Supporting Information to:

UV-induced functionalization of poly(divinylbenzene) nanoparticles via efficient [2+2] cycloadditions

Anitha Ethirajan[a], Linny Baeten,[a,b] Matthias Conradi[b], Kayte Ranieri,[b] Bert Conings[a], Hans-Gerd Boyen[a] and Tanja Junkers[b*]*

[a] Institute for Materials Research, Hasselt University, Wetenschapspark 1, 3590 Diepenbeek, Belgium.
Tel: + 32 (0)11 268877; E-mail: anitha.ethirajan@uhasselt.be

[b] Polymer Reaction Design Group, Institute for Materials Research (imo-imomec), Hasselt University,
Agoralaan D, 3590 Diepenbeek, Belgium. Fax: + 32 (0)11 26 8399;
Tel: + 32 (0)11 268318; E-mail: tanja.junkers@uhasselt.be

www.imo.uhasselt.be and www.polymatter.net

Figure S1 shows the reaction scheme for the surface functionalization of polyDVB nanoparticles with an ATRP initiator via [2+2]-cycloaddition and ATRP polymerization. In a first step, the divinylbenzene monomer is polymerized via miniemulsion technique to form polydivinylbenzene nanoparticles with vinyl groups at the surface. In a second step, a UV-induced [2+2] cycloaddition of aldehyde functionalized ATRP initiator is performed, followed by an ATRP polymerization on the surface.
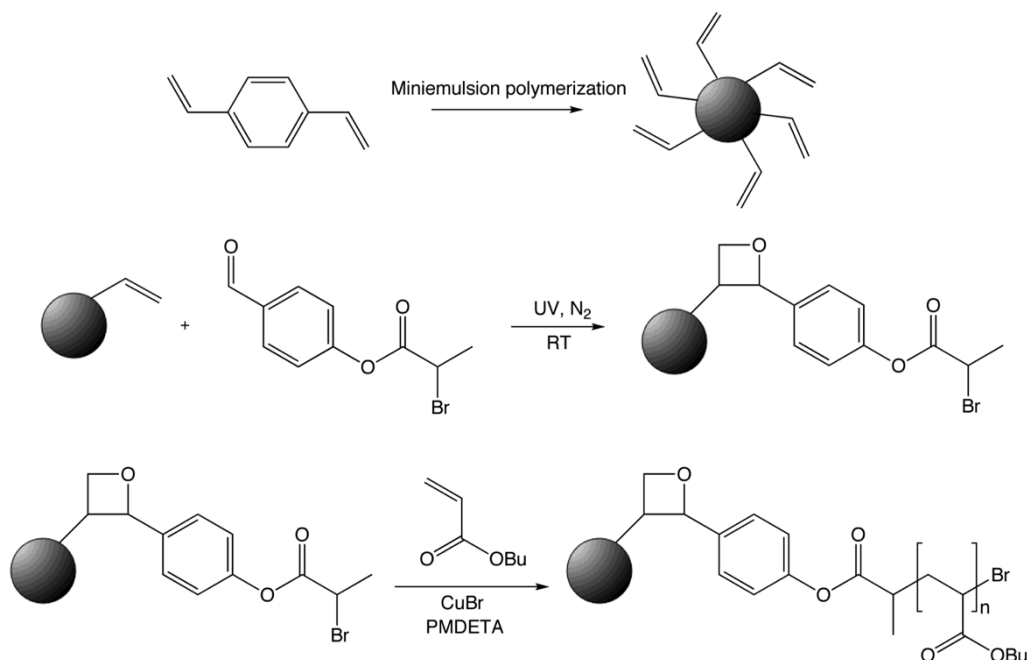

Figure S1: Chemical reaction scheme of [2+2] cycloaddition of aldehyde functionalized ATRP initiator and ATRP polymerization.

Figure S2 shows the reaction scheme for the surface functionalization of polydivinylbenzene nanoparticles with N-succinimidyl-p-formylbenzoate (NHS-aldehyde) via [2+2]-cycloaddition. In a first step, the divinylbenzene monomer is polymerized via miniemulsion technique to form nanoparticles with vinylgroups at the surface. In a second step, a UV-induced [2+2] cycloaddition reaction is performed, to introduce NHS groups on the surface. The latter is then used for the coupling of gold labeled antibodies in a third step.
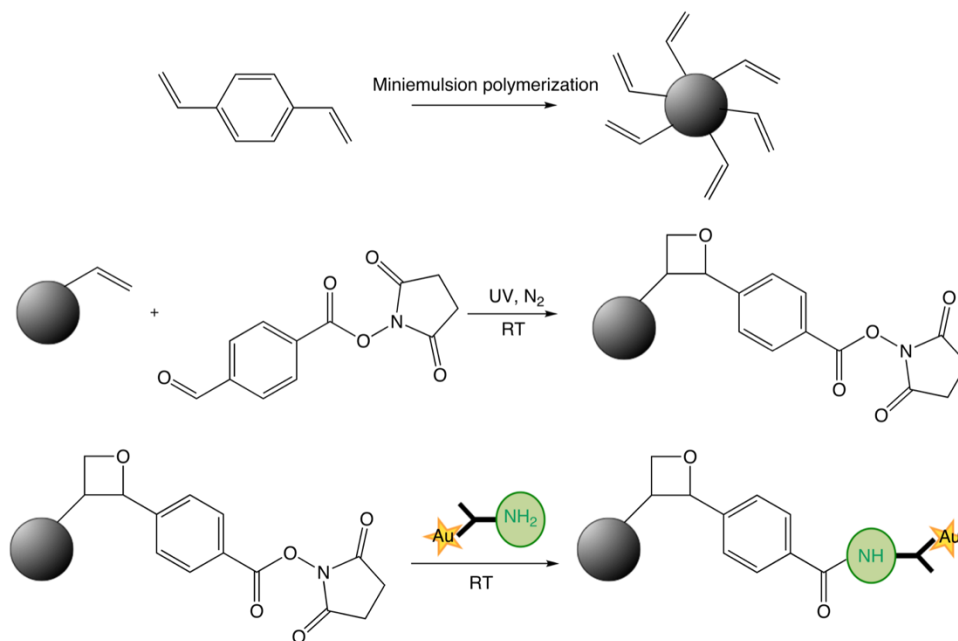

Figure S2: Chemical reaction scheme of [2+2]-cycloaddition of aldehyde functionalized NHS and coupling of gold labeled antibodies.

Figure S3 shows the TEM–images of a) core polyDVB particles after 30 minutes of polymerization, b) after surface functionalization with the ATRP initiator and c) after ATRP polymerization on the surface of the polyDVB particles and d) PSD chart obtained by counting particles in TEM image a). The average diameter determined was 110 ± 30 nm (222 particles counted) and ImageJ program was used for processing.
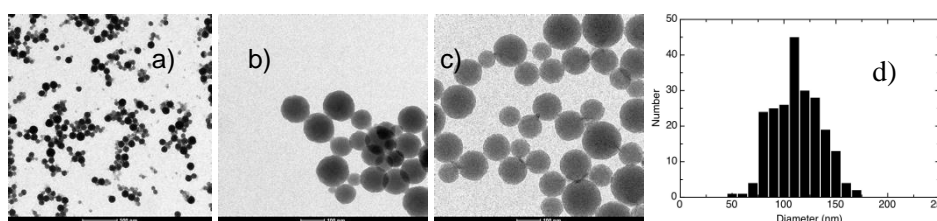

Figure S3: TEM images of a) core polyDVB particles, b) UV-treated polyDVB particles and c) ATRP-treated polyDVB particles and d) PSD chart of particles in TEM image a).

Figure S4 shows the TEM-images of core polyDVB particles with longer polymerization times, respectively 1 and 3 hours. It can be seen that particle size increases with longer polymerization times.
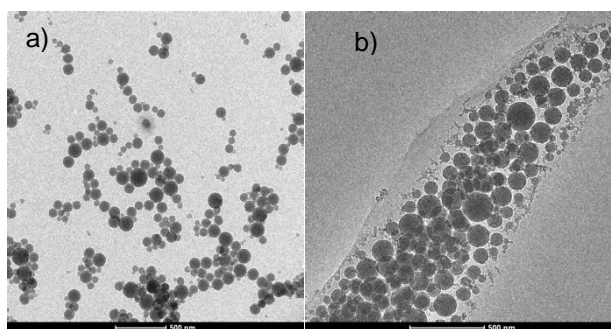

Figure S4: TEM images of core polyDVB particles after a) 1h of polymerization and b) 3h of polymerization.

Figure S5 shows the ATR-FTIR spectra of the polyDVB nanoparticles before and after surface functionalization (introduction of ATRP initiator and grafting-from of pBA chains) . The black spectrum represents the core polyDVB particles synthesized via miniemulsion. After the UV-treatment, when the [2+2] cycloaddition of 4-Formylphenyl 2-bromopropanoate is performed (green spectrum) on the particle surface, a decrease in intensity for C=C the peak positions is observed. After ATRP polymerization with butyl acrylate, a clear C=O peak appears in the red spectrum.
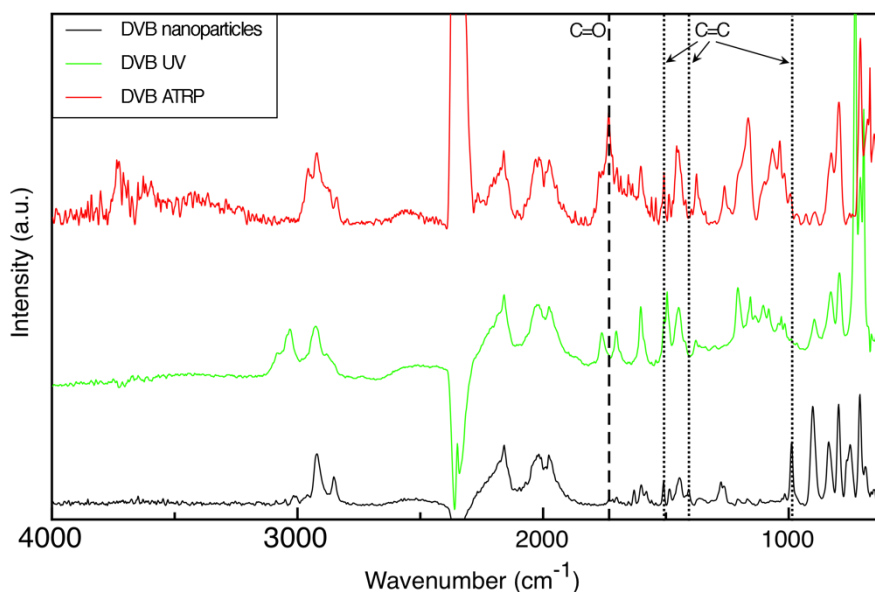

Figure S5: ATR-FTIR spectra of unmodified core polyDVB particles (black), UV-treated polyDVB particles (green) and ATRP-treated polyDVB particles (red).

Figure S6 shows the ATR-FTIR spectra of the polyDVB nanoparticles before and after surface functionalization (introduction of NHS ester). The black ATR-FTIR spectrum represents the core polyDVB particles without any surface functionalization. The green ATR-FTIR spectrum represents the N-succinimidyl-p-formylbenzoate (NHS-aldehyde). After the UV-induced cycloaddition of the NHS-aldehyde to the core polyDVB nanoparticles, the red ATR-FTIR shows successful surface modification.
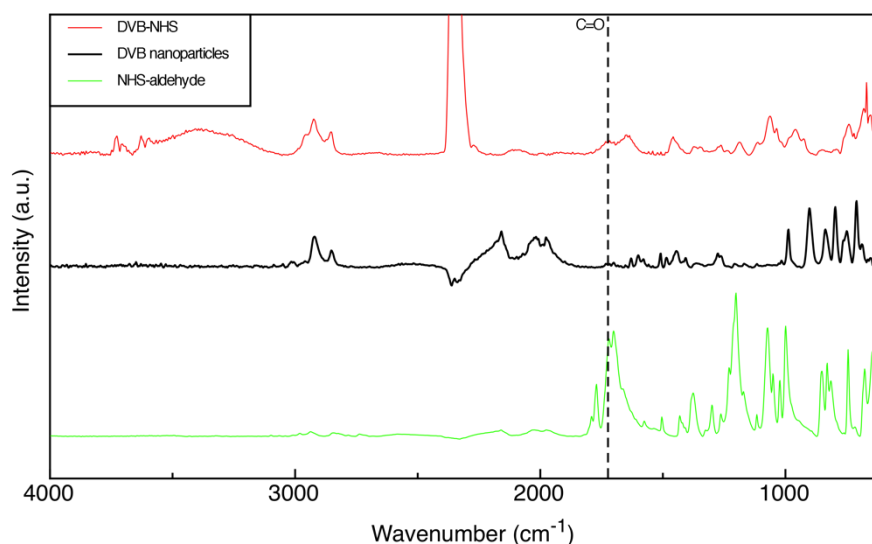

Figure S6: ATR-FTIR spectra of N-succinimidyl-p-formylbenzoate (NHS-aldehyde) (green), umnofified polyDVB particles (black) and polyDVB particles functionalized with N-succinimidyl-p-formylbenzoate after UV-treatment (red).

Table S1: Quantification of core level peak intensities (corrected for photoionization cross sections) for the different sample stages as expressed by the intensity ratios between the Br 3d signal and the C1s main line.

	$I_{Br3d} / I_{C1s\_C-C/C-H}$
Before UV reaction	0
After UV reaction	0.0045
After polymerization	0.0042

Calculations grafting density

The grafting density (GD) can be calculated by means of different methods. The first method is based on the weight increase of the particles after ATRP grafting in which the weight increase is solely from the grafted polymer chains on the photo-functionalized nanoparticles. The second method is based on the increase of the oxygen content in the elemental analysis of the photo-functionalized particles and the ATRP grafted particles. The third is based on XPS core level spectra

Method 1: GD based on weighing of the particles after ATRP grafting

Starting with 10 mg of photo-functionalized nanoparticles, 20 mg of particles were obtained after grafting with BuA. The weight increase of 10 mg can be directly attributed to the total mass of grafted pBuA chains on the surface. The number of chains per gram of core pDVB nanoparticles can be calculated by dividing the weight of the pBuA by the number average molecular weight of the polymer obtained in the sacrificial initiator approach, multiplying by the number of Avogadro, leading to $2.0 \cdot 10^{20}$ pBuA chains per gram of core pDVB nanoparticles.

$$Number\ of\ pBuA\ chains\ _{per\ gram\ core\ DVB\ nanoparticle} = \frac{w_{pBuA}}{\overline{M}_{n\ (pBuA)}} \cdot N_A \qquad (1)$$

Based on the known volume of the core pDVB nanoparticles ($6.97 \cdot 10^5$ nm^3 per particle) and the density of the core pDVB nanoparticles (1.18 g·cm^{-3}), the weight of one core nanoparticle is determined to $8.22 \cdot 10^{-16}$ g. Moreover, each nanoparticle has on average a particle surface area (SA) of $3.80 \cdot 10^4$ nm^2, leading to a total SA of $4.62 \cdot 10^{19}$ nm^2·g^{-1} per gram of DVB. The GD can now be calculated by dividing the number of chains per gram of core nanoparticles ($2.0 \cdot 10^{20}$ chains·g^{-1}) by the surface area per gram of core nanoparticles ($4.62 \cdot 10^{19}$ nm^2·g^{-1}), which results in a GD of 4.34 chains·nm^{-2}.

	BuA grafted pDVB nanoparticles
Diameter core pDVB nanoparticle	110 nm
Radius core pDVB nanoparticle	55.0 nm
Density core pDVB nanoparticles	1.18 g·cm^{-3}
M$_n$	3000 g·mol^{-1}
SA of one core pDVB nanoparticle	$3.80 \cdot 10^4$ nm^2
Volume of one core pDVB nanoparticle	$6.97 \cdot 10^5$ nm^3
Number of chains per gram of core pDVB nanoparticles	$2.0 \cdot 10^{20}$ chains·g^{-1}
SA per gram of core pDVB nanoparticles	$4.62 \cdot 10^{19}$ nm^2·g^{-1}
GD	4.34 chains·nm^{-2}

Method 2: GD based on elemental analysis

	Oxygen content via elemental analysis	Grafting density
Core DVB nanoparticles	1.05%	/
Photo-functionalized nanoparticles	3.3%	7.4 chains·nm^{-2}
ATRP grafted nanoparticles	12.65%	3.6 chains·nm^{-2}

The oxygen detected in the core pDVB particles can be attributed to oxygen adsorbed on the porous surface topology of the nanoparticles.[1]

- *GD of photo-functionalized nanoparticles*

After the photo-functionalization and taking into account the oxygen already present in the core DVB particles, a 2.25% O weight increase is obtained. This means that 1g of photo-functionalized nanoparticles contain 0.0225 g of O. Each photo-functionalized chain contains 3 O atoms, thus the loading capacity (LC) per gram of nanoparticles can be calculated as followed (equation (2)):

$$LC = \frac{w_O}{n_O.MW_O} \tag{2}$$

where LC is the loading capacity in mole, w_O is the weight of the oxygen per gram of nanoparticles obtained via elemental analysis, n_O is the number of oxygen atoms per chain and MW_O is the molecular weight of oxygen. The loading capacity after photo-functionalization is $0.45 \cdot 10^{-3}$ mol per gram of nanoparticles. This value can be transformed into the number of moieties per gram of nanoparticles by multiplying this number by the number of Avogadro (N_A), leading to $2.74 \cdot 10^{20}$ chains per gram of nanoparticles. Based on the volume of one core pDVB nanoparticle ($1.44 \cdot 10^6$ nm^3) and the density of the core pDVB nanoparticles (1.18 g·cm^{-3}), the average weight of one core nanoparticle is $1.69 \cdot 10^{-15}$ g, which corresponds to $5.89 \cdot 10^{14}$ core nanoparticles per gram material. One nanoparticle has a surface area (SA) of $6.16 \cdot 10^4$ nm^2. Thus the surface area per gram nanoparticles is $3.63 \cdot 10^{19}$ nm^2·g^{-1}. Slightly different values are obtained compared to the gravimetry method since new batch of partciles was resynthesized for the elemental analysis. The grafting density can be calculated by dividing the number of chains per gram ($2.74 \cdot 10^{20}$ chains g^{-1}) by the SA per gram material ($3.63 \cdot 10^{19}$ nm^2·g^{-1}). Therefore the grafting density of the particles is 7.4 chains nm^{-2}. This value is associated with a considerable error since the exact amount of adsorbed oxygen in the DVB matrix is unknown for the grafted particles. Using the absolute oxygen content obtained for the ungrafted chains as a direct indicator for the error, an error margin of 32% and thus 7.4 ± 2.4 is obtained.

	BuA grafted pDVB nanoparticles
Diameter core pDVB nanoparticle	140 nm
Radius core pDVB nanoparticle	70.0 nm
Density core pDVB nanoparticles	1.18 g·cm^{-3}
SA of one core pDVB nanoparticle	$6.16 \cdot 10^4$ nm^2
Volume of one core pDVB nanoparticle	$1.44 \cdot 10^6$ nm^3
Number of chains per gram of core pDVB nanoparticles	$2.74 \cdot 10^{20}$ chains·g^{-1}
SA per gram of core pDVB nanoparticles	$3.63 \cdot 10^{19}$ nm^2·g^{-1}
GD	7.43 chains·nm^{-2}

[1] L. Nebhani, D; Schmiedl, L. Barner, C. Barner-Kowollik, *Adv. Funct. Mat.* **2010**, *20*, 2010-2020.

- *GD of ATRP grafted nanoparticles*

After grafting with pBuA, the elemental analysis showed an increase of 11.6 w% in O% per gram material (or 0.116 g O per gram material). Based on the theoretical composition of the monomer, this value corresponds to 0.464 g of pBuA per gram material. Therefore, one gram of material contains 0.464g pBuA and 0.536g pDVB. Based on the molecular weight of pBuA (4000 g·mol^{-1}) and N_A, the number of chains per gram material can be calculated by equation (1), leading to $6.98 \cdot 10^{19}$ chains per gram material. Taking into account that one gram of material corresponds to 0.536 g starting pDVB, a number of $1.30 \cdot 10^{20}$ chains per gram pDVB is deduced.

Based on the volume of one core pDVB nanoparticle ($1.44 \cdot 10^6$ nm^3) and the density of the core pDVB nanoparticles (1.18 g·cm^{-3}), the weight of one core nanoparticle is $1.69 \cdot 10^{-15}$ g. Moreover, one core nanoparticle has a particle surface area (SA) of $6.16 \cdot 10^4$ nm^2, leading to a SA per gram of DVB of $3.63 \cdot 10^{19}$ nm$^2 \cdot$g^{-1}. The GD can now be calculated by dividing the number of chains per gram of core nanoparticles ($1.30 \cdot 10^{20}$ chains·g^{-1}) by the surface area per gram of core nanoparticles ($3.63 \cdot 10^{19}$ nm$^2 \cdot$g^{-1}). This method yields a GD of 3.6 ($\pm$ 0.4) chains·nm^{-2}.

	BuA grafted pDVB nanoparticles
Diameter core pDVB nanoparticle	140 nm
Radius core pDVB nanoparticle	70.0 nm
Density core pDVB nanoparticles	1.18 g·cm^{-3}
M_n	4000 g·mol^{-1}
SA of one core pDVB nanoparticle	$6.16 \cdot 10^4$ nm^2
Volume of one core pDVB nanoparticle	$1.44 \cdot 10^6$ nm^3
Number of chains per gram of core pDVB nanoparticles	$1.30 \cdot 10^{20}$ chains·g^{-1}
SA per gram of core pDVB nanoparticles	$3.63 \cdot 10^{19}$ nm$^2 \cdot$g^{-1}
GD	3.59 chains·nm^{-2}

Method 3: XPS analysis after surface functionalization

In order to estimate the thickness as well as the areal density of the initiator moieties attached to the nanoparticle surface after UV treatment, the C1s and Br3d core level spectra have been integrated using a Shirley background.[2] Taking atomic subshell photoionization cross sections of 0.013 Mb (C1s) and 0.039 Mb (Br3d) into account[3] a relative intensity ratio of I_{Br3d}/I_{C1s} = 0.0045 can be derived. This value now serves as input for a core/shell model simulation. Here, the nanoparticle is divided into a large number of identical cells, each contributing to the photoelectron current an intensity that is affected by the attenuation in cells being closer to the surface according to the mean free path within these cells. In general, the latter depends on the material a photoelectron is travelling through, as well as on its kinetic energy. Taking a mean free path value of 3.3 nm for C1s photoelectrons while travelling through the polymer particles[4] and assuming the same value for C1s photoelectrons travelling through the functionalized surface layer (the shell) together with a slightly increased mean free path value of 3.6 nm for Br3d

[2] D. A. Shirley, *Phys. Rev. B* 1972, **5**, 4709.
[3] J. Yeh, I. Lindau, *Atomic Data and Nuclear Data Tables*, 1985, **32**, 1.
[4] NIST Electron inelastic-mean-free-path database version 1.2, http://www.nist.gov/srd/nist71.cfm

photoelectrons (arising from their higher kinetic energy), the shell thickness can systematically be varied until the measured Br3d/C1s intensity ratio is reproduced. This yields an estimated shell thickness of 1.6 ± 0.2 nm. The uncertainty has been estimated by accounting for errors induced by the integration of the individual core level spectra (5%), the accuracy of the photoionization cross sections (5%) as well as the distribution in particle radius (55 ± 15 nm). Assuming now a similar density for the polymer particle and its shell (1.18g/cm^{3}) the number of initiator molecules forming the shell can be derived. This way, an areal density of 4.7 ± 0.7 molecules/nm^2 is obtained.