

# Mitigating of Arsenic Accumulation in Rice (Oryza sativa L.) from Typical Arsenic Contaminated Paddy Soil of Southern China Using Nanostructured $\alpha$ -MnO2: Pot Experiment and Field Application

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1	Mitigating of Arsenic Accumulation in Rice (Oryza sativa L.)
2	from Typical Arsenic Contaminated Paddy Soil of Southern
3	China Using Nanostructured α-MnO <sub>2</sub> : Pot Experiment and
4	Field Application
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# 20 Abstract

Manganese oxides are naturally occurring powerful oxidants and scavenger which can control the mobility and bioavailability of arsenic (As). However, the effect of synthetic nanostructured manganese oxides on the mobilization and transportation of As at actual paddy soils are poorly understood, especially in the low or medium background Mn concentration soil. In the present study, a novel Nano manganese

26	oxide with higher reactivity and surface area has been synthesized. A 90-d soil
27	incubation experiment combined with pot and field rice cultivation trials were
28	designed to evaluate the effectiveness of exogenous $\alpha$ -MnO <sub>2</sub> nanorods on the
29	mobilization and transportation of As in soil-rice systems. Our results proved that the
30	addition of $\alpha$ -MnO <sub>2</sub> nanorods can effectively control the soil-to-solution partitioning
31	of As under anaerobic conditions. After treatment with different amounts of $\alpha$ -MnO <sub>2</sub>
32	nanorods, the content of effective As decreased with the increasing of residual As and
33	insoluble binding As (Ca-As and Fe-As). Besides, the enhanced oxidation of As (III)
34	into As(V) by $\alpha$ -MnO <sub>2</sub> nanorods increased the adsorption of As onto indigenous
35	iron(hydr) oxides which greatly reduced the soil porewater As content. Additionally,
36	pot experiment and filed applications are further proved that the influx of As into
37	aerial parts of rice plants (stems, husk and leaves) was strictly prohibited after
38	treatments with different amount of $\alpha$ -MnO <sub>2</sub> nanorods; more interestingly,
39	significantly negative correlations have been observed between As and Mn in rice,
40	which indicated that as Mn is increased in soil, As in brown rice decreases. Our
41	results demonstrated that the use of $\alpha$ -MnO <sub>2</sub> nanorods in As polluted paddy soil
42	containing low levels of background Mn oxides can be a promising remediation
43	strategy.

44

# Keywords: Arsenic, Nanostructured-MnO<sub>2</sub>, Rice, Accumulation, Paddy soil

45 **1. Introduction** 

46 Hunan province is world-renowned for its luxuriant deposit of non-ferrous metal

ores (tungsten, bismuth, realgar) (Lei et al., 2015; Okkenhaug et al., 2012; Williams et 47 al., 2009). In the past several decades, intensive mineral exploitation, ore extraction 48 49 and refining activities have caused a large amount of toxic trace elements (Cd, Hg, Pb and As) to be discharged into farmland which has greatly affected the local soil and 50 water environment(Li et al., 2017; Zhao et al., 2015). Among them, arsenic is a 51 ubiquitous and highly toxic metalloid element that has caused severe contamination in 52 Hunan province(Lei et al., 2013; Lei et al., 2015). It is reported that the 53 arsenic-contaminated farmland in Hunan province has already seriously impaired the 54 development of agriculture and posed a serious threat to the health of local residents, 55 because rice is a dominant staple food (Li et al., 2016; Liao et al., 2005). 56

The existence forms (speciation) of arsenic may be more important than the total 57 58 arsenic in the soil, which determine its effectiveness and toxicity to organisms. It is generally established that trivalent As species are more toxic than their pentavalent 59 counterparts because they binds to sulfhydryl groups(-SH), impairing the function of 60 many proteins (Fu et al., 2016; Liao et al., 2005; Liu, 2005). In paddy soils, arsenic is 61 predominantly present as the inorganic species arsenate and arsenite (Takahashi et al., 62 2004). The extent of As mobility and bioavailability in paddy soil is, in part, regulated 63 by the type of minerals exist in the soil system and the oxidation state of As (Fendorf 64 and Kocar, 2009; Ying et al., 2012). Generally, As(III) are far more mobile than 65 As(V); and the relative content of arsenate and arsenite in paddy soil are primarily 66 depending on the redox status of soil (Yamaguchi et al., 2011). Arsenate often exists 67 in anionic forms (e.g.,  $H_2AsO_4^{-}$ ,  $HAsO_4^{2-}$ ) under aerobic conditions with the content 68

69	can account for 65-98% of total arsenic (Ohtsuka et al., 2013). On the contrary,
70	arsenite takes an electrically uncharged molecule form(H <sub>3</sub> AsO <sub>3</sub> ) under anaerobic
71	reducing conditions (Eh<100mV; pH<9) (Han et al., 2011). The amount of Fe oxides
72	in the soil plays an important role in controlling the concentration of As species in the
73	soil solution; typically, As(V) is strongly adsorbed with metal-(oxyhydr) oxides,
74	whereas As(III) is poorly associated with soil minerals owing to its feature of
75	charge-neutral (Chen et al., 2006; Ehlert et al., 2014); and thus rendering it
76	comparatively effective towards to plants uptake than the As(V) (Xu et al., 2017).
77	However, it is well recognized that during the drastically aerobic-anaerobic transition
78	within paddy fields, the absorbed As will released into soil porewater (Ohtsuka et al.,
79	2013; Xu et al., 2017). The mobilization of As in flooded paddy fields is because of
80	two main processes. Firstly, the reductive dissolution of iron(oxyhydr) oxides have
81	been triggered by soil flooding which caused the sorbed solid phase arsenic releasing
82	into the liquid phase (Lemonte et al., 2017; Weber et al., 2010; Yamaguchi et al.,
83	2011). Secondly, the adsorbed As(V) is reduced to As(III) under the reductive
84	conditions and the latter has a greater tendency to partitioning into the liquid phase
85	than As(V) (Liu et al., 2015; Takahashi et al., 2004). Compared with other terrestrial
86	plant, rice (Oryza sativa L.) is efficient in As uptake and translocation, because of the
87	flooded conditions and highly expressed arsenic transporter (Si transporter,
88	aquaporins and phosphate transporters) (Ma et al., 2008; Meharg, 2004; Meharg and
89	Jardine, 2003). Thus, effective measures must be taken to reduce the bio-availability
90	and mobility of As(III) in paddy soil during rice cultivation.

91	In current literature, several measures have been proposed for reducing the
92	bio-availability of As in soils, such as amendments stabilization (biochar, natural
93	minerals, etc.) (Kumpiene et al., 2008; Li et al., 2018), electro-kinetics
94	(Balasubramanian et al., 2009), acid flushing (Beiyuan et al., 2017; Tokunaga and
95	Hakuta, 2002), phytoremediation (Gilloaiza et al., 2016; Jankong et al., 2007) and
96	agronomic mitigation strategies (Limmer et al., 2018; Seyfferth et al., 2018). However,
97	those methods are hard to meet the actual demand of paddy fields remediation. Due to
98	either their high-cost (Liu et al., 2018), vast energy requirements (Villen-Guzman et
99	al., 2017), or long treatment times (Wan et al., 2016); above all, high cost (or
100	unsustainability) hinder the application of many technologies in polluted farmland
101	(Bontempi, 2017). Chemical stabilization methods, in particular, have been widely
102	accepted in the remediation of As-contaminated soils because they are relatively cost
103	effective (sustainability) and easy to operate and management. Recently, engineered
104	nanoparticles stabilizer such as zero valent iron (Gil-Díaz et al., 2017; Gil-Díaz et al.,
105	2016) and iron phosphate (vivianite) nanoparticles (Liu and Zhao, 2007) has been
106	proved to be an advanced environmental remediation technologies, which could
107	provide cost-effective solutions to some of the most intractable environmental restore
108	problems due to their large surface areas and high surface reactivity (Zhang, 2003).
109	For the variable valence elements (As), by consideration of regulatory measures (in
110	situ oxidation by chemical amendments) to induction the transformation of As(III) to
111	As(V) is considered to be a promising approach which can alleviate the associated
112	environmental risks of As in paddy soil (Lin et al., 2017; Suda and Makino, 2016; Xu

et al., 2017). However, to our knowledge, there are few related studies focused on the induction of As to be transformed into low-effective and low-toxicity forms using oxidants in paddy soil.

Manganese oxides are naturally occurring powerful oxidants that can effectively 116 117 catalyze the oxidation of As(III) to As(V) under natural circumstances (Bruce A. 118 Manning et al., 2002; Ehlert et al., 2014; Han et al., 2011; Lafferty et al., 2010). The As(III) oxidation by manganese oxides can occur across a wide pH ranged from 119 4.0~8.2, however, the oxidation rates are deeply associated with their structure, 120 surface charge properties, mineral crystallinity and abundance (Oscarson et al., 1983; 121 Scott and Morgan, 1995). The study conducted by (Bruce A. Manning et al., 2002) 122 showed that As(III) can be quickly oxidized into As(V) in the presence of MnO<sub>2</sub> with 123 124 only about 10% of As(III) was remined after 10 hours reaction. Besides, another profound and detailed researches conducted by Scott and Morgan (Scott and Morgan, 125 1995) who found that birnessite ( $\delta$ -MnO<sub>2</sub>) can quickly oxidize As(III) to As(V), about 126 80% of the reaction can be accomplished within 1h and this process was accompanied 127 by the release of  $Mn^{2+}$ . Apart from the oxidation ability towards to As(III) by 128 manganese oxides, it was also reported that As(III), after being oxidized by 129 Mn-oxides, can subsequently be adsorbed onto the surfaces of MnOOH (oxidation 130 intermediates) and ferric-(oxyhydr) oxide (Ehlert et al., 2014; Nesbitt et al., 1998); 131 thus the partitioning of As into solution was restrained. Although there are many 132 studies (BA et al., 2002; Ehlert et al., 2014) have been conducted on the oxidation of 133 As(III) by manganese oxides, however, most of these studies are concentrated on pure 134

135 minerals in aqueous solution.

To the best of our knowledges, there are very few related studies on the 136 oxidation of As(III) by synthetic nanostructured-MnO<sub>2</sub> in actual paddy soils; 137 especially in the low or medium background Mn concentration soil. In China, it has 138 been reported that the content of Mn in soil was varied between 10-5532 mg  $kg^{-1}$  with 139 an average amount of 710 mg·kg<sup>-1</sup>(Liu et al., 1983). The Mn concentrations which 140 below the average value of 710mg kg<sup>-1</sup> can be classified as low manganese soil. 141 Recently, a soil incubation experiment conducted by Xu et al., 2017) showed 142 that additions of synthetic Mn oxide (hausmannite) in low background Mn content 143 paddy soils can effectively control the partitioning of As from solid phase to liquid 144 phase due to the oxidation of As(III) However, they only have considered the 145 146 efficiency of micrometer scale Mn oxides under the laboratory conditions but did not test it under complex field trials. Therefore, we have proposed a hypothesis that the 147 endogenous iron oxides in paddy soil can be used to retain the oxidized As(V) after 148 incorporation with nanostructured manganese oxides under flooded conditions, 149 thereby reducing the bioavailability of As towards to rice. The major objectives of this 150 present study are therefore to (i) investigated the potential of synthetic  $\alpha$ -MnO<sub>2</sub> 151 nanorods on the control of solid to solution distribution of As under flooded 152 conditions; (ii) determined whether synthetic a-MnO<sub>2</sub> nanorods could reduce As 153 uptake into rice grow in paddy soil with low endogenous Mn concentration at fields 154 scales; and (iii) elucidated the associated mechanisms regarding to the reduced of As 155 uptake by rice. 156

## 157 2. Material and methods

#### 158 **2.1 Chemicals and reagents**

All chemicals, including manganese sulfate (MnSO<sub>4</sub>·H<sub>2</sub>O), potassium persulfate 159  $(K_2S_2O_8)$ , ammonium phosphate  $((NH_4)_3PO_4 \cdot 3H_2O)$ , urea  $(CO(NH_2)_2)$ , potassium 160 carbonate (K<sub>2</sub>CO<sub>3</sub>), hydrochloric acid (HCl), nitric acid (HNO<sub>3</sub>) and sulfuric acid 161 (H<sub>2</sub>SO<sub>4</sub>) used in this study were of analytical grade without any further purification. 162 The  $\alpha$ -MnO<sub>2</sub> nanorods was synthesized following the protocol previously outlined 163 by (Yu et al., 2013) with minor modification; Briefly, the  $\alpha$ -MnO<sub>2</sub> nanorods were 164 hydrothermally synthesized using a solution containing a certain amount of 165 MnSO<sub>4</sub>·H<sub>2</sub>O (0.3415 g, 2 mmol) and  $K_2S_2O_8$  (0.5434 g, 2 mmol), the detailed 166 167 procedures can be found in supplementary material. The obtained a-MnO<sub>2</sub> nano materials are a very stable black powdery solid. The powder X-ray diffraction (XRD) 168 patterns of the obtained materials were recorded on a Bruker D8 Advance XRD 169 170 diffractometer with Cu Ka radiation (Voltage: 40 kV; Current: 40 mA; Scanning rate: 10°/min). The morphologies of the samples were observed by emission scanning 171 electron microscopy (SEM, Quanta F250, FEI, USA) Ultrapure water (18.2 MΩ cm) 172 was used in all experiments, unless otherwise stated. All experimental containers were 173 174 soaked with 10% HNO<sub>3</sub> overnight and rinsed several times with deionized water before use. 175

#### 176 **2.2 Site characterization and soil sampling**

177 Chenzhou City lies between 24°53' and 26°50' latitudes and between 112°13' 178 and 114°14' longitudes(Lei et al., 2015). The research paddy field is located in 179 Dengjiatang (25°36'N, 113°00'E) village, Su Xian district, Chenzhou City. An 180 As-product factory was located at Dengjiatang in 1992, but it has been out of 181 production since 1999(Lei et al., 2015; Liao et al., 2004; Liao et al., 2005).

Bulk arsenic contaminated soil samples were collected from the plow layers (0-20cm). The soil samples were air-dried at room temperature, ground, and passed through a 5-mm nylon sieve. The elementary physicochemical properties were analyzed, and the results were shown in Table S1 in supplementary material.

#### 186 **2.3 Soil incubation and As sequential extraction experiment**

Soil incubation experiments were designed to explore the effect of  $\alpha$ -MnO<sub>2</sub> 187 nanorods on the variations of As fractionation in flooded paddy soil. To prepare the 188  $\alpha$ -MnO<sub>2</sub> treated samples, 15 kg sieved soil was weighted carefully and packed into a 189 190 polyethylene pot (50 cm  $\times$  22 cm). Subsequently, the soil was amended with  $\alpha$ -MnO<sub>2</sub> nanorods to maintain the rates of 0.2%, 0.5%, 1.0% and 2.0% of soil weight. The pot 191 was first pre-incubated for 24 h in the dark with soil moisture content being 192 maintained at 70% field water holding capacity. After that, the pots were incubated at 193 25°C in the growth chamber, with daily additions of ultra-pure water to maintain the 194 water level of 3cm above the soil surface. 195

196 After 90 days, the soil samples were collected from the surface (0-20 cm depth)

of the soil profile. After being air-dried at ambient temperature, obtained soil will first 197 ground to pass through 1mm screen, and then ground again using agate mortar and 198 199 passed through 0.15 mm screen prior to analysis. A sequential As fractionation schemes was employed to determine the operationally defined As fractionation(Van et 200 al., 2003; Wu et al., 2006). The detail operation procedure was described in 201 supplementary material. (Table S2). All the treatments and extractions procedures 202 were run in triplicates unless stated otherwise. The extraction efficiency of arsenic 203 fractionation was presented in Table S3. 204

205 2.4 Pot experiment designs

206 The pot experiment was carried out in a greenhouse of Hunan agricultural university. Firstly, 15.0 kg homogenized arsenic contaminated soil was packed in 207 208 each polyethylene pot with a height of 50 cm and a diameter of 22 cm. Ammonium phosphate ((NH<sub>4</sub>)<sub>3</sub>PO<sub>4</sub>·3H<sub>2</sub>O), urea (CO(NH<sub>2</sub>)<sub>2</sub>), potassium carbonate (K<sub>2</sub>CO<sub>3</sub>), were 209 added to each pot as basal fertilizers at dosage of 4.29, 2.93, and 3.30 g for N, P, and 210 K supply, respectively. Then the soil was amended with  $\alpha$ -MnO<sub>2</sub> nanorods at rates of 211 212 0.2%, 0.5%, 1.0% and 2.0% of soil weight. Each pot was then saturated with distilled water and drained down to an equilibrium state for 7 days under natural conditions. 213 All the treatments were triplicated and randomly arrangement and three blank controls 214 215 (without  $\alpha$ -MnO<sub>2</sub> nanorods addition) were provided.

The rice seeds (*O. sativa L* Yuzhenxiang, obtained from Hunan Rice Research
Institute) were disinfected in 30% H<sub>2</sub>O<sub>2</sub> solution for 10 minutes, followed by thorough

washing with deionized water and soaking in deionized water for 24 h. Rice seeds 218 were germinated in moist vermiculite trays until the three-leaf stage and then 219 220 transplanted into pots. During the whole growth period, all pots were irrigated with distilled water daily to maintain the water level of 3 cm above the soil surface. 221 Porewater samples were collected at a 15-days interval after tillering stage (after 45 222 223 days) using a porous fiber tube. The pH, Mn and As concentrations in soil solutions were analyzed. Rice was harvested at the 105th day. At each growth stage (tillering, 224 heading and maturing stages) rice plants were collected, digested, and then the 225 concentrations of Mn and As in the organs (roots, stems, leaves, husk and grains) of 226 rice plants were analyzed. 227

#### 228 **2.5 Field application experiment designs**

#### 229 2.5.1 Experimental design

The field experiment has carried out in May 2013 at a paddy field in Dengjiatang, 230 Chenzhou City, Hunan Province, (25°36'N, 113°00'E). The paddy fields were divided 231 232 into 1.5 m ×1.5 m sub-plots with a 40-cm buffer zone between each. Before rice planting, the top soil (0–20cm depth) was subject to manual plowing.  $\alpha$ -MnO<sub>2</sub> 233 nanorods was then added into the soil at a rate of 0.2%, 0.5%, 1.0% and 2.0% of soil 234 weight (0-20 cm), respectively, and thoroughly mixed with top soil. All treatment was 235 conducted in triplicate with a completely randomized factorial design, and three blank 236 controls (without  $\alpha$ -MnO<sub>2</sub> nanorods addition) were provided. Each plot was then let to 237 equilibrium for 7 days under natural conditions. Rice cultivars used in the pot 238

experiment (Yuzhenxiang) were also used in the field experiment. Rice seedlings 239 were transplanted after germination for 30 days (on June 6, 2013) and harvested on 240 241 September 30, 2013. To facilitate in situ sampling of pore water, during the growth of rice, 'Rhizon' soil solution samplers (Rhizon Research Products, Wageningen, The 242 Netherlands) were buried in each plot while the rice was transplanted. During the 243 whole growth period, water layer of about 3.0 cm above the topsoil of the paddy field 244 was maintained. The other cultivation methods were the same as the local paddy 245 cultivation methods until the rice has matured. 246

247 2.5.2 Sampling and, analyses

248 Extractions of pore water were conducted in the tillering stage (45 days after germination) and were extracted every 15 days during the last period (days 45-105). 249 250 The soil solution pH was recorded at the same time intervals as for pore water sampling. The concentrations of Mn in pore water were measured in acidified 251 subsamples by inductively coupled Plasma optical emission spectrometer (ICP-OES, 252 PerkinElmer Optima 8300, USA). As concentrations in soil pore water was 253 254 determined by atomic fluorescence spectrometer (AFS-920, Beijing Titan Instruments Co., Ltd.). At each specifically growth period, the plants sample was collected at the 255 tillering, heading and maturing stages. Rice plant samples were separated into roots, 256 257 stems, leaves and grain. And washed three times using distilled water, the cleaned plant samples were placed in an oven at 105°C for 2 h and dried at 70°C for 3 days to 258 constant weight, and then ground to pass a 100-mesh sieve with a micro plant 259

260 grinding machine.

The method for total As digestion and determination in rice plants was conducted 261 following the protocol of GB/T 5009.11-2003 which issued by Ministry of 262 Environmental Protection (MEP) of China (Geng et al., 2017). Typically, 1.0 g of dry 263 rice sample was digested using a mixture of acids (4:1 HNO<sub>3</sub>: HClO<sub>4</sub>, v/v) at 180°C 264 on a graphite digestion furnace. After digestion, the solution will let to cool down to 265 ambient temperature and made to 25 mL using UP water, each digested solution will 266 store in 50mL polyethylene bottle at 4°C before analysis. As concentrations in plants 267 tissues will determined by atomic fluorescence spectrometer (AFS-920, Beijing Titan 268 Instruments Co., Ltd.), Mn concentrations will measure by ICP-OES (Optima8300 269 PerkinElmer). For quality assurance and quality control purposes, blanks and standard 270 271 plant reference material (shrub branches and leaves GBW07603 (GSV-2), rice material (GBW10010 (GSB-1) were obtained from China Standard Materials 272 Research Center, Beijing, P.R. China and digested along with the unknown samples 273 and used for the QA/QC program. All the glassware was washed with detergent firstly, 274 soaked with 20% HNO<sub>3</sub> solution for 24 h, and then rinsed with UP water for three 275 times before use. 276

#### 277 2.6 Statistical analyses

All statistical analyses were performed with SPSS 22.0 software (SPSS Inc., Chicago, IL, USA). Differences between the control and treatment samples were determined using ANOVA and Tukey multiple comparisons analysis with p<0.05 indicating statistical significance. Correlations were obtained by Pearson correlationcoefficient in bivariate correlations.

283

### 284 **3. Results**

#### 285 3.1 Characterization of synthesized α-MnO<sub>2</sub> nanorods

The x-ray diffraction (XRD) pattern and scanning transmission electron 286 microscopy (SEM) images of the synthesized  $\alpha$ -MnO<sub>2</sub> nanorods are shown in Fig.S1, 287 Fig.S2, respectively. It can be seen that manganese dioxide is clustered; the structure 288 289 is uniformly well dispersed spherical agglomerate particles. High-magnification SEM images showed that the  $\alpha$ -MnO<sub>2</sub> is an urchin-like spherical with a diameter of 1-1.5 290  $\mu$ m, which consists of several straight and radially grown nanorods with uniform 291 diameter around 30-40nm (Fig.S2a, b). The synthesized  $\alpha$ -MnO<sub>2</sub> nanorods has an 292 obvious characteristic diffraction peak in the XRD pattern (Fig.S1), which indicated 293 that the high crystallization degree of nanostructured manganese dioxide, and the 294 295 characteristic peak was also in agreement with the standard data given in its JCPDS card (24-0072). 296

# 297 **3.2 Effects of different amount of α-MnO<sub>2</sub> nanorods on distribution of arsenic** 298 fractionation

The As sequential extraction procedure is widely used to evaluate As distribution within soil fractionation, which can help us to understand the mobility and bioavailability of As in soil(Jin et al., 2011; Wenzel et al., 2001; Zhang et al., 2017).The effect of different dosages of  $\alpha$ -MnO<sub>2</sub> nanorods on the distribution and percentages of Arsenic fractionation in paddy soil are presented in Fig.1.

In general, the application of  $\alpha$ -MnO<sub>2</sub> amendment increased the residual 304 fractionation and reduced the effective forms of As to some extent, indicating that 305  $\alpha$ -MnO<sub>2</sub> can effectively control the bioavailability of As in soil. As shown in Fig.1, in 306 the treated and untreated soil, loosely bound As comprised the smallest proportions of 307 all the As fractionation (<3%); what's more, the content of loosely bound As in the 308 309 soil with 1.0% MnO<sub>2</sub> treatment was significantly lower than that in other treatment. However, residual fractionation contained the most proportions of As (about 50%), 310 whether or not to add  $\alpha$ -MnO<sub>2</sub> into soil. Besides, proportions of residual As in the 311 312 1.0% MnO<sub>2</sub> treated soil was increased to 57.08% which indicated that the addition of 1.0% MnO<sub>2</sub> increased the residual As and reduced the other forms of As in the soil. 313 For binding state-As (Al-As, Ca-As and Fe-As), there was no significant 314

difference in the content of Al-As in the soil after treatment with different amounts of  $\alpha$ -MnO<sub>2</sub>, whereas Ca-As decreased to 10.23% after treatment with 2%  $\alpha$ -MnO<sub>2</sub>; and with the increase of concentration, Ca-As content showed an obviously decreasing trend, which indicated that the addition of  $\alpha$ -MnO<sub>2</sub> in paddy soil has a certain inhibitory effect on the binding of As and Ca.

320

321



Fig.1. Fraction distributions of arsenic in paddy soil amended with different amounts of α-MnO<sub>2</sub>; different
 letters indicate significant difference between different treatments (*P* < 0.05), arsenic fraction tested was</li>
 separately from each other.

For Fe-bound As, there was no regular change of Fe-As in the soil after the addition of  $\alpha$ -MnO<sub>2</sub>. However, it can be seen that the content of Fe-As in soils increased to 24.53% and 27.16% at rates of 0.5% and 2.0%, respectively. However, at dosages of 0.2% and 1%, the amount of Fe-As in the soil has reduced, and the addition of 1% was more obvious, which was 20.62%.

#### 330 **3.3 Pot experiments**

#### 331 **3.3.1 Effect of α-MnO<sub>2</sub> on the dynamic pH variations of soil solution**

The dynamics variations of pH in the pore water during growth of rice under treatments of different amounts of  $\alpha$ -MnO<sub>2</sub> were presented in Fig.S3. In general, during the whole growth stage (45-105 days), the soil pore water pH either increased to or remained stable in the near-neutral range after amended with different amounts of  $\alpha$ -MnO<sub>2</sub>. The pH range of the soil solution was around 7.36-7.55 for the 45th day, which was higher than the pH (7.25) of CK (Control treatments). The highest pH value of the pore water was recorded at the 45th day (7.25-7.55). Afterwards, the pH of the pore water decreased with the rice growth and reached the lowest value (6.28-6.76) at the 105th days. Compared to the control,  $\alpha$ -MnO<sub>2</sub> treatments slightly reduced the pH of the paddy soil under flooded condition, the pH was reduced by 0.58-1.06 unit after supplementation with  $\alpha$ -MnO<sub>2</sub>.

#### 343 **3.3.2 Effect of α-MnO<sub>2</sub> on the dynamic variations of As and Mn in pore water**



Fig.2. Dynamics variations of pore water As (a) and Mn (b) in the α-MnO<sub>2</sub> treatments throughout the rice
 cultivation period. Data are means SE (n= 3).

After treatments with different dosage of  $\alpha$ -MnO<sub>2</sub> the dynamic variation of arsenic and manganese content in pore water at different growth period of rice is shown in Fig.2. Different reduction patterns of arsenic in soil pore water were observed among with different treatments. Regardless of the addition of  $\alpha$ -MnO<sub>2</sub>, the arsenic content in soil pore water of CK was decreased from the 45th day to the 105th

day, and reaching the minimum value of  $13.26\pm0.94 \text{ µg} \cdot \text{L}^{-1}$  at the 105th day; possibly 351 due to the adsorbed by newly formed endogeneity amorphous iron oxides (Amstaetter 352 et al., 2012; Cismasu et al., 2015). However, compared with CK, the content of 353 arsenic in pore water after adding  $\alpha$ -MnO<sub>2</sub> with different dosages decreased markedly 354 355 at the 45th day. All treatments showed an obvious dosage dependent reduction trend 356 of arsenic in soil pore water. Similarly, the decline trend was observed between CK and 0.2% treatments; however, with the increasing of dosages the content of arsenic 357 began to rebound after 45 days. Arsenic concentrations peaked at 60 days of reaction 358 at 0.5% treatments; 90 day for 1.0% treatments and 60 day of 2.0% treatments. In 359 spite of fluctuation, what worth affirming was that arsenic in soil pore water still 360 decreased to a constant level (10.94-14.69  $\mu$ g·L<sup>-1</sup>) at the end of rice cultivation period 361 (90-105 day), except for 1% treatment, nearly 2-fold (20.49 µg·L<sup>-1</sup>) increased in 362 porewater As was observed during 90-105 day. 363

Unlike As, α-MnO<sub>2</sub> treatments increased porewater Mn levels (Fig.2b), four 364 treatments (0.2%, 0.5%, 1.0% and 2.0%) showed that substantial increased in the 365 porewater Mn concentration at the 45th day, whereas Mn concentration in the CK soil 366 porewater remained low level throughout the whole cultivation period (45-105 days). 367 What's more, Mn concentrations kept increasing after another 15 days; with 17-33 368 folds augment compared with that of control soil. This is more likely coupled with the 369 enhanced dissolution of manganese resulting from oxidation of As(III) to As(V) by 370 manganese oxide (Xu et al., 2017; Z et al., 2017). Yet, effluent Mn(II) concentrations 371 in soil porewater began to drop at the 60th day, with concentrations declining to a 372

373 constant level of 0.13-0.60 mg·L<sup>-1</sup> after 90 days of cultivation. Furthermore, among 374 all the four treatments, 1.0 % retained the highest porewater Mn dissolution rate 375 compared with other treatments during day 45-75 days and reaching a peak of 49.77 376 mg·L<sup>-1</sup> on the 60th day. In the meantime, however, the other three treatments have 377 already dropped to low levels (5.2-12.6 mg·L<sup>-1</sup>).

# 378 **3.3.3 Effect of α-MnO<sub>2</sub> on the accumulation of As and Mn in rice plants parts**379 during different growth stage



380 Fig.3. As and Mn concentrations in the root (a, b), straw (c, d), leaf (e, f) in the α-MnO<sub>2</sub> treatments; Data are

means SE (n= 3), different letters indicate significant difference between different treatments (P < 0.05),</li>
 significant tests are separate from each other (black letter indicates tillering stage, red letter indicates
 heading stage, blue letter indicates maturation stage)

384 The contents of As and Mn in different parts of rice at various growth stages are shown in Fig.3. Three different rice growth stages (Tillering, heading and mature 385 stage) were selected to estimate the efficiency of  $\alpha$ -MnO<sub>2</sub> in controlling the 386 accumulation of As and Mn in rice. In general, regardless of the addition of  $\alpha$ -MnO<sub>2</sub>, 387 the distribution pattern of arsenic in rice roots, stems and leaves follows the order of 388 roots > stems > leaves. However, with the addition of  $\alpha$ -MnO<sub>2</sub>, the arsenic content in 389 various parts of rice at each growth period was reduced to varying degrees compared 390 with the CK. That is, the addition of  $\alpha$ -MnO<sub>2</sub> can effectively impede the migration of 391 arsenic to rice plants. In α-MnO<sub>2</sub> treatments, root As was decreased by 10.63-77.51% 392 at tillering stage (Fig.3a), 52.71-81.94% at heading stage compared to the control. 393 394 However, the interesting thing is that the reduction rate of arsenic in the mature stage was much lower than that of the other two growth period, which was indicated that 395 the heading and tillering stage may be the key time period to control the transportation 396 397 and migration of As (Li et al., 2015; Zheng et al., 2011).

On the contrary, the distribution pattern of Mn content in rice roots was clearly different from that of As in rice. Compared with CK, the Mn content in various parts of rice plants has significantly increased (p<0.05) at different growth stages after addition of  $\alpha$ -MnO<sub>2</sub> and the increasing trend was obviously dosage dependent, which illustrated that the order of Mn in roots, stems and leaves of rice was: 2.0% > 1.0% > 0.5% > 0.2% > CK (Fig.3b). The overall Mn concentration in rice roots was increased by 1.9-15.87 times at tillering stage, 4.58-39.46 times at heading stage and 6.5-49.89 405 times at the mature stage, respectively.

Although  $\alpha$ -MnO<sub>2</sub> treatments were effective in reducing and accumulating of As 406 407 and Mn in rice roots; their effects, however, on As, Mn transportation in the straw and leaf was less satisfactory. For the re-distribution of As in rice stalk and leaf, regardless 408 409 of the addition of  $\alpha$ -MnO<sub>2</sub> and rice growth period sequences, the higher accumulation 410 efficiency were observed at mature stage for both stalk and leaf; which indicated that mature stage were the crucial period for the translocation and migration of As in rice 411 (Fig.3ce). However, the accumulation of Mn in stalks and leaves showed different 412 413 patterns (Fig.3df), rice tended to accumulate more Mn during tillering stage, which was probably due to the enhanced dissolution of Mn in soil porewater at early stages 414 (45-60 days) of rice growth after amended with  $\alpha$ -MnO<sub>2</sub> nanorods. 415

In addition, the observed results also implied that the re-distribution of As in rice part did not interfered by the added  $\alpha$ -MnO<sub>2</sub> after the tillering stage and this phenomenon was less time-dependent; which also confirmed that the As was sequestered in rice roots and thus the subsequent influx of As into other rice parts was restrained (Fig.3ce).

The content of As and Mn in brown rice and husk can further prove this phenomenon (Fig.4). The addition of  $\alpha$ -MnO<sub>2</sub> Nano-rods significantly reduced the total arsenic content in brown rice and husk (p<0.05), while the Mn content was significantly increased (p<0.05). Compared with CK, the contents of total arsenic in the husks decreased by 36.4%, 24.0%, 12.6% and 15.5%, respectively. Furthermore, the content of total arsenic in brown rice was decreased by 17.8%, 36.4%, 65.4% and 60.7%, respectively, which demonstrated that the addition of α-MnO<sub>2</sub> nanorods can effectively prevent arsenic uptake by brown rice. Meanwhile, Mn concentrations in rice husk and brown rice increased in a dosage dependent way, which demonstrated that the Mn concentrations increased with increasing dosages of α-MnO<sub>2</sub> nanorods. The content of Mn in the husk increased by 55.8%, 79.3%, 102.0% and 133.3%, respectively; and the content of Mn in brown rice increased by 148.7%, 174.6%, 295.5% and 310.4%, respectively (Fig.4).



Treatments

Fig.4 Concentrations of As and Mn in rice husk and brown rice (Pot experiment),(a) As-rice
husk, (b) As-brown rice,(c) Mn-rice husk,(d) Mn-brown rice; Data are means SE (n= 3),
different letters indicate significant difference between different treatments (P < 0.05).</li>

# 437 3.3.4 Relationships between As and Mn concentration in rice plants parts under 438 different growth stages

To further identify the relationship between the concentrations of As and Mn in rice (Fig.S4) and different growth stages (Fig.5), we performed a correlation analysis; and the results were presented in Fig.S4 and Fig.5 The concentration of As showed an obviously significant negative correlation (p<0.05) with the amount of Mn in the rice, indicating that the As content of rice plants decreased significantly with the increase of Mn concentrations. And this also indicated that the application of  $\alpha$ -MnO<sub>2</sub> nanorods was extremely effective at decreasing the available As in the soil



#### Mn concentration (mg/kg)

446 Fig.5. Relationships between As and Mn concentration in rice plants during different growth stage (a, b, c,

447 root-As-Mn), (d, e, f, stalk -As-Mn) and (g, h, I, leaf-As-Mn); Data are means SE (n= 3). In the meantime, the relationships between As and Mn in different rice parts at 448 different growth stages were also analyzed, and the correlations were presented in 449 Fig.5, as can be seen from the above picture, overall As and Mn correlations in 450 various parts showed obviously negative correlations. Especially for root As and Mn, 451 their correlation showed higher  $R^2$  value compared with other two parts, regardless of 452 the growth stages ( $R^2=0.71-0.88$ , p<0.05; Fig.5abc). In contrast, although at certain 453 growth stages, the As and Mn in rice parts showed extremely high significance; their 454 overall relationships in stalks ( $R^2=0.20-0.63$ ; Fig.5def) and leaves ( $R^2=0.44-0.87$ ; 455 Fig.5ghi), however, were poor than roots As-Mn. 456

### 457 **3.4 Field applications**





Fig.6. As and Mn concentrations in the root (a, d), straw (b, e), and leaf (c, f) of rice in the α-MnO<sub>2</sub>
 treatments (Field application); Data are means SE (n= 3), different letters indicate significant difference
 between different treatments (P < 0.05), significant tests are separate from each other (black letter indicates</li>
 tillering stage, red letter indicates heading stage, blue letter indicates maturation stage)

463 Previous studies have shown that the addition of  $\alpha$ -MnO<sub>2</sub> nanorods in 464 arsenic-contaminated soil can effectively control the transport of arsenic from soil to 465 rice in pot experiments. This result provides a theoretical basis for the application of 466 α-MnO<sub>2</sub> nanorods in field trials. With this in mind, field trials were designed to

467	further evaluate the effectiveness of $\alpha$ -MnO <sub>2</sub> nanorods in controlling the mobility and
468	bio-availability of arsenic in soil-rice interfaces under complex natural systems. The
469	content of arsenic and manganese in different parts of rice at various stages in the
470	field experiment was shown in Fig.6. With the addition of $\alpha$ -MnO <sub>2</sub> nanorods, the
471	arsenic content in various parts of rice at each period was reduced to varying degrees
472	compared with the CK group (Fig.6b). It was also can be seen that the arsenic
473	concentration in rice roots was much higher than that in the stalks and leaves.
474	Furthermore, the similar results have been observed in stalks and leaves compared
475	with pot experiment, in which rice tended to accumulate more As at mature stage.
476	And the observed results also indicated that most of the As was sequestered in rice
477	roots (Fig.6bc). Meanwhile, after treatments with different amounts of $\alpha$ -MnO <sub>2</sub>
478	nanorods, the content of Mn in various parts of rice plants increased significantly

(p<0.05) at different growth periods compared with the CK. And this result was 479 consistent with the results obtained in the pot experiment, which also proved that the 480 481  $\alpha$ -MnO<sub>2</sub> nanorods can indeed reduce the bio-availability of arsenic in soil. After treatment with  $\alpha$ -MnO<sub>2</sub> nanorods, total As content in brown rice and husk was 482 significantly reduced (Fig.7; p<0.05). Arsenic content in the husk of CK was 483 3.06±0.41mg·kg<sup>-1</sup> and 0.96±0.08mg·kg<sup>-1</sup> in brown rice. Compared with CK, the 484 contents of total As in husks decreased by 60.5%, 79.6%, 65.7%, and 56.9%, 485 respectively; and the content of total As in brown rice was decreased by 61.5%, 486 60.4%, 43.4%, and 77.1%, respectively. Among them, the lowest content 487





489 treatments.

490 Fig.7. Concentrations of As in rice husk and brown rice (Field application), (a) rice husk, (b) brown rice; 491 Data are means SE (n= 3), different letters indicate significant difference between different treatments (P < 492

### 493 **4. Discussion**

# 494 4.1 Effects of different dosages of α-MnO<sub>2</sub> nanorods on the solid-liquid 495 partitioning of As in paddy soil

Since long is it established that the toxicity, activity and bioavailability of As in 496 497 paddy soil are closely related to its presence in the soil (Yamaguchi et al., 2011). The hazards of As in soil are not only related to its content, but also related to its 498 effectiveness in the soil and its associated binding forms (classification). In the 499 500 present study, a 90d soil incubation experiment combined with sequential extraction procedure, therefore, was conducted to study the arsenic fractionation transformed in 501 soil under flooded conditions after amended with  $\alpha$ -MnO<sub>2</sub> nanorods; because it can 502 503 furnish us an indication for the mobility and bioavailability of As in paddy soil after amended with  $\alpha$ -MnO<sub>2</sub> nanorods. As can be draw from the above results (Fig.1), after 504 treatment with different amounts of  $\alpha$ -MnO<sub>2</sub> nanorods the content of effective As 505 decreased with the increasing of residual As and insoluble binding As (Ca-As and 506 Fe-As), indicating that supplementation of  $\alpha$ -MnO<sub>2</sub> nanorods can effectively control 507 the bio-availability of As in soil and reduced the associated influx of As into rice 508 plants. Generally, loosely bound As (water soluble and exchangeable As) are highly 509 bio-availability, and they are easily absorbed by organisms, resulting in greater 510 toxicity. The Fe-As and other encapsulated state As are not easily absorbed by 511 organisms and enter the water body, their harmfulness is relatively low. Fe-As, Al-As 512 513 bind closely to soil, their toxicity to organisms are generally less than Ca-As.

514	(Herreweghe et al., 2003)found that water-soluble As and exchangeable As are
515	soluble As in the soil or As adsorbed on the surface of soil particles, which accounts
516	for less than 3% of total As. Similar results have been observed in our experiment, the
517	proportion of loosely bound As in the soil was less than 3% regardless of the addition
518	of $\alpha$ -MnO <sub>2</sub> nanorods, which indicated that the mobilization pool of As in paddy soil
519	depends on other forms. Thus, the content of binding As (such as Fe-As, Al-As) may
520	be the most contributing component in soil. Under aerobic conditions, As(V) is
521	strongly adsorbed on most mineral constituents, including Fe and Al(hydr)oxides
522	(Goldberg, 2002) whereas, arsenite is more mobile because of its poor affinity for
523	mineral surfaces (Han et al., 2011). Manganese oxide has long be regarded as natural
524	occurring powerful oxidants (Han et al., 2011; Ren et al., 2013), which can rapidly
525	convert As(III) to As(V) over the pH range of 4~8.2 under natural conditions (Bruce
526	A. Manning et al., 2002). In the present study, in order to simulate an authentic rice
527	growing environment, the soil have been flooded with water along with the whole
528	incubation period. It is well accepted that incubation under flooded conditions can
529	cause solid As to be distributed to liquid phase(Lemonte et al., 2017; Xu et al., 2017),
530	in which the dominating As species was arsenite ; so As can be re-mobilized into soil
531	when As(V) is reduced to As(III). In spite of this circumstance, as can be seen from
532	the Fig.1, the content of Fe-As still increased after treatment with 0.5% and 2.0% of
533	$\alpha$ -MnO <sub>2</sub> nanorods, which proved that the increased binding state As was more likely
534	due to the oxidation of As(III) into As(V) by $\alpha$ -MnO <sub>2</sub> nanorods and subsequently
535	resulted to the enhanced adsorption onto ferric(hydr)oxides and Al mineral. The

subsequently re-allocation of As into the soil solution was, thus effectively reduced.
Besides, the content of Ca-As was decreased with an increase in the associated
residual As at the dosage of 1%, which indicated that the other encapsulated As
fractionation was also transformed to less effective forms.

Combined the obtained results in soil incubation experiments, which can 540 provide us with a reliable theoretical basis for our subsequent experiment. The 541 hypothesis of the present study was proposed that in the rice rhizosphere 542 micro-environment, addition of α-MnO<sub>2</sub> nanorods can affect the chemical speciation 543 of As in the soil solution through in situ oxidation which in turn affect the 544 bio-availability and mobility of As for rice uptake. As already discussed in soil 545 incubation experiment, the partitioning of As into the soil solution was significantly 546 547 controlled by As fractionation transformed. This part of observed results can be further proved by the dynamic monitoring of As variations in soil solution (Fig.2ab). 548 As can be seen from Fig.2a, the content of porewater As was significantly decreased 549 as a dosage dependent way at the 45th day with the increasing content of  $\alpha$ -MnO<sub>2</sub> 550 nanorods. And during the whole growth stages (45-105 days), in spite of slightly 551 fluctuation, the overall concentrations of As was decreased. Several studies have been 552 deciphered that the effluent of Mn is attributed to the process of arsenite oxidation by 553 manganese oxides (Lafferty et al., 2010; Oscarson et al., 1983; Tournassat et al., 554 2002); and it is widely accepted that the oxidation of As(III) to As(V) involves in a 555 two-step process, including the release of Mn(II) and sorption of Mn(II) on oxidation 556 intermediate (Nesbitt et al., 1998). 557

Our results are extremely consistent with those previous studies. As can be 558 drawn from the above picture (Fig.2b) the content of Mn has been increased to an 559 560 extraordinary magnitude (17-33 folds) compared with the untreated soil. Nevertheless, unfortunately the persistence increasing of Fe(II) and Mn(II) in soil porewater under 561 reduced conditions at early growth stages (45-60 days) can inhibited the abiotic 562 oxidation of As(III) by  $\alpha$ -MnO<sub>2</sub> nanorods (Ehlert et al., 2014). Another study reported 563 by Chen et al. (2006) showed that the oxidation rates could be also impaired by soil 564 organic matter(Chen et al., 2006). Those reported studies explained that why the Mn 565 566 concentrations in the soil porewater tend to decrease with the rice growth time prolonged (60-105days) (Fig.2b). 567

#### 568 **4.2 Enhanced impeded of As accumulation in rice by α-MnO<sub>2</sub> nanorods.**

569 Combined the results which obtained from soil incubation experiments, we can draw a conclusion that the partitioning of As into soil porewater has been rigorous 570 hindered (Fig.2a). Moreover, the impeded As elution has also in turn affected the 571 accumulation of As into rice. But the interplay between the addition of  $\alpha$ -MnO<sub>2</sub> 572 573 nanorods and As in plant cultivation systems was far more complexity than the pure soil incubation. Pot and field rice cultivation together with soil incubation 574 experiments were therefore combined to clarify the effects of different amounts of 575 576  $\alpha$ -MnO<sub>2</sub> nanorods on the accumulation and translocation of As into rice (Fig.3-7). On the basis of the present study, we believe that there are at least two factors appear to 577 play crucial roles in controlling the As influx into rice plants. 578

579	Firstly, in situ oxidation of As(III) to As(V) in the presence of $\alpha$ -MnO <sub>2</sub> nanorods
580	leads to the subsequently enhanced adsorption of As onto endogenous iron(oxyhydr)
581	oxides; greatly reduced the re-allocation of As into soil porewater, and thus directly
582	cut down the total As transportation into rice parts. As can be seen from the above
583	picture (Fig.3 and Fig.6) The addition of Mn oxides significantly impeded the
584	accumulation of As into subterranean parts (roots $p < 0.05$ ); and the associated influx
585	of As into aerial parts (stems, leaves, husk and brown rice) has been also obstructed
586	(Fig.3 and Fig.6; $p < 0.05$ ). Correlations analysis (Fig.S4 and Fig.5) between As and
587	Mn in rice parts can also prove that the transportation of As has been greatly limited.
588	Secondly, apart from those existing fact, As bioavailability could be also
589	mediated through iron and manganese plaque formation on the rice roots and so
590	influence As uptake by rice plants (Liu, 2005; Liu et al., 2005). Therefore, enhanced
591	sequestrate of As by rice roots were probably due to the iron and manganese plaque
592	formation of the rice roots (Fig.3a and Fig.5a). Besides, the addition of manganese
593	oxides can also promote Fe(II) oxidation (Postma and Appelo, 2000); Ehlert et al.
594	(Ehlert et al., 2014) reported that birnessite additions can promote Fe(II) oxidation to
595	Fe(III), thereby creating newly-formed Fe(III) hydroxides which could serve as
596	efficient sorbents for As(III).
597	Hence, by combination of the obtained facts, we can draw a conclusion that the
598	addition of $\alpha$ -MnO <sub>2</sub> nanorods served as a multifunctional role on the As mobilization

soil leads to the less dissolution of As in soil porewater. Secondly, As(III) oxidation

599

and transportation in paddy fields. Firstly, the effects on the As fractionation in paddy

trigged by  $\alpha$ -MnO<sub>2</sub> nanorods resulted to the adsorption of water soluble As(V) onto iron oxides. Lastly, the enhanced dissolution of Mn(II) and Fe(III) can lead to the more iron and manganese plaque formation, which can sequester more As on the surface of rice roots.

# 605 **5 Conclusions**

In the present study, soil incubation experiments which combined with pot and 606 607 field rice cultivation trials were designed to evaluate the effectiveness of exogenous  $\alpha$ -MnO<sub>2</sub> nanorods on the mobilization and transportation of As in soil-rice systems. 608 Our results proved that the addition of  $\alpha$ -MnO<sub>2</sub> nanorods can effectively control the 609 610 soil-to-solution partitioning of As under anaerobic conditions. The As fractionation can be transformed from effective forms into less effective forms impeded the 611 re-allocation of As into soil porewater, and thus reduced the total amounts of As 612 influx into rice. Besides, the enhanced oxidation of As(III) into As(V) by α-MnO<sub>2</sub> 613 nanorods greatly increased the adsorption of As onto indigenous iron(hydr) oxides, 614 thus, reduced the soil porewater As. Furthermore, with the simultaneous 615 co-occurrences of the oxidation intermediates of Mn(II) and Fe(III), which can 616 probably lead to the enhanced formation of iron and manganese plaque on the surface 617 of rice roots. Combined, the influx of As into aerial parts of rice plants (stems, husk 618 and leaves) was strictly prohibited by rice roots. Nevertheless, it should be noted that 619 the abiotic oxidation of As by  $\alpha$ -MnO<sub>2</sub> nanorods are greatly impaired by various 620 environmental factors (such as DOM, microbial activities and ligands), thus, further 621

622 work is still needed to verify these results under fields or laboratory scales.

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### 629 **References**

- Amstaetter K, Borch T, Kappler A. Influence of humic acid imposed changes of
   ferrihydrite aggregation on microbial Fe(III) reduction. Geochimica Et
   Cosmochimica Acta 2012; 85: 326-341.
- BA M, SE F, B B, DL S. Arsenic(III) oxidation and arsenic(V) adsorption reactions
  on synthetic birnessite. Environmental Science & Technology 2002; 36: 976-81.
- Balasubramanian N, Kojima T, Srinivasakannan C. Arsenic removal through
  electrocoagulation: kinetic and statistical modeling. Chemical Engineering
  Journal 2009; 155: 76-82.
- Beiyuan J, Li JS, Dcw T, Wang L, Poon CS, Li XD, et al. Fate of arsenic before and
  after chemical-enhanced washing of an arsenic-containing soil in Hong Kong.
  Science of the Total Environment 2017; 599-600: 679-688.
- Bontempi E. A new approach for evaluating the sustainability of raw materials
  substitution based on embodied energy and the CO 2 footprint. Journal of
  Cleaner Production 2017; 162.
- Bruce A. Manning, ‡ SEF, Benjamin Bostick A, Suarez§ DL. Arsenic(III) Oxidation
  and Arsenic(V) Adsorption Reactions on Synthetic Birnessite. Environmental
  Science & Technology 2002; 36: 976.
- 647 Chen Z, Kim KW, Zhu YG, Mclaren R, Liu F, He JZ. Adsorption (AsIII,V) and
  648 oxidation (AsIII) of arsenic by pedogenic Fe–Mn nodules. Geoderma 2006; 136:
  649 566-572.
- Cismasu AC, Williams KH, Nico PS. Iron and carbon dynamics during aging and
   reductive transformation of biogenic ferrihydrite. Environmental Science &
   Technology 2015; 50.

- Ehlert K, Mikutta C, Kretzschmar R. Impact of birnessite on arsenic and iron
  speciation during microbial reduction of arsenic-bearing ferrihydrite.
  Environmental Science & Technology 2014; 48: 11320-11329.
- Fendorf S, Kocar BD. Biogeochemical processes controlling the fate and transport of
  arsenic: implications for South and Southeast Asia. Advances in Agronomy
  2009; 104: 137-164.
- Fu QL, Liu C, Achal V, Wang YJ, Zhou DM. Aromatic Arsenical Additives (AAAs)
  in the Soil Environment: Detection, Environmental Behaviors, Toxicities, and
  Remediation. Vol 140, 2016.
- Geng A, Wang X, Wu L, Wang F, Chen Y, Yang H, et al. Arsenic accumulation and
  speciation in rice grown in arsanilic acid-elevated paddy soil. Ecotoxicology &
  Environmental Safety 2017; 137: 172.
- Gil-Díaz M, Alonso J, Rodríguez-Valdés E, Gallego JR, Lobo MC. Comparing
  different commercial zero valent iron nanoparticles to immobilize As and Hg in
  brownfield soil. Science of the Total Environment 2017; 584-585.
- Gil-Díaz M, Diez-Pascual S, González A, Alonso J, Rodríguez-Valdés E, Gallego JR,
  et al. A nanoremediation strategy for the recovery of an As-polluted soil.
  Chemosphere 2016; 149: 137-145.
- Gilloaiza J, White SA, Root RA, Solísdominguez FA, Hammond CM, Chorover J, et
  al. Phytostabilization of Mine Tailings Using Compost-Assisted Direct Planting:
  Translating Greenhouse Results to the Field. Science of the Total Environment
  2016; 565: 451.
- Goldberg S. Competitive Adsorption of Arsenate and Arsenite on Oxides and Clay
   Minerals. Soil Science Society of America Journal 2002; 66: 413-421.
- Han X, Li YL, Gu JD. Oxidation of As(III) by MnO 2 in the absence and presence of
  Fe(II) under acidic conditions. Geochimica Et Cosmochimica Acta 2011; 75:
  368-379.
- Herreweghe SV, Swennen R, Vandecasteele C, Cappuyns V. Solid phase speciation
  of arsenic by sequential extraction in standard reference materials and
  industrially contaminated soil samples. Environmental Pollution 2003; 122:
  323-342.
- Jankong P, Visoottiviseth P, Khokiattiwong S. Enhanced phytoremediation of arsenic
   contaminated land. Chemosphere 2007; 68: 1906-1912.
- Jin HP, Choppala GK, Bolan NS, Chung JW, Chuasavathi T. Biochar reduces the
  bioavailability and phytotoxicity of heavy metals. Plant & Soil 2011; 348: 439.
- Kumpiene J, Lagerkvist A, Maurice C. Stabilization of As, Cr, Cu, Pb and Zn in soil
  using amendments--a review. Waste Management 2008; 28: 215.
- Lafferty BJ, Gindervogel M, Sparks DL. Arsenite Oxidation by a Poorly Crystalline
   Manganese-Oxide 1. Stirred-Flow Experiments. Environmental Science &
   Technology 2010; 44: 8460-6.
- Lei M, Tie B, Zeng M, Qing P, Song Z, Williams PN, et al. An arsenic-contaminated
  field trial to assess the uptake and translocation of arsenic by genotypes of rice.
  Environ Geochem Health 2013; 35: 379-390.
- 696 Lei M, Tie BQ, Song ZG, Liao BH, Lepo JE, Huang YZ. Heavy metal pollution and

- 697 potential health risk assessment of white rice around mine areas in Hunan
  698 Province, China. Food Security 2015; 7: 45-54.
- Lemonte JJ, Stuckey JW, Sanchez JZ, Tappero RV, Rinklebe J, Sparks DL. Sea level
  rise induced arsenic release from historically contaminated coastal soils.
  Environmental Science & Technology 2017; 51.
- Li B, Peng L, Wei D, Lei M, Liu B, Lin Y, et al. Enhanced flocculation and
  sedimentation of trace cadmium from irrigation water using phosphoric fertilizer.
  Science of the Total Environment 2017; 601-602: 485.
- Li JS, Wang L, Cui JL, Poon CS, Beiyuan J, Dcw T, et al. Effects of low-alkalinity
  binders on stabilization/solidification of geogenic As-containing soils:
  Spectroscopic investigation and leaching tests. Science of the Total Environment
  2018; 631-632: 1486.
- Li M, Wang L, Jia Y, Yang Z. Arsenic speciation in locally grown rice grains from
  Hunan Province, China: Spatial distribution and potential health risk. Science of
  the Total Environment 2016; s 557–558: 438-444.
- Li R, Zhou Z, Zhang Y, Xie X, Li Y, Shen X. Uptake and Accumulation
  Characteristics of Arsenic and Iron Plaque in Rice at Different Growth Stages.
  Communications in Soil Science & Plant Analysis 2015; 46: 2509-2522.
- Liao X, Chen T, Xie H, Xiao X. Effect of application of P fertilizer on efficiency of
  As removal from As-contaminated soil using phytoremediation: Field study.
  Acta Scientiae Circumstantiae 2004.
- Liao XY, Chen TB, Xie H, Liu YR. Soil As contamination and its risk assessment in
  areas near the industrial districts of Chenzhou City, Southern China.
  Environment International 2005; 31: 791-798.
- Limmer MA, Wise P, Dykes GE, Seyfferth AL. Silicon Decreases Dimethylarsinic
   Acid Concentration in Rice Grain and Mitigates Straighthead Disorder.
   Environmental Science & Technology 2018; 52: 4809-4816.
- Lin L, Gao M, Qiu W, Wang D, Huang Q, Song Z. Reduced arsenic accumulation in
   indica rice (Oryza sativa L.) cultivar with ferromanganese oxide impregnated
   biochar composites amendments. Environmental Pollution 2017; 231: 479.
- Liu C, Yu HY, Liu C, Li F, Xu X, Wang Q. Arsenic availability in rice from a mining
  area: is amorphous iron oxide-bound arsenic a source or sink? Environmental
  Pollution 2015; 199: 95-101.
- Liu L, Li W, Song W, Guo M. Remediation techniques for heavy metal-contaminated
  soils: Principles and applicability. Science of the Total Environment 2018; 633:
  206-219.
- Liu R, Zhao D. Reducing leachability and bioaccessibility of lead in soils using a new
  class of stabilized iron phosphate nanoparticles. Water Research 2007; 41:
  2491-2502.
- Liu WJ. Direct evidence showing the effect of root surface iron plaque on arsenite and
   arsenate uptake into rice (Oryza sativa) roots. New Phytologist 2005; 165: 91-97.
- Liu WJ, Zhu YG, Smith FA. Effects of Iron and Manganese Plaques on Arsenic
  Uptake by Rice Seedlings (Oryza sativa L.) Grown in Solution Culture Supplied
  with Arsenate and Arsenite. Plant & Soil 2005; 277: 127-138.

- Liu Z, Zhu QQ, Tang LH. Microelements in the main soils of China. Soil Science
  1983; 135: 40-46.
- Ma JF, Yamaji N, Mitani N, Xu XY, Su YH, Mcgrath SP, et al. Transporters of
  Arsenite in Rice and Their Role in Arsenic Accumulation in Rice Grain.
  Proceedings of the National Academy of Sciences of the United States of
  America 2008; 105: 9931-9935.
- Meharg AA. Arsenic in rice--understanding a new disaster for South-East Asia.
  Trends in Plant Science 2004; 9: 415-417.
- Meharg AA, Jardine L. Arsenite transport into paddy rice (Oryza sativa) roots. New
  Phytologist 2003; 157: 39-44.
- Nesbitt HW, Canning GW, Bancroft GM. XPS study of reductive dissolution of
  752 7?9-birnessite by H3AsO3, with constraints on reaction mechanism Part 1.
  753 EXAFS studies of the geometry of coprecipitated and adsorbed arsenate.
  754 Geochimica Et Cosmochimica Acta 1998: 2097-2110.
- Ohtsuka T, Yamaguchi N, Makino T, Sakurai K, Kimura K, Kudo K, et al. Arsenic
  dissolution from Japanese paddy soil by a dissimilatory arsenate-reducing
  bacterium Geobacter sp. OR-1. Environmental Science & Technology 2013; 47:
  6263-6271.
- Okkenhaug G, Zhu YG, He J, Li X, Luo L, Mulder J. Antimony (Sb) and arsenic (As)
  in Sb mining impacted paddy soil from Xikuangshan, China: differences in
  mechanisms controlling soil sequestration and uptake in rice. Environmental
  Science & Technology 2012; 46: 3155.
- Oscarson DW, Huang PM, Liaw WK, Hammer UT. Kinetics of Oxidation of Arsenite
   by Various Manganese Dioxides1. Soil Sci.soc.am.j 1983; 47: 644-648.
- Postma D, Appelo CAJ. Reduction of Mn-oxides by ferrous iron in a flow system:
  column experiment and reactive transport modeling. Geochimica Et
  Cosmochimica Acta 2000; 64: 1237-1247.
- Ren HT, Jia SY, Wu SH, Liu Y, Hua C, Han X. Abiotic oxidation of Mn(II) induced
  oxidation and mobilization of As(III) in the presence of magnetite and hematite.
  Journal of Hazardous Materials 2013; s 254–255: 89-97.
- Scott MJ, Morgan JJ. Reactions at Oxide Surfaces. 1. Oxidation of As(III) by
   Synthetic Birnessite. Environmental Science & Technology 1995; 29: 1898.
- Seyfferth AL, Limmer MA, Dykes GE. On the Use of Silicon as an Agronomic
  Mitigation Strategy to Decrease Arsenic Uptake by Rice. Advances in
  Agronomy 2018.
- Suda A, Makino T. Functional effects of manganese and iron oxides on the dynamics
  of trace elements in soils with a special focus on arsenic and cadmium: A review.
  Geoderma 2016; 270: 68-75.
- Takahashi Y, Minamikawa R, Hattori KH, Kurishima K, Kihou N, Yuita K. Arsenic
  Behavior in Paddy Fields during the Cycle of Flooded and Non-flooded Periods.
  Environmental Science & Technology 2004; 38: 1038-1044.
- Tokunaga S, Hakuta T. Acid washing and stabilization of an artificial
  arsenic-contaminated soil. Chemosphere 2002; 46: 31-38.
- 784 Tournassat C, Charlet L, Bosbach D, Manceau A. Arsenic(III) oxidation by birnessite

- and precipitation of manganese(II) arsenate. Environmental Science &
  Technology 2002; 36: 493.
- Van HS, Swennen R, Vandecasteele C, Cappuyns V. Solid Phase Speciation Of
  Arsenic By Sequential Extraction In Standard Reference Materials And
  Industrially Contaminated Soil Samples. Environmental Pollution 2003; 122:
  323-42.
- Villen-Guzman M, Gomez-Lahoz C, Garcia-Herruzo F, Vereda-Alonso C, Paz-Garcia
   JM, Rodriguez-Maroto JM. Specific Energy Requirements in Electrokinetic
   Remediation. Transport in Porous Media 2017: 1-11.
- Wan X, Lei M, Chen T. Cost-benefit calculation of phytoremediation technology for
   heavy-metal-contaminated soil. Science of the Total Environment 2016; s
   563–564: 796-802.
- Weber FA, Hofacker AF, Voegelin A, Kretzschmar R. Temperature dependence and
  coupling of iron and arsenic reduction and release during flooding of a
  contaminated soil. Environmental Science & Technology 2010; 44: 116-122.
- Wenzel WW, Kirchbaumer N, Prohaska T, Stingeder G, Lombi E, Adriano DC.
  Arsenic fractionation in soils using an improved sequential extraction procedure.
  Analytica Chimica Acta 2001; 436: 309-323.
- Williams PN, Lei M, Sun G, Huang Q, Lu Y, Deacon C, et al. Occurrence and
  partitioning of cadmium, arsenic and lead in mine impacted paddy rice: Hunan,
  China. Environmental Science & Technology 2009; 43: 637.
- Wu B, Liao XY, Chen TB. Comparison of five methods for fractionation of
  calcareous soil contaminated with arsenic. Acta Scientiae Circumstantiae 2006;
  26: 1467-1473.
- Xu X, Chen C, Wang P, Kretzschmar R, Zhao FJ. Control of arsenic mobilization in
  paddy soils by manganese and iron oxides. Environmental Pollution 2017; 231:
  37-47.
- Yamaguchi N, Nakamura T, Dong D, Takahashi Y, Amachi S, Makino T. Arsenic
  release from flooded paddy soils is influenced by speciation, Eh, pH, and iron
  dissolution. Chemosphere 2011; 83: 925-932.
- Ying SC, Kocar BD, Fendorf S. Oxidation and competitive retention of arsenic
  between iron- and manganese oxides. Geochimica Et Cosmochimica Acta 2012;
  96: 294-303.
- Yu P, Zhang X, Wang D, Wang L, Ma Y. Shape-Controlled Synthesis of 3D
  Hierarchical MnO2 Nanostructures for Electrochemical Supercapacitors. Crystal
  Growth & Design 2013; 9: 528-533.
- Z Y, W Q, F W, M L, D W, Z S. Effects of manganese oxide-modified biochar
  composites on arsenic speciation and accumulation in an indica rice (Oryza sativa L.) cultivar. Chemosphere 2017; 168: 341-349.
- Zhang RH, Li ZG, Liu XD, Wang BC, Zhou GL, Huang XX, et al. Immobilization
  and bioavailability of heavy metals in greenhouse soils amended with rice
  straw-derived biochar. Ecological Engineering 2017; 98: 183-188.
- Zhang WX. Nanoscale Iron Particles for Environmental Remediation: An Overview.
  Journal of Nanoparticle Research 2003; 5: 323-332.

- Zhao FJ, Ma Y, Zhu YG, Tang Z, Mcgrath SP. Soil contamination in China: current
  status and mitigation strategies. Environmental Science & Technology 2015; 49:
  750.
- Zheng MZ, Cai C, Hu Y, Sun GX, Williams PN, Cui HJ, et al. Spatial distribution of
  arsenic and temporal variation of its concentration in rice. New Phytologist 2011;
  189: 200.
- 835