

In situ reduction of Ag on magnetic nanoparticles with gallic acid: effect of the synthesis parameters on morphology

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1 ***In situ* reduction of Ag on magnetic nanoparticles with gallic acid: effect of the**  
2 **synthesis parameters on morphology**

3  
4 **Abstract**

5 Fe<sub>3</sub>O<sub>4</sub>—Ag composite nanoparticles (NPs) were prepared by a new *in situ* reduction of  
6 Ag on the surface of Fe<sub>3</sub>O<sub>4</sub> NPs using gallic acid as reducing agent in different NPs/gallic  
7 acid ratio. The obtained NPs were characterized by Field-Emission Scanning/Scanning  
8 Transmission Electron Microscopy and Fourier-transform infrared spectroscopy in order  
9 to evaluate their crystal structure, their morphology, the effective functionalization and  
10 the presence of Ag. Different morphologies were obtained: polydispersed aggregates,  
11 nanoflower-like and nanodumbbell nanocomposites, depending on the synthesis  
12 conditions. Well-defined Fe<sub>3</sub>O<sub>4</sub>-Ag nanodumbbells were obtained in aqueous media, with  
13 NPs/gallic acid ratio of 10:1. It was also assessed that silica shell is not a key issue to  
14 optimize the morphology of Ag NPs nucleated on the Fe<sub>3</sub>O<sub>4</sub> core.

15  
16 **Structured Abstract**

17 **Aim:** Synthesis of Fe<sub>3</sub>O<sub>4</sub>—Ag composite nanoparticles by a new *in situ* reduction of Ag  
18 NPs on the surface of Fe<sub>3</sub>O<sub>4</sub> NPs using gallic acid as reducing agent.

19 **Materials & Methods:** The influence of process parameters on NPs morphology and  
20 functionalization was evaluated by means Field-Emission Scanning/Scanning  
21 Transmission Electron Microscopy and Fourier-transform infrared spectroscopy.

22 **Results and Conclusions:** The synthesis conditions affect the morphology of the  
23 obtained nanoparticles, evidencing the formation of polydispersed aggregates,  
24 nanoflower-like or nanodumbbell nanocomposites. In particular, well-defined

25 nanodumbbells were obtained in aqueous media, with NPs/gallic acid ratio of 10:1; while  
26 the presence of a silica shell did not improve the morphology of Ag NPs nucleated on the  
27 Fe<sub>3</sub>O<sub>4</sub> core.

28

### 29 **Lay abstract:**

30 Nanoparticles (NPs) are extensively investigated in the biomedical field for the diagnosis  
31 and the treatment of tumors. The aim of this work is to develop a simple and green method  
32 to produce composite nanoparticles, formed by iron oxides (magnetic) and silver NPs.

33 Magnetic NPs can be moved using an external magnetic field and localized in the tumor;  
34 silver NPs can generate useful heat to kill tumor cells when exposed to light and possess  
35 antibacterial properties. The performed study showed that by varying the process  
36 parameters (e.g. amount of reagents, presence of coatings on the particles ...), it is possible  
37 to obtain multifunctional NPs with different shapes and sizes, potentially useful for the  
38 treatment of tumors.

39

### 40 **Keywords**

41 Magnetic nanoparticles, silver nanoparticles, *in situ* reduction, gallic acid.

42

### 43 **Introduction**

44 Magnetic nanoparticles (MNPs) are one of main objects of study in the current literature  
45 concerning nanotechnology [1, 2]. MNPs are often suggested in biomedical applications  
46 as drug carriers due to their ability, when exposed to an external magnetic field, to be  
47 moved purposely to a target site in the human body. Most of the MNPs described in  
48 literature are composed of iron oxide (mainly Fe<sub>3</sub>O<sub>4</sub> or  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>) often used as magnetic

49 cores enclosed into an organic or inorganic shell (such as silica, gold, fatty acid, citric  
50 acid and others) [3-5]. Iron oxide MNPs are among the most appropriate nanoplatforms  
51 for biomedical applications due to their chemical stability, superparamagnetism,  
52 biocompatibility, easiness of synthesis and since they can be further functionalized with  
53 molecules of particular interest, such as anticancer drug or specific bioactive molecules  
54 [3-5]. Iron oxide MNPs are used as contrast agents in magnetic resonance imaging  
55 (MRI), drug delivery systems [6, 7] and deeply investigated for applications in the field  
56 of cancer treatment by hyperthermia [2, 8, 9].

57 The decoration of the magnetic core with ultra-small metal nanoparticles can further  
58 enhance the variety of potential applications of this versatile category of nanoplatform in  
59 biomedical field, merging the magnetic properties of the core with the peculiar properties  
60 of metal nanoparticles [10, 11].

61 For example, silver nanoparticles are of great interest because they exhibit peculiar  
62 functionalities, such as, among others, antimicrobial, antiviral, optical and catalytic  
63 properties [12-15]. Their chemical stability, biocompatibility and long-term efficacy  
64 toward a broad bacterial spectrum make them suitable in many biomedical applications  
65 as targeted tool for *in situ* infection treatment [16].

66 Besides the well-known antimicrobial activity [14, 17, 18], silver nanoparticles are of  
67 great interest in the field of surface-enhanced Raman spectroscopy [19] and photothermal  
68 cancer therapy [20]. Silver nanoparticles exhibit strong absorption band at about 400 nm.  
69 The position of the absorption band can vary depending on the size and shape of silver  
70 nanoparticles.

71 Magnetic iron oxide nanoparticles and silver nanoparticles can be combined into  
72 nanocomposite platforms implementing the above mentioned properties [6]. For example,

73 the decoration of Fe<sub>3</sub>O<sub>4</sub> nanoparticles with Ag nanoparticles gives rise to enhanced optical  
74 responses [21]. Moreover, the combination of magnetic (Fe<sub>3</sub>O<sub>4</sub>) and antibacterial (Ag)  
75 properties, can be exploited for the magnetic targeting and removal of antimicrobial  
76 agent.

77 Since magnetic nanoparticles are very sensitive to agglomeration, many methods have  
78 been investigated in literature to impart electrostatic or steric repulsion, such as the  
79 coating with organic (e.g. surfactants, polymers) or inorganic materials (e.g. silica, metal  
80 oxides, metals) [22, 23]. Moreover, MNPs have been decorated with noble metals NPs,  
81 especially Au NPs, by using different approaches [24]. One of the proposed methods to  
82 decorate MNPs with metallic NPs is to cover the magnetic NPs with a silica layer, which  
83 can be easily functionalized and acts as intermediate layer between the magnetic core and  
84 the metallic NPs [25]. Noble metal NPs can be also directly attached to the magnetic core  
85 using silanes or coupling and reducing agents such as NaBH<sub>4</sub>, hydroxylamine, plant  
86 extracts, or by the metal NPs adsorption on magnetic NPs surface [11, 24, 26-30].

87 In the present work we report a new method to prepare Fe<sub>3</sub>O<sub>4</sub>—Ag composite  
88 nanoparticles by *in situ* reduction of Ag on a magnetic core of Fe<sub>3</sub>O<sub>4</sub>, focusing the  
89 attention on the effect of process parameters (synthesis media and functionalization steps)  
90 on the nanoparticles nucleation and morphology. In particular, Gallic Acid (GA) has been  
91 used for the first time, on the basis of authors knowledge, as *in situ* reducing agent for Ag  
92 nanoparticles directly on Fe<sub>3</sub>O<sub>4</sub> NPs' surface. GA has been chosen for its above  
93 mentioned double action, as well as for its intrinsic anti-inflammatory and anti-tumor  
94 action, and for its potential sustainability, due to the possibility to be extracted from  
95 vegetal wastes coming from the food production chains [31]. The obtained composite  
96 NPs, due to both the magnetic properties of Fe<sub>3</sub>O<sub>4</sub> and the optical/antibacterial properties

97 of Ag NPs, can be a useful tool for targeting bacterial infection as well as for treating  
98 tumor and associated complications.

99

## 100 **2. Experimental**

101 Each experimental step is described in the following paragraphs. All reactants were  
102 purchased from Sigma Aldrich®/Merck.

103

### 104 **2.1 Synthesis of CA stabilized MNPs**

105 The synthesis of Fe<sub>3</sub>O<sub>4</sub> NPs (Magnetite-based - M NPs) was carried out by co-  
106 precipitation [32-34]. As reported also in [10, 11], M NPs were synthesized by mixing  
107 aqueous solution of Fe<sup>2+</sup> and Fe<sup>3+</sup> salts in alkaline conditions for ammonium hydroxide  
108 NH<sub>4</sub>OH (Merck, 25% aqueous solution) [5]. Two solutions 0.1 M of ferrous chloride  
109 FeCl<sub>2</sub> · 4 H<sub>2</sub>O and ferric chloride FeCl<sub>3</sub> · 6H<sub>2</sub>O (Sigma Aldrich, > 99%) were prepared  
110 separately by dissolving the required quantities of FeCl<sub>2</sub> · 4H<sub>2</sub>O and FeCl<sub>3</sub> · 6H<sub>2</sub>O in 100  
111 ml of bi-distilled water, magnetically stirred in two distinct beakers. The pH was around  
112 3 for the ferrous chloride solution, and around 2 for the ferric chloride solution. After the  
113 complete dissolution of the salts, 50 ml of 0.1M FeCl<sub>3</sub> · 6H<sub>2</sub>O and 37.5 ml of the 0.1M  
114 FeCl<sub>2</sub> · 4H<sub>2</sub>O were mixed in a beaker, reaching a pH value around 1.7.

115 The pH was then adjusted to about 10 by adding NH<sub>4</sub>OH dropwise to the iron salts  
116 solution, under continuous mechanical stirring: the mixture turned to black, due to the  
117 formation of a suspension of iron oxide NPs (M NPs).

118 The as obtained suspension was then sonicated for 20 minutes and washed in bi-distilled  
119 water, using a magnet to induce sedimentation. The as obtained M NPs were finally re-  
120 suspended in 100 ml of bi-distilled water reaching a pH between 7-8.

121 Aiming to improve the MNPs suspension stability, the synthesis was followed by the M  
122 NPs stabilization with Citric Acid (CA) [32, 33]. 120 ml of a 0.05 M of CA (Sigma  
123 Aldrich, > 99.5%) solution were added to the M NPs, previously separated by  
124 sedimentation, adjusting the pH to a value of 5.2 by adding dropwise a concentrated  
125 NH<sub>4</sub>OH solution. The new suspension was then heated to 80 °C and stirred (150 rpm) for  
126 90 minutes. The duration of the heating step was optimized in order to promote the CA  
127 adsorption on the M NPs while minimizing the magnetite decomposition to maghemite.  
128 Finally, the CA-capped MNPs were rinsed by means of an ultrafiltration device (Solvent  
129 Resistant Stirred Cells - Merck Millipore)(4 steps in 50 ml of bi-distilled water at pH =  
130 10.2) and suspended in 120 ml of bi-distilled water, adjusting the pH of the final water  
131 suspension to 10.2 with diluted ammonia.

132

## 133 **2.2 Silica coating on MNPs**

134 A part of CA-capped MNPs was coated with a silica shell following the Stöber method  
135 [34] by suspending them in a mixture of Tetraethyl orthosilicate (TEOS) as silica  
136 precursor, ethanol and water (ethanol:water volume ratio of 4:1), under stirring in orbital  
137 shaker at 37 °C and 150 rpm for 3 hours. Approximately 0.002 ml of TEOS for each mg  
138 of magnetic nanoparticles was used. Then, the obtained MS NPs were double-washed  
139 with bi-distilled water using an ultrafiltration device and re-dispersed in water.

140

## 141 **2.3 Functionalization with aminopropyltriethoxysilane (APTES)**

142 To enhance the interaction between Ag NPs and magnetic NPs, terminal -NH<sub>2</sub> groups  
143 were introduced on the surface of both M NPs (route A) and MS NPs (route B) by means  
144 of functionalization with APTES (> 98%, Merck).

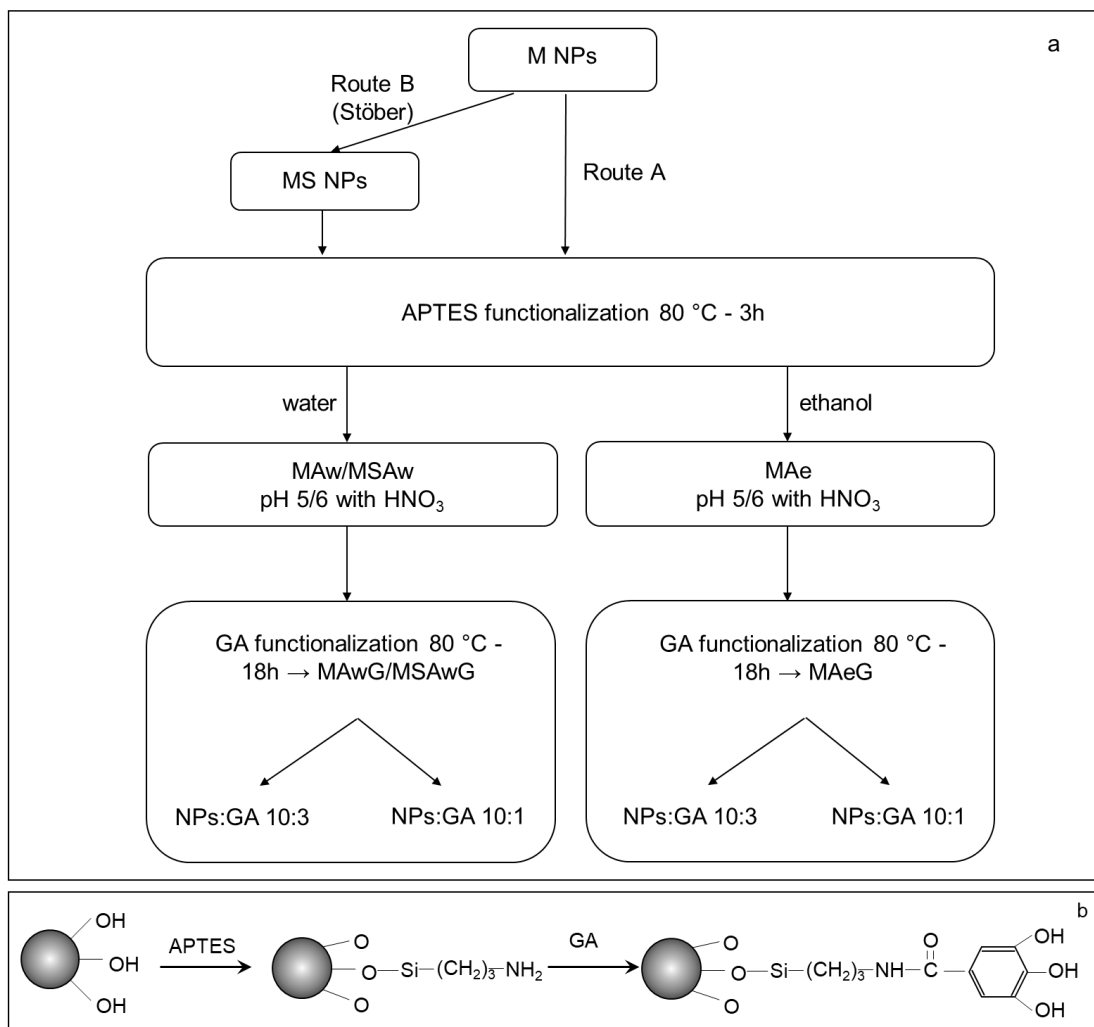
145 The functionalization with APTES on the M NPs was performed including a final step of  
146 washing and re-suspension in two different media (ethylic alcohol or water) in order to  
147 verify the role of the medium on GA grafting and, in turn, on the nucleation of Ag NPs.  
148 MS NPs were resuspended only in water.  
149 For this step, 3 ml of each magnetic NPs suspension were diluted in 50 ml of absolute  
150 ethanol (anhydrous, >99.9%, Carlo Erba Reagents) and sonicated for 3 minutes. 10  $\mu$ l of  
151 APTES were then added to the resulting colloidal suspensions. The obtained suspensions  
152 were then heated at 80 °C under mechanical stirring for three hours. The stirring time has  
153 been prolonged respect to the conventional synthesis reported in literature, to enhance the  
154 APTES grafting and assure a uniform presence of terminal amine group on the magnetic  
155 nanoparticles surface, as reported in previous works [10, 11, 35]. Subsequently, the  
156 mixtures were cooled at room temperature, the nanoparticles were magnetically separated  
157 and centrifuged (7500 rpm /20 minutes) for three times, washed in distilled water and re-  
158 suspended in water or ethanol (M NPs) or only in water (MS NPs) up to a volume of 50  
159 ml. To induce positive charge at the surface of the APTES-coated nanoparticles, a diluted  
160 HNO<sub>3</sub> solution (0.05 ml of 6 M HNO<sub>3</sub> with 20 ml of water) was added drop by drop until  
161 reaching a pH value  $\approx$  5-6, and stirred for 10 minutes.

162

#### 163 **2.4 Functionalization with GA**

164 Gallic acid (GA) was added to both ethanol and water-based M NPs suspensions using  
165 two different NPs/GA weight ratio (10:3 and 10:1) [37] and to water-based MS NPs  
166 suspension using a NPs/GA weight ratio of 10:1. The mixtures were stirred for 18 h at 80  
167 °C, at the end of the treatment the NPs dispersed in ethanol were washed and centrifuged

168 three times with ethanol and re-suspended in ethanol; the same procedure was adopted  
 169 for NPs dispersed in water, using bi-distilled water as washing and dispersion medium.  
 170 Figure 1 resumes the steps of the functionalization process.



171

172 **Figure 1:** functionalization process (a) and scheme (b) of MAwG and MAeG synthesis.

173

## 174 2.5 Synthesis of Fe<sub>3</sub>O<sub>4</sub>—Ag composite nanoparticles

175 The reduction of Ag NPs was performed both for ethanol and water dispersed M NPs and  
 176 MS NPs, using a AgNO<sub>3</sub>/GA weight ratio of 1.7. AgNO<sub>3</sub> was dissolved in water and  
 177 mixed with the NPs suspension at 37 °C for 15 minutes at 150 rpm; the pH of the mixture  
 178 was adjusted to about 11 using NH<sub>4</sub>OH. At the end of the synthesis the mixture was

179 washed with bi-distilled water, NPs were magnetically separated and re-dispersed in bi-  
 180 distilled water for further analysis.

181 Table 1 reports the acronyms of the obtained NPs and resume their synthesis steps.

	<b>Magnetite</b>	<b>Silica shell</b>	<b>Synthesis in water (w) or ethanol (e)</b>	<b>APTES</b>	<b>Gallic acid (GA)</b>	<b>Silver</b>
<b>M</b>	✓					
<b>MAw</b>	✓		w	✓		
<b>MAe</b>	✓		e	✓		
<b>MAwG</b>	✓		w	✓	✓	
<b>MAeG</b>	✓		e	✓	✓	
<b>MAwGAg</b>	✓		w	✓	✓	✓
<b>MAeGAg</b>	✓		e	✓	✓	✓
<b>MS</b>	✓	✓				
<b>MSAw</b>	✓	✓	w	✓		
<b>MSAwG</b>	✓	✓	w	✓	✓	
<b>MSAwGAg</b>	✓	✓	w	✓	✓	✓

182

183 **Table 1:** acronyms and resume of NPs synthesis steps.

184

## 185 **2.6 Characterization**

186 The structural characterization of M NPs was carried out by means of wide-angle (2θ  
 187 within 10-70°) X-ray diffraction analysis (XRD – X’Pert Philips diffractometer) using  
 188 the Bragg Brentano camera geometry and CuKα incident radiation. The identification of

189 crystalline phases was performed by using X'Pert HighScore program equipped with  
190 PCPDFWIN database.

191 To evaluate the effective functionalization with APTES and GA, all NPs before the *in*  
192 *situ* reduction of silver were analyzed by means of Fourier-transform infrared  
193 spectroscopy (FTIR Tensor 27, Bruker Optics S.p.A, Ettlingen, Germany) from 4500 to  
194 300 cm<sup>-1</sup> and with 2 cm<sup>-1</sup> resolution. OPUS software (v. 6.5, Bruker S.p.A) was used for  
195 instrumental control and spectral acquisition.

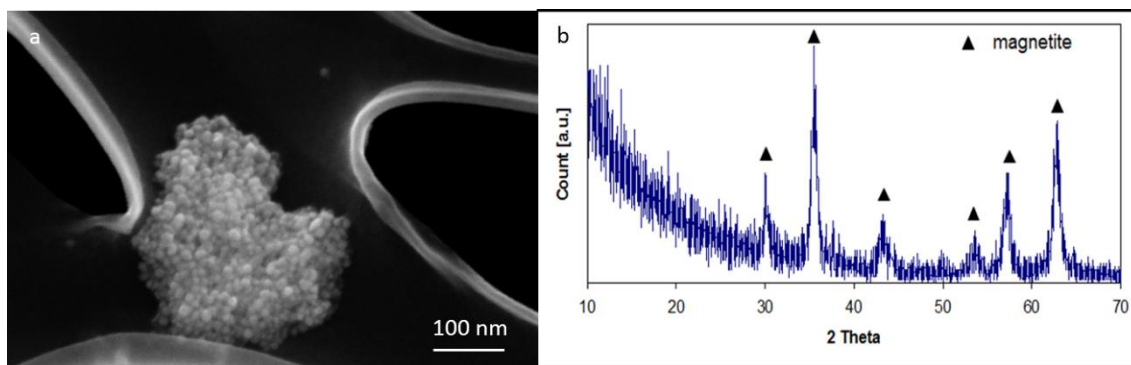
196 In order to investigate the NPs functionalization after each synthesis step, the formation  
197 of Ag NPs and their dimension and distribution, all nanoparticles were characterized in  
198 terms of morphology and composition by means of FESEM (Field-Emission Scanning  
199 Electron Microscope - SUPRATM 40, Zeiss, Germany) and STEM (Scanning  
200 Transmission Electron Microscopy - Merlin Gemini Zeiss, Germany), equipped with  
201 Energy Dispersive Spectroscopy (EDS). For STEM observation a drop of each NPs  
202 suspension was placed on a copper grid with carbon film (SPI Supplies® Brand Lacey  
203 Carbon Coated 200 Mesh Copper Grids – JEOL S.p.A.). STEM analyses were carried out  
204 using different detectors and signals (secondary and backscattered electrons, the Bright  
205 Field - BF and Dark Field – DF) in order to better distinguish the metallic NPs.

206

### 207 **3. Results**

#### 208 **3.1 Route A**

209 Morphological (FESEM) and phase (XRD) analyses of M NPs (Figure 2) revealed the  
210 presence of NPs with spherical morphology and a diameter of about 5-20 nm. The XRD  
211 pattern reports the presence of only magnetite/maghemite phase, in accordance with the  
212 synthesis performed in air.



213

214 **Figure 2:** CA stabilized M NPs: a) morphological evaluation (FESEM), b) XRD  
 215 analysis

216

217 Table 2 reports the peaks and bands identified by FTIR analysis of M NPs (stabilized with  
 218 CA), MAw, MAwG, MAe and MAeG. Figure 3 shows as example the curves obtained  
 219 for M, MAw and MAwG NPs/GA 10:3. As it can be noticed, M NPs spectrum shows the  
 220 presence of Fe-O bond between 500-600  $\text{cm}^{-1}$  typical of iron-oxide NPs [38], the  
 221 asymmetric stretching of C-O from the COOH group of CA at 1310  $\text{cm}^{-1}$ , the large peak  
 222 between 1400  $\text{cm}^{-1}$  and 1660  $\text{cm}^{-1}$  can be assigned both to COO-Fe bonding (in literature  
 223 1431  $\text{cm}^{-1}$ ) and to the shift of peak characteristic of symmetric vibration of C=O due to  
 224 the CA interaction with magnetite; moreover a band at about 1700  $\text{cm}^{-1}$ , relative to the  
 225 C=O stretching vibration from the free COOH group of CA, is still visible. In the curve a  
 226 band between 2600-2950  $\text{cm}^{-1}$  ascribable to  $\text{CH}_2$  stretching and a barely resolvable band  
 227 at about 3300  $\text{cm}^{-1}$  due to OH stretching are also observable [38-40]. The MAw NPs  
 228 spectrum shows a shoulder at 850-950  $\text{cm}^{-1}$  ascribable to the stretch of Si-OH symmetric  
 229 vibration or Si-O-Fe bond, and two bands around 960 and 1000-1150  $\text{cm}^{-1}$  belonging to  
 230 the stretching of Si-O and Si-O-Si groups. The peaks at about 1330 and 1620  $\text{cm}^{-1}$   
 231 relative to C-N and NH stretching vibrations respectively, the band between 2920-2850  
 232  $\text{cm}^{-1}$  ascribable to the stretching and vibration of C-H and the bands at 3445 and 1640  $\text{cm}^{-1}$

233 <sup>1</sup>, attributable to the N–H stretching vibration and NH<sub>2</sub> bending mode of free NH<sub>2</sub> groups  
 234 [41-43], are hardly attributable due to the overlapping with CA peaks.  
 235 The functionalization with GA introduces three peaks between 1600-1200 cm<sup>-1</sup> ascribable  
 236 to the C=C bond, a small peak at about 1700 cm<sup>-1</sup> relative to the C=O stretch of carboxylic  
 237 group and a band centered at about 3400 cm<sup>-1</sup> relative to OH groups of GA [44, 45].  
 238 FTIR spectra of M, MAe and MAeG (obtained peaks reported in table 2) evidence a band  
 239 between 400 and 500 cm<sup>-1</sup> ascribable to Fe-O bond and the peak at 1310 cm<sup>-1</sup> due to the  
 240 asymmetric stretching of -CO from the COOH group, the peak probably due to both the  
 241 COO-Fe bond and to the C=O of COOH group that interact with magnetite NPs, the C=O  
 242 vibration from the free COOH group of CA at about 1700 cm<sup>-1</sup> [38, 40]. The  
 243 functionalization with APTES is confirmed by the presence of a shoulder at about 970  
 244 cm<sup>-1</sup> ascribable to Si-O stretching, a peak at about 1100 cm<sup>-1</sup> due to the Si-O-Si stretching  
 245 and the shoulder at about 2870 cm<sup>-1</sup> is related to the stretching vibration C-H. Even if  
 246 after the synthesis the NPs were washed, some characteristic peaks of ethanol (not  
 247 reported in table 2) can be observed in the spectrum of MAe at about 1260-1000 cm<sup>-1</sup>  
 248 ascribable to the stretching of C-O and between 3400-3300 cm<sup>-1</sup> related to the OH  
 249 stretching. Both the influence of the residual ethanol and the CA disturb the identification  
 250 of the peaks relative to the C-N stretching vibration and, the free NH stretching vibration  
 251 [41-43]. The spectrum of MAeG shows three peaks between 1600-1200 cm<sup>-1</sup> ascribable  
 252 to the C=C bond and a very small peak at about 1700 cm<sup>-1</sup> relative to the C=O stretch of  
 253 carboxylic group, the band at about 3400 cm<sup>-1</sup> relative to OH groups of GA in this case  
 254 is poor visible [44, 45].  
 255

Assignment	Position (cm <sup>-1</sup> )	Remarks
------------	------------------------------	---------

Fe-O	500-600	Typical of iron-oxide NPs (M, MAw, MAwG, MAe, MAeG)
Si-OH, Si-O-Fe	850-950	A shoulder related to the stretch of Si-OH symmetric vibration or Si-O-Fe bond (MS, MSAw, MSAwG)
Si-O	960-970	A band due to the Si-O stretching (MAw, MAwG, MAe, MAeG)
Si-O-Si	1000-1150	A band belonging to the stretching of Si-O-Si groups (MAw, MAwG, MAe, MAeG)
C=C	1200-1600	A peak ascribable to the C=C bond (MAeG, MAeG)
C-O	1310	The asymmetric stretching of C-O from the COOH group of CA (M, MAw, MAwG, MAe, MAeG)
C-N	1330	The peak related to C-N stretching vibration (MAw, MAwG, MAe, MAeG)
COO-Fe, C=O	1400-1600	The large peak can be assigned both to COO-Fe bonding (in literature 1431 cm <sup>-1</sup> ) and to the shift of peak characteristic of symmetric vibration of C=O due to the CA interaction with magnetite (M, MAw, MAwG, MAe, MAeG)
NH	1620	A broad band at 1628 cm <sup>-1</sup> can be ascribed to the N-H stretching vibration (MAw, MAe)

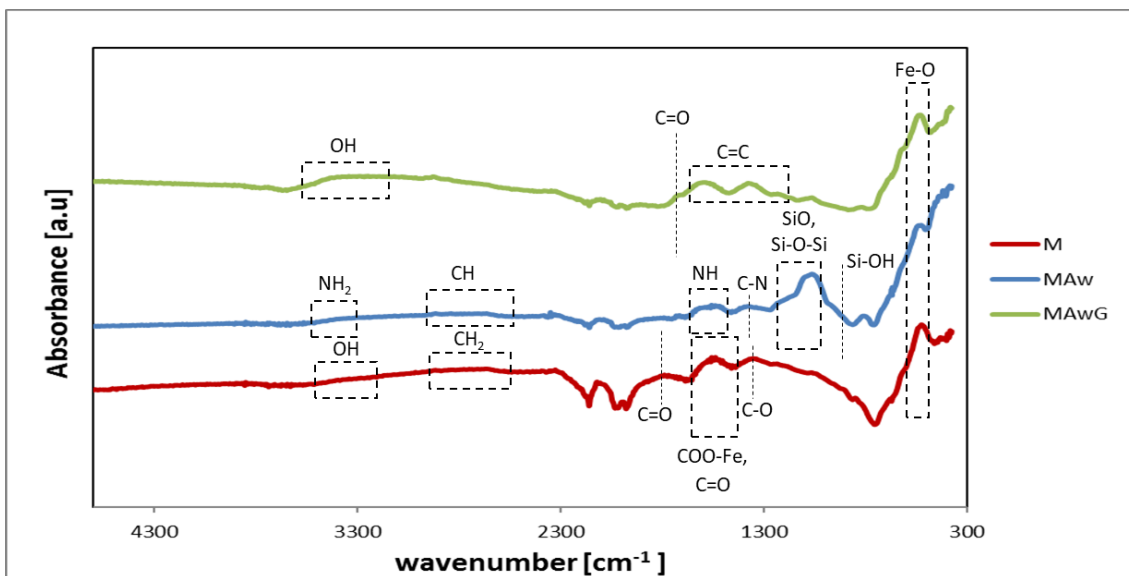
NH <sub>2</sub>	1640	The band attributable to NH <sub>2</sub> bending mode of free NH <sub>2</sub> groups (MAw, MAe)
C=O	1700	Stretching vibration from the free COOH group of CA (M, MAw, MAwG, MAe, MAeG)
C=O	1700	A small peak related to the C=O stretch of carboxylic group (MAwG, MAeG)
CH <sub>2</sub>	2600-2950	A band ascribable to CH <sub>2</sub> stretching (M, MAw, MAwG, MAe, MAeG)
C-H	2850-2920	The band ascribable to the stretching and vibration of C-H (MAw, MAeG, MAe, MAeG)
OH	3300-3400	A band related to OH groups (M, MAw, MAwG, MAe, MAeG)
N-H	3445	The bands attributable to the N-H stretching vibration (MAw, MAwG, MAe, MAeG)

256

257 **Table 2:** List of the main peaks detected in the FTIR spectra of M, MAw, MAwG,

258 MAe, MAeG, MS, MSAw and MSAwG NPs.

259

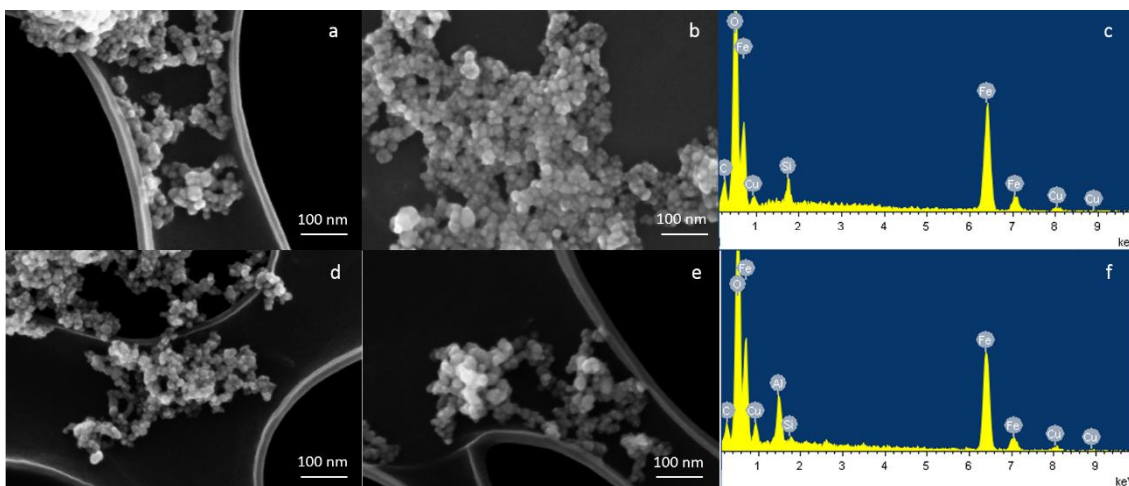


260

261 **Figure 3:** FTIR spectra of M, MAw and MAwG (NPs/GA 10:3) NPs.

262

263 The morphological and compositional analysis of MAeG (Figure 4a-c) shows the  
 264 presence of spherical particles with a diameter between 5-20 nm. The EDS analysis  
 265 (Figure 4c, MAeG with NPs/GA 10:1 as example) shows the Si peak ascribable to APTES  
 266 functionalization. Analogously, the morphological analysis of MAwG (Figure 4d-e)  
 267 reveals the presence of NPs with pseudo-spherical morphology and a diameter of about  
 268 5-20 nm. Also in this case the compositional analysis performed on MAwG with NPs/GA  
 269 10:1 (Figure 4f) evidences the peak of Si, due to the APTES functionalization.



270

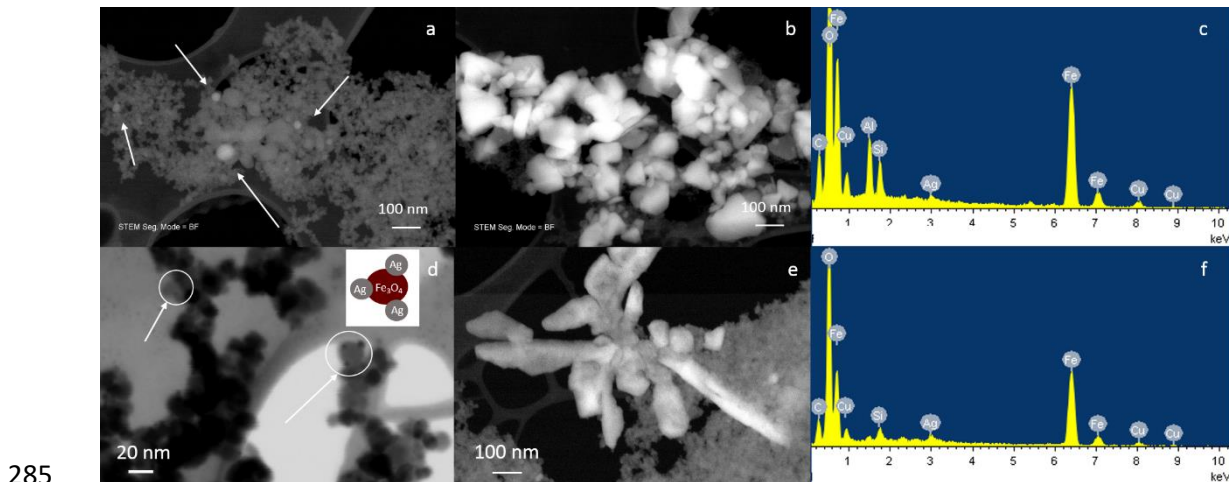
271 **Figure 4:** FESEM-EDS analysis of a) MAeG 10:1, b) MAeG 10:3, c) MAeG 10:1, d)  
272 MAwG 10:1, e) MAwG 10:3, f) MAwG 10:1.

273

274 The STEM-EDS analysis of MAeGAg is reported in figure 5; as it can be observed  
275 MAeGAg performed using NPs/GA ratio 10:1 show the presence of Ag nanometric  
276 particles (< 50 nm) agglomerated with magnetite nanoparticles, evidenced with arrows in  
277 figure 5a together with big silver particles (100-150 nm) with different shape (Figure 5b).

278 The EDS of area (Figure 5c) shows the peak of Ag, together with elements characteristic  
279 of MAeG (Fe, Si and O) and elements acribable to sample holder (Cu, Al, C).

280 MAeGAg performed using NPs/GA ratio 10:3 show the presence of very small Ag NPs  
281 (< 10 nm), which forms a nanoflower structure with magnetic NPs (Figure 5d, evidenced  
282 by arrows and schematized in the inset). However, also in this case, in some area of the  
283 sample big silver crystals were observed (Figure 5f and e) as confirmed by the EDS  
284 analysis (Figure 5f).

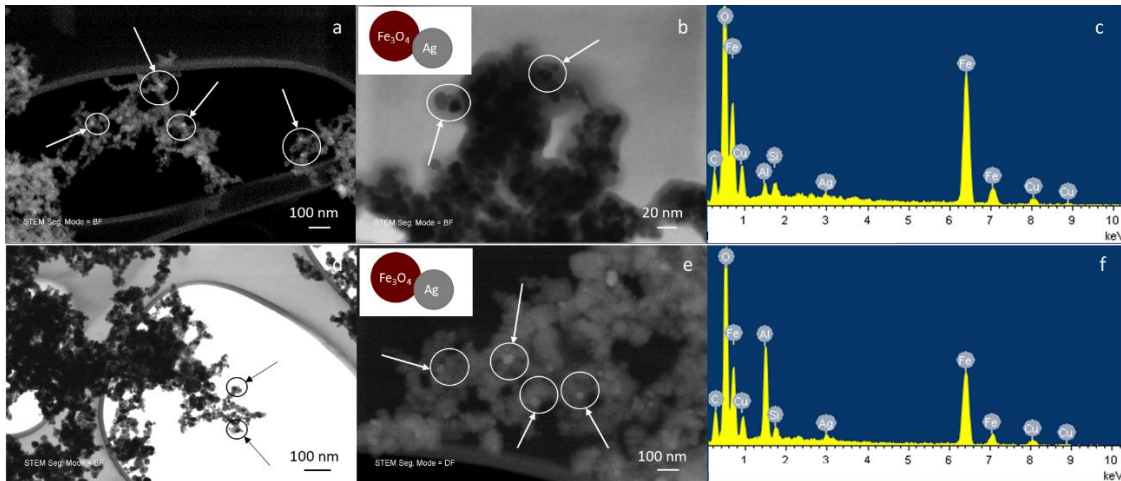


285

286 **Figure 5:** FESEM-EDS analysis of MAeGAg 10:1 (a-c) and MAeGAg 10:3 (d-f).

287

288 Figure 6 shows the STEM-EDS analysis of MAwGAg; in this case only Ag nanoparticles  
 289 with dimension comparable with magnetite NPs were observed both for NPs/GA ratio  
 290 10:1 (Figure 6a and b) and 10:3 (Figure 6d and e). Thus, in both NPs/GA ratio conditions,  
 291 the synthesis in water allowed to obtain nanodumbbell structures with magnetite NPs  
 292 connected with Ag NPs (Figure 6b and 6e). EDS analysis (Figure 6c and f) shows the Ag  
 293 peak in addition to magnetic NPs peaks. As far as concerned the stability of composite  
 294 NPs, their visual inspection suggests that they form a fairly stable suspension. Future test  
 295 will be carried out to experimentally evaluate the stability of the nanostructures over time.  
 296

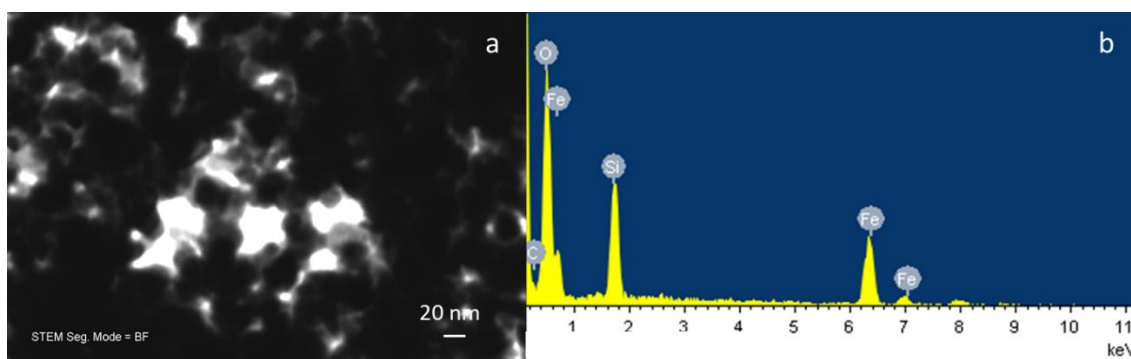


297  
 298 **Figure 6:** STEM-EDS analysis of MAwGAg 10:1 (a-c) and MAwGAg 10:3 (d-f).  
 299

### 300 **3.2 Route B**

301 The peaks obtained by means of FTIR analysis of MS, MSAw and MSAwG nanoparticles  
 302 are reported in table 2. MS NPs evidence both the peak ascribable to Fe-O bond at about  
 303  $500-600\text{ cm}^{-1}$  and the peak characteristic of silica shell: at  $780\text{ cm}^{-1}$  the stretch of Si-OH,  
 304 the broad band at about  $850-990\text{ cm}^{-1}$  can be attributed to the Si-O symmetric vibration  
 305 or to the Si-O-Fe bond and the peak at  $1060\text{ cm}^{-1}$  the stretch of Si-O-Si [46, 47]. The

306 spectrum of MSAw shows an increase of the intensity of the peak related to Si-O-Si bond  
307 attributable to the APTES functionalization, together with the peak of Fe-O bond. The  
308 spectrum of MSAwG shows the peaks between 1600-1200  $\text{cm}^{-1}$  ascribable to the C=C  
309 bond, the peak at about 1700  $\text{cm}^{-1}$  relative to the C=O stretch of carboxylic group, the  
310 band at about 3400  $\text{cm}^{-1}$  relative to OH groups of GA [44, 45].  
311 The morphological analysis of MSAwG (Figure 7a) evidences the presence of pseudo-  
312 spherical nanoparticles in the range 10-20 nm. The EDS spectrum (Figure 7b) shows a  
313 very intense peak of Si ascribable to the silica shell, together with Fe and O peak; C peak  
314 is due to the grid.



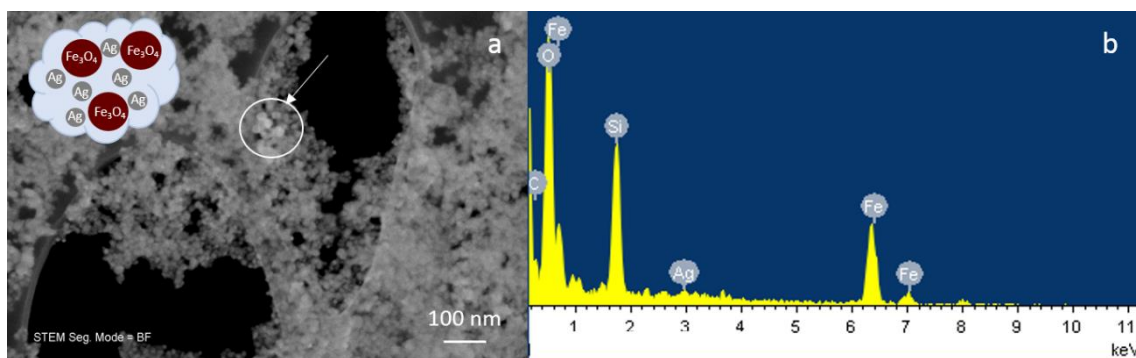
315

**Figure 7:** STEM image and EDS analysis of MSAwG.

316

317

318 The STEM analysis of MSAwGAg (Figure 8a) shows the formation of Ag NPs of  $\leq 20$   
319 nm not well dispersed among magnetic NPs, but aggregated in some area (schematic  
320 representation in Figure 8a); compositional analysis of an area (Figure 8b) shows the  
321 effective presence of the Ag peak.



322

323 **Figure 8:** STEM-EDS analysis of MSAwGAg obtained using a NPs/GA ratio of 10:1.

324

#### 325 4. Discussion

326 Aiming to improve the MNPs suspension stability, useful also for further  
 327 functionalization steps, the prevention of their aggregation is a crucial aspect. In fact, the  
 328 stability of MNPs in water is not optimal, due to anisotropic dipolar attraction [48] that  
 329 could limit the nucleation of the metal nanoparticles on their surface [49]. For this  
 330 purpose, CA was added to the MNPs suspension as stabilizing agent, promoting  
 331 electrostatic repulsion, as reported also by several authors [48, 50, 51].

332 Concerning the functionalization with APTES and GA, no significant differences were  
 333 noticed by FTIR analysis, although in the synthesis performed in ethanol it was difficult  
 334 to identify some characteristic groups due to probable traces of residual ethanol.

335 The morphological and compositional analysis of samples synthesized in water evidenced  
 336 the nucleation of smaller Ag nanoparticles by using a NPs/GA ratio of 10:3. We supposed  
 337 that the higher amount of GA used in this synthesis introduced more nucleation sites  
 338 allowing the formation of a lot of very small Ag NPs. In any case, even in this synthesis  
 339 some larger Ag crystals, with a multibranch morphology, were noticed. These crystals  
 340 could have been originated due to the seeding role of the nanoflowers-like structures, as

341 reported for ternary heterostructures by H. Zeng and S. Sun [52], further enhanced by  
342 the excess of reducing agent in the synthesis environment.

343 The syntheses performed in water revealed the formation of small Ag nanoparticles both  
344 using a NPs/GA ratio of 110:1 and 10:3. This result could be explained considering that,  
345 as reported by [52], the partial exposure of Fe(II) on the nanoparticles to aqueous phase  
346 could act as self-catalytic center for the reduction of  $\text{Ag}^+$  ions to  $\text{Ag}^\circ$  nanoparticles,  
347 leading to the heterodimer morphology.

348 In summary, the synthesis in water allows to obtain more homogenous heterodimer  
349 nanostructures, while the ethanol-based synthesis seems to have some critical aspects in  
350 the control of the dimension and structure of silver NPs. Moreover, in the synthesis in  
351 water, the different ratio between NPs and GA amount seems to not significantly  
352 influence the amount and dimension of Ag NPs. For this reason, the route B was performed  
353 only in water using a NPs/GA ratio of 10:1.

354 The NPs coating with a shell of silica seems to not improve the NPs functionalization and  
355 most of all the *in situ* reduction of Ag NPs; in this case some small aggregates of Ag NPs,  
356 not uniformly distributed were observed. As reported in other studies [47, 53], the  
357 structure of nanocomposites is significantly influenced by charges, reactive groups or  
358 functional moieties on the nanoparticles surface. The role of silica layer in the  
359 precipitation of the above mentioned aggregates can be explained considering that bi-  
360 component hybrid nanoparticles are generally obtained by sequential nucleation and  
361 growth of the second component on a preformed seed of the first one [52]. The successful  
362 synthesis of heterodimers strongly depends on the ability of suppressing the formation of  
363 dispersed NPs of the second component (homogeneous nucleation) and promote their  
364 seed-mediated heterogeneous nucleation on the first component NPs. This could be

365 achieved if the lattice spacing between the two components is well-matched to ensure  
366 epitaxial growth of the second component on the first one, which often participate to the  
367 reaction as catalyst, lowering the activating energy for heterogeneous nucleation. The  
368 obtained results demonstrated that the silica shell seems to not positively influence the  
369 decoration of MS NPs, probably due to its shield effect on the heterogeneous nucleation  
370 of Ag NPs, while promoting their homogeneous nucleation in the synthesis medium and  
371 their aggregated with the MS NPs, due to electrostatic interactions. Thus, the silver *in situ*  
372 reduction by GA and the heterogeneous nucleation are promoted when magnetic NPs are  
373 not coated by the silica shell; these two mechanisms are synergistic and lead to a better  
374 decoration of magnetite NPs. In particular, if magnetic NPs are not coated by silica, they  
375 are able to expose Fe(II) supporting the heterogeneous nucleation of Ag NPs, in addition  
376 to the *in situ* reduction due to GA.

377

## 378 **5. Conclusions**

379 A novel method to prepare composite nanoparticles Fe<sub>3</sub>O<sub>4</sub>—Ag, by *in situ* reduction of  
380 Ag on a magnetic core of Fe<sub>3</sub>O<sub>4</sub> was developed. The synthesis allowed at first to prepare  
381 Fe<sub>3</sub>O<sub>4</sub> NPs stabilized by CA with spherical shape and size in the range 5 – 20 nm. The  
382 obtained CA capped Fe<sub>3</sub>O<sub>4</sub> NPs were successfully functionalized with APTES to graft  
383 GA, as reducing agent for Ag NPs, in different media and with different NP/GA ratios.  
384 Depending on the investigated synthesis conditions, different morphologies were  
385 obtained, from polydispersed aggregates to nanoflower-like and nanodumbbell  
386 nanocomposites. The best results were obtained in aqueous media, with NPs/GA ratio of  
387 10:1, producing Fe<sub>3</sub>O<sub>4</sub>—Ag well defined nanodumbbells of about 20 nm size.

388 A further sol-gel coating of the CA-capped Fe<sub>3</sub>O<sub>4</sub> NPs with a silica shell, functionalized  
389 with APTES and GA in aqueous medium, was performed to assess its influence on the  
390 nucleation of Ag NPs. In the presence of the silica coating, Ag NPs with size < 20 nm  
391 nucleated as polydispersed aggregated among Fe<sub>3</sub>O<sub>4</sub> NPs, evidencing that the silica shell  
392 is not a key issue to optimize the morphology of Ag NPs nucleated on the magnetic core.  
393 Further optimizations of synthesis parameters (such as time and reagent ratio) are still  
394 necessary to improve the size and distribution of Ag NPs on Fe<sub>3</sub>O<sub>4</sub> NPs. The  
395 electromagnetic and optical characterizations are still in progress, as well as the  
396 evaluation of suspensions stability.

397

#### 398 **Summary Points**

- 399 1. Development of a novel method to prepare composite nanoparticles Fe<sub>3</sub>O<sub>4</sub>—Ag,  
400 by in situ reduction of Ag on a magnetic core
- 401 2. Use of gallic acid, a green anti-inflammatory and anti-tumor phenolic compound,  
402 as *in situ* reducing agent for Ag NPs
- 403 3. Evaluation of different synthesis media and functionalization steps
- 404 4. Estimation of different gallic acid/NPs ratios
- 405 5. Successfully functionalization of magnetic NPs with APTES and grafting with  
406 GA, both in water and ethanol
- 407 6. Polydispersed aggregates, nanoflower-like and nanodumbbell nanocomposites  
408 were obtained according to the synthesis conditions
- 409 7. Well-defined Fe<sub>3</sub>O<sub>4</sub>—Ag nanodumbbells (about 20 nm) were obtained using  
410 aqueous media, a NPs/GA ratio of 10:1

411 8. The presence of a silica coating was not found to be useful to optimize the  
412 morphology of Ag NPs nucleated on the magnetic core

413

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417

#### 418 **References**

419 [1] Rahman MM, *Nanomaterials*, 356. ISBN: 978-953-307-913-4, (2011). DOI:

420 10.5772/1371

421 [2] Wu W, Wu Z, Yu T, Jiang C, Kim WS. Recent progress on magnetic iron oxide

422 nanoparticles: synthesis, surface functional strategies and biomedical applications. *Sci.*

423 *Technol. Adv. Mater.* 16, 023501(2015). DOI: 10.1088/1468-6996/16/2/023501.

424 [3] Tartaj P, Morales MP, Veintemillas-Verdaguer S, Gonzalez-Carreno T, Serna CJ.

425 The preparation of magnetic nanoparticles for applications in biomedicine *J. Phys. D.*

426 *Appl. Phys.* 36, 182-197 (2003). DOI: 10.1088/0022-3727/36/13/202

427 [4] Veiseh O, Gunn J, Zhang M, Design and fabrication of magnetic nanoparticles for

428 targeted drug delivery and imaging. *Adv. Drug. Deliv. Rev.* 62(3), 284–304 (2010). doi:

429 10.1016/j.addr.2009.11.002

430 [5] Li Z, Kawashita M, Araki N, Mitsumori M, Hiraoka M, Do M. Magnetite

431 nanoparticles with high heating efficiencies for application in the hyperthermia of

432 cancer. *Mater. Sci. Eng. C.* 30, 990–996 (2010). Doi:10.1016/j.msec.2010.04.016

433 [6] Prucek R, Kilianová TJ, Panáček M et al. The targeted antibacterial and antifungal  
434 properties of magnetic nanocomposite of iron oxide and silver nanoparticles.  
435 *Biomaterials* 32, 4704-4713 (2011). doi: 10.1016/j.biomaterials.2011.03.039

436 [7] Kluchova K, Zboril R, Tucek J, et al.  
437 Superparamagnetic maghemite nanoparticles from solid-state synthesis - their  
438 functionalization towards per oral MRI contrast agent and magnetic carrier for trypsin  
439 immobilization. *Biomaterials* 30, 2855 (2009). doi: 10.1016/j.biomaterials.2009.02.023

440 [8] Corr SA, Rakovich YP, Gun'ko YK. Multifunctional magnetic-  
441 fluorescent nanocomposites for biomedical applications. *Nanoscale Res. Lett.* 3, 87  
442 (2008). doi: 10.1007/s11671-008-9122-8

443 [9] Srivastava P, Sharma PK, Muheem A, Warsi MH. Magnetic Nanoparticles: A  
444 Review on Stratagems of Fabrication and its Biomedical Applications. *Recent Patents*  
445 *on Drug Delivery & Formulation*, 11, 101-113 (2017).  
446 doi.:10.2174/1872211311666170328150747

447 [10] Miola M, Ferraris S, Pirani F. et al. Reductant-free synthesis of magnetoplasmonic  
448 iron oxide-gold nanoparticles *Ceramics International* 43, 15258–15265 (2017). DOI:  
449 10.1016/j.ceramint.2017.08.063

450 [11] Multari C, Miola M, Laviano F, et al. Magnetoplasmonic nanoparticles for  
451 photothermal therapy. *Nanotechnology* 30, 255705 (9pp) (2019). doi:10.1088/1361-  
452 6528/ab08f7

453 [12] Mohan YM, Lee K, Premkumar T, Geckeler KE. Hydrogel networks as nano-  
454 reactors: a novel approach to silver nanoparticles for antibacterial applications. *Polymer*  
455 48, 158 (2007). Doi:10.1016/j.polymer.2006.10.045

- 456 [13] Wang AQ, Liu JH, Lin SD, Lin TS, Mou CY. A novel efficient Au-Ag alloy  
457 catalyst system: preparation, activity, and characterization. *J Catal* 233, 186. (2005)  
458 doi.org/10.1016/j.jcat.2005.04.028
- 459 [14] Lee D, Cohen RE, Rubner MF. Antibacterial properties of Ag nanoparticle loaded  
460 multilayers and formation of magnetically directed antibacterial microparticles.  
461 *Langmuir* 21, 9651 (2005). Doi:10.1021/la0513306
- 462 [15] Zhang XF, Liu ZG, Shen W, Gurunathan S. Silver Nanoparticles: Synthesis,  
463 Characterization, Properties, Applications, and Therapeutic Approaches. *Int. J. Mol. Sci.*  
464 17(9) 1534 (2016). doi: 10.3390/ijms17091534
- 465 [16] Markova Z, Šišková KM, J. Filip et al. Air Stable Magnetic Bimetallic Fe–Ag  
466 Nanoparticles for Advanced Antimicrobial Treatment and Phosphorus Removal.  
467 *Environ. Sci. Technol.* 47, 5285–5293 (2013). Doi:10.1021/es304693g
- 468 [17] Lee HJ, Yeo SY, Jeong SH. Antibacterial effect of nanosized silver colloidal  
469 solution on textile fabrics. *J. Mater. Sci.* 38, 2199 (2003).  
470 Doi:10.1023/A:1023736416361
- 471 [18] Pal S, Tak YK, Song JM. Does the antimicrobial activity of silver nanoparticles  
472 depend on the shape of the nanoparticle? a study of the gram-negative  
473 bacterium *Escherichia coli*. *Appl. Environ. Microbiol.* 73, 1712. (2007) DOI:  
474 10.1128/AEM.02218-06
- 475 [19] Wu YX, Liang P, Dong Q, et al. Design of a silver nanoparticle for sensitive  
476 surface enhanced Raman spectroscopy detection of carmine dye. *Food Chemistry* 237,  
477 974-980 (2017). Doi: 10.1016/j.foodchem.2017.06.057

- 478 [20] Zhang H, Chen G, Yu B, Cong H. Emerging advanced nanomaterials for Cancer  
479 photothermal therapy. *Rev. Adv. Mater. Sci.* 53, 131-146 (2018). DOI: 10.1515/rams-  
480 2018-0010
- 481 [21] Ching G, Jiang J, Ying JY, Ji W. Fe<sub>3</sub>O<sub>4</sub>-Ag nanocomposites for optical limiting:  
482 broad temporal response and low threshold. *Opt Express* 18, 6183 (2010). DOI:  
483 10.1364/OE.18.006183
- 484 [22] Laurent S, Forge D, Port M, et al. Magnetic Iron Oxide Nanoparticles: Synthesis,  
485 Stabilization, Vectorization, Physicochemical Characterizations, and Biological  
486 Applications. *Chem. Rev.*, 108(6), 2064–2110 (2008). DOI: 10.1021/cr068445e
- 487 [23] Kharisov BI, Rasika Dias HV, Kharissova OV, Vázquez A, Peña Y Gómez I.  
488 Solubilization, dispersion and stabilization of magnetic nanoparticles in water and non-  
489 aqueous solvents: recent trends. *RSC Adv* ,4, 45354-45381 (2014). DOI:  
490 10.1039/C4RA06902A
- 491 [24] Stafford S, Serrano Garcia R, Gun'ko YK. Multimodal Magnetic-Plasmonic  
492 Nanoparticles for Biomedical Applications. *Appl. Sci.* 8, 97 (2018).  
493 doi:10.3390/app8010097
- 494 [25] Abbas M, Ramulu Torati S, Kim C. A novel approach for the synthesis of ultrathin  
495 silica-coated iron oxide nanocubes decorated with silver nanodots (Fe<sub>3</sub>O<sub>4</sub>/SiO<sub>2</sub>/Ag) and  
496 their superior catalytic reduction of 4-nitroaniline. *Nanoscale*, 7, 12192-12204 (2015).  
497 DOI: 10.1039/C5NR02680F
- 498 [26] Lee B, Koo S. Silver reduction on the surface of magnetite nanoparticles using a  
499 coupling agent. *J. Ind. and Eng. Chem.* 18(3), 1191–1195(2012). Doi:  
500 10.1016/j.jiec.2012.01.009

501 [27] Ballesteros CAS, Cancino J, Marangoni VS, Zucolotto V. Nanostructured Fe<sub>3</sub>O<sub>4</sub>  
502 satellite gold nanoparticles to improve biomolecular detection. *Sensors and Actuators B*  
503 198, 377–383 (2014). doi.: 10.1016/j.snb.2014.03.079

504 [28] Fantechi E, Castillo PM, Conca E, Cugia F, Sangregorio C, Casula MF, Assessing  
505 the hyperthermic properties of magnetic heterostructures: The case of gold–iron oxide  
506 composites. *Interface Focus* 6, 20160058 (2016). doi: 10.1098/rsfs.2016.0058

507 [29] Ivashchenko O, Gapinski J, Peplińska B. et al. Self-organizing silver and ultrasmall  
508 iron oxide nanoparticles prepared with ginger rhizome extract: Characterization,  
509 biomedical potential and microstructure analysis of hydrocolloids. *Mater. Design* 133,  
510 307–324 (2017). doi.: 10.1016/j.matdes.2017.08.001

511 [30] Amala Jayanthi S, Manovah David T, Jayashainy J, Muthu Gnana D, Nathan T,  
512 Sagayaraj P. A convenient two-step bottom-up approach for developing Au/Fe<sub>3</sub>O<sub>4</sub>  
513 nanocomposites with useful optical and magnetic properties. *J. Alloys and Compounds*  
514 606, 254–226 (2014). DOI: 10.1016/j.jallcom.2014.03.186

515 [31] Ferraris S, Miola M, Cochis A. et al. In situ reduction of antibacterial silver ions to  
516 metallic silver nanoparticles on bioactive glasses functionalized with Polyphenols. *Appl.*  
517 *Surf.Sci.* 396, 461–470 (2017). doi.: 10.1016/j.apsusc.2016.10.177

518 [32] Borroni E, Miola M, Ferraris S. et al. Tumor targeting by lentiviral vectors  
519 combined with magnetic nanoparticles in mice. *Acta Biomaterialia* 59, 303–316 (2017).  
520 doi: 10.1016/j.actbio.2017.07.007.

521 [33] Multari C, Miola M, Ferraris S. et al. Synthesis and characterization of silica-coated  
522 superparamagnetic iron oxide nanoparticles and interaction with pancreatic cancer cells.  
523 *Int J Appl Ceram Technol.* 15, 947–960 (2018). Doi: 10.1111/ijac.12897

524 [34] Muzio G, Miola M, Ferraris S, et al. A. Follenzi. Innovative superparamagnetic  
525 iron-oxide nanoparticles coated with silica and conjugated with linoleic acid: Effect on  
526 tumor cell growth and viability. *Mat. Sci. Eng. C* 76, 439–447 (2017). doi:  
527 10.1016/j.msec.2017.03.063.

528 [35] Stöber W, Fink A, Controlled growth of monodisperse silica spheres in the micron  
529 size range, *J. Colloid Interface Sci.* 26, 62–69 (1968).

530 [36] Miola M, Multari C, Debellis D, Laviano F, Gerbaldo R, Vernè E,  
531 Magneto-plasmonic heterodimers: Evaluation of different synthesis approaches. *J Am*  
532 *Ceram Soc.* 1–10 (2021).

533 [37] Panagiota S, Louloudi M, Deligiannakis Y, EPR study of phenolic radical  
534 stabilization by grafting on SiO<sub>2</sub>. *Chemical Physics Letters* 472, 85-89 (2009) . Doi:  
535 10.1016/j.cplett.2009.02.080

536 [38] Cheraghipour E, Javadpour S, Mehdizadeh, AR, Citrate capped superparamagnetic  
537 iron oxide nanoparticles used for hyperthermia therapy *J. Biomedical Science and*  
538 *Engineering*, 5, 715-719 (2012). doi: 10.4236/jbise.2012.512089.

539 [39] Nigam S, Barick KC, Bahadur D. Development of citrate-stabilized Fe<sub>3</sub>O<sub>4</sub>  
540 nanoparticles: Conjugation and release of doxorubicin for therapeutic applications a, *J.*  
541 *Magn. Magn. Mat.* 323, 237–243 (2011). Doi: 10.1016/j.jmmm.2010.09.009

542 [40] Kulshrestha P, Gogoi M, Bahadur D, Banerjee R. In vitro application of paclitaxel  
543 loaded magnetoliposomes for combined chemotherapy and hyperthermia. *Colloids Surf*  
544 *B Biointerfaces* 96, 1-7( 2012). doi: 10.1016/j.colsurfb.2012.02.029.

545 [41] Bini RA, Marques RFC, Santos FJ, Chaker JA, Jafelicci Jr M,. Synthesis and  
546 functionalization of magnetite nanoparticles with different amino-functional

547 alkoxy silanes. *J. Magn. Magn. Mat.* 324, 534–539 (2012). Doi:  
548 10.1016/j.jmmm.2011.08.035

549 [42] Khatiri R, Reyhani A, Mortazavi SZ, Hossainipour M. Preparation and  
550 characterization of Fe<sub>3</sub>O<sub>4</sub> /SiO<sub>2</sub> / APTES core-shell nanoparticles. Proceedings of the  
551 4th *International Conference on Nanostructures*. (2012).

552 [43] Kanimozhi S, Perinbam K. Synthesis of amino-silane modified superparamagnetic  
553 Fe<sub>3</sub>O<sub>4</sub> nanoparticles and its application in immobilization of lipase from *Pseudomonas*  
554 *fluorescens* Lp1. *Materials Research Bulletin*, 48, 1830-1836 (2013). Doi:  
555 10.1016/j.materresbull.2013.01.024Get rights and content

556 [44] Wang W, Chen Q, Jiang C, Yang D, Liu X, Xu S, One-step synthesis of  
557 biocompatible gold nanoparticles using gallic acid in the presence of poly-(N-vinyl-2-  
558 pyrrolidone), *Colloids and Surfaces A: Physicochem. Eng. Aspects* 301, 73–79 (2007).  
559 doi:10.1016/j.colsurfa.2006.12.037

560 [45] Deligiannakis Y, Sotiriou GA, Pratsinis SE. Antioxidant and Antiradical SiO<sub>2</sub>  
561 Nanoparticles Covalently Functionalized with Gallic Acid. *ACS Appl. Mater. Interfaces*  
562 4, 6609–6617 (2012). doi: 10.1021/am301751s.

563 [46] Nadeema K, Ali L, Gul I, Rizwand S, Mumtaz M, Effect of silica coating on the  
564 structural, dielectric, and magnetic properties of maghemite nanoparticles. *J. Non-*  
565 *Crystal. Sol.* 404, 72–77 (2014). Doi: 10.1016/j.jnoncrysol.2014.07.036

566 [47] Emadi M, Shams E, Kazem Amini M, Removal of Zinc from Aqueous Solutions  
567 by Magnetite Silica Core-Shell Nanoparticles. *Journal of Chemistry* 2012, ID 787682  
568 (2013). Doi: 10.1155/2013/787682

569 [48] Cheraghipour E, Javadpour S, Mehdizadeh AR, Citrate capped superparamagnetic  
570 iron oxide nanoparticles used for hyperthermia therapy, *J. Biomed. Sci. Eng.* 5, 715–719  
571 (2012) . DOI: 10.4236/jbise.2012.512089.

572 [49] Cozzoli PD, Pellegrino T, Manna L, Synthesis, properties and perspectives of  
573 hybrid nanocrystal structures, *Chem. Soc. Rev.* 35, 1195–1208 (2006).

574 Doi:10.1039/B517790C

575 [50] Goetze TC, GansauBuske N, Roeder M, Gornert P, Bahr M, Biocompatible  
576 magnetic core/shell nanoparticles, *J. Magn. Magn. Mater.* 252, 399–402 (2002).

577 Doi:10.1016/S0304-8853(02)00624-8

578 [51] Campelj S, Makovec D, Drogenik M, Preparation and properties of water-based  
579 magnetic fluids, *J. Phys. Condens. Matter* 20, 204101 (2008). doi: 10.1088/0953-

580 8984/20/20/204101 \* One of the most significant examples of the use of citric acid as  
581 stabilizing and dispersant agent.

582 [52] Zeng H, Sun S. Syntheses, Properties, and Potential Applications of  
583 Multicomponent Magnetic Nanoparticles. *Adv. Func. Mat.* 18, 391-400 (2008). Doi:

584 10.1002/adfm.200701211 \* Complete and detailed review concerning the development  
585 of multicomponent nanoparticles.

586 [53] Cham-Fai Leung K, Xuan S, Zhu X, et al. Gold and iron oxide hybrid  
587 nanocomposite materials. *Chem. Soc. Rev.* 41, 1911–1928(2013). Doi:

588 10.1039/C1CS15213K

589

590 **Table of Contents graphic**

**Gallic acid functionalized  
magnetic NPs**

