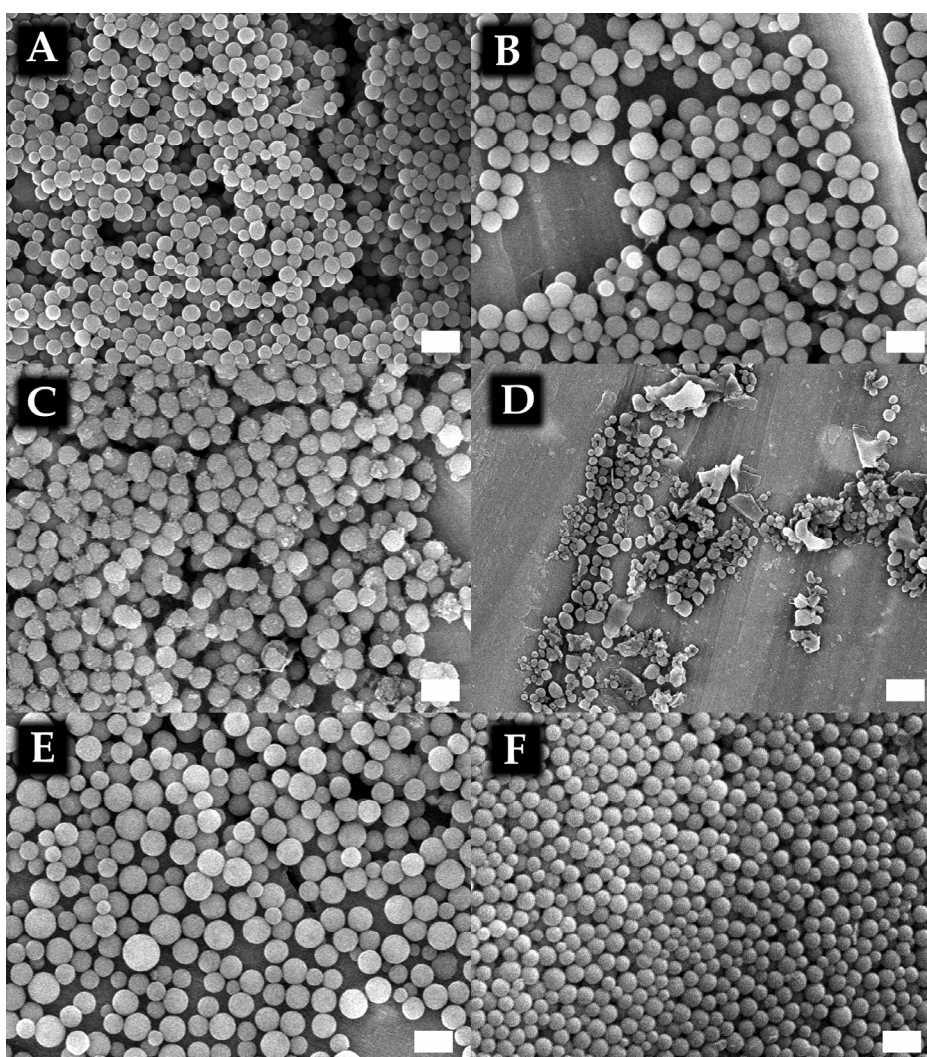


# Non-destructive horseradish peroxidase immobilization in porous silica nanoparticles

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## Supplementary information



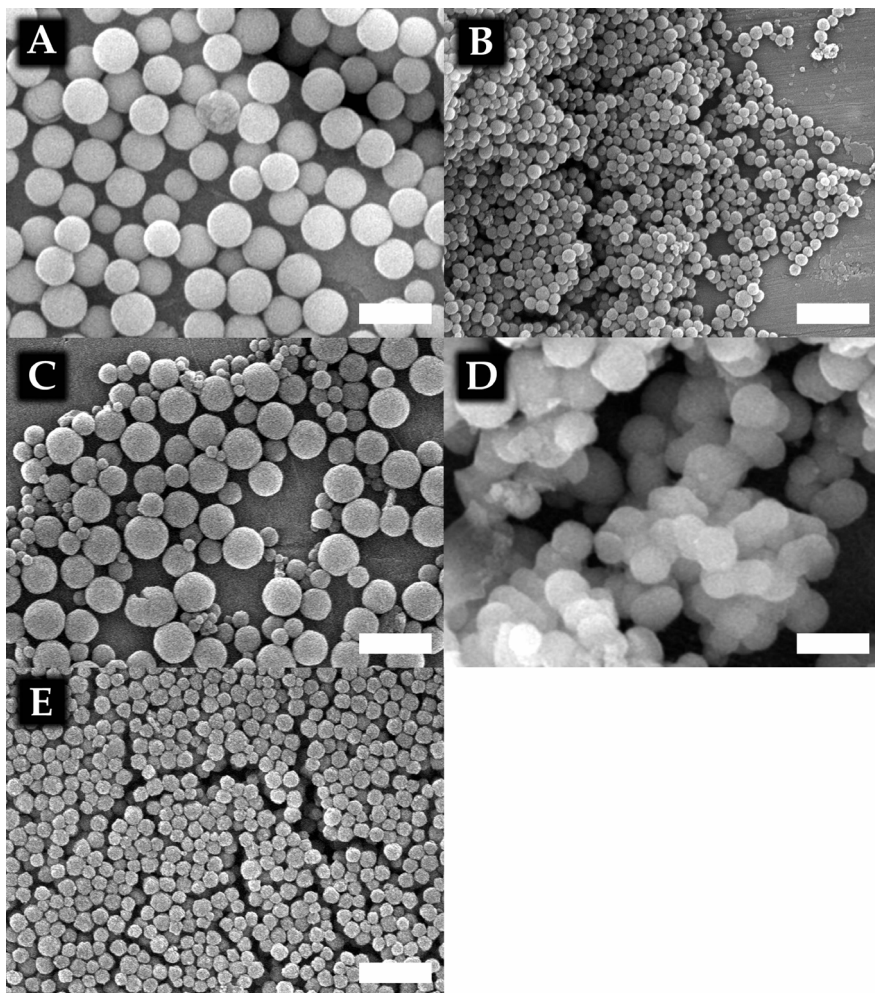
**Figure 1S:** Silica particles prepared under stirring with different molecular weights. Showing the influence of the molecular weight, the type end group and their number as well as the stability of the particles to calcination. A: 2,000 g/mol HO-PEG-OH; B: 2,000 g/mol A-PEG-A, C: 1,000 g/mol A-PEG-A, D: 1,000 g/mol S-PEG-S, E: 550 g/mol S-PEG-OH and F: 2,000 g/mol HO-PEG-OH after calcination. All scale bars are 1  $\mu\text{m}$ .

**Table 1S:** Relationship between formulation and silica morphology in water. This table provides additional information to that provided in Table 1, and shows more formulations with DGS and PEG in water that will lead to either monolithic fragments, fused particles or spherical particles. In the latter case, the mean particle diameter as found by SEM is given.

PEG MW [g/mol]	end-group	microscopic (SEM) appearance <sup>a</sup>	Stirring (S) or Ultra-sound (U)	Particle size (SEM) nm <sup>b</sup>
200	HO-PEG-OH	no material	S	
600		no material	S	
8,000		spherical particles	S	415±56
100,000		spherical particles	S	152±25
200	A-PEG-A	monolith fragments	S	
600		monolith fragments	S	
2,000		spherical particles	U	225±39
8,000		spherical particles monolith fragments plus very few particles	S	454±80
200	S-PEG-S	monolith fragments	S	
600		monolith fragments nearly spherical particles	S	~180
1,000		spherical particles	S	275±36
8,000		spherical particles	S	
550	S-PEG-OH	monolith fragments	U	
1,000		spherical particles	S	323±64

<sup>a</sup> For examples of these structures, see Figure 1C,D,E in the paper.

<sup>b</sup> Particles sizes were calculated from SEM images by measuring the particle diameter manually, STD were calculated from 150 measured particles. Where 2 numbers appear, the particle sizes were bimodal, and the means of both modalities are reported.



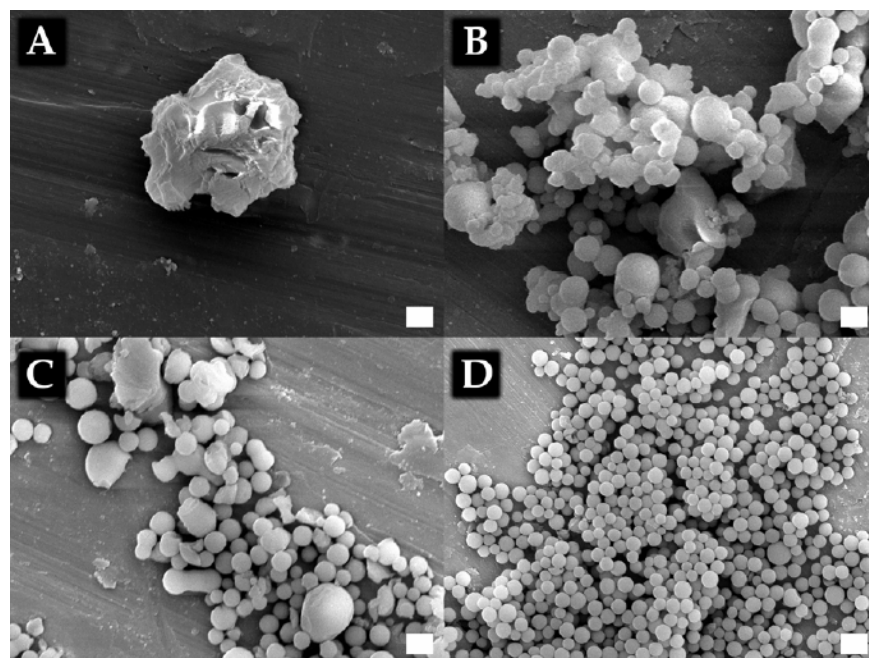
**Figure 2S:** Influence of ultrasound on the formation of silica particles using different PEG. Silica prepared with 10,000 g/mol HO-PEG-OH using A: stirring, B: ultrasound. Silica prepared with 2,000 g/mol HO-PEG-OH using C: ultrasound (for the complementary experiment with stirring see Figure 1S in the supplementary information). Silica prepared with 1,000 g/mol A-PEG-A using D: stirring, E: ultrasound. All Scale bars are 1  $\mu\text{m}$ .

**Table 2S:** Effect of buffer and glycerol on the preparation of silica particles. All formulations are done with stirring and aging at room temperature. For more examples, see Table 2 in the paper.

Entry	PEG MW [g/mol]	end-group	Ionic strength (mM)	pH	glycerol [mg]	microscopic (SEM) appearance <sup>a</sup>	Particle size (SEM) nm <sup>b</sup>
							697±119
1	2,000	A-PEG-A	10	5.1		particles	148±35
2	2,000	A-PEG-A	10	9.0		fused particles	
3	1,100	S-PEG-OH	100	5.1		particles	329±68
4	1,100	S-PEG-OH		7.0	1150	fused particles	
5	1,100	S-PEG-OH		7.0	4600	fused particles	
6	2,000	S-PEG-OH	10	7.4	1500	fused particles	
7	8,000	S-PEG-OH	50	7.4	1500	fused particles	
8	1,100	S-PEG-OH	100	7.4	1500	monolith fragments	
9	2,000	A-PEG-OH	10	7.4	500	fused particles	
10	2,000	A-PEG-OH	10	7.4	1000	fused particles	
11	8,000	A-PEG-OH	10	7.4		monolith fragments	

<sup>a</sup> All samples were prepared using stirring rather than ultrasound

<sup>b</sup> Particles sizes were calculated from SEM images by measuring the particle diameter manually, STD were calculated from 150 measured particles. Where two numbers appear, the particle sizes were bimodal, and the means of both modalities are reported.



**Figure 1S:** SEM images of 10,000 g/mol HO-PEG-OH with increasing amounts of water. A: 250  $\mu$ l, B: 500  $\mu$ l, C: 1 ml, D: 1.25 ml, respectively. All scale bars are 1  $\mu$ m.