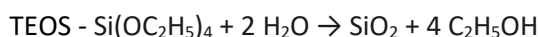
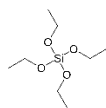





<b>Protocol</b>	#2.1
<b>Title</b>	<b>BOMB coating ferrite MNPs with silica oxide</b>
<b>Keywords</b>	magnetic nanoparticles, SiO <sub>2</sub> , magnetic separation, silica coating
<b>Authors</b>	Oberacker P*, Stepper P*, Bond DM*, Höhn S, Focken J, Meyer V, Schelle L, Sugrue VJ, Jeunen GJ, Moser T, Hore SR, von Meyenn F, Hipp K, Hore TA# and Jurkowski TP#
<b>Citation</b>	<i>Oberacker et al., Bio-On-Magnetic-Beads (BOMB): Open platform for high-throughput nucleic acid manipulation. PLOS Biology, 17(1), <a href="https://doi.org/10.1371/journal.pbio.3000107">https://doi.org/10.1371/journal.pbio.3000107</a></i>
<b>Online</b>	<a href="https://bomb.bio/protocols/">https://bomb.bio/protocols/</a>
<b>Revision</b>	V1.3 (27 <sup>th</sup> February 2019)

## Summary

Here we provide a simple protocol for silica-coating of ferrite MNPs (BOMB protocol #1.1). The silica-coated magnetic beads are synthesised by a modified protocol including the hydrolysis of tetraethyl orthosilicate (TEOS) on the surface of ferrite magnetic core particles according to Stöber et al. 1968 [1].



## Chemicals

Name	Provider	PN	MW [g/mol]		Safety codes
<b>Ethanol (C<sub>2</sub>H<sub>6</sub>O, 99.9 %)</b>	Riedel-de Haën	34963	46.07	 Danger	H: 225-319 P: 210-280-305+351+338-308+313
<b>Tetraethyl orthosilicate (≥99%) (GC)</b>	Aldrich	86578	208.33	 Danger	H: 226+319+332+335
<b>Ammonia solution (NH<sub>4</sub>OH, 25%)</b>	EMD Millipore	1.05432	n.a.	 Corrosive Danger	H: 290+314+335+400 P: 273+280+301+330+331+305+351+338+308+310

Please consult appropriate MSDS information before working with these chemicals! Use lab coat, gloves and eye protection at all times! The chemicals are available from other providers as well. No preference is given to the indicated vendors.

## Equipment and setup



Fume hood

Heated magnetic stirrer (e.g. IKAMAG REO)

Strong neodymium permanent magnet (e.g. NdFeB N45 40x40x20 mm)

Sterile plastic bottles

## BOMB Silica-coating

Step	Task	Time	✓
	<i>All procedures can be performed under inert N<sub>2</sub> atmosphere or standard conditions</i>		
1	Prewash 22.5 g (wet mass ~1.2 g dry) of magnetic core particles with ethanol	5 min	<input type="checkbox"/>
2	Add 2 L of 99% ethanol in a large bottle (2.5 litre), place it on a magnetic stirrer (300-400 rpm) and heat it up to ~80 °C.	30 min	<input type="checkbox"/>
3	Add 50 ml of a 25% ammonia solution and the prewashed beads and stir for 30 minutes	30 min	<input type="checkbox"/>
4	Add 45 ml Tetraethyl orthosilicate (TEOS) under constant stirring and incubate for another 30 minutes	30 min	<input type="checkbox"/>
5	Add 400 ml of ddH <sub>2</sub> O to the solution		<input type="checkbox"/>
6	Allow the reaction to proceed for >4 h (ideally overnight)	4 hours to o/n	<input type="checkbox"/>
7	Cool down the solution to RT	15 min	<input type="checkbox"/>
8	Separate the coated MNPs using a strong neodymium magnet.	5 min	<input type="checkbox"/>
9	Wash twice with pure water	15 min	<input type="checkbox"/> <input type="checkbox"/>
10	Wash twice with pure ethanol	15 min	<input type="checkbox"/> <input type="checkbox"/>
11	Wash with pure water until the pH of the solution becomes neutral (3-4 times)	30 min	<input type="checkbox"/> <input type="checkbox"/>
<b>End</b>	Check the yield by weighing the wet mass of the beads	~12 h (2 h hands-on)	
	Store @ RT for up to 1 year		

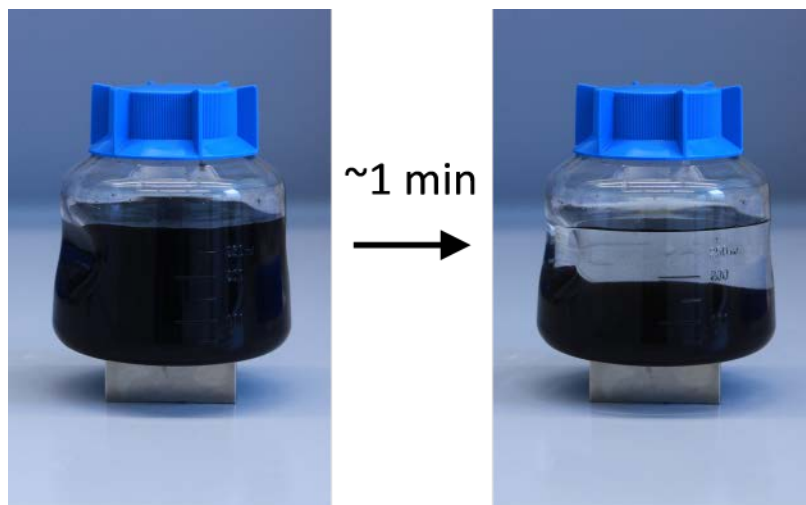
## Modifications

By using different ratios of core particles and TEOS one can control the thickness of the glass layer and consequently the size of the particles. The standard ratio used in the above protocol is: 1 g of magnetic core particles, 2 ml TEOS, 20 ml of ethanol, 0.5 ml of 25 % ammonia solution, 4 ml of water, which results in particles with an average size of ~400 nm. Increasing or decreasing the TEOS to beads ratio results in formation of other sizes of coated particles.

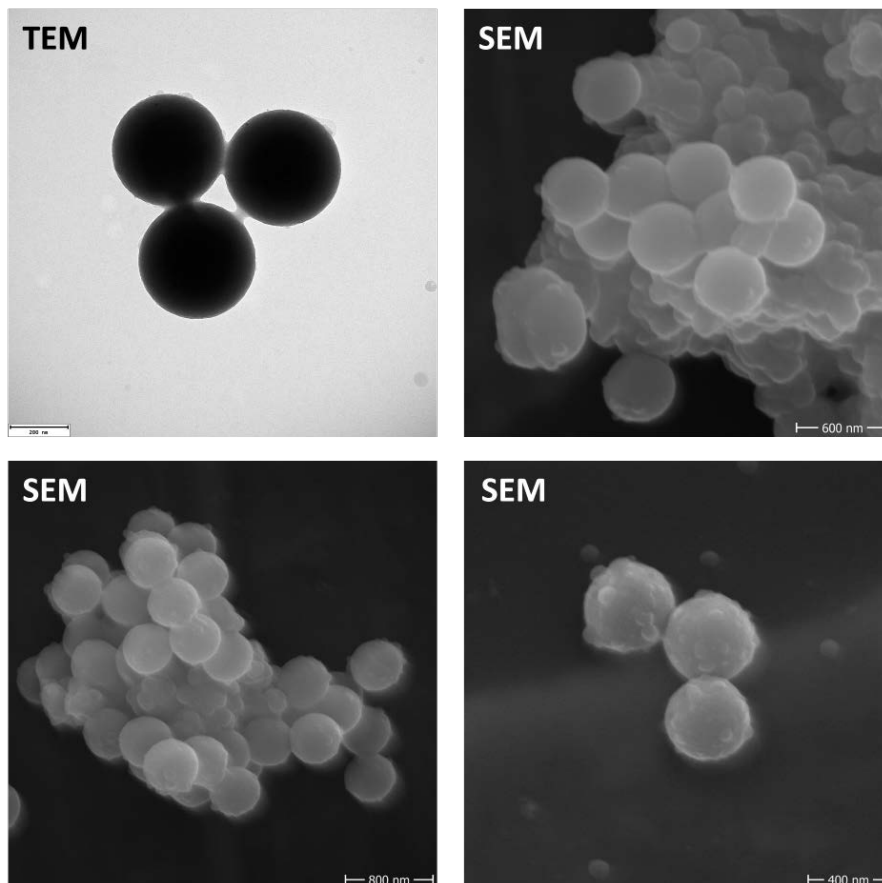
## Troubleshooting

Problem	Solution
Brown colour of the reaction, low yield of beads retained	<ul style="list-style-type: none"> <li>The uncoated magnetic core particles get slowly oxidized during storage, prepare fresh core particles and redo the reaction</li> </ul>
Large amounts of white precipitate	<ul style="list-style-type: none"> <li>TEOS hydrolyses spontaneously in water solution, thus forming silica nanoparticles. Use fresh TEOS for the reaction</li> </ul>

## Exemplary results



**Fig 1: Silica coated MNPs.** Magnetic decantation happens within a minute. The water appears clear indicating no iron oxidation occurring during storage.



**Fig 2: Electron micrographs of silica coated MNPs.**

## References

1. Stöber W, Fink A, Bohn E. Controlled growth of monodisperse silica spheres in the micron size range. *J Colloid Interface Sci.* 1968;26: 62–69. doi:10.1016/0021-9797(68)90272-5