



HAL
open science

Carbon based nanomaterial interactions with metals and metalloids in terrestrial environment: A review

Mathieu Leroy, Emmanuel Flahaut, Camille Larue

► **To cite this version:**

Mathieu Leroy, Emmanuel Flahaut, Camille Larue. Carbon based nanomaterial interactions with metals and metalloids in terrestrial environment: A review. Carbon, 2023, 206, pp.325-339. <10.1016/j.carbon.2023.02.047>. <hal-04006124>

HAL Id: hal-04006124

<https://hal.science/hal-04006124v1>

Submitted on 18 Oct 2023

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire HAL, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



HAL Authorization

152 **Carbon based nanomaterial interactions with metals and metalloids in terrestrial**
153 **environment: a review**

154
155 **Mathieu Leroy^{1,2}, Emmanuel Flahaut^{2*}, Camille Larue^{1*}**

156
157 ¹Laboratoire écologie fonctionnelle et environnement, Université de Toulouse, CNRS,
158 Toulouse, France, ²CIRIMAT, Université de Toulouse, CNRS, INPT, UPS, UMR CNRS-
159 UPS-INP N°5085, Université Toulouse 3 Paul Sabatier, Bât. CIRIMAT, 118, route de
160 Narbonne, 31062 Toulouse cedex 9, France

161
162 *Corresponding authors: emmanuel.flahaut@univ-tlse3.fr; +00-33-(0)5-61-55-69-70;
163 camille.larue@ensat.fr; +00-33-(0)5 34-32-37-55

164
165 **Abstract:** The use of carbon based nanomaterials (CNMs) is diversifying and is being
166 considered in so many sectors that their release into the environment is inevitable. Terrestrial
167 ecosystems and particularly agrosystems are considered a major sink for CNMs due to their
168 accumulation in sewage sludge widely used for crop production. These ecosystems already
169 accumulated other contaminants such as metals and metalloids which are more closely
170 monitored to date. Although reviews about these contaminants considered individually have
171 already been published, no review focused on their interactions and the impact of these
172 interactions on their respective transfer and impacts on dwelling organisms (bacteria,
173 earthworms and plants). Indeed, CNMs and metal(loid)s can interact together as metal(loid)s
174 are able to adsorb on the surface of CNMs under the influence of diverse parameters such as
175 pH, organic matter content or intrinsic CNM properties, among others. In ecotoxicity studies,
176 interactive mechanisms (synergy and antagonism) were most of the time highlighted even

177 though the underlying mechanisms are not yet fully understood. This work questions the
178 current paradigm in use for risk assessment related to the additivity of the risks in presence of
179 a cocktail of contaminants and *in fine* on the strategies to be applied in order to guarantee
180 food safety.

181

182 **Keywords:** adsorption, carbon nanotube, cocktail effect, organism, metal(loid), toxicity, soil

183

184 **Highlights:**

185 - Metal(loid) adsorption on CNM depends on pH, contact time, temperature, CNM properties.

186 - Most studies demonstrate a change in metal transfer in organisms due to CNM presence.

187 - Most studies showed synergic and/or antagonist effect on the toxicity of the cocktail.

188 - Additive effect model for risk assessment is not appropriate in many cases.

189 - More integrative markers should be considered to properly evaluate ecosystem health.

190

191 **1. Introduction**

192 Scientific and industrial interest in carbon-based nanomaterials (CNMs) continues to grow
193 because of their remarkable properties and the applications they make possible¹. A CNM is a
194 nanomaterial essentially composed of carbon with any external dimension in the nanoscale or
195 having internal structure or surface structure in the nanoscale (*i.e.* nanometric scale particles
196 aggregated together)¹. Among CNMs, most have a structure based on graphene, a sheet of
197 carbon atoms joined together in a hexagonal lattice, one atom thick, sp² bonded²⁻⁴. Over the
198 years, the number of derived materials using graphene as a precursor grew up⁵. Examples
199 include multilayer graphene, functionalized graphene materials such as graphene oxide (GO),
200 carbon nanotubes (CNTs), fullerene, and many metal-CNT composite

201 The potential applications of CNMs are such that models already predict the evolution of their
202 concentrations in natural ecosystems after release at the end of life of commercial goods in
203 which they are included, or expected during their life cycle. For example, Sun al.⁶ predicted
204 that agricultural soils treated with sewage sludge are the main sink for CNTs with an annual
205 increase of 74 ng.kg⁻¹⁶. Furthermore, the use of CNMs as nanofertilizers⁷, nanopesticides⁸ and
206 as decontamination agents⁹ is also increasingly being considered.

207 Indeed, CNMs are capable of adsorbing different compounds on their surface. Among these
208 many compounds are metal(loid)s, which are elements present in natural conditions in biotope
209 at concentrations below 100 mg.kg⁻¹ including copper (Cu), zinc (Zn), iron (Fe), manganese
210 (Mn), molybdenum (Mo), boron (B), cobalt (Co), nickel (Ni), lead (Pb), cadmium (Cd),
211 chromium (Cr), arsenic (As), and selenium (Se)¹⁰. Cu, Zn, Fe, Mn, Mo, and B are essential for
212 plant growth, and Cu, Zn, Fe, Mn, Mo, Co and Se are essential to the growth and health of
213 animals including human beings¹¹ while others are toxic (Ni, Pb, Cd, As). For example, it has
214 been largely reported that CNTs can adsorbed metal(loid)s in aqueous solution¹²⁻¹⁷ and this

215 ability raises questions about its influence on heavy metal transfer and subsequent impacts
216 along food webs.

217 Agrosystems seems to be particularly suitable for the coexistence of both CNMs and
218 metal(loid)s (among others) due to cultural practices. Indeed, the continuous spreading of
219 organic and inorganic fertilizers and the application of sludge from wastewater treatment
220 plants have progressively led to the accumulation of both CNMs and metal(loid)s in
221 agrosystems¹⁸⁻²⁰.

222 Toxicological risk assessment of contaminants is usually carried out using additive models
223 that do not take into account the interactions between contaminants and their underlying
224 toxicological effects. For several years, metal(loid)s are submitted to a close monitoring in
225 sludge and food to ensure safety for populations, but the emerging contaminants such as
226 CNMs are not yet regulated. Studies report that interactions (both synergy and/or antagonism)
227 between contaminants can modify their transfer and toxicity to different organisms²¹⁻³⁰
228 (Figure 1).

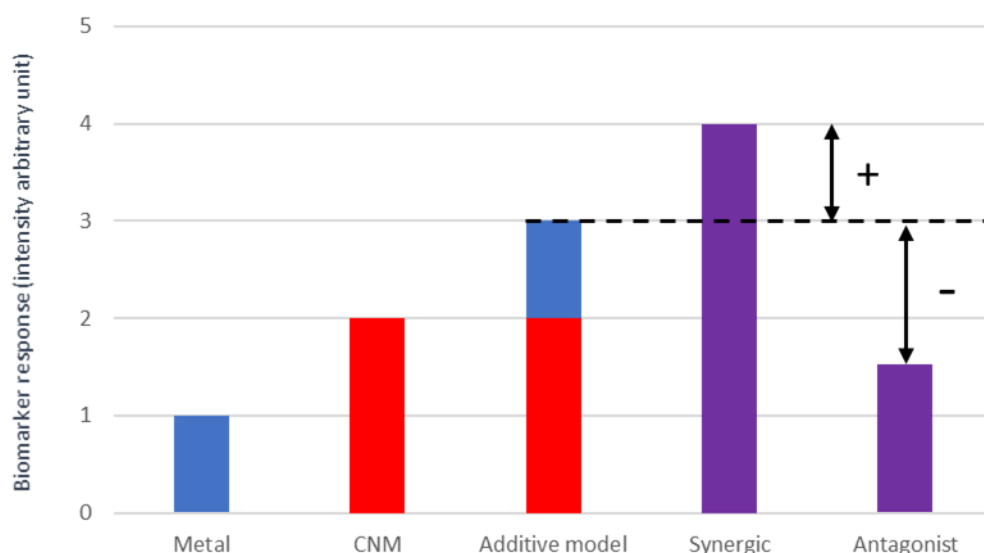


Figure 1 : Schematic differences on additive model compared to synergic and antagonist due to exposure to a cocktail of metal(loid) and CNM substances.

229

230 Tools have been developed to reveal possible interactions between stressors such as mixture
231 of substances³¹⁻³³. A meta-analysis by Vijver *et al.* aimed at predicting the response of
232 organisms exposed to mixtures of metals (Cd, Cu and Zn) and showed that the frequency of
233 occurrence of antagonism was 51%, and a synergistic response was detected in 28% of
234 cases³¹. Thus, in nearly 80% of the cases, interactive and non-additive patterns were
235 demonstrated. This finding may lead to a profound questioning of the current risk assessment
236 performed by the authorities.

237 The main goal of this work was to extensively review articles related to 1) CNM-metal(loid)
238 interactions in aqueous medium to better understand implied mechanisms and the influencing
239 parameters and 2) ecotoxicological studies related to simultaneous CNM-metal(loid)
240 contamination and their consequences on the transfer and impacts on agrosystem organisms to
241 reveal the extent of the interactive phenomenon.

242

243 **2. Interaction CNMs - metal(loid)s**

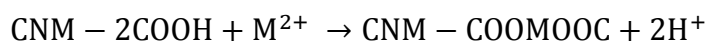
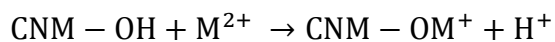
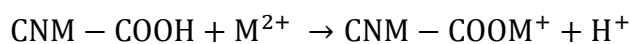
244 **2.1. Mechanisms of interactions between CNMs and metal(loid)s**

245

246 Several interaction mechanisms have been described between CNMs and metal(loid)s and
247 their predominance depends on the material considered. Interactions can be related to physical
248 adsorption, chemical interaction, electrostatic attraction and sorption-precipitation³⁴⁻³⁶.

249 For CNMs with surface functional groups, the chemical interactions between CNMs and
250 metal cations is the most observed mechanism¹⁶. Metal cations are adsorbed onto the surface
251 of CNMs via oxygenated functional groups (*e.g.*, -COOH, -OH, or C=O) and then release a
252 H⁺ ion from the surface of the CNMs (Figure 2, example for GO and CNT). The release of H⁺
253 causes a decrease in the pH of the medium. The adsorption mechanism of divalent metal ions

254 on CNMs is illustrated according to the following equations (where CNM stands for carbon-
 255 based nanomaterials, and M for metal(loid)):



256

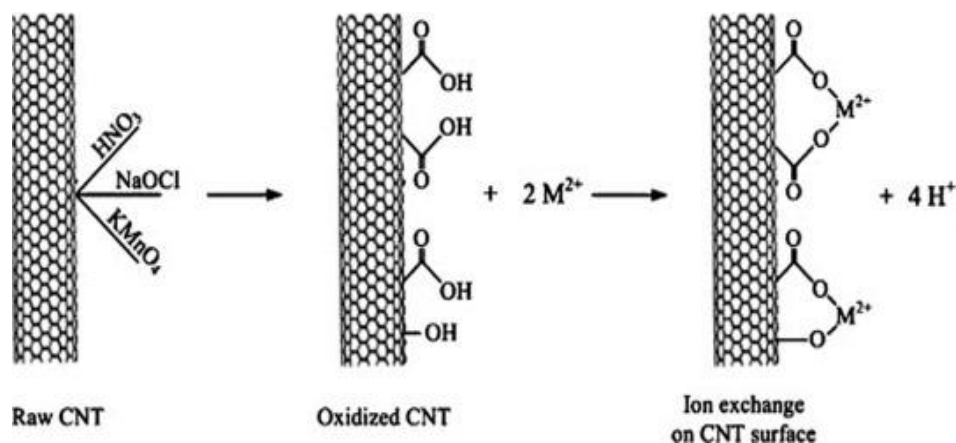


Figure 2 : Structure of carbon nanotubes (CNT) and their interactions with metal cations ($=\text{M}^{2+}$) (Figure adapted from Rao et al. (2007)³⁷)

257

258

259 Metal ion adsorption can also result from the combined effects of electrostatic attraction and
 260 sorption-precipitation processes^{38,39}. Liang *et al.*³⁹ observed that Cd^{2+} ions were adsorbed on
 261 the surface of modified Al_2O_3 -CNTs *via* physical interactions as electrostatic interactions and
 262 van der Waals interactions occurred between Cd^{2+} ions and carbon atoms in the CNTs³⁹. On
 263 another study, following an increase in the pH value of the medium, Zn^{2+} ions were adsorbed
 264 more on the surface of the CNTs due to the prevalence of carboxylate functions on their
 265 surface. The authors attributed this observation to the electrostatic interactions between the
 266 surface and divalent Zn^{2+} ions⁴⁰.

267 In addition, CNMs have also been explored for the removal of anionic pollutants, such as
 268 phosphates (PO_4^{3-}), perchlorates (ClO_4^-) and fluorides (F^-), from aqueous solutions⁴¹⁻⁴³. In
 269 contrast to the immobilization of cationic metal species, the mechanism of anion adsorption

270 was previously attributed to electronic interactions with π electrons of the carbon sp^2
271 network⁴³. This anion $-\pi$ association is based on the interaction between the negatively
272 charged anion with an electron-deficient aromatic ring on a graphene layer⁴³.

273 Finally, new adsorbents have also been prepared by modifying CNMs with organic molecules
274 that have a natural ability to complex metal ions. For instance, intrinsic functional groups on
275 GO present a large variety and thus the specificity of interaction with given species of interest
276 may be limited. To counteract this, the number of adsorption sites may be increased by
277 grafting compounds such as ethylenediaminetetraacetic acid (EDTA)⁴⁴ and chitosan⁴⁵.

278

279 **2.2. Factors influencing the CNM - metal(loid) interactions in simplified media**

280 **2.2.1. CNM intrinsic properties**

281 Due to the predominance of the adsorption mechanism by chemical interaction, it is obvious
282 that the oxidation state of CNMs influences the sorption of metal(loid)s on their surface. The
283 greater the oxidation of the CNMs, the more it provides oxygenated functional groups capable
284 of interacting with the surrounding metal ions, regardless of CNM shape and size^{9,46,47}.

285 Similarly, following the same idea, the degree of reduction of the CNMs acts on their
286 adsorption capacity. The maximum adsorption capacity of rGO (12.6% of O) was much lower
287 than that of GO (51.6% of O) due to the decrease in available oxygenated functional groups⁴⁸.

288 If the oxidation state can be tuned upon synthesis, it is very unlikely that it evolves in the
289 natural environment (with the exception of fullerenes). Their reduction may however be easier
290 to occur in environmental conditions. There are many examples of partial reduction of GO by
291 natural molecules extracted from plant leaves⁴⁹. However, enzymatic transformations of
292 CNMs are unfortunately often investigated using only pure enzymes and under laboratory

293 conditions and they are consequently not very representative of enzymatic transformations in
294 the natural environment⁵⁰

295 The specific surface area (SSA, m².g⁻¹) strongly influences the adsorption of metals. In the
296 case of graphene, most interaction sites with pollutants are present at the top and bottom
297 faces^{51,52}, even if sites located at the edge may also play some role. While all the surface of
298 monolayer graphene is available for the adsorption, multilayer graphene and CNTs have
299 internal layers respectively little or not accessible by adsorbates⁵¹. Similarly, it has been
300 observed that single walled CNTs are good adsorbents, which can be attributed to their high
301 SSA. Practically, the measured SSA of graphene-based CNMs is generally very disappointing
302 (often below a few tens of m².g⁻¹) when compared to the expected theoretical values (> 2600
303 m².g⁻¹ for graphene)⁵³ due to their significant agglomeration.

304

305 **2.2.2. Environmental parameters**

306 The relative concentrations of CNMs and metal ions in the medium is an important parameter
307 controlling adsorption phenomenon. To better understand these interactions, some theoretical
308 model isotherms were developed, the most common being the Langmuir and the Freundlich
309 models⁵⁴ according to the following equations:

$$\text{Langmuir} : q_e = k_L q_m C_e / (1 + k_L C_e)$$

$$\text{Freundlich} : q_e = k_F C_e^{1/n}$$

310 where q_e and q_m (mg.g⁻¹) represent equilibrium and maximum adsorption capacities,
311 respectively; C_e (mg.L⁻¹) is the equilibrium aqueous concentration; k_L (L.mg⁻¹) the
312 equilibrium constant and k_F and n the Freundlich constant.

313 These two models differ in particular by the fact that Langmuir model is considering the
314 adsorption of a monolayer of atoms on a homogeneous surface and is empirical while
315 Freundlich considers the adsorption of a multilayer of atoms on a heterogeneous surface and
316 is a theoretical model.

317 Adsorption kinetics are generally described by diffusion models such as the pseudo-first-order
318 and pseudo-second-order following the equations⁵⁴:

$$\textit{Pseudo first order} : \log(q_e - q_t) = \log q_e - k_1 t / 2.303$$

$$\textit{Pseudo second order} : q_t = k_2 q_e^2 t / (t k_2 q_e + 1)$$

319 where q_t and q_e are the adsorption capacities ($\text{mg}\cdot\text{g}^{-1}$) at time t and equilibrium, k_1 (min^{-1}) and
320 k_2 (min^{-1}) are the rate constants of the pseudo-first-order, pseudo-second-order, respectively.

321 Results extracted from these models show that the main environmental factors affecting the
322 adsorption of metal ions on the surface of CNMs include the initial metal(loid) ion and CNM
323 concentrations, pH, contact time, temperature, ionic strength and competition with other
324 molecules. Table S1 summarizes the data found in the literature regarding CNM interactions
325 with different metal(loid)s.

326 Regarding the intrinsic properties of the CNMs and the properties of the experimental
327 medium, which both influence the adsorption of metal(loid)s, a criticism can be raised related
328 to the lack of reported experimental details in the literature. Indeed, out of the 102 CNMs
329 tested, the SSA was reported for only 48 of them. Furthermore, as the Freundlich and
330 Langmuir models are thermodynamic processes, it is unfortunate that the study temperature
331 was provided only in 56 cases out of 102. This lack of reporting makes the intercomparability
332 of the studies impossible and thus weakens the conclusions and slows down the scientific
333 progress, especially for the safer-by-design and nanoecotoxicity fields.

334 The average adsorption values from the Langmuir model for CNTs, oxidized CNTs and
 335 graphene oxide have been extracted from related articles for different metal(loid)s (Cd, Cr,
 336 Cu, Hg, Ni, Pb and Zn; Table 1).

337

338 *Table 1 : Mean of maximum adsorption ($q_m = \text{mg.g}^{-1}$) of metal(loid)s (Cd, Cr, Cu, Hg, Ni, Pb and Zn) on the*
 339 *surface of carbon nanotubes (CNT), oxidized carbon nanotubes (CNT-oxidized) and graphene oxide (GO) in*
 340 *mono-elemental conditions extracted from the reported studies in Table S1 described with Langmuir model.*
 341 *Data were selected on restricted pH range ($4 < \text{pH} < 8$) in which adsorption is favored ($n_{\text{tot}}=117$).*

	Cd (n=20)	Cr (n=13)	Cu (n=17)	Hg (n=19)	Ni (n=9)	Pb (n=20)	Zn (n=19)
$q_{m_{\text{CNT}}}$	12.4 (± 9.9)	3.1 (n=1)	22.9 (± 2.6)	24.4 (± 38.9)	11.1 (± 3.8)	30.9 (± 2.6)	35.6 (± 9.4)
$q_{m_{\text{CNT-oxidized}}}$	35.9 (± 28.2)	21.3 (± 11.7)	37.4 (± 27.3)	27.3 (n = 1)	49.2 (n = 1)	75.7 (± 31.3)	24.4 (± 28.8)
$q_{m_{\text{GO}}}$	306.9 (± 315.7)	-	183.3 (± 156.6)	-	84.4 (± 19.7)	1280.7 (± 516.5)	345.0 (n=1)

342

343 The adsorption of each metal(loid) appears to be always higher for GO than for CNTs and
 344 oxidized CNTs. Similarly, the oxidation of CNTs promotes the adsorption of metal(loid)s
 345 since the average adsorption of metal(loid)s on the surface of oxidized CNTs is higher for
 346 each metal (except Zn, probably related to the high variability for Zn adsorption onto oxidized
 347 CNTs) than on the surface of raw CNTs. On the basis of the data in Table 1, the sequence of
 348 adsorption affinities in mono-elemental conditions can be drawn up:

349 - for CNTs: $\text{Zn} \geq \text{Pb} > \text{Hg} \geq \text{Cu} > \text{Cd} \geq \text{Ni} > \text{Cr}$

350 - for CNTs oxidized: $\text{Pb} > \text{Ni} > \text{Cu} \geq \text{Cd} > \text{Hg} \geq \text{Cr} \geq \text{Zn}$.

351 It is also important to note that these adsorption experiments were carried out in mono-
 352 elemental conditions (one adsorbent + one ionic species) thus not considering potential ionic
 353 repulsions possibly induced if several ionic species would be present in the solution.

354

355 2.2.2.1. pH

356 The pH is a parameter that greatly influences the interactions between CNMs and metal
357 cations because it determines not only metal speciation but also surface charge of CNMs and
358 complexation behavior of functional groups^{55,56}. Most of the studies in Table S1 evidenced
359 that the adsorption capacity of CNMs was affected by variations in the pH of the
360 solution^{34,37,57,58}. Adsorption of the metal(loid) is partly related to the point of zero charge
361 (pH_{PZC}) of a given CNM; when $\text{pH} > \text{pH}_{\text{PZC}}$, cation adsorption is higher due to the
362 electrostatic interactions between the cations and the negative surface charge of CNM. On the
363 other hand, the adsorption of cations is lower when $\text{pH} \leq \text{pH}_{\text{PZC}}$ due to the neutralization of
364 the surface charge of CNM, or even the presence of repulsive positive charges. A change in
365 pH also affects competing complexing reactions and metal ion species³⁶. Generally, the state
366 of the divalent metal ions in solution depends on the pH. The most common species would be
367 present in the form of M^{2+} , $\text{M}(\text{OH})^+$, $\text{M}(\text{OH})_2$ et $\text{M}(\text{OH})_3^-$ depending on the pH of the
368 medium due to hydrolysis^{16,34,36}. It is important to note that precipitation of metal cations
369 may occur when the pH increases, which also affects the removal of metal ions⁵⁹.

370

371 2.2.2.2. CNM concentration

372 Increasing the concentration of CNM up to a certain point, increases the overall surface
373 available for reactions and thus increases the sorption of metal(loid)s. For instance, adsorption
374 of Ni^{2+} , Cd^{2+} , Cu^{2+} and Pb^{2+} increased with increasing CNT concentration in different
375 experiments^{35,37,38,60}. Similar results were also observed for GO^{61} . However, beyond a
376 threshold mass (specific to the CNM studied), the adsorption capacity reaches saturation and
377 then decreases^{15,61,62}. For a exposition of 50 mg.L^{-1} of oxidized SWCNT, Moradi et al.¹⁵
378 observed a maximum adsorption capacity up to 55, 68 and 90 mg.g^{-1} for Cd, Cu and Pb

379 respectively, while this adsorption decreased at 45, 58 and 82 mg.g⁻¹ with a exposition to 100
380 mg.L⁻¹. Indeed, beyond the threshold, an increase in the concentration of adsorbent could
381 reduce the available sites because the effective surface area is likely to decrease due to homo-
382 agglomeration, entanglement and/or folding of the CNMs (depending on its morphology)^{15,63}.
383 In addition, metal ions can induce CNM agglomeration due to their interactions with
384 carboxylic groups⁶⁴.

385

386 **2.2.2.3. Ionic strength**

387 Increasing the ionic strength of the solution can be achieved by increasing the concentration
388 of an ion or in multi-elemental conditions by having several types of metal ions in the
389 medium. However, this last experimental set-up even though more relevant in environmental
390 context has been much less studied. The adsorption capacities of ions by CNMs decreased
391 with increasing ionic strength for two reasons : (i) the formation of double-layer electrical
392 complexes between cations and CNMs and (ii) the influence of the ionic strength on
393 the activity coefficients of ions, which limit their transfer to the CNM surfaces³⁸. In multi-
394 elemental conditions, the increase in number of ions present in the media decreases the part of
395 metal ions adsorbed onto CNMs. Affinity sequences of metal ions for adsorption sites could
396 be deduced from these multi-elemental adsorption experiments (Table 2). For instance, Salam
397 *et al.*⁶⁵ showed that ion binding affinity followed the order of Cu(II) > Zn(II) > Pb(II) > Cd(II)
398 for MWCNTs (initial metal concentrations = 1 mg.L⁻¹, pH = 7, contact time = 2h). For GO,
399 Sitko *et al.*⁶⁶ observed another ranking of affinity: Pb(II) > Cu(II) > Cd(II) > Zn(II) (initial
400 concentration = 1 mg.L⁻¹, GO = 0.1 g.L⁻¹, pH = 5, contact time = 2h). Metal precipitation
401 related to the pH of the solution could explain differences observed on affinity sequences⁶⁷.
402 Furthermore, the addition of metals in the medium could increase the co-precipitation⁶⁸ and

403 could modified the fitting of experimental adsorption measured with Langmuir and/or
404 Freundlich modelling. To date, very little studies dealing with competitive adsorptions are
405 available and more work needs to be carried out to understand the underlying mechanisms
406 responsible for the establishment of these adsorption affinity sequences.

Table 2: Maximum adsorption capacity of different CNMs established by adsorption models for various metal(loid)s in multi-elemental conditions extracted from the cited articles (SSA = specific area ($m^2.g^{-1}$), q_m = maximum modelled adsorption ($mg.g^{-1}$), R^2 = correlation coefficient, C = metal concentration range ($mg.L^{-1}$), A = Adsorbent dosage ($mg.L^{-1}$), T = temperature ($^{\circ}C$), Ct = Contact time (hours), RPM = agitation (Rotation Per Minutes))

M ⁿ⁺	Adsorbent	SSA	Isotherm model		C	A	Seq affinity	Parameters				References
			Langmuir					T	Ct	pH	RPM	
			qm	R2								
Cd(II), Cu(II), Pb(II), Zn(II)	MWCNT	61.5			1	2500 to 15 000	Cu>Zn>Pb>Cd		2	7	400	Salam <i>et al.</i> ⁶⁵
Cu(II), Pb(II), phenol	N ₂ H ₄ -SH-Fe ₃ O ₄ /o- MWCNT		Pb: 40, Zn: 21, Phenol: 56	Pb: 0.91, Zn: 0.96, Phenol: 0.9	20 to 60	200	Pb>Cu	25	12	6		Jiang <i>et al.</i> ⁶⁹
Pb(II), Cu(II), Ni(II)	carbonaceous nanofibers	150.3	Pb: 470.3, Cu: 98.5, Ni: 64.6		0.1 to 0.6 mmol.L ⁻¹ (Pb: 20.7 to 124.3 ; Cu: 6.5 to 39.3 ; Ni: 5.9 to 35 mg.L ⁻¹)	200	Pb>Cu>Ni	25		5.5		Ding <i>et al.</i> ⁷⁰
Cd(II), Hg(II), As(II)	GO		Cd: 66.8, Hg: 55.4, As: 23.7			333.3	Cd>Hg>As					Alimohammady <i>et al.</i> ⁷¹
Cd(II), Hg(II), As(II)	GO-3- aminopyrazole		Cd: 85.3, Hg: 74.6, As: 38.3		20	333.3	Cd>Hg>As		2	8	250	
Cd(II), Cu(II), Pb(II), Zn(II)	GO				5 μ mol.g ⁻¹ of GO	100	Pb>Cu>Cd>Zn					Sitko <i>et al.</i> ⁶⁶
Cd(II), Cu(II), Ni(II)	GO membrane				0.1 to 1 mmol.L ⁻¹ (Cd: 11 to 112 ; Cu: 6 to 64 ; Ni: 6 to 59 mg.L ⁻¹)	200	Cu>Cd>Ni	30	12	5.5	150	Tan <i>et al.</i> ⁷²

Cd(II), Cu(II), Pb(II)	O-MWCNT		Cd: 3.3, Cu: 17.0, Pb: 34.0	Cd: 0.21, Cu: 0.97, Pb: 0.97	5 to 30	1000	Pb>Cu>Cd			5		Li <i>et al.</i> ³⁸
Ni(II), Zn(II)	SWCNT	700	Ni: 28.4, Zn: 29.5	Ni: 0.98, Zn : 0.98	10 to 80	500	Ni=Zn		12		180	Lu <i>et al.</i> ⁷³
Ni(II) and Zn(II)	MWCNT	459	Ni: 18.6, Zn: 19.4	Ni: 0.98, Zn: 0.98	10 to 80	500	Ni=Zn		12		180	

210 **2.2.2.4. Contact time**

211 As the contact time increases, the percentage of metal removal increases until an equilibrium
212 is established^{16,36}. The equilibrium depends on all the experimental parameters of the medium,
213 including the initial concentration of metal ions. However, the two most important parameters
214 on the adsorption kinetics of metals is the number of adsorption sites available for a given
215 metal concentration and contact time^{16,36}.

216

217 **2.2.2.5. Temperature**

218 The most common observation is that sorption capacity increases with rising temperature^{16,36}.
219 For example, the sorption of Pb^{2+} , Cu^{2+} and Zn^{2+} on CNTs is an endothermic process and
220 increases with increasing temperature^{14,74}. Similar results were observed for the same metals
221 for GO⁶⁶. Authors explained that the increase in temperature could lead to an increase in the
222 rate of diffusion of ions through the outer boundary layer and into the pores of CNMs due to a
223 temperature-induced decrease in the viscosity of the solution^{36,66}.

224

225 **2.3. Metal(loid) adsorption by CNMs in complex media**

226 Due to the increasing use of CNMs, their potential in the agricultural and environmental fields
227 and their interactions with metal(loid)s in simplified environments (aqueous solution), it is
228 important to investigate how interactions investigated in simplified medium can be transposed
229 in more complex environments. Various studies have thus recently been carried out (i) to
230 characterize interactions of CNMs with compounds that are widely found in ecosystems, such
231 as organic matter (OM) and sedimentary matrices and (ii) to investigate the role of some key
232 parameters.

233

234 **2.3.1. Organic Matter**

235 Humic substances generally represent a predominant part of the natural OM distributed in
236 soils, sediments and water⁷⁵. It has been reported that 3 to 28% of the OM dissolved in
237 wastewater treatment plant effluents are humic substances⁷⁶. They are hydrophobic organic
238 acids mainly derived from soil humus and plants. These humic substances are often
239 encountered in agrosystems especially due to the use of wastewater treatment plant effluents
240 for fertilizing agricultural soils. Humic acid (HA) and fulvic acid (FA) are the two most
241 important components of humic substances. It is expected that HA/FA will have a significant
242 influence on the sorption of metal ions by CNMs because the hydrophobic parts as well as the
243 aromatic monomers of HA/FA have a strong affinity for CNMs via hydrophobic and $\pi - \pi$
244 interactions. At the same time, the hydrophilic fractions of HA/FA contain various functional
245 groups (carboxyl, phenol, hydroxyl, amine and quinine) which can bind to metal(loid)s⁷⁷. For
246 instance, Lin *et al.*⁷⁷ observed that HA bound to the surface of MWCNTs significantly
247 favoured Pb(II) sorption (pH = 5.0) (Figure 3A). Similar results were also observed for the
248 sorption of Cd(II) (pH = 6.0) on HA-coated MWCNTs⁷⁸. Additionally, OM can also increase
249 the stability of CNMs in suspension as the OM polymeric chains adsorbed to their surface
250 tend to avoid each other in solution so as not to constrain the conformational space available
251 to each of them inducing repulsive forces and thus decreasing the chances of homo-
252 agglomeration^{79,80} (and in turn increasing the potential available sites for metal(loid)
253 adsorption) (Figure 3B and 3C). These interactions also depend on both pH and OM
254 composition. Indeed, Sheng *et al.* reported an increase in the adsorption of Cu(II) on
255 MWCNTs due to dissolved HA/FA at pH < 7.5 (up to 90%), while a decrease was found at
256 pH > 7.5 (to 80%)⁸¹. In parallel, Sun *et al.*⁸² found that there was a higher binding affinity
257 between Cu(II) and FA compared to HA, due to more oxygenated functional groups present

258 in FA that could also complex with Cu(II) (pH = 6). Thus, it seems that there are different
259 OM-metal(loid)- CNM interaction processes.

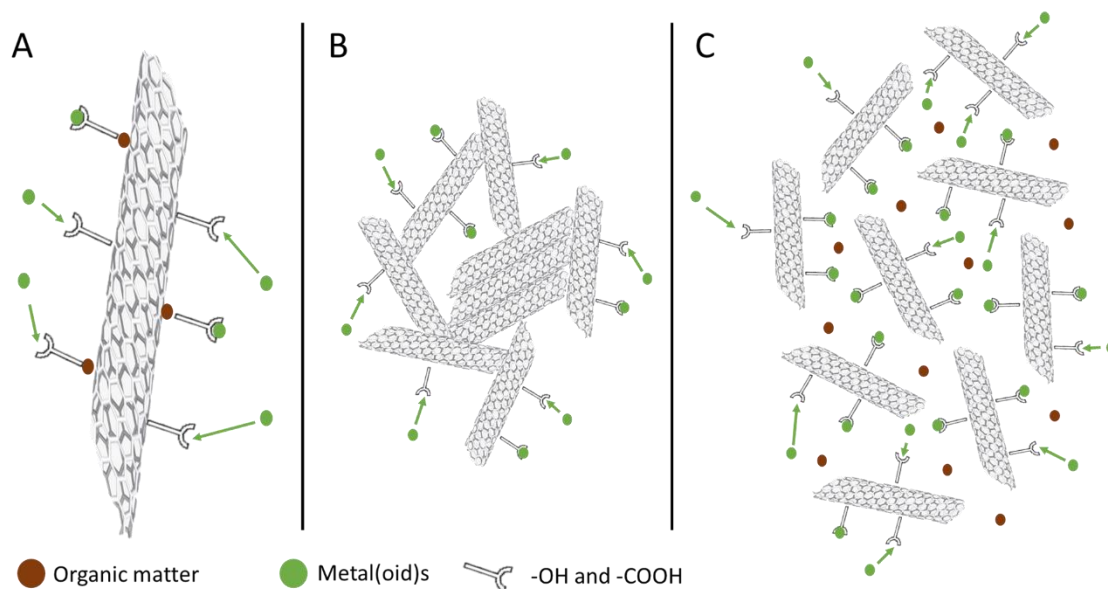


Figure 3 : Effects of organic matter (A) bound on CNTs which increase metal(loid) adsorption onto CNTs and (B-C) on the dispersion of CNTs in media (B : without OM, C : with OM)

260

261 2.3.2. Soil

262 First of all, it should be reminded that due to the complexity of both soil and sediment, very
263 few studies have been conducted to understand (i) what are the properties of these matrices
264 that influence the mobility and/or agglomeration of CNMs and (ii) the related mechanisms.

265 However, as in simplified conditions, CNMs influence the fate of metal(loid)s in soil. For
266 instance, a study revealed that Pb and Cu immobilization in soil increased with increasing
267 MWCNT concentration correlated with the increased adsorption sites on the surface of CNTs
268 available for Pb and Cu ion adsorption⁸³.

269 These interactions between CNMs and metal(loid)s in soil vary according to CNM intrinsic
270 properties such as their morphology (diameter, length) and their surface chemistry (surface

271 charge and functionalization)⁸⁴⁻⁸⁶. While it can be expected that the smaller the CNM, the
272 higher their mobility in soils, it remains difficult to support this assumption from available
273 data in the literature.

274 In addition, properties such as soil texture, structure, porosity, permeability, or type of clay
275 contribute to the actual flow of CNMs through the soil⁸⁴. It appears that MWCNTs^{87,88} and
276 GO⁸⁹ generally tend to have higher retention (low transport) in clay than in sandy soils. For
277 example, the retention profile indicated that retention was higher in soil with smaller size sand
278 particles (240 μm) than soil with higher size sand particles (607 μm)⁹⁰. Lu *et al.* showed that
279 the type of clay influenced CNM transport because the addition of 5% of kaolinite inhibited
280 more graphene transport in soil column than montmorillonite and illite which presented the
281 same particle size (Kaolinite: 28.0 μm , montmorillonite: 20.0 μm and illite: 23.9 μm) due to
282 greater amounts of positively charged edge sites. Furthermore, the ionic strength of the soil
283 solution is also a key parameter to consider. Indeed, Qi *et al.* demonstrated that the addition of
284 0.5 mM of Ca^{2+} significantly inhibited GO transport in clay soil in contrast to sandy soil by
285 complexing with the surface oxygen of GO and soil components leading to enhance GO
286 agglomeration and link GO with soil grains⁹¹.

287 Several studies using Toxicity Characteristic Leaching Procedures (TCLP) determined the
288 leachability of metal(loid)s in CNT-amended soils. For instance, Yang *et al.*⁹² observed that
289 the bioaccessible Sb content in a loamy soil was significantly reduced (20.7%) with the
290 addition of 0.9 wt.% MWCNTs compared to control (without CNT addition). In sandy soil,
291 Vithanage *et al.*⁹³ did not observe any significant difference in extractable Sb at the tested
292 MWCNT concentrations (0.5, 1 and 2.5 wt.%). The immobilisation and, consequently, the
293 bioavailability of Sb differed between the two studies, which could be related to different
294 texture and other related parameters (cation exchange capacity, structure, water
295 holding capacity).

296 As in simplified medium, the interaction of CNMs and metal(loid)s is metal dependent.
297 Indeed, a study using 0.2, 1 and 5 wt.% GO in a historically polluted soil showed that the
298 availability of Pb and Cu followed a dose-dependent relationship so that at the highest
299 concentration of GO, their availabilities relative to the controls were found to be reduced by
300 2.5% and 5.6% respectively. This decrease was much smaller for Zn and Cd (*ca.* 1%) and As
301 availability was increased by 191%⁹⁴. Regarding the kinetics, it seems that reaching the
302 equilibrium takes much longer in more complex environments than in simplified medium. For
303 example, Song *et al.*⁹⁵ observed that in soil solution the adsorption of Cd²⁺ on MWCNTs
304 reached 65% after 7 days and up to 87% and 88% after 30 and 60 days, respectively. To
305 compare with aqueous media, the time to reach equilibrium does not exceed 12 hours (Table
306 S1).

307 Overall, more studies involving solid matrices under environmentally relevant conditions
308 need to be carried out as the risk of CNM release into the environment is increasing. It is
309 important to understand all the physico-chemical characteristics of soils affecting the
310 interactions among contaminants in order to better predict their transfer and toxicity in the
311 environment and along trophic chains.

312

313 **3. Metal(loid)s, CNM and CNM-metal(loid) complex uptake and impacts on agrosystem** 314 **organisms**

315 **3.1. Uptake and impacts of metal(loid)s and CNMs as single contaminant**

316 The toxicological impacts of metal(loid)s and CNMs and their potential consequences on
317 agrosystems have been widely studied. Thus, the scientific corpus available is abundant and
318 compiled in several scientific articles for microorganisms⁹⁶⁻¹⁰¹, invertebrates^{96,102-106} and crop
319 plants^{19,96,101,102,107-110} which are the major organisms of agrosystems due to their functions

320 and interactions. This corpus is regularly updated according to the technical advances,
321 offering a step further on the scientific knowledge in these fields of research especially for
322 metal(loid)s due to older knowledge.

323 Microorganisms are essential for many agrosystem services like mineralization, C, N and P
324 biogeochemical cycles and because of their interactions with plants like mycorrhizal
325 association¹¹¹. Microorganisms are capable of accumulating metal(loid)s, this accumulation is
326 generally governed by the sorptivity of the cell envelope and the ability to absorb metal(loid)s
327 into the cytosol¹¹². The metal membrane transporters are not highly selective and thus
328 metal(loid)s can be taken up through this pathway^{112,113} leading to alterations in the
329 microorganisms at different scales. Metal(loid) toxicity could occur through displacement of
330 essential metals from their original binding sites or through ligand interactions and
331 conformational structure alterations with oxidative phosphorylation and osmotic balance¹¹⁴.
332 Damage to the DNA structure, the cell membrane and the activity of some enzymes was also
333 reported^{97,98,115-117}. Thus, metal(loid)s can affect the microbial population by decreasing their
334 growth/proliferation, and altering their morphology and biochemical activities^{100,118}. Due to
335 these alterations, the functions performed by microorganisms such as the decomposition of
336 organic matter^{97,98}, C sequestration or N fixation could be negatively impacted^{98,99,119}.
337 According to a study that compiled the lowest metal(loid) amount added that resulted in a
338 decrease in the rate of basal respiration (LOAED), the mean LOAED for Hg, C, Cu, Pb, Ni,
339 Cd, Zn was 257, 411, 500, 506, 576, 594 and 697 mg.kg⁻¹, respectively (Table S2)⁹⁸.

340 In parallel, for CNMs, transmission electron microscopy observations have evidenced that
341 they may interact strongly with bacterial cell membranes. These interactions may disrupt the
342 integrity of the membrane structure by oxidative stress and/or physical damage, allowing
343 CNMs to be internalized in bacteria¹²⁰⁻¹²². In addition, a majority of studies has observed a
344 decrease in soil microbial activity following exposure to CNMs¹²²⁻¹²⁷. CNMs would present

345 an antimicrobial potential since a whole set of studies reported a loss of viability following
346 exposure^{123,124,128–130}. Changes in microbial community structure have also been
347 reported^{123,126,131,132}. Finally, microbial functions such as mineralization, N and CO₂ fixation
348 could be decreased¹³³. The effect of CNMs on microbial activity¹²² and biomass appeared at
349 85 mg.L⁻¹ and 1-5 mg.g⁻¹, respectively^{124,126,127}.

350 Oligochaetes are considered as soil engineers because of their involvement in physical and
351 chemical processes such as litter mixing, soil porosity, active soil water transport, OM
352 mineralization and nutrient mobilization and cycles^{134–136}. These organisms present two
353 pathways of contamination: the oral pathway by ingestion of soil and the tegumentary
354 pathway by contact with soil. After absorption, metal(loid)s can accumulate in the posterior
355 alimentary canal¹³⁷. Metal(loid)s may decrease the growth of earthworms through delayed
356 biomass gain^{138–141} and act on energy reserves by decreasing the production of lipid
357 reserves^{142,143} and modifying the gut microbiota¹⁴⁴. In addition, morphological and
358 behavioural changes have been demonstrated following exposure to metal(loid)s¹⁴⁵. It was
359 observed that metal(loid)s can interfere with earthworm reproduction by reducing the number
360 of juveniles produced^{138,139,146} induced by a reduction of number of sperm produced¹⁴⁷, the
361 production of cocoons^{138,141,146,148–150} and the success of cocoon hatching^{139,151,152}. Median
362 lethal concentrations (LC₅₀) of metal(loid) after a 17 day exposure were on average 97, 663,
363 756, 791, 836, 1415, 7542 mg.kg⁻¹ for As, Cu, Ni, Cd, Zn, Cr(III) and Pb, respectively¹⁰³.

364 For CNM exposure, it has been demonstrated that earthworms are able to eliminate orally
365 ingested CNMs by depuration thus limiting the toxicity of these contaminants^{153–155}. As a
366 result, many studies have not observed any apparent effects on earthworms following CNM
367 exposure^{154,156,157}. However, few works reported DNA damages^{158,159} and a decreased cocoon
368 production¹⁶⁰ (up to -60% and -78% at 495 and 1000 mg DWCNT.kg⁻¹ food). According to
369 Scott-Fordsmand *et al.*¹⁶⁰ and Hu *et al.*¹⁵⁸, the median effective concentration (EC₅₀) for

370 earthworms exposed to DWCNT and MWCNT would be >500 and $1480.3 \text{ mg.kg}^{-1}$ for
371 growth and mortality, respectively.

372 Plants can take up metal(loid)s from the soil through their roots, transport them through the
373 xylem and accumulate them in the different tissues of all organs (roots, stem, leaves, grains
374 and fruits) by symplastic and apoplastic pathways¹⁶¹. Generally, the roots tend to accumulate
375 metal(loid)s such as Al, Cd, Cr, Cu, Hg, Ni, P and Zn, vegetative parts have a lower
376 accumulation level and the reproductive parts tend to exclude them^{162,161}. During their
377 transport in the plant, metal(loid)s are largely bound to the cell walls, which explains why
378 most of the metal(loid)s are bound in the roots (about 75-90%) and a smaller fraction in the
379 shoots¹⁶¹. However, some particularities exist and accumulation is a function of the stage of
380 development of the plant and the metal(loid) considered¹⁶¹. The stress imposed by meta(loid)l
381 accumulation can induce reactive oxygen species (ROS), promote DNA damage, impair
382 functional membrane integrity, nutrient homeostasis and disrupt protein and enzyme function
383 and activity¹⁶³⁻¹⁶⁵. These toxic effects lead to a slowing down of cellular parameters, *i.e.* a
384 reduction of photosynthetic activity, lipid production^{107,166}, plant growth parameters, and
385 consequently of the yield^{107,166-172}. The phytotoxicity of the metal(loid)s under their ionic
386 form follows the trend (from most to least toxic): $\text{Pb} \approx \text{Hg} > \text{Cu} > \text{Cd} \approx \text{As} > \text{Co} \approx \text{Ni} > \text{Zn} > \text{Mn}$,
387 with median toxic concentrations of : 0.06, 0.09, 0.13, 0.56, 0.67, 1.00, 1.12, 1.63 and 2.53
388 mg.L^{-1} (hydroponic exposure)¹⁷³.

389 Although the entry mechanisms of CNMs are not well known, it is established that they can
390 interact with plants at the root system level. Once absorbed, they can share the vascular
391 system with water and nutrients and can be transported by transpiration in the upper part of
392 the plants⁹⁶. Several studies have observed CNM accumulations in all parts of the
393 plant^{110,174,175}. CNMs can act on plants starting from the seed germination process with a trend
394 to activate and increase the seed germination rate¹⁷⁶⁻¹⁷⁹. The scientific corpus of research on

395 the effects of CNMs reports contrasting effects on plant growth and physiological status. It
396 reports an increase, a decrease or even a neutral effect on growth parameters (root system,
397 biomass production, flowers and fruits)^{110,174}. In case of toxicity, it was associated with
398 generation of ROS, antioxidant activity, necrotic lesions of cells and leaf tissues as well as
399 root and leaf morphological changes, decreased chlorophyll, and programmed cell death^{180–}
400 ¹⁸³. The inherent properties of CNMs (functionalization, length, diameter) and experimental
401 designs (exposure medium, duration, plant species) could be a reason for the observed
402 variability in CNM toxicity. The mean “lowest observed effect level” (LOEC) for plants
403 exposed to CNTs was 389 mg.L⁻¹ according to Jackson *et al.*¹²⁰.

404

405 Regardless to LOAED, NOEC, LOEC and EC₅₀, it seems that metal(loid) concentrations
406 needed to induce toxicity were much lower than those required for CNMs.

407 Table 3: Effects of simultaneous CNM-metal(loid) exposure on transfer and associated toxicity for three representative agrosystem organisms (bacteria, earthworms and
 408 plants). The light blue lines represent studies with hydroponic exposure and the dark blue lines represent studies with soil exposure (SOD = superoxide dismutase, POD =
 409 peroxidase, MDA = malondialdehyde, CAT = catalase, GR = glutathione reductase, GP = glutathione peroxidase, Chlo = chlorophyll, n.p = data not provided).

Type and CNM concentration	CNM size	Type and metal(loid) concentration	Organisms	Exposure medium	Exposure duration	Transfer effects	Interactive toxicological effects	References
GO (0, 1 and 10 mg.kg ⁻¹)	n.p	Cd (0 and 1 mg.L ⁻¹)	Plant: <i>Oryza sativa</i>	Culture solution	30 days	Increase of Cd transfer: +15% and +32% of Cd in roots and shoots with addition of 10 mg.L ⁻¹ GO compared to control (without GO with Cd)	No interactive effects on germination and dry biomass of roots and shoots	Li <i>et al.</i> ²³
GO (0.1, 1, 10 mg.L ⁻¹)	n.p	As (0 and 10 mg.L ⁻¹)	Plant: <i>Triticum aestivum</i>	Water	n.p	Compared with the 19.9% rate of As uptake recorded without GO exposure, the uptake rate of As increased to 24.6% and 32.1%, then decreased to 16.1% in the presence of GO concentrations of 0.1, 1 and 10 mg.L ⁻¹ , respectively	Synergic effects in the presence of both contaminants: reduction of root number and fresh biomass, reduction of root length, increase of antioxidant enzymes activity (SOD and POD), decrease of photosynthetic pigments (chlo a/b)	Hu <i>et al.</i> ²⁴
GO (0, 1, 2.5, 5, 10, 20, and 40 mg L ⁻¹)	n.p	Cd (0, 1, and 5 mg.L ⁻¹)	Plant: <i>Triticum aestivum</i>	Water	7 days	Increase of Cd transfer in roots and leaves at every GO and Cd concentrations tested.	Synergic effects: decrease in dry mass of roots and shoots, decrease in chlorophyll concentration and rubisco, increase in H ₂ O ₂ and O ₂ concentration in roots and leaves, increase in antioxidant enzymes activity (MDA, POD and SOD)	Gao <i>et al.</i> ²⁵

GO (0, 30, 60 mg.L ⁻¹)	n.p	Cd (0 and 2 mg.L ⁻¹)	Plant: <i>Lactuca sativa</i>	Culture solution. Addition of GO on foliar way.	28 days	n.p	Antagnist effects: increase in dry mass, photosynthetic parameters (quantum yield, chlorophyll a and total chlorophyll content and electron transfer rate), decrease in O ₂ concentration in leaves and roots, activity of antioxidant enzymes (SOD, CAT, GR, GP) and glutathione concentration	Gao <i>et al.</i> ¹⁸⁴
GO (0, 30, 60 mg.L ⁻¹)	n.p	Cd (0 and 2 mg.L ⁻¹)	Plant: <i>Lactuca sativa</i>	Culture solution. Addition of GO on foliar way.	28 days	Decrease of Cd transfer: - 19 and -9% of Cd in roots with the addition of 30 and 60 mg.L ⁻¹ of GO respectively, - 33% of Cd in leaves with 30 mg.L ⁻¹ of GO	Antagnist effect on the number and diameter of roots and the concentration of vitamin C	Gao <i>et al.</i> ²⁶
GO (1.0, 2.5, 5.0, 10, 20, and 40 mg.L ⁻¹)	n.p	Cd solution (1.0 and 5.0 mg.L ⁻¹)	Plant : wheat seedlings	Culture solution	n.p	n.p	Decrease in roots growth and microtubule protein. Increase on cytochrome P450	Gao <i>et al.</i> ¹⁸⁵
MWCNT (0, 100, 500, 1000 mg.kg ⁻¹)	Length: 3–12 μm, diameter: 12–15 nm, SSA : 245,28 m ² .g ⁻¹	Cd and As (Cd: 1.41 mg kg ⁻¹ , As: 54.80 mg.kg ⁻¹)	Plant : <i>Solanum nigrum</i> L.	Natural soil	60 days	No effect on Cd transfer with the addition of MWCNT. Decrease on As in stem (-25%) at 100 mg.kg-1, in leaf (-31%) at 1000 mg.kg-1	Increase in antioxidant enzymes activities (MDA, PRO and APX)	Chen <i>et al.</i> ¹⁸⁶

FNCB (10 mg.L ⁻¹)	Diameter : 47.35 ± 1.67 nm, SSA: 635.96 m ² .g ⁻¹	Cd (20 and 60 mg.L ⁻¹)	Worms (<i>Eisenia fetida</i>) cells (coelomocytes)	Culture plate	12H	Decrease on Cd transfer (-31%) in supernatants at 60 mg.L ⁻¹ of FNCB. Increase on Cd transfer (+37%) in free Cd content inside cells	No effect on viability of coelomocytes	Xu et al. ¹⁸⁷
GO (1000 and 2000 mg.kg ⁻¹)	n.p	Cd (0.1 mg.kg ⁻¹)	Bacteria	Soil	3-15-60 days	n.p	No Interactive effect but the addition of GO changes in the composition and diversity of the bacterial community	Xiong et al. ¹⁸⁸
MWCNT (0, 10, 50, 100 mg.kg ⁻¹)	Length = 5–30 μm, diameter < 8 nm	Cd (0, 2.0 and 10 mg.kg ⁻¹)	Earthworms: <i>Eisenia fetida</i>	Soil	14 days	Decrease in Cd concentration: at 2 mg.L ⁻¹ of Cd -24%, -18%, and -12% with addition of 10, 50 and 100 mg.kg ⁻¹ of MWCNT. Decrease more pronounced at 10 mg.L ⁻¹ of Cd (-42%, -34%, -26% of Cd with addition of 10, 50 and 100 mg.kg ⁻¹ of MWCNT).	There appear to be an interaction: decreased percentage of inhibition of worm growth, detoxifying enzymes, and metabolites for the lowest the MWCNT concentration.	Yang et al. ³²
GO spheres (5 mg.L ⁻¹)	n.p	Cu (0 and 240 mg.L ⁻¹)	Plant: <i>Triticum aestivum</i>	Soil	14 days	Reduction of transfer: decrease of Cu concentration in roots and stems by 60% and 67% and it was similar in leaves (increase only in 2%) with addition of GO compared to the control (without GO with Cu)	No interactive effects growth parameters (biomass, root, stem and leaf lengths). cellular: photosynthesis, transpiration and stomatal conductance	Zhao et al. ²¹
GO (1000 and 3000 mg.kg ⁻¹)	n.p	Cd (0 and 10 mg.kg ⁻¹)	Plant: <i>Oryza sativa</i>	Soil	40 days	Increase in Cd with 3000 mg.kg ⁻¹ addition of 12.5% in total rice biomass compared to control (no GO with Cd)	No interactive effects on the growth parameters studied (fresh biomass and size)	He et al. ²²
MWCNT (0, 100, 500, 1000 and 5000 mg.kg ⁻¹)	n.p	Cd (22.4 mg.kg ⁻¹)	Plant: <i>Boehmeria nivea</i>	Natural soil	60 days	Increase in Cd concentration: in leaves by +15%, +12% and +9% at 100, 500 and 1000 mg.kg ⁻¹ , in stems by +33% and +28% at 100 and 500 mg.kg ⁻¹ . Decreased Cd transfer: in stems by -10% at 5000 mg.kg ⁻¹ , in roots by -22%, -21%, -29% and -30% at 100, 500, 1000, 5000 mg.kg ⁻¹	Interactive effects: increase in leaf dry mass with the addition of 1000 mg.kg ⁻¹ of MWCNT, stem dry mass at 500 mg.kg ⁻¹ of MWCNT and root dry mass at 500 and 1000 mg.kg ⁻¹ of MWCNT, increase in H ₂ O ₂ , MDA concentration and SOD activity from the addition of 100 mg.kg ⁻¹ of MWCNT and decrease of POD activity from 100 mg.kg ⁻¹ of MWCNT	Gong et al. ²⁷

MWCNT (0, 100, 500, 1000 mg.kg ⁻¹)	Length: 3–12 µm, diameter: 12–15 nm, SSA : 245,28 m ² .g ⁻¹	Cd and As (Cd: 1.41 mg kg ⁻¹ , As: 54.80 mg.kg ⁻¹)	Plant : Solanum nigrum L.	Natural soil	60 days	No effect on Cd transfer with the addition of MWCNT. Decrease on As in stem (-25%) at 100 mg.kg ⁻¹ , in leaf (-31%) at 1000 mg.kg ⁻¹	Increase in antioxidant enzymes activities (MDA, PRO and APX)	Chen et al. ¹⁸⁶
MWCNT (0, 0.1 and 10 mg.kg ⁻¹)	Length = 1 to 20 µm, average diameter of 16.8 ± 6.7 nm	Two types of soil with two contamination level on 3 metals: 1 = Cd : 13, Pb : 524 and Zn : 689 mg.kg ⁻¹ and 2 = Cd : 28, Pb : 1471 and Zn 1825 mg.kg ⁻¹)	Plant: <i>Lactuca sativa</i> , earthworms: <i>Eisenia fetida</i> and native soil bacteria	Natural soil	42 days	In lower contaminated soil: no effects on metals transfer for earthworms. Increase in Cd (+37%) and Zn (+21%) transfer with 10 mg.kg ⁻¹ of MWCNT for lettuces. In higher contaminated soil: increase in Pb (+34%) and Zn (+25%) transfer with 10 mg.kg ⁻¹ of MWCNT for earthworms. Decrease in Cd (-29%) and Zn (-30%) transfer with 0.1 mg.kg ⁻¹ of MWCNT for lettuces.	Interactive effects on higher contaminated soil: biomass reduction for lettuces exposed to 0.1 mg.kg ⁻¹ of MWCNT. Increase in flavonoid concentration with 10 mg.kg ⁻¹ of MWCNT.	Leroy et al. ³⁰

410

411 **3.2. Impacts of the interaction between metal(loid)s and CNMs**

412 After an extensive literature survey on the topic, only articles focusing on CNTs, graphene
413 and GO together with a proper statistical analysis were kept for this review. Considering
414 studies evaluating both the transfer and the toxicity of metal(loid)s and CNMs (Table 3) (11
415 studies), 18% focused on bacteria, 18% on earthworms and 82% on plants. Among these
416 studies, about 64% reported interactive toxicological effects due to the simultaneous presence
417 of CNMs and metal(loid)s. Within these interactive effects, when all control conditions were
418 set-up (without metal(loid) and without CNM contamination, metal(loid) alone, CNM alone),
419 we could distinguish antagonist (18%) and synergic (18%) effects (Figure 1).

420

421 **3.2.1. Bacteria**

422 For bacteria, very few studies have been conducted. Furthermore, in the available articles the
423 experimental design did not permit to highlight the existence of interactive effects since the
424 impact of CNM alone or metal(loid)s alone were not tested (not the purpose of the study).
425 However, a study using soil contaminated with Cd (0.1 mg.kg^{-1}) and GO (1000 and 2000
426 mg.kg^{-1}) evidenced a decrease in soil microbial diversity after addition of GO, indicating that
427 the microbial communities in the Cd-contaminated group without GO were more diverse than
428 those in the treated groups (Cd x GO)¹⁸⁸. In addition, authors observed some changes in
429 bacterial communities after addition of GO with an increase in several bacterial abundances
430 (*Acidobacteria* and *Actinobacteria*) compared to single Cd contamination and a decrease in
431 phyla such as *WD272* and *TM6* revealing phylum-specific sensitivities¹⁸⁸. Interestingly, the
432 abundance of *Rhodospirillales* and *Nitrospirae* which are symbiotic N₂-fixing bacteria was

433 reduced at 3 days and then restored at 15 and 60 days evoking some resilience in the
434 ecosystem.

435 These few studies alone do not allow generalization to be made and highlight the need to
436 initiate more studies on microorganisms with simultaneous CNM and metal(loid) exposure.
437 Indeed, due to adsorption of metal(loid)s on CNM and to CNM strong interactions with
438 bacterial cell membranes, CNMs could be a vector to increase metal(loid) transfer to
439 microorganisms and associated toxicity or depending on their interactions with soil particle
440 they could also mitigate the metal(loid) toxicity (*i.e.* the complex CNM-metal(loid) could
441 adsorb to soil particles making metal less bioavailable to bacteria cells and thus less toxic).

442

443 **3.2.2. Earthworms**

444 Studies focused on earthworms report contrasting results of the addition of CNMs on
445 metal(loid) transfer and toxicity. Yang *et al.* observed a decrease of Cd transfer (antagonist
446 effect) ranging from -43% to -31% with the addition of 10 mg.kg⁻¹ and 100 mg.kg⁻¹ of
447 MWCNTs, respectively after 14 days of exposure in the model *Eisenia fetida* (Cd: 10mg.kg⁻¹
448)³². The decrease in Cd transfer was thus inversely proportional to the dose of MWCNTs.
449 Additionally, the change in transfer was also metal dose-dependent: at 10 mg.kg⁻¹ MWCNTs,
450 a -43% decrease was noticed at 10 mg.kg⁻¹ of Cd but decreased to -28% at 2 mg.kg⁻¹ of Cd. In
451 agreement with transfer results, the addition of MWCNTs decreased earthworm growth
452 inhibition caused by Cd, detoxifying enzymes and metabolites³². At the highest Cd
453 concentration (10 mg.kg⁻¹), a growth inhibition rate of 27% was registered in comparison with
454 earthworms exposed to control conditions (no Cd, no MWCNTs), this rate was decreased to
455 8% and 22% at 10 and 100 mg.kg⁻¹ of MWCNTs, respectively.

456 In contrast, Leroy *et al.* rather observed synergic effects of metals and CNTs when
457 earthworms were exposed to two contaminated soils (Zn, Pb and Cd) and demonstrated either
458 no impact of the addition of MWCNTs on metal uptake in one soil or an increase up to +34%
459 and +25% of Pb and Zn in the second soil (with no impact on Cd transfer) after 6 weeks³⁰.
460 Furthermore, when an increased uptake was detected it was CNT dose dependent. Even
461 though earthworms are able to eliminate orally ingested CNMs by depuration, it appears that CNM
462 ingestion could lead to metal release in earthworm tissues, maybe due to changing physico-chemical
463 conditions in the gut (especially pH) leading to desorption. Despite transfer increase, no
464 toxicological effects were found.

465 The difference between these articles could partly be explained by different soil texture.
466 Indeed, the first one which induced Pb and Zn transfer increase upon CNT addition had a
467 lower OM content than the other soil. Authors reminded that a low OM is less favorable for
468 earthworm feeding and burrowing activities¹⁸⁹ and thus metal transfer^{30,190}.

469

470 **3.2.3. Plants**

471 Among studies on the transfer and toxicological impacts of simultaneous contamination with
472 CNMs and metal(loid)s on plants (9 studies reviewed), 89% present results on the transfer of
473 metal(loid)s from the exposure medium to the plants and 100% on toxicity. In the transfer
474 studies, 91% of the studies showed CNM - metal(loid) interactions with either an
475 increase^{24,23,25,21,22,27,30,191,192} and/or a decrease^{21,24,26,27,29,30} of metal(loid) transfer differing
476 from the additive risk model.

477 Furthermore in the studies evidencing a synergic effect with an increased uptake of
478 metal(loid)s, some authors demonstrated a dose dependent effect of the addition of CNMs on
479 the transfer of metal(loid)s both in hydroponics and in soil²¹⁻²⁹. For example, Gao *et al.* found

480 that the addition of GO to a Cd contaminated medium (1 mg.L^{-1} Cd) led to a dose-dependent
481 increase in Cd concentration in both roots and leaves: the addition of 5 mg.L^{-1} of GO
482 increased by 12% the Cd concentration in leaves while an addition of 40 mg.L^{-1} led to a 50%
483 increase²⁵. Hypotheses such as facilitating metal entry into root cells due to interactions
484 between CNMs and root cells and/or uptake of CNMs with surface adsorbed metals may
485 explain the increased transfer observed in these examples. Additionally, it seems that at high
486 concentrations, CNMs may have a different impact than at lower concentrations in the
487 medium^{24,30}. Indeed, wheat plants exposed to As(III) took up 20% of this As while the uptake
488 rate of As increased to 25% and 32% in presence of GO concentrations of 0.1 and 1 mg.L^{-1} ,
489 then decreased to 16% at 10 mg.L^{-1} ²⁴. Likewise, Leroy *et al.* also observed contrasted results
490 depending on CNM concentrations and explained these results by CNM dispersion in the
491 medium (soil): at low concentrations (0.1 mg.kg^{-1}), CNTs may be better dispersed in the soil
492 than at higher concentration (10 mg.kg^{-1})^{193,194} offering thus, a higher exchange surface and
493 higher adsorption. This hypothesis is supported also by the study of Zhang *et al.* in which the
494 authors observed that the agglomeration of CNTs decreased the adsorption of some organic
495 compounds¹⁹³. Several hypotheses can be suggested for this increased uptake: CNMs with
496 adsorbed metal(loid)s can act as vectors by damaging root cell walls and facilitate
497 metal(loid)s entry in crops tissues or this complex might accumulate in the rhizosphere and
498 lead to an enhanced metal release with the pH modification at the vicinity of the root surface.

499

500 When an increased transfer of metals due to the presence of CNMs in the medium was
501 detected, it was mostly accompanied by an increased toxicity. Indeed, among the studies that
502 investigated the toxicological impacts of simultaneous contamination, 75% of them found
503 interactive effects highlighting an increase in oxidative stress^{24,25,27-29,184}, decrease in different
504 plant components^{24-26,29,184,195} (chlorophyll, protein, sugar) and in plant biomass^{24-29,184,195,196}

505 (Table 3). For instance, synergistic effects were evidenced on rubisco concentration : indeed a
506 decrease by 53% was detected in the presence of both Cd (5 mg.L⁻¹) and GO (40 mg.L⁻¹)
507 while Cd alone reduced it by 14.5% and GO alone by 18.5%; additive model would have
508 suggested a decrease by 33% ((-14.5%) + (-18.5%)) = -33%) much lower than the 53%
509 observed here²⁵. This observation was similar for other parameters (root and shoot biomass,
510 chlorophyll, O₂ and H₂O₂ concentrations) and more pronounced with higher Cd and GO
511 contamination. Similarly, Hu *et al.* demonstrated that the addition of GO (10 mg.L⁻¹)
512 decreased plant development and photosynthetic pigments concentrations together with an
513 increase in antioxidant enzymes activities (SOD and POD) in *Triticum aestivum L.* exposed to
514 As solution (10 mg.L⁻¹)²⁴; for instance after exposure to the co-contamination SOD increased
515 by 426%, while it increased by 56% after exposure to As alone and decreased by 4% after
516 exposure to GO alone (additive model would suggest a 52% increase).

517 However, some studies also highlighted antagonist interactions between metal(loid)s and
518 CNMs with a decrease in the transfer of metal(loid)s from soil to plants and finally lower or
519 no toxicity symptoms^{26-29,184}. These effects can also be organ dependent. Indeed, Zhao *et al.*
520 showed that GO-chitosan spheres significantly decreased Cu bioaccumulation in roots (-60%)
521 and stems (-67%) of wheat compare to single metal contamination, but not in leaves (almost
522 similar with an increase of Cu bioaccumulation around +2%)²¹. Despite changes in Cu
523 transfer with the addition of GO, the co-exposure did not impact growth parameters studied
524 (root, stem and leaf length) but decreased the photosynthetic rate and stomatal conductance up
525 to -58% and -34%, respectively. Likewise, Gong *et al.* found similar pattern with a decrease
526 in Cd bioaccumulation in roots up to -22% upon MWCNT addition (100 mg.kg⁻¹) and an
527 increase in Cd accumulation in stem and leaves up to +15% and +33%, respectively²⁷. These
528 differences on metal bioaccumulation according to plant organ could reflect a plant defence
529 strategy. It is known that plants have defence mechanisms against metal(loid) exposure like

530 accumulating metal(loid)s in specific compartments of the plant such as cell walls and leaves
531 to reduce their toxicity¹⁹⁷.

532 There are some disparities on transfer and toxicity of CNM-metal(loid) simultaneous
533 exposure. The adsorption mechanism alone does not seem to explain such disparities and the
534 concentration in CNMs, which directly affects their dispersion and aggregation in the
535 medium, could be a possible explanation.

536 Although the studies carried out in liquid media (water, culture solution, soil solution, etc.)
537 are a first approach privileged at the beginning of new researches on new substances, they do
538 not allow to apprehend the complex relations between solid matrix and contaminants since
539 some studies show their importance to determine the fate of contaminants in organisms³⁰.
540 Indeed, the type of soil could explain differences observed on interactive effects on transfer
541 and toxicity due to the strong influence of soil physico-chemical parameters (clay, OM, pH
542 *etc.*). Whatever the types of interactive effects, these largely dominate in the studies compared
543 to additive effects used in risk assessment.

544

545 **4. Conclusion – Knowledge gaps and Perspectives**

546 In 2012, the European Commission stated that they are “committed to ensuring that the risks
547 associated with chemical mixtures are properly considered and assessed”. In parallel, in 2019,
548 the European Environmental Agency warned again about the "cocktail effect" of chemical
549 pollutants, especially those found in European waters, whose cocktail effects are assumed or
550 proven¹⁹⁸. However up to date lots of progress remain to be done on that matter.

551 Indeed, after reviewing extensively the literature, we can conclude that strong interactions
552 exist between CNMs and metal(loid)s which have been demonstrated in simplified medium
553 (solution) and whose impacts are visible in the environment by modifying the behaviour of

554 contaminants in cocktail and their impact on organisms. These interactions include physical
555 adsorption, chemical interaction, electrostatic attraction and sorption-precipitation. In the
556 environment, the vast majority of studies concluded that these interactions modify the fate
557 (uptake and toxicity) of these contaminants in cocktail in comparison with an additive model.
558 The nature of the impact (synergic or antagonist) is influenced by many parameters : 1) CNM
559 nature which influences metal(loid) adsorption capabilities, 2) CNM concentration which
560 influences their behaviour, in particular their agglomeration, 3) CNM morphology and SSA as
561 well as adsorption abilities and CNM transfer through biological barriers, 4) metal(loid)
562 nature and number of different metallic species because of their specific affinity and potential
563 competition and repulsion for adsorption sites onto CNMs, 5) exposure medium which
564 influences CNM and metal(loid) behaviour, 6) soil characteristics (clay content, OM and pH)
565 which influence metal(loid) adsorption onto CNMs as well as organism behaviour and 7) type
566 of organisms (crops, earthworms or bacteria) and environmental relevance of exposure
567 conditions (exposure pathways, organism lifestyle, organism interactions, ...). So, in
568 conclusion, the additive model used so far for risk assessment of chemical substances in
569 mixture is not relevant; instead a multiplicative model which integrates synergy and
570 antagonism would seem more suitable with correction factors for the different influencing
571 parameters highlighted in this review to encompass processes which remain unclear so far and
572 leading to seemingly contradictory results in the literature.

573

574 **Knowledge gaps**

575 The knowledge gaps we identified through this literature survey are listed below:

576 (1) More information on CNM characteristics are needed in the articles to further understand
577 contrasted results observed on transfer and toxicity.

578 (2) The use of element speciation models (such as Minteq or ChemEQL among others) in
579 solution and their comparison with experimental data and their fitting with Langmuir or
580 Freundlich equations would be interesting to identify if these models could give relevant
581 predictions, especially in multi-metallic conditions.

582 (3) More studies need to be conducted on agrosystem organisms to have more information on
583 the toxicological effects resulting from exposure to a mixture of contaminants. For instance,
584 only 3 metal(loid)s (Cu = 1, As = 1 and Cd = 8, studies and one study with Cd, Pb and Zn
585 simultaneously) were tested in interactions with CNMs, which represents a small proportion of
586 the existing metals.

587 (4) Another important point is the difficulty to properly analyse the results in many studies
588 due to incomplete experimental designs that do not permit to highlight possible interactions.
589 On several occasions, the studies did not investigate the effects of CNMs alone or
590 metal(loid)s alone (missing control conditions). Thus, recurrent increases and decreases of the
591 studied biomarkers of toxicity were attributed to the addition of CNMs in the presence of
592 metal(loid)s, but these interpretations are only partial in the absence of tests with single
593 contaminant.

594 (5) The statistics are sometimes performed only by modality of metal(loid) contamination, not
595 allowing to establish possible interactions between CNMs and metal(loid)s (ANOVA 1-way
596 *vs* ANOVA 2-ways).

597 (6) More studies on ecosystem functions (mineralization and recycling) and biogeochemical
598 cycles (C, N and P) are needed to have a more integrative view of these contaminant impacts
599 on agrosystems. So far, these markers have only been studied after exposure to a single
600 contaminant even though they are very good indicators of an ecosystem health.

601 (7) Studies are also needed to identify a potential bioaccumulation along food webs. Studies
602 in micro-mesocosms with trophic chains seem to be a relevant experimental set-up to answer
603 this question¹⁹⁹. Due to CNM-metal(loid) interactions, the exposure duration in micro-
604 mesocosms needs to be adapted to organism cycles, especially to know if CNM-metal(loid)
605 exposure is putting at risk crop productions and human health.

606 (8) Technical development is also mandatory as the mechanisms explaining transfer and
607 toxicity results are still not well understood, in particular because data about CNMs actual
608 concentrations in the organisms are rarely available.

609

610 **Perspectives**

611 Using combined toxicity interaction indices could be a practice to generalize to gain a deeper
612 understanding of these interactions among contaminants. For example, Yang *et al.*³²
613 demonstrated synergic and antagonist effects using indices such as the Biomarker Response
614 Index (BRI) coupled with the Concentration Addition Index (CAI) and the Effect Addition
615 Index (EAI), which together quantify the joint effects of mixtures for a range of effect levels.
616 BRI is an index based on the level of alteration of the response of the biomarkers tested in the
617 treatments with contamination(s) compared to the control (without contamination). The level
618 of biomarker impairment is classified into four distinct levels, each corresponding to a score:
619 mild = 4, moderate = 3, major = 2 and severe = 1²⁰⁰. The score for each biomarker is
620 multiplied by the corresponding weighting to form an integrated index that is used to assess
621 the overall effect of contaminants in a system submitted to multiple stress²⁰¹. This allows the
622 integration of complex biomarker responses into a single predefined class. It is thus possible
623 to establish a dose-response relationship between contamination levels and biomarker
624 responses²⁰². This index has been used in the risk assessment of chemicals under the Water

625 Framework Directive in Wales²⁰⁰. The BRI coupled with the CAI and EAI allow to determine
626 respectively additive (=0), synergic (>0) or antagonist (<0) effects²⁰³. The CAI and EAI are
627 derived from the commonly used concentration addition (CA) and independent action (IA)
628 models that assess the toxicity of substances in non-interactive mixtures.

629 Another factor which is usually not considered is contamination aging. Indeed, the impact of
630 metal(loid)s is influenced by their origin and aging^{204–207}. The toxicological effects of
631 metal(loid)s spiked in soil in laboratory experiments (in salt form) differ with soils with an
632 aged contamination under the influence of natural processes^{204–207}. For example, oxidation
633 and precipitation of some metal(loid)s can affect their bioavailability and consequently their
634 toxicity²⁰⁸. This parameter should be more considered in future experiments.

635 Although micro and nanoplastics have different properties than CNMs, they are principally made of
636 carbon and also display adsorption capacities leading to similar effects on transfer and toxicity (Table
637 S3). Plastic concentration in agricultural soils could be much higher than the predicted CNM
638 concentrations related to sludge application or mulching practices in particular. Special attention
639 should thus be paid to the potential transfer and toxicological impacts of these micro and nanoplastics
640 in co-exposure situations with other contaminants such as metal(loid)s.

641 Finally, agrosystems interact closely with other organisms such as insects, small mammals
642 and birds on which they depend for part or all of their life cycles. Considering how these
643 organisms rely on agrosystems, they may be an entry point for the contamination to spread up
644 to other ecosystems.

645

646 **Acknowledgement**

647 The authors would like to acknowledge the Région Occitanie, Toulouse Federal University,
648 and EC2CO program CARBOSTRESS (CNRS-INSU) for their funding support.

Bibliography:

- (1) ISO/TS 80004-3:2020(fr), *Nanotechnologies — Vocabulaire — Partie 3: Nano-objets carbonés*. <https://www.iso.org/obp/ui/#iso:std:iso:ts:80004:-3:ed-2:v1:fr> (accessed 2021-05-09).
- (2) Singh, V.; Joung, D.; Zhai, L.; Das, S.; Khondaker, S. I.; Seal, S. Graphene Based Materials: Past, Present and Future. *Progress in Materials Science* **2011**, *56* (8), 1178–1271. <https://doi.org/10.1016/j.pmatsci.2011.03.003>.
- (3) Bianco, A.; Cheng, H.-M.; Enoki, T.; Gogotsi, Y.; Hurt, R. H.; Koratkar, N.; Kyotani, T.; Monthieux, M.; Park, C. R.; Tascon, J. M. D.; Zhang, J. All in the Graphene Family – A Recommended Nomenclature for Two-Dimensional Carbon Materials. *Carbon* **2013**, *65*, 1–6. <https://doi.org/10.1016/j.carbon.2013.08.038>.
- (4) Wick, P.; Louw-Gaume, A. E.; Kucki, M.; Krug, H. F.; Kostarelos, K.; Fadeel, B.; Dawson, K. A.; Salvati, A.; Vázquez, E.; Ballerini, L.; Tretiach, M.; Benfenati, F.; Flahaut, E.; Gauthier, L.; Prato, M.; Bianco, A. Classification Framework for Graphene-Based Materials. *Angewandte Chemie International Edition* **2014**, *53* (n° 30), 7714–7718. <https://doi.org/10.1002/anie.201403335>.
- (5) Huang, X.; Yin, Z.; Wu, S.; Qi, X.; He, Q.; Zhang, Q.; Yan, Q.; Boey, F.; Zhang, H. Graphene-Based Materials: Synthesis, Characterization, Properties, and Applications. *Small* **2011**, *7* (14), 1876–1902. <https://doi.org/10.1002/sml.201002009>.
- (6) Gottschalk, F.; Sun, T.; Nowack, B. Environmental Concentrations of Engineered Nanomaterials: Review of Modeling and Analytical Studies. *Environmental Pollution* **2013**, *181*, 287–300. <https://doi.org/10.1016/j.envpol.2013.06.003>.
- (7) Butt, B. Z.; Naseer, I. Nanofertilizers. In *Nanoagronomy*; Javad, S., Ed.; Springer International Publishing: Cham, 2020; pp 125–152. https://doi.org/10.1007/978-3-030-41275-3_8.
- (8) Paramo, L. A.; Feregrino-Pérez, A. A.; Guevara, R.; Mendoza, S.; Esquivel, K. Nanoparticles in Agroindustry: Applications, Toxicity, Challenges, and Trends. *Nanomaterials* **2020**, *10* (9), 1654. <https://doi.org/10.3390/nano10091654>.
- (9) Perreault, F.; Faria, A. F. de; Elimelech, M. Environmental Applications of Graphene-Based Nanomaterials. *Chem. Soc. Rev.* **2015**, *44* (16), 5861–5896. <https://doi.org/10.1039/C5CS00021A>.
- (10) Ramade, F. *Dictionnaire Encyclopédique Des Sciences De La Nature Et De La Biodiversité*, Dunod.; Paris, 2008.
- (11) Marschner, H. *Marschner's Mineral Nutrition of Higher Plants*; Academic Press, 2011.
- (12) Addo Ntim, S.; Mitra, S. Adsorption of Arsenic on Multiwall Carbon Nanotube–Zirconia Nanohybrid for Potential Drinking Water Purification. *Journal of Colloid and Interface Science* **2012**, *375* (1), 154–159. <https://doi.org/10.1016/j.jcis.2012.01.063>.
- (13) Tofighy, M. A.; Mohammadi, T. Adsorption of Divalent Heavy Metal Ions from Water Using Carbon Nanotube Sheets. *Journal of Hazardous Materials* **2011**, *185* (1), 140–147. <https://doi.org/10.1016/j.jhazmat.2010.09.008>.
- (14) Moradi, O. The Removal of Ions by Functionalized Carbon Nanotube: Equilibrium, Isotherms and Thermodynamic Studies. *Chemical and Biochemical Engineering Quarterly* **2011**, *25* (2), 229–240.
- (15) Moradi, O.; Zare, K. Adsorption of Pb(II), Cd(II) and Cu(II) Ions in Aqueous Solution on SWCNTs and SWCNT –COOH Surfaces: Kinetics Studies. *Fullerenes, Nanotubes and Carbon Nanostructures* **2011**, *19* (7), 628–652. <https://doi.org/10.1080/1536383X.2010.504955>.
- (16) Peng, W.; Li, H.; Liu, Y.; Song, S. A Review on Heavy Metal Ions Adsorption from Water by Graphene Oxide and Its Composites. *Journal of Molecular Liquids* **2017**, *230*, 496–504. <https://doi.org/10.1016/j.molliq.2017.01.064>.
- (17) Gao, X.; Hassan, I.; Peng, Y.; Huo, S.; Ling, L. Behaviors and Influencing Factors of the Heavy Metals Adsorption onto Microplastics: A Review. *Journal of Cleaner Production* **2021**, *319*, 128777. <https://doi.org/10.1016/j.jclepro.2021.128777>.

- (18) Nicholson, F. A.; Smith, S. R.; Alloway, B. J.; Carlton-Smith, C.; Chambers, B. J. An Inventory of Heavy Metals Inputs to Agricultural Soils in England and Wales. *Science of The Total Environment* **2003**, *311* (1), 205–219. [https://doi.org/10.1016/S0048-9697\(03\)00139-6](https://doi.org/10.1016/S0048-9697(03)00139-6).
- (19) Nagajyoti, P. C.; Lee, K. D.; Sreekanth, T. V. M. Heavy Metals, Occurrence and Toxicity for Plants: A Review. *Environmental Chemistry Letters* **2010**, *8* (3), 199–216. <https://doi.org/10.1007/s10311-010-0297-8>.
- (20) *Toxic Metals in Soil-Plant Systems*; Ross, S., Ed.; Wiley: Chichester, West Sussex, England ; New York, 1994.
- (21) Zhao, L.; Guan, X.; Yu, B.; Ding, N.; Liu, X.; Ma, Q.; Yang, S.; Yilihamu, A.; Yang, S.-T. Carboxylated Graphene Oxide-Chitosan Spheres Immobilize Cu²⁺ in Soil and Reduce Its Bioaccumulation in Wheat Plants. *Environment International* **2019**, *133*, 105208. <https://doi.org/10.1016/j.envint.2019.105208>.
- (22) He, Y.; Qian, L.; Zhou, K.; Hu, R.; Huang, M.; Wang, M.; Zhao, G.; Liu, Y.; Xu, Z.; Zhu, H. Graphene Oxide Promoted Cadmium Uptake by Rice in Soil. *ACS Sustainable Chem. Eng.* **2019**, *7* (12), 10283–10292. <https://doi.org/10.1021/acssuschemeng.8b06823>.
- (23) Li, J.; Wu, F.; Fang, Q.; Wu, Z.; Duan, Q.; Li, X.; Ye, W. The Mutual Effects of Graphene Oxide Nanosheets and Cadmium on the Growth, Cadmium Uptake and Accumulation in Rice. *Plant Physiology and Biochemistry* **2020**, *147*, 289–294. <https://doi.org/10.1016/j.plaphy.2019.12.034>.
- (24) Hu, X.; Kang, J.; Lu, K.; Zhou, R.; Mu, L.; Zhou, Q. Graphene Oxide Amplifies the Phytotoxicity of Arsenic in Wheat. *Scientific Reports* **2014**, *4* (1), 6122. <https://doi.org/10.1038/srep06122>.
- (25) Gao, M.; Yang, Y.; Song, Z. Effects of Graphene Oxide on Cadmium Uptake and Photosynthesis Performance in Wheat Seedlings. *Ecotoxicology and Environmental Safety* **2019**, *173*, 165–173. <https://doi.org/10.1016/j.ecoenv.2019.01.093>.
- (26) Gao, M.; Xu, Y.; Chang, X.; Dong, Y.; Song, Z. Effects of Foliar Application of Graphene Oxide on Cadmium Uptake by Lettuce. *Journal of Hazardous Materials* **2020**, *398*, 122859. <https://doi.org/10.1016/j.jhazmat.2020.122859>.
- (27) Gong, X.; Huang, D.; Liu, Y.; Zeng, G.; Wang, R.; Xu, P.; Zhang, C.; Cheng, M.; Xue, W.; Chen, S. Roles of Multiwall Carbon Nanotubes in Phytoremediation: Cadmium Uptake and Oxidative Burst in *Boehmeria Nivea* (L.) Gaudich. *Environ. Sci.: Nano* **2019**, *6* (3), 851–862. <https://doi.org/10.1039/C8EN00723C>.
- (28) Lian, J.; Wu, J.; Zeb, A.; Zheng, S.; Ma, T.; Peng, F.; Tang, J.; Liu, W. Do Polystyrene Nanoplastics Affect the Toxicity of Cadmium to Wheat (*Triticum Aestivum* L.)? *Environmental Pollution* **2020**, *263*, 114498. <https://doi.org/10.1016/j.envpol.2020.114498>.
- (29) Dong, Y.; Gao, M.; Song, Z.; Qiu, W. Microplastic Particles Increase Arsenic Toxicity to Rice Seedlings. *Environmental Pollution* **2020**, *259*, 113892. <https://doi.org/10.1016/j.envpol.2019.113892>.
- (30) Leroy, M.; Pey, B.; Jasse, V. E. J.; Liné, C.; Elger, A.; Probst, A.; Flahaut, E.; Silvestre, J.; Larue, C. Interactive Effects of Metals and Carbon Nanotubes in a Microcosm Agrosystem. *Journal of Hazardous Materials* **2022**, *431*, 128613. <https://doi.org/10.1016/j.jhazmat.2022.128613>.
- (31) Vijver, M. G.; Elliott, E. G.; Peijnenburg, W. J. G. M.; de Snoo, G. R. Response Predictions for Organisms Water-Exposed to Metal Mixtures: A Meta-Analysis. *Environmental Toxicology and Chemistry* **2011**, *30* (6), 1482–1487. <https://doi.org/10.1002/etc.499>.
- (32) Yang, X.; Gong, J.; Zhang, X.; Huang, Y.; Zhang, W.; Yang, J.; Lin, J.; Chai, Y.; Liu, J. Evaluation of the Combined Toxicity of Multi-Walled Carbon Nanotubes and Cadmium on Earthworms in Soil Using Multi-Level Biomarkers. *Ecotoxicology and Environmental Safety* **2021**, *221*, 112441. <https://doi.org/10.1016/j.ecoenv.2021.112441>.
- (33) Rodea-Palomares, I.; González-Pleiter, M.; Martín-Betancor, K.; Rosal, R.; Fernández-Piñas, F. Additivity and Interactions in Ecotoxicity of Pollutant Mixtures: Some Patterns, Conclusions, and Open Questions. *Toxics* **2015**, *3* (4), 342–369. <https://doi.org/10.3390/toxics3040342>.

- (34) Ren, X.; Chen, C.; Nagatsu, M.; Wang, X. Carbon Nanotubes as Adsorbents in Environmental Pollution Management: A Review. *Chemical Engineering Journal* **2011**, *170* (2), 395–410. <https://doi.org/10.1016/j.cej.2010.08.045>.
- (35) Gupta, V. K.; Moradi, O.; Tyagi, I.; Agarwal, S.; Sadegh, H.; Shahryari-Ghoshekandi, R.; Makhlof, A. S. H.; Goodarzi, M.; Garshasbi, A. Study on the Removal of Heavy Metal Ions from Industry Waste by Carbon Nanotubes: Effect of the Surface Modification: A Review. *Critical Reviews in Environmental Science and Technology* **2016**, *46* (2), 93–118. <https://doi.org/10.1080/10643389.2015.1061874>.
- (36) Ihsanullah; Abbas, A.; Al-Amer, A. M.; Laoui, T.; Al-Marri, M. J.; Nasser, M. S.; Khraisheh, M.; Atieh, M. A. Heavy Metal Removal from Aqueous Solution by Advanced Carbon Nanotubes: Critical Review of Adsorption Applications. *Separation and Purification Technology* **2016**, *157*, 141–161. <https://doi.org/10.1016/j.seppur.2015.11.039>.
- (37) Rao, G. P.; Lu, C.; Su, F. Sorption of Divalent Metal Ions from Aqueous Solution by Carbon Nanotubes: A Review. *Separation and Purification Technology* **2007**, *58* (1), 224–231. <https://doi.org/10.1016/j.seppur.2006.12.006>.
- (38) Li, Y.-H.; Ding, J.; Luan, Z.; Di, Z.; Zhu, Y.; Xu, C.; Wu, D.; Wei, B. Competitive Adsorption of Pb²⁺, Cu²⁺ and Cd²⁺ Ions from Aqueous Solutions by Multiwalled Carbon Nanotubes. *Carbon* **2003**, *41* (14), 2787–2792. [https://doi.org/10.1016/S0008-6223\(03\)00392-0](https://doi.org/10.1016/S0008-6223(03)00392-0).
- (39) Liang, J.; Liu, J.; Yuan, X.-Z.; Dong, H.; Zeng, G.; Wu, H.; Hou, W.; Liu, J.; Hua, S.; Zhang, S.; Zhigang, Y.; He, X.; He, Y. Facile Synthesis of Alumina-Decorated Multi-Walled Carbon Nanotubes for Simultaneous Adsorption of Cadmium Ion and Trichloroethylene. *Chemical Engineering Journal* **2015**, *273*. <https://doi.org/10.1016/j.cej.2015.03.069>.
- (40) Lu, C.; Chiu, H. Adsorption of Zinc(II) from Water with Purified Carbon Nanotubes. *Chemical Engineering Science* **2006**, *61* (4), 1138–1145. <https://doi.org/10.1016/j.ces.2005.08.007>.
- (41) Vasudevan, S.; Lakshmi, J. The Adsorption of Phosphate by Graphene from Aqueous Solution. *RSC Adv.* **2012**, *2* (12), 5234–5242. <https://doi.org/10.1039/C2RA20270K>.
- (42) Zhang, S.; Shao, Y.; Liu, J.; Aksay, I. A.; Lin, Y. Graphene–Polypyrrole Nanocomposite as a Highly Efficient and Low Cost Electrically Switched Ion Exchanger for Removing ClO₄[–] from Wastewater. *ACS Appl. Mater. Interfaces* **2011**, *3* (9), 3633–3637. <https://doi.org/10.1021/am200839m>.
- (43) Shi, G.; Ding, Y.; Fang, H. Unexpectedly Strong Anion– π Interactions on the Graphene Flakes. *Journal of Computational Chemistry* **2012**, *33* (14), 1328–1337. <https://doi.org/10.1002/jcc.22964>.
- (44) Madarang, C. J.; Kim, H. Y.; Gao, G.; Wang, N.; Zhu, J.; Feng, H.; Goring, M.; Kasner, M. L.; Hou, S. Adsorption Behavior of EDTA-Graphene Oxide for Pb (II) Removal. *ACS Appl. Mater. Interfaces* **2012**, *4* (3), 1186–1193. <https://doi.org/10.1021/am201645g>.
- (45) Liu, L.; Li, C.; Bao, C.; Jia, Q.; Xiao, P.; Liu, X.; Zhang, Q. Preparation and Characterization of Chitosan/Graphene Oxide Composites for the Adsorption of Au(III) and Pd(II). *Talanta* **2012**, *93*, 350–357. <https://doi.org/10.1016/j.talanta.2012.02.051>.
- (46) Shim, J.-W.; Park, S.-J.; Ryu, S.-K. Effect of Modification with HNO₃ and NaOH on Metal Adsorption by Pitch-Based Activated Carbon Fibers. *Carbon* **2001**, *39* (11), 1635–1642. [https://doi.org/10.1016/S0008-6223\(00\)00290-6](https://doi.org/10.1016/S0008-6223(00)00290-6).
- (47) Bian, Y.; Bian, Z.-Y.; Zhang, J.-X.; Ding, A.-Z.; Liu, S.-L.; Wang, H. Effect of the Oxygen-Containing Functional Group of Graphene Oxide on the Aqueous Cadmium Ions Removal. *Applied Surface Science* **2015**, *329*, 269–275. <https://doi.org/10.1016/j.apsusc.2014.12.090>.
- (48) Li, Z.; Chen, F.; Yuan, L.; Liu, Y.; Zhao, Y.; Chai, Z.; Shi, W. Uranium(VI) Adsorption on Graphene Oxide Nanosheets from Aqueous Solutions. *Chemical Engineering Journal* **2012**, *210*, 539–546. <https://doi.org/10.1016/j.cej.2012.09.030>.
- (49) Khan, J.; Jaafar, M. Reduction Efficiencies of Natural Substances for Reduced Graphene Oxide Synthesis. *J Mater Sci* **2021**, *56* (33), 18477–18492. <https://doi.org/10.1007/s10853-021-06492-y>.

- (50) Sigmund, G.; Jiang, C.; Hofmann, T.; Chen, W. Environmental Transformation of Natural and Engineered Carbon Nanoparticles and Implications for the Fate of Organic Contaminants. *Environmental Science: Nano* **2018**, *5* (11), 2500–2518. <https://doi.org/10.1039/C8EN00676H>.
- (51) Sitko, R.; Zawisza, B.; Malicka, E. Graphene as a New Sorbent in Analytical Chemistry. *TRAC Trends in Analytical Chemistry* **2013**, *51*, 33–43. <https://doi.org/10.1016/j.trac.2013.05.011>.
- (52) Zhao, J.; Wang, Z.; White, J. C.; Xing, B. Graphene in the Aquatic Environment: Adsorption, Dispersion, Toxicity and Transformation. *Environ. Sci. Technol.* **2014**, *48* (17), 9995–10009. <https://doi.org/10.1021/es5022679>.
- (53) Peigney, A.; Laurent, C.; Flahaut, E.; Bacsca, R.; Rousset, A. Specific Surface Area of Carbon Nanotubes and Bundles of Carbon Nanotubes. *Carbon* **2001**, *39*, 507–514. [https://doi.org/10.1016/S0008-6223\(00\)00155-X](https://doi.org/10.1016/S0008-6223(00)00155-X).
- (54) Xu, J.; Cao, Z.; Zhang, Y.; Yuan, Z.; Lou, Z.; Xu, X.; Wang, X. A Review of Functionalized Carbon Nanotubes and Graphene for Heavy Metal Adsorption from Water: Preparation, Application, and Mechanism. *Chemosphere* **2018**, *195*, 351–364. <https://doi.org/10.1016/j.chemosphere.2017.12.061>.
- (55) Suzuki, S. *Physical and Chemical Properties of Carbon Nanotubes*; BoD – Books on Demand, 2013.
- (56) Zhu, Y.; Murali, S.; Cai, W.; Li, X.; Suk, J. W.; Potts, J. R.; Ruoff, R. S. Graphene and Graphene Oxide: Synthesis, Properties, and Applications. *Advanced Materials* **2010**, *22* (35), 3906–3924. <https://doi.org/10.1002/adma.201001068>.
- (57) Marques, P. A. S. S.; Rosa, M. F.; Pinheiro, H. M. PH Effects on the Removal of Cu²⁺, Cd²⁺ and Pb²⁺ from Aqueous Solution by Waste Brewery Biomass. *Bioprocess Engineering* **2000**, *23* (2), 135–141. <https://doi.org/10.1007/PL00009118>.
- (58) Fu, F.; Wang, Q. Removal of Heavy Metal Ions from Wastewaters: A Review. *Journal of Environmental Management* **2011**, *92* (3), 407–418. <https://doi.org/10.1016/j.jenvman.2010.11.011>.
- (59) Yu, J.-G.; Zhao, X.-H.; Yu, L.-Y.; Jiao, F.-P.; Jiang, J.-H.; Chen, X.-Q. Removal, Recovery and Enrichment of Metals from Aqueous Solutions Using Carbon Nanotubes. *J Radioanal Nucl Chem* **2014**, *299* (3), 1155–1163. <https://doi.org/10.1007/s10967-013-2818-y>.
- (60) Ihsanullah; Al-Khaldi, F. A.; Abusharkh, B.; Khaled, M.; Atieh, M. A.; Nasser, M. S.; laoui, T.; Saleh, T. A.; Agarwal, S.; Tyagi, I.; Gupta, V. K. Adsorptive Removal of Cadmium(II) Ions from Liquid Phase Using Acid Modified Carbon-Based Adsorbents. *Journal of Molecular Liquids* **2015**, *204*, 255–263. <https://doi.org/10.1016/j.molliq.2015.01.033>.
- (61) Cui, L.; Wang, Y.; Gao, L.; Hu, L.; Yan, L.; Wei, Q.; Du, B. EDTA Functionalized Magnetic Graphene Oxide for Removal of Pb(II), Hg(II) and Cu(II) in Water Treatment: Adsorption Mechanism and Separation Property. *Chemical Engineering Journal* **2015**, *281*, 1–10. <https://doi.org/10.1016/j.cej.2015.06.043>.
- (62) Wang, H.; Yuan, X.; Wu, Y.; Huang, H.; Zeng, G.; Liu, Y.; Wang, X.; Lin, N.; Qi, Y. Adsorption Characteristics and Behaviors of Graphene Oxide for Zn(II) Removal from Aqueous Solution. *Applied Surface Science* **2013**, *279*, 432–440. <https://doi.org/10.1016/j.apsusc.2013.04.133>.
- (63) Sen, T. K.; Sarzali, M. V. Removal of Cadmium Metal Ion (Cd²⁺) from Its Aqueous Solution by Aluminium Oxide (Al₂O₃): A Kinetic and Equilibrium Study. *Chemical Engineering Journal* **2008**, *142* (3), 256–262. <https://doi.org/10.1016/j.cej.2007.12.001>.
- (64) Yang, S.-T.; Chang, Y.; Wang, H.; Liu, G.; Chen, S.; Wang, Y.; Liu, Y.; Cao, A. Folding/Aggregation of Graphene Oxide and Its Application in Cu²⁺ Removal. *J Colloid Interface Sci* **2010**, *351* (1), 122–127. <https://doi.org/10.1016/j.jcis.2010.07.042>.
- (65) Salam, M. A.; Al-Zhrani, G.; Kosa, S. A. Simultaneous Removal of Copper(II), Lead(II), Zinc(II) and Cadmium(II) from Aqueous Solutions by Multi-Walled Carbon Nanotubes. *Comptes Rendus Chimie* **2012**, *15* (5), 398–408. <https://doi.org/10.1016/j.crci.2012.01.013>.
- (66) Sitko, R.; Turek, E.; Zawisza, B.; Malicka, E.; Talik, E.; Heimann, J.; Gagor, A.; Feist, B.; Wrzalik, R. Adsorption of Divalent Metal Ions from Aqueous Solutions Using Graphene Oxide. *Dalton Trans.* **2013**, *42* (16), 5682–5689. <https://doi.org/10.1039/C3DT33097D>.

- (67) Lewis, A. Precipitation of Heavy Metals. In *Sustainable Heavy Metal Remediation: Volume 1: Principles and Processes*; Rene, E. R., Sahinkaya, E., Lewis, A., Lens, P. N. L., Eds.; Environmental Chemistry for a Sustainable World; Springer International Publishing: Cham, 2017; pp 101–120. https://doi.org/10.1007/978-3-319-58622-9_4.
- (68) Patnaik, P. *Dean's Analytical Chemistry Handbook*, 2nd edition.; McGraw Hill: New York, 2004.
- (69) Jiang, L.; Yu, H.; Zhou, X.; Hou, X.; Zou, Z.; Li, S.; Li, C.; Yao, X. Preparation, Characterization, and Adsorption Properties of Magnetic Multi-Walled Carbon Nanotubes for Simultaneous Removal of Lead(II) and Zinc(II) from Aqueous Solutions. *Desalination and Water Treatment* **2016**, *57* (39), 18446–18462. <https://doi.org/10.1080/19443994.2015.1090924>.
- (70) Ding, C.; Cheng, W.; Wang, X.; Wu, Z.-Y.; Sun, Y.; Chen, C.; Wang, X.; Yu, S.-H. Competitive Sorption of Pb(II), Cu(II) and Ni(II) on Carbonaceous Nanofibers: A Spectroscopic and Modeling Approach. *Journal of Hazardous Materials* **2016**, *313*, 253–261. <https://doi.org/10.1016/j.jhazmat.2016.04.002>.
- (71) Alimohammady, M.; Jahangiri, M.; Kiani, F.; Tahermansouri, H. Highly Efficient Simultaneous Adsorption of Cd(II), Hg(II) and As(III) Ions from Aqueous Solutions by Modification of Graphene Oxide with 3-Aminopyrazole: Central Composite Design Optimization. *New J. Chem.* **2017**, *41* (17), 8905–8919. <https://doi.org/10.1039/C7NJ01450C>.
- (72) Tan, P.; Hu, Y.; Bi, Q. Competitive Adsorption of Cu²⁺, Cd²⁺ and Ni²⁺ from an Aqueous Solution on Graphene Oxide Membranes. *Colloids and Surfaces A: Physicochemical and Engineering Aspects* **2016**, *509*, 56–64. <https://doi.org/10.1016/j.colsurfa.2016.08.081>.
- (73) Lu, C.; Liu, C.; Su, F. Sorption Kinetics, Thermodynamics and Competition of Ni²⁺ from Aqueous Solutions onto Surface Oxidized Carbon Nanotubes. *Desalination* **2009**, *249* (1), 18–23. <https://doi.org/10.1016/j.desal.2009.06.009>.
- (74) Lu, C.; Chiu, H.; Liu, C. Removal of Zinc(II) from Aqueous Solution by Purified Carbon Nanotubes: Kinetics and Equilibrium Studies. *Ind. Eng. Chem. Res.* **2006**, *45* (8), 2850–2855. <https://doi.org/10.1021/ie051206h>.
- (75) Morales, J.; Manso, J. A.; Cid, A.; Mejuto, J. C. Degradation of Carbofuran and Carbofuran-Derivatives in Presence of Humic Substances under Basic Conditions. *Chemosphere* **2012**, *89* (11), 1267–1271. <https://doi.org/10.1016/j.chemosphere.2012.05.018>.
- (76) Imai, A.; Fukushima, T.; Matsushige, K.; Kim, Y.-H.; Choi, K. Characterization of Dissolved Organic Matter in Effluents from Wastewater Treatment Plants. *Water Research* **2002**, *36* (4), 859–870. [https://doi.org/10.1016/S0043-1354\(01\)00283-4](https://doi.org/10.1016/S0043-1354(01)00283-4).
- (77) Lin, D.; Tian, X.; Li, T.; Zhang, Z.; He, X.; Xing, B. Surface-Bound Humic Acid Increased Pb²⁺ Sorption on Carbon Nanotubes. *Environmental Pollution* **2012**, *167*, 138–147. <https://doi.org/10.1016/j.envpol.2012.03.044>.
- (78) Tian, X.; Li, T.; Yang, K.; Xu, Y.; Lu, H.; Lin, D. Effect of Humic Acids on Physicochemical Property and Cd(II) Sorption of Multiwalled Carbon Nanotubes. *Chemosphere* **2012**, *89* (11), 1316–1322. <https://doi.org/10.1016/j.chemosphere.2012.05.082>.
- (79) Bandyopadhyaya, R.; Nativ-Roth, E.; Regev, O.; Yerushalmi-Rozen, R. Stabilization of Individual Carbon Nanotubes in Aqueous Solutions. *Nano Lett.* **2002**, *2* (1), 25–28. <https://doi.org/10.1021/nl010065f>.
- (80) Hyung, H.; Fortner, J. D.; Hughes, J. B.; Kim, J.-H. Natural Organic Matter Stabilizes Carbon Nanotubes in the Aqueous Phase. *Environ. Sci. Technol.* **2007**, *41* (1), 179–184. <https://doi.org/10.1021/es061817g>.
- (81) Sheng, G.; Li, J.; Shao, D.; Hu, J.; Chen, C.; Chen, Y.; Wang, X. Adsorption of Copper(II) on Multiwalled Carbon Nanotubes in the Absence and Presence of Humic or Fulvic Acids. *Journal of Hazardous Materials* **2010**, *178* (1–3), 333–340. <https://doi.org/10.1016/j.jhazmat.2010.01.084>.
- (82) Sun, W.-L.; Xia, J.; Li, S.; Sun, F. Effect of Natural Organic Matter (NOM) on Cu(II) Adsorption by Multi-Walled Carbon Nanotubes: Relationship with NOM Properties. *Chemical Engineering Journal* **2012**, *200–202*, 627–636. <https://doi.org/10.1016/j.cej.2012.06.118>.

- (83) Rizwan, M. S.; Imtiaz, M.; Huang, G.; Chhajro, M. A.; Liu, Y.; Fu, Q.; Zhu, J.; Ashraf, M.; Zafar, M.; Bashir, S.; Hu, H. Immobilization of Pb and Cu in Polluted Soil by Superphosphate, Multi-Walled Carbon Nanotube, Rice Straw and Its Derived Biochar. *Environ Sci Pollut Res* **2016**, *23* (15), 15532–15543. <https://doi.org/10.1007/s11356-016-6695-0>.
- (84) Jaisi, D. P.; Elimelech, M. Single-Walled Carbon Nanotubes Exhibit Limited Transport in Soil Columns. *Environ. Sci. Technol.* **2009**, *43* (24), 9161–9166. <https://doi.org/10.1021/es901927y>.
- (85) Kasel, D.; Bradford, S. A.; Šimůnek, J.; Pütz, T.; Vereecken, H.; Klumpp, E. Limited Transport of Functionalized Multi-Walled Carbon Nanotubes in Two Natural Soils. *Environmental Pollution* **2013**, *180*, 152–158. <https://doi.org/10.1016/j.envpol.2013.05.031>.
- (86) Lu, Y.; Yang, K.; Lin, D. Transport of Surfactant-Facilitated Multiwalled Carbon Nanotube Suspensions in Columns Packed with Sized Soil Particles. *Environmental Pollution* **2014**, *192*, 36–43. <https://doi.org/10.1016/j.envpol.2014.05.008>.
- (87) Zhang, W.; Zhang, F.; Han, Z.; Lü, X.; Lin, D.; Werner, D. Effect of Clay Minerals on Transport of Surfactants Dispersed Multi-walled Carbon Nanotubes in Porous Media. *Acta Geologica Sinica - English Edition* **2017**, *1* (91), 135–144. <https://doi.org/10.1111/1755-6724.13067>.
- (88) Han, B.; Liu, W.; Zhao, X.; Cai, Z.; Zhao, D. Transport of Multi-Walled Carbon Nanotubes Stabilized by Carboxymethyl Cellulose and Starch in Saturated Porous Media: Influences of Electrolyte, Clay and Humic Acid. *Science of The Total Environment* **2017**, *599–600*, 188–197. <https://doi.org/10.1016/j.scitotenv.2017.04.222>.
- (89) Lu, T.; Xia, T.; Qi, Y.; Zhang, C.; Chen, W. Effects of Clay Minerals on Transport of Graphene Oxide in Saturated Porous Media. *Environ Toxicol Chem* **2017**, *36* (3), 655–660. <https://doi.org/10.1002/etc.3605>.
- (90) Kasel, D.; Bradford, S. A.; Šimůnek, J.; Heggen, M.; Vereecken, H.; Klumpp, E. Transport and Retention of Multi-Walled Carbon Nanotubes in Saturated Porous Media: Effects of Input Concentration and Grain Size. *Water Research* **2013**, *47* (2), 933–944. <https://doi.org/10.1016/j.watres.2012.11.019>.
- (91) Qi, Z.; Zhang, L.; Chen, W. Transport of Graphene Oxide Nanoparticles in Saturated Sandy Soil. *Environ. Sci.: Processes Impacts* **2014**, *16* (10), 2268–2277. <https://doi.org/10.1039/C4EM00063C>.
- (92) Vithanage, M.; Herath, I.; Almaroai, Y. A.; Rajapaksha, A. U.; Huang, L.; Sung, J.-K.; Lee, S. S.; Ok, Y. S. Effects of Carbon Nanotube and Biochar on Bioavailability of Pb, Cu and Sb in Multi-Metal Contaminated Soil. *Environ Geochem Health* **2017**, *39* (6), 1409–1420. <https://doi.org/10.1007/s10653-017-9941-6>.
- (93) Yang, Z.; Xu, Z.; Geng, L.; Shu, W.; Zhu, T. Effect of Multi-Walled Carbon Nanotubes on Extractability of Sb and Cd in Contaminated Soil. *Ecotoxicology and Environmental Safety* **2020**, *205*, 111316. <https://doi.org/10.1016/j.ecoenv.2020.111316>.
- (94) Baragaño, D.; Forján, R.; Welte, L.; Gallego, J. L. R. Nanoremediation of As and Metals Polluted Soils by Means of Graphene Oxide Nanoparticles. *Scientific Reports* **2020**, *10* (1), 1896. <https://doi.org/10.1038/s41598-020-58852-4>.
- (95) Song, B.; Zeng, G.; Gong, J.; Zhang, P.; Deng, J.; Deng, C.; Yan, J.; Xu, P.; Lai, C.; Zhang, C.; Cheng, M. Effect of Multi-Walled Carbon Nanotubes on Phytotoxicity of Sediments Contaminated by Phenanthrene and Cadmium. *Chemosphere* **2017**, *172*, 449–458. <https://doi.org/10.1016/j.chemosphere.2017.01.032>.
- (96) Liné, C.; Larue, C.; Flahaut, E. Carbon Nanotubes: Impacts and Behaviour in the Terrestrial Ecosystem - A Review. *Carbon* **2017**, *123*, 767–785. <https://doi.org/10.1016/j.carbon.2017.07.089>.
- (97) Baath, E. Effects of Heavy Metals in Soil on Microbial Processes and Populations (a Review). *Water, Air, and Soil Pollution* **1989**, *47* (3–4), 335–379. <https://doi.org/10.1007/BF00279331>.
- (98) Giller, K. E.; Witter, E.; Mcgrath, S. P. Toxicity of Heavy Metals to Microorganisms and Microbial Processes in Agricultural Soils: A Review. *Soil Biology and Biochemistry* **1998**, *30* (10), 1389–1414. [https://doi.org/10.1016/S0038-0717\(97\)00270-8](https://doi.org/10.1016/S0038-0717(97)00270-8).

- (99) Giller, K. E.; Witter, E.; McGrath, S. P. Heavy Metals and Soil Microbes. *Soil Biology and Biochemistry* **2009**, *41* (10), 2031–2037. <https://doi.org/10.1016/j.soilbio.2009.04.026>.
- (100) Abdu, N.; Abdullahi, A. A.; Abdulkadir, A. Heavy Metals and Soil Microbes. *Environ Chem Lett* **2017**, *15* (1), 65–84. <https://doi.org/10.1007/s10311-016-0587-x>.
- (101) Jastrzębska, A. M.; Kurtycz, P.; Olszyna, A. R. Recent Advances in Graphene Family Materials Toxicity Investigations. *J Nanopart Res* **2012**, *14* (12), 1320. <https://doi.org/10.1007/s11051-012-1320-8>.
- (102) Gall, J. E.; Boyd, R. S.; Rajakaruna, N. Transfer of Heavy Metals through Terrestrial Food Webs: A Review. *Environ Monit Assess* **2015**, *187* (4), 201. <https://doi.org/10.1007/s10661-015-4436-3>.
- (103) Sivakumar, S. Effects of Metals on Earthworm Life Cycles: A Review. *Environmental monitoring and assessment* **2015**, *187*, 4742. <https://doi.org/10.1007/s10661-015-4742-9>.
- (104) Tyler, G.; Pålsson, A.-M. B.; Bengtsson, G.; Bååth, E.; Tranvik, L. Heavy-Metal Ecology of Terrestrial Plants, Microorganisms and Invertebrates. *Water Air Soil Pollut* **1989**, *47* (3), 189–215. <https://doi.org/10.1007/BF00279327>.
- (105) Nahmani, J.; Hodson, M. E.; Black, S. A Review of Studies Performed to Assess Metal Uptake by Earthworms. *Environmental Pollution* **2007**, *145* (2), 402–424. <https://doi.org/10.1016/j.envpol.2006.04.009>.
- (106) Sizmur, T.; Hodson, M. E. Do Earthworms Impact Metal Mobility and Availability in Soil? – A Review. *Environmental Pollution* **2009**, *157* (7), 1981–1989. <https://doi.org/10.1016/j.envpol.2009.02.029>.
- (107) Shahid, M.; Khalid, S.; Abbas, G.; Shahid, N.; Nadeem, M.; Sabir, M.; Aslam, M.; Dumat, C. Heavy Metal Stress and Crop Productivity; 2015; pp 1–25.
- (108) Kumar, S.; Trivedi, P. K. Chapter 25 - Heavy Metal Stress Signaling in Plants. In *Plant Metal Interaction*; Ahmad, P., Ed.; Elsevier, 2016; pp 585–603. <https://doi.org/10.1016/B978-0-12-803158-2.00025-4>.
- (109) Vardhan, K. H.; Kumar, P. S.; Panda, R. C. A Review on Heavy Metal Pollution, Toxicity and Remedial Measures: Current Trends and Future Perspectives. *Journal of Molecular Liquids* **2019**, *290*, 111197. <https://doi.org/10.1016/j.molliq.2019.111197>.
- (110) Wang, Q.; Li, C.; Wang, Y.; Que, X. Phytotoxicity of Graphene Family Nanomaterials and Its Mechanisms: A Review. *Front. Chem.* **2019**, *7*. <https://doi.org/10.3389/fchem.2019.00292>.
- (111) Rovira, A. D. Interactions Between Plant Roots and Soil Microorganisms. *Annual Review of Microbiology* **1965**, *19* (1), 241–266. <https://doi.org/10.1146/annurev.mi.19.100165.001325>.
- (112) Haferburg, G.; Kothe, E. Microbes and Metals: Interactions in the Environment. *Journal of Basic Microbiology* **2007**, *47* (6), 453–467. <https://doi.org/10.1002/jobm.200700275>.
- (113) Gadd, G. M. 2010. Metals, Minerals and Microbes: Geomicrobiology and Bioremediation. *Microbiology* *156* (3), 609–643. <https://doi.org/10.1099/mic.0.037143-0>.
- (114) Bruins, M. R.; Kapil, S.; Oehme, F. W. Microbial Resistance to Metals in the Environment. *Ecotoxicology and Environmental Safety* **2000**, *45* (3), 198–207. <https://doi.org/10.1006/eesa.1999.1860>.
- (115) I.v.n, R.; M, M.; N, B. TOLERANCE OF HEAVY BY GRAM POSITIVE SOIL BACTERIA. **2009**, *2* (4), 1185–1189.
- (116) Gadd, G. M. Heavy Metal Accumulation by Bacteria and Other Microorganisms. *Experientia* **1990**, *46* (8), 834–840. <https://doi.org/10.1007/BF01935534>.
- (117) Chander, K.; Brookes, P. C.; Harding, S. A. Microbial Biomass Dynamics Following Addition of Metal-Enriched Sewage Sludges to a Sandy Loam. *Soil Biology and Biochemistry* **1995**, *27* (11), 1409–1421. [https://doi.org/10.1016/0038-0717\(95\)00074-O](https://doi.org/10.1016/0038-0717(95)00074-O).
- (118) Roane, T. M.; Pepper, I. L. Microbial Responses to Environmentally Toxic Cadmium. *Microbial Ecology* **1999**, *38* (4), 358–364.
- (119) Giller, K. E.; McGrath, S. P.; Hirsch, P. R. Absence of Nitrogen Fixation in Clover Grown on Soil Subject to Long-Term Contamination with Heavy Metals Is Due to Survival of Only Ineffective

- Rhizobium. *Soil Biology and Biochemistry* **1989**, *21* (6), 841–848.
[https://doi.org/10.1016/0038-0717\(89\)90179-X](https://doi.org/10.1016/0038-0717(89)90179-X).
- (120) Jackson, P.; Jacobsen, N. R.; Baun, A.; Birkedal, R.; Kühnel, D.; Jensen, K. A.; Vogel, U.; Wallin, H. Bioaccumulation and Ecotoxicity of Carbon Nanotubes. *Chem Cent J* **2013**, *7*, 154.
<https://doi.org/10.1186/1752-153X-7-154>.
- (121) Zheng, X.; Su, Y.; Chen, Y.; Wan, R.; Li, M.; Wei, Y.; Huang, H. Carboxyl-Modified Single-Walled Carbon Nanotubes Negatively Affect Bacterial Growth and Denitrification Activity. *Sci Rep* **2014**, *4*. <https://doi.org/10.1038/srep05653>.
- (122) Hu, W.; Peng, C.; Luo, W.; Lv, M.; Li, X.; Li, D.; Huang, Q.; Fan, C. Graphene-Based Antibacterial Paper. *ACS Nano* **2010**, *4* (7), 4317–4323. <https://doi.org/10.1021/nn101097v>.
- (123) Jin, L.; Son, Y.; DeForest, J. L.; Kang, Y. J.; Kim, W.; Chung, H. Single-Walled Carbon Nanotubes Alter Soil Microbial Community Composition. *Sci. Total Environ.* **2014**, *466–467*, 533–538.
<https://doi.org/10.1016/j.scitotenv.2013.07.035>.
- (124) Chung, H.; Son, Y.; Yoon, T. K.; Kim, S.; Kim, W. The Effect of Multi-Walled Carbon Nanotubes on Soil Microbial Activity. *Ecotoxicology and Environmental Safety* **2011**, *74* (4), 569–575.
<https://doi.org/10.1016/j.ecoenv.2011.01.004>.
- (125) Jin, L.; Son, Y.; Yoon, T. K.; Kang, Y. J.; Kim, W.; Chung, H. High Concentrations of Single-Walled Carbon Nanotubes Lower Soil Enzyme Activity and Microbial Biomass. *Ecotoxicology and Environmental Safety* **2013**, *88*, 9–15. <https://doi.org/10.1016/j.ecoenv.2012.10.031>.
- (126) Ge, Y.; Priester, J. H.; Mortimer, M.; Chang, C. H.; Ji, Z.; Schimel, J. P.; Holden, P. A. Long-Term Effects of Multiwalled Carbon Nanotubes and Graphene on Microbial Communities in Dry Soil. *Environ. Sci. Technol.* **2016**, *50* (7), 3965–3974. <https://doi.org/10.1021/acs.est.5b05620>.
- (127) Chung, H.; Kim, M. J.; Ko, K.; Kim, J. H.; Kwon, H.; Hong, I.; Park, N.; Lee, S.-W.; Kim, W. Effects of Graphene Oxides on Soil Enzyme Activity and Microbial Biomass. *Science of The Total Environment* **2015**, *514*, 307–313. <https://doi.org/10.1016/j.scitotenv.2015.01.077>.
- (128) Liu, S.; Zeng, T. H.; Hofmann, M.; Burcombe, E.; Wei, J.; Jiang, R.; Kong, J.; Chen, Y. Antibacterial Activity of Graphite, Graphite Oxide, Graphene Oxide, and Reduced Graphene Oxide: Membrane and Oxidative Stress. *ACS Nano* **2011**, *5* (9), 6971–6980.
<https://doi.org/10.1021/nn202451x>.
- (129) Giraud, L.; Tourrette, A.; Flahaut, E. Carbon Nanomaterials-Based Polymer-Matrix Nanocomposites for Antimicrobial Applications: A Review. *Carbon* **2021**.
<https://doi.org/10.1016/j.carbon.2021.06.002>.
- (130) Akhavan, O.; Ghaderi, E. Toxicity of Graphene and Graphene Oxide Nanowalls Against Bacteria. *ACS Nano* **2010**, *4* (10), 5731–5736. <https://doi.org/10.1021/nn101390x>.
- (131) Kerfahi, D.; Tripathi, B. M.; Singh, D.; Kim, H.; Lee, S.; Lee, J.; Adams, J. M. Effects of Functionalized and Raw Multi-Walled Carbon Nanotubes on Soil Bacterial Community Composition. *PLOS ONE* **2015**, *10* (3), e0123042.
<https://doi.org/10.1371/journal.pone.0123042>.
- (132) Evariste, L.; Braylé, P.; Mouchet, F.; Silvestre, J.; Gauthier, L.; Flahaut, E.; Pinelli, E.; Barret, M. Graphene-Based Nanomaterials Modulate Internal Biofilm Interactions and Microbial Diversity. *Front. Microbiol.* **2021**, *12*. <https://doi.org/10.3389/fmicb.2021.623853>.
- (133) Wu, F.; You, Y.; Werner, D.; Jiao, S.; Hu, J.; Zhang, X.; Wan, Y.; Liu, J.; Wang, B.; Wang, X. Carbon Nanomaterials Affect Carbon Cycle-Related Functions of the Soil Microbial Community and the Coupling of Nutrient Cycles. *Journal of Hazardous Materials* **2020**, *390*, 122144.
<https://doi.org/10.1016/j.jhazmat.2020.122144>.
- (134) Lavelle, P.; Spain, A. *Soil Ecology*; Springer Science & Business Media, 2001.
- (135) Kristensen, E.; Penha-Lopes, G.; Delefosse, M.; Valdemarsen, T.; Quintana, C. O.; Banta, G. T. What Is Bioturbation? The Need for a Precise Definition for Fauna in Aquatic Sciences. *Marine Ecology Progress Series* **2012**, *446*, 285–302. <https://doi.org/10.3354/meps09506>.
- (136) Medina-Sauza, R. M.; Álvarez-Jiménez, M.; Delhal, A.; Reverchon, F.; Blouin, M.; Guerrero-Analco, J. A.; Cerdán, C. R.; Guevara, R.; Villain, L.; Barois, I. Earthworms Building Up Soil Microbiota, a Review. *Front. Environ. Sci.* **2019**, *7*. <https://doi.org/10.3389/fenvs.2019.00081>.

- (137) Morgan, J. E.; Morgan, A. J. The Distribution of Cadmium, Copper, Lead, Zinc and Calcium in the Tissues of the Earthworm *Lumbricus Rubellus* Sampled from One Uncontaminated and Four Polluted Soils. *Oecologia* **1990**, *84* (4), 559–566.
- (138) Spurgeon, D. J.; Hopkin, S. P.; Jones, D. T. Effects of Cadmium, Copper, Lead and Zinc on Growth, Reproduction and Survival of the Earthworm *Eisenia Fetida* (Savigny): Assessing the Environmental Impact of Point-Source Metal Contamination in Terrestrial Ecosystems. *Environmental Pollution* **1994**, *84* (2), 123–130. [https://doi.org/10.1016/0269-7491\(94\)90094-9](https://doi.org/10.1016/0269-7491(94)90094-9).
- (139) van Gestel, C. A. M.; Dirven-van Breemen, E. M.; Baerselman, R. Accumulation and Elimination of Cadmium, Chromium and Zinc and Effects on Growth and Reproduction in *Eisenia Andrei* (Oligochaeta, Annelida). *Science of The Total Environment* **1993**, *134*, 585–597. [https://doi.org/10.1016/S0048-9697\(05\)80061-0](https://doi.org/10.1016/S0048-9697(05)80061-0).
- (140) Neuhauser, E. F.; Loehr, R. C.; Milligan, D. L.; Malecki, M. R. Toxicity of Metals to the Earthworm *Eisenia Fetida*. *Biol Fert Soils* **1985**, *1* (3), 149–152. <https://doi.org/10.1007/BF00301782>.
- (141) Ma, W. Sublethal Toxic Effects of Copper on Growth, Reproduction and Litter Breakdown Activity in the Earthworm *Lumbricus Rubellus*, with Observations on the Influence of Temperature and Soil PH. *Environmental Pollution Series A, Ecological and Biological* **1984**, *33* (3), 207–219. [https://doi.org/10.1016/0143-1471\(84\)90011-4](https://doi.org/10.1016/0143-1471(84)90011-4).
- (142) Li, M.; Liu, Z.; Xu, Y.; Cui, Y.; Li, D.; Kong, Z. Comparative Effects of Cd and Pb on Biochemical Response and DNA Damage in the Earthworm *Eisenia Fetida* (Annelida, Oligochaeta). *Chemosphere* **2009**, *74* (5), 621–625. <https://doi.org/10.1016/j.chemosphere.2008.10.048>.
- (143) Beaumelle, L.; Vile, D.; Lamy, I.; Vandenbulcke, F.; Gimbert, F.; Hedde, M. A Structural Equation Model of Soil Metal Bioavailability to Earthworms: Confronting Causal Theory and Observations Using a Laboratory Exposure to Field-Contaminated Soils. *Science of The Total Environment* **2016**, *569–570*, 961–972. <https://doi.org/10.1016/j.scitotenv.2016.06.023>.
- (144) Wang, H.-T.; Ding, J.; Chi, Q.-Q.; Li, G.; Pu, Q.; Xiao, Z.-F.; Xue, X.-M. The Effect of Biochar on Soil-Plant-Earthworm-Bacteria System in Metal(Loid) Contaminated Soil. *Environmental Pollution* **2020**, *263*, 114610. <https://doi.org/10.1016/j.envpol.2020.114610>.
- (145) GOPAL, V.; CLEMENT, T.; NAGARAJAN, K. Potential of *Megascolex Pumilio* in Biomonitoring Environmental Pollution. *Indian J. Environ. Health* **1988**, *28* (3), 194–199.
- (146) Abbasi, S. A.; Soni, R. Stress-induced Enhancement of Reproduction in Earthworm *Octochaetus Pattoni* Exposed to Chromium (vi) and Mercury (II)— Implications in Environmental Management. *International Journal of Environmental Studies* **1983**, *22* (1), 43–47. <https://doi.org/10.1080/00207238308710100>.
- (147) Cikutovic, M. A.; Fitzpatrick, L. C.; Venables, B. J.; Goven, A. J. Sperm Count in Earthworms (*Lumbricus Terrestris*) as a Biomarker for Environmental Toxicology: Effects of Cadmium and Chlordane. *Environmental Pollution* **1993**, *81* (2), 123–125. [https://doi.org/10.1016/0269-7491\(93\)90076-Z](https://doi.org/10.1016/0269-7491(93)90076-Z).
- (148) Spurgeon, D. J.; Hopkin, S. P. Comparisons of Metal Accumulation and Excretion Kinetics in Earthworms (*Eisenia Fetida*) Exposed to Contaminated Field and Laboratory Soils. *Applied Soil Ecology* **1999**, *11* (2), 227–243. [https://doi.org/10.1016/S0929-1393\(98\)00150-4](https://doi.org/10.1016/S0929-1393(98)00150-4).
- (149) Lock, K.; Janssen, C. R. Toxicity of Arsenate to the Compostworm *Eisenia Fetida*, the Potworm *Enchytraeus Albidus* and the Springtail *Folsomia Candida*. *Bull. Environ. Contam. Toxicol.* **2002**, *68* (5), 760–765. <https://doi.org/10.1007/s001280320>.
- (150) Lock, K.; Janssen, C. R. Ecotoxicity of Nickel to *Eisenia Fetida*, *Enchytraeus Albidus* and *Folsomia Candida*. *Chemosphere* **2002**, *46* (2), 197–200. [https://doi.org/10.1016/S0045-6535\(01\)00112-6](https://doi.org/10.1016/S0045-6535(01)00112-6).
- (151) Bengtsson, G.; Gunnarsson, T.; Rundgren, S. Effects of Metal Pollution on the Earthworm *Dendrobaena Rubida* (Sav.) in Acidified Soils. *Water Air Soil Pollut* **1986**, *28* (3), 361–383. <https://doi.org/10.1007/BF00583501>.

- (152) van Gestel, C. A. M.; van Dis, W. A.; van Breemen, E. M.; Sparenburg, P. M. Development of a Standardized Reproduction Toxicity Test with the Earthworm Species *Eisenia Fetida* Andrei Using Copper, Pentachlorophenol, and 2,4-Dichloroaniline. *Ecotoxicology and Environmental Safety* **1989**, *18* (3), 305–312. [https://doi.org/10.1016/0147-6513\(89\)90024-9](https://doi.org/10.1016/0147-6513(89)90024-9).
- (153) Petersen, E. J.; Pinto, R. A.; Zhang, L.; Huang, Q.; Landrum, P. F.; Weber, W. J. Effects of Polyethyleneimine-Mediated Functionalization of Multi-Walled Carbon Nanotubes on Earthworm Bioaccumulation and Sorption by Soils. *Environ. Sci. Technol.* **2011**, *45* (8), 3718–3724. <https://doi.org/10.1021/es103004r>.
- (154) Li, S.; Irin, F.; Atore, F. O.; Green, M. J.; Cañas-Carrell, J. E. Determination of Multi-Walled Carbon Nanotube Bioaccumulation in Earthworms Measured by a Microwave-Based Detection Technique. *Science of The Total Environment* **2013**, *445–446*, 9–13. <https://doi.org/10.1016/j.scitotenv.2012.12.037>.
- (155) Eom, H.-J.; Roca, C. P.; Roh, J.-Y.; Chatterjee, N.; Jeong, J.-S.; Shim, I.; Kim, H.-M.; Kim, P.-J.; Choi, K.; Giralt, F.; Choi, J. A Systems Toxicology Approach on the Mechanism of Uptake and Toxicity of MWCNT in *Caenorhabditis Elegans*. *Chemico-Biological Interactions* **2015**, *239*, 153–163. <https://doi.org/10.1016/j.cbi.2015.06.031>.
- (156) Petersen, E. J.; Pinto, R. A.; Landrum, P. F.; Weber, Jr., Walter J. Influence of Carbon Nanotubes on Pyrene Bioaccumulation from Contaminated Soils by Earthworms. *Environ. Sci. Technol.* **2009**, *43* (11), 4181–4187. <https://doi.org/10.1021/es803023a>.
- (157) Calisi, A.; Grimaldi, A.; Leomanni, A.; Lionetto, M. G.; Dondero, F.; Schettino, T. Multibiomarker Response in the Earthworm *Eisenia Fetida* as Tool for Assessing Multi-Walled Carbon Nanotube Ecotoxicity. *Ecotoxicology* **2016**, *25* (4), 677–687. <https://doi.org/10.1007/s10646-016-1626-x>.
- (158) Hu, C. W.; Zhang, L. J.; Wang, W. L.; Cui, Y. B.; Li, M. Evaluation of the Combined Toxicity of Multi-Walled Carbon Nanotubes and Sodium Pentachlorophenate on the Earthworm *Eisenia Fetida* Using Avoidance Bioassay and Comet Assay. *Soil Biology and Biochemistry* **2014**, *70*, 123–130. <https://doi.org/10.1016/j.soilbio.2013.12.018>.
- (159) Zhao, S.; Wang, Y.; Duo, L. Biochemical Toxicity, Lysosomal Membrane Stability and DNA Damage Induced by Graphene Oxide in Earthworms. *Environmental Pollution* **2021**, *269*, 116225. <https://doi.org/10.1016/j.envpol.2020.116225>.
- (160) Scott-Fordsmand, J. J.; Krogh, P. H.; Schaefer, M.; Johansen, A. The Toxicity Testing of Double-Walled Nanotubes-Contaminated Food to *Eisenia Veneta* Earthworms. *Ecotoxicology and Environmental Safety* **2008**, *71* (3), 616–619. <https://doi.org/10.1016/j.ecoenv.2008.04.011>.
- (161) Tremel-Schaub, A.; Feix, I. *Contamination des sols: Transferts des sols vers les plantes*; EDP Sciences: Les Ulis France; Angers, 2005.
- (162) Comité Européen de Normalisation (CEN). Standards for the Safety of Users, the Environment and Plants 1995 CEN/TC 223/WG2 N.136 Version 2. 1995.
- (163) Assche, F. V.; Clijsters, H. Effects of Metals on Enzyme Activity in Plants. *Plant, Cell & Environment* **1990**, *13* (3), 195–206. <https://doi.org/10.1111/j.1365-3040.1990.tb01304.x>.
- (164) Jadia, C. D.; Fulekar, M. H. Phytoremediation of Heavy Metals: Recent Techniques. *African Journal of Biotechnology* **2009**, *8* (6).
- (165) Tamás, M. J.; Sharma, S. K.; Ibstedt, S.; Jacobson, T.; Christen, P. Heavy Metals and Metalloids As a Cause for Protein Misfolding and Aggregation. *Biomolecules* **2014**, *4* (1), 252–267. <https://doi.org/10.3390/biom4010252>.
- (166) Marin, A. R.; Pezeshki, S. R.; Masschelen, P. H.; Choi, H. S. Effect of Dimethylarsenic Acid (DMAA) on Growth, Tissue Arsenic, and Photosynthesis of Rice Plants. *Journal of Plant Nutrition* **1993**, *16* (5), 865–880. <https://doi.org/10.1080/01904169309364580>.
- (167) Panda, S. K.; Patra, H. K. Nitrate and Ammonium Ions Effect on the Chromium Toxicity in Developing Wheat Seedlings. *Proceedings of the National Academy of Sciences India. Section B, Biological Sciences* **2000**, *70* (1), 75–80.

- (168) Abedin, Md. J.; Cotter-Howells, J.; Meharg, A. A. Arsenic Uptake and Accumulation in Rice (*Oryza Sativa* L.) Irrigated with Contaminated Water. *Plant and Soil* **2002**, *240* (2), 311–319. <https://doi.org/10.1023/A:1015792723288>.
- (169) Sheldon, A. R.; Menzies, N. W. The Effect of Copper Toxicity on the Growth and Root Morphology of Rhodes Grass (*Chloris Gayana* Knuth.) in Resin Buffered Solution Culture. *Plant Soil* **2005**, *278* (1), 341–349. <https://doi.org/10.1007/s11104-005-8815-3>.
- (170) Du, X.; Zhu, Y.-G.; Liu, W.-J.; Zhao, X.-S. Uptake of Mercury (Hg) by Seedlings of Rice (*Oryza Sativa* L.) Grown in Solution Culture and Interactions with Arsenate Uptake. *Environmental and Experimental Botany* **2005**, *54* (1), 1–7. <https://doi.org/10.1016/j.envexpbot.2004.05.001>.
- (171) Wang, M.; Zou, J.; Duan, X.; Jiang, W.; Liu, D. Cadmium Accumulation and Its Effects on Metal Uptake in Maize (*Zea Mays* L.). *Bioresource Technology* **2007**, *98* (1), 82–88. <https://doi.org/10.1016/j.biortech.2005.11.028>.
- (172) Chibuike, G. U.; Obiora, S. C. *Heavy Metal Polluted Soils: Effect on Plants and Bioremediation Methods*. Applied and Environmental Soil Science. <https://doi.org/10.1155/2014/752708>.
- (173) Kopittke, P. M.; Blamey, F. P. C.; Asher, C. J.; Menzies, N. W. Trace Metal Phytotoxicity in Solution Culture: A Review. *Journal of Experimental Botany* **2010**, *61* (4), 945–954. <https://doi.org/10.1093/jxb/erp385>.
- (174) Liné, C.; Larue, C.; Flahaut, E. Carbon Nanotubes: Impacts and Behaviour in the Terrestrial Ecosystem - A Review. *Carbon* **2017**, *123*, 767–785. <https://doi.org/10.1016/j.carbon.2017.07.089>.
- (175) Larue, C.; Pinault, M.; Czarny, B.; Georgin, D.; Jaillard, D.; Bendiab, N.; Mayne-L’Hermite, M.; Taran, F.; Dive, V.; Carrière, M. Quantitative Evaluation of Multi-Walled Carbon Nanotube Uptake in Wheat and Rapeseed. *Journal of Hazardous Materials* **2012**, *227–228*, 155–163. <https://doi.org/10.1016/j.jhazmat.2012.05.033>.
- (176) Oleszczuk, P.; Joško, I.; Xing, B. The Toxicity to Plants of the Sewage Sludges Containing Multiwalled Carbon Nanotubes. *Journal of Hazardous Materials* **2011**, *186* (1), 436–442. <https://doi.org/10.1016/j.jhazmat.2010.11.028>.
- (177) Ratnikova, T. A.; Podila, R.; Rao, A. M.; Taylor, A. G. *Tomato Seed Coat Permeability to Selected Carbon Nanomaterials and Enhancement of Germination and Seedling Growth*. The Scientific World Journal. <https://doi.org/10.1155/2015/419215>.
- (178) Lahiani, M. H.; Chen, J.; Irin, F.; Poretzky, A. A.; Green, M. J.; Khodakovskaya, M. V. Interaction of Carbon Nanohorns with Plants: Uptake and Biological Effects. *Carbon* **2015**, *81*, 607–619. <https://doi.org/10.1016/j.carbon.2014.09.095>.
- (179) Lahiani, M. H.; Dervishi, E.; Ivanov, I.; Chen, J.; Khodakovskaya, M. Comparative Study of Plant Responses to Carbon-Based Nanomaterials with Different Morphologies. *Nanotechnology* **2016**, *27* (26), 265102. <https://doi.org/10.1088/0957-4484/27/26/265102>.
- (180) Lin, C.; Fugetsu, B.; Su, Y.; Watari, F. Studies on Toxicity of Multi-Walled Carbon Nanotubes on Arabidopsis T87 Suspension Cells. *Journal of Hazardous Materials* **2009**, *170* (2), 578–583. <https://doi.org/10.1016/j.jhazmat.2009.05.025>.
- (181) Shen, C.-X.; Zhang, Q.-F.; Li, J.; Bi, F.-C.; Yao, N. Induction of Programmed Cell Death in Arabidopsis and Rice by Single-Wall Carbon Nanotubes. *American Journal of Botany* **2010**, *97* (10), 1602–1609. <https://doi.org/10.3732/ajb.1000073>.
- (182) Mondal, A.; Basu, R.; Das, S.; Nandy, P. Beneficial Role of Carbon Nanotubes on Mustard Plant Growth: An Agricultural Prospect. *J Nanopart Res* **2011**, *13* (10), 4519. <https://doi.org/10.1007/s11051-011-0406-z>.
- (183) Begum, P.; Fugetsu, B. Phytotoxicity of Multi-Walled Carbon Nanotubes on Red Spinach (*Amaranthus Tricolor* L) and the Role of Ascorbic Acid as an Antioxidant. *Journal of Hazardous Materials* **2012**, *243*, 212–222. <https://doi.org/10.1016/j.jhazmat.2012.10.025>.
- (184) Gao, M.; Chang, X.; Yang, Y.; Song, Z. Foliar Graphene Oxide Treatment Increases Photosynthetic Capacity and Reduces Oxidative Stress in Cadmium-Stressed Lettuce. *Plant Physiology and Biochemistry* **2020**, *154*, 287–294. <https://doi.org/10.1016/j.plaphy.2020.06.021>.

- (185) Gao, M.; YujuanYang; Song, Z. Toxicity of Cadmium to Wheat Seedling Roots in the Presence of Graphene Oxide. *Chemosphere* **2019**, *233*, 9–16.
<https://doi.org/10.1016/j.chemosphere.2019.05.127>.
- (186) Chen, X.; Wang, J.; Hayat, K.; Zhang, D.; Zhou, P. Small Structures with Big Impact: Multi-Walled Carbon Nanotubes Enhanced Remediation Efficiency in Hyperaccumulator Solanum Nigrum L. under Cadmium and Arsenic Stress. *Chemosphere* **2021**, *276*, 130130.
<https://doi.org/10.1016/j.chemosphere.2021.130130>.
- (187) Xu, K.; Liu, Y.-X.; Wang, X.-F.; Li, S.-W.; Cheng, J.-M. Combined Toxicity of Functionalized Nano-Carbon Black and Cadmium on Eisenia Fetida Coelomocytes: The Role of Adsorption. *Journal of Hazardous Materials* **2020**, *398*, 122815. <https://doi.org/10.1016/j.jhazmat.2020.122815>.
- (188) Xiong, T.; Yuan, X.; Wang, H.; Leng, L.; Li, H.; Wu, Z.; Jiang, L.; Xu, R.; Zeng, G. Implication of Graphene Oxide in Cd-Contaminated Soil: A Case Study of Bacterial Communities. *Journal of Environmental Management* **2018**, *205*, 99–106.
<https://doi.org/10.1016/j.jenvman.2017.09.067>.
- (189) Capowiez, Y.; Gilbert, F.; Vallat, A.; Poggiale, J.-C.; Bonzom, J.-M. Depth Distribution of Soil Organic Matter and Burrowing Activity of Earthworms—Mesocosm Study Using X-Ray Tomography and Luminophores. *Biol Fertil Soils* **2021**, *57* (3), 337–346.
<https://doi.org/10.1007/s00374-020-01536-y>.
- (190) Wen, B.; Hu, X.; Liu, Y.; Wang, W.; Feng, M.; Shan, X. The Role of Earthworms (Eisenia Fetida) in Influencing Bioavailability of Heavy Metals in Soils. *Biol Fertil Soils* **2004**, *40* (3), 181–187.
<https://doi.org/10.1007/s00374-004-0761-3>.
- (191) Manjate, E.; Ramos, S.; Almeida, C. M. R. Potential Interferences of Microplastics in the Phytoremediation of Cd and Cu by the Salt Marsh Plant Phragmites Australis. *Journal of Environmental Chemical Engineering* **2020**, *8* (2), 103658.
<https://doi.org/10.1016/j.jece.2020.103658>.
- (192) Li, M.; Liu, Y.; Xu, G.; Wang, Y.; Yu, Y. Impacts of Polyethylene Microplastics on Bioavailability and Toxicity of Metals in Soil. *Science of The Total Environment* **2021**, *760*, 144037.
<https://doi.org/10.1016/j.scitotenv.2020.144037>.
- (193) Zhang, S.; Shao, T.; Bekaroglu, S. S. K.; Karanfil, T. The Impacts of Aggregation and Surface Chemistry of Carbon Nanotubes on the Adsorption of Synthetic Organic Compounds. *Environmental Science & Technology* **2009**, *43* (15), 5719–5725.
<https://doi.org/10.1021/es900453e>.
- (194) Petosa, A. R.; Jaisi, D. P.; Quevedo, I. R.; Elimelech, M.; Tufenkji, N. Aggregation and Deposition of Engineered Nanomaterials in Aquatic Environments: Role of Physicochemical Interactions. *Environmental Science & Technology* **2010**, *44* (17), 6532–6549.
<https://doi.org/10.1021/es100598h>.
- (195) Dong, Y.; Gao, M.; Qiu, W.; Song, Z. Uptake of Microplastics by Carrots in Presence of As (III): Combined Toxic Effects. *Journal of Hazardous Materials* **2021**, *411*, 125055.
<https://doi.org/10.1016/j.jhazmat.2021.125055>.
- (196) Wang, F.; Zhang, X.; Zhang, S.; Zhang, S.; Adams, C. A.; Sun, Y. Effects of Co-Contamination of Microplastics and Cd on Plant Growth and Cd Accumulation. *Toxics* **2020**, *8* (2), 36.
<https://doi.org/10.3390/toxics8020036>.
- (197) Singh, S.; Zacharias, M.; Kalpana, S.; Mishra, S. Heavy Metals Accumulation and Distribution Pattern in Different Vegetable Crops. *JECE* **2012**, *4* (4), 75–81.
<https://doi.org/10.5897/JECE11.076>.
- (198) Whalley, C.; Mohaupt, V. *Chemicals in European Waters — European Environment Agency*; Publication 18; European Environment Agency, 2019; p 80.
<https://www.eea.europa.eu/publications/chemicals-in-european-waters> (accessed 2022-05-10).
- (199) Evariste, L.; Mottier, A.; Lagier, L.; Cadarsi, S.; Barret, M.; Sarrieu, C.; Soula, B.; Mouchet, F.; Flahaut, E.; Pinelli, E.; Gauthier, L. Assessment of Graphene Oxide Ecotoxicity at Several

- Trophic Levels Using Aquatic Microcosms. *Carbon* **2020**, *156*, 261–271.
<https://doi.org/10.1016/j.carbon.2019.09.051>.
- (200) Hagger, J. A.; Jones, M. B.; Lowe, D.; Leonard, D. R. P.; Owen, R.; Galloway, T. S. Application of Biomarkers for Improving Risk Assessments of Chemicals under the Water Framework Directive: A Case Study. *Marine Pollution Bulletin* **2008**, *56* (6), 1111–1118.
<https://doi.org/10.1016/j.marpolbul.2008.03.040>.
- (201) Broeg, K.; Westernhagen, H. v.; Zander, S.; Körting, W.; Koehler, A. The “Bioeffect Assessment Index” (BAI): A Concept for the Quantification of Effects of Marine Pollution by an Integrated Biomarker Approach. *Marine Pollution Bulletin* **2005**, *50* (5), 495–503.
<https://doi.org/10.1016/j.marpolbul.2005.02.042>.
- (202) Li, X.; Wang, M.; Chen, W.; Jiang, R. Evaluation of Combined Toxicity of Siduron and Cadmium on Earthworm (*Eisenia Fetida*) Using Biomarker Response Index. *Science of The Total Environment* **2019**, *646*, 893–901. <https://doi.org/10.1016/j.scitotenv.2018.07.380>.
- (203) Zhang, J.; Liu, S.-S.; Zhang, J.; Qin, L.-T.; Deng, H.-P. Two Novel Indices for Quantitatively Characterizing the Toxicity Interaction between Ionic Liquid and Carbamate Pesticides. *Journal of Hazardous Materials* **2012**, *239–240*, 102–109.
<https://doi.org/10.1016/j.jhazmat.2012.07.063>.
- (204) Lock, K.; Janssen, C. R. Influence of Aging on Metal Availability in Soils. In *Reviews of Environmental Contamination and Toxicology*; Ware, G. W., Ed.; Reviews of Environmental Contamination and Toxicology; Springer: New York, NY, 2003; pp 1–21.
https://doi.org/10.1007/0-387-21728-2_1.
- (205) Ma, Y.; Lombi, E.; Oliver, I. W.; Nolan, A. L.; McLaughlin, M. J. Long-Term Aging of Copper Added to Soils. *Environ. Sci. Technol.* **2006**, *40* (20), 6310–6317.
<https://doi.org/10.1021/es060306r>.
- (206) Schreck, E.; Foucault, Y.; Geret, F.; Pradère, P.; Dumat, C. Influence of Soil Ageing on Bioavailability and Ecotoxicity of Lead Carried by Process Waste Metallic Ultrafine Particles. *Chemosphere* **2011**, *85*, 1555–1562. <https://doi.org/10.1016/j.chemosphere.2011.07.059>.
- (207) McBride, M. B.; Cai, M. Copper and Zinc Aging in Soils for a Decade: Changes in Metal Extractability and Phytotoxicity. *Environ. Chem.* **2015**, *13* (1), 160–167.
<https://doi.org/10.1071/EN15057>.
- (208) Martínez, C. E.; McBride, M. B. Cd, Cu, Pb, and Zn Coprecipitates in Fe Oxide Formed at Different PH: Aging Effects on Metal Solubility and Extractability by Citrate. *Environmental Toxicology and Chemistry* **2001**, *20* (1), 122–126. <https://doi.org/10.1002/etc.5620200112>.