight contribution of the -CH_2-$CH(CH_3)$-O- and -CH_2-CH_2-O- fragments divided by $MW_{relative}$ will then give the weight percentage pluronic. The weight percentages PPS and initiator are calculated in the same way. In this calculation the end groups on pluronic and PPS are ignored.

Supporting 2: NMR spectra Pluronic-pyridyldisulfide (6) and 100% Pyridyldisulfide-NP



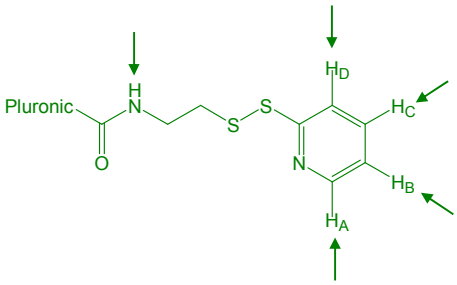


Fig. 1 NMR spectra of Pluronic-pyridyldisulfide (**6**) and 100% Pyridyldisulfide-NP in deuterated chloroform in the region between 8.6 and 6.8 ppm together with the assignment of the signals.

Supporting 3: Estimation of number of biotin, peptide and OVA molecules per NP

Table 1: Data used to calculate number of biotin, peptide and ovalbumin molecules per particle

Sample	Size[c] [nm]	Conc [mg/ml]	Loading [mM]	Copies/particle
Biotin	42.0	35.2	1.9[b]	776
Peptide[a]	35.1	58.8	0.93[b]	158
Ovalbumin[a]	39.6	41.0	0.24[b]	73

[a] These (different) NP batches were made from 25%-carboxylate-NP and are different from the 25% batch mentioned in Table 1 and Figs. 2-4.

[b] For calculating the number of ovalbumin molecules, the actual ovalbumin concentration (0.24 mM) instead of the pyridyldisulfide loading (0.55 mM) was used. For biotin and peptide, the pyridyldisulfide loading was used for calculation.

[c] Size for Pyridyldisulfide-NP.

Calculations were done with the following assumptions:

1. The NPs have uniform size
2. The diameter of the NPs equals the one measured by DLS
3. The weight percentage Pluronic of the NPs is 60%
4. The surface of the NP is only covered with Pluronic polymers
5. The surface occupied by one Pluronic polymer is 8.16 nm^2 (*1*)
6. The molecular weight for Pluronic and OVA are 12700 and 44000 respectively

From the diameter the radius (r) and the surface area ($4\pi r^2$) of one NP can be calculated. Dividing this number by the surface area occupied by one Pluronic polymer (8.16 nm^2) gives the number of Pluronic polymers per particle. With the NP concentration and the mass percentage Pluronic known, the number of particles in 1 mL can be calculated. With the loading in mM, the number of biotin, peptide and ovalbumin molecules per particle can be estimated, for which the results are given in Table 1. These values are estimates, and are presented suitably rounded in the main manuscript.

(1) Rehor, A., Hubbell, J. A., and Tirelli, N. (2004) Oxidation-Sensitive Polymeric Nanoparticles. *Langmuir 21*, 411-417.