Oznaka poročila: ARRS-RPROJ-ZP-2015/91

ZAKLJUČNO POROČILO RAZISKOVALNEGA PROJEKTA

A. PODATKI O RAZISKOVALNEM PROJEKTU

1.Osnovni podatki o raziskovalnem projektu

Šifra projekta	J1-4140
Naslov projekta	Strupene kovine in organokovinske spojine v kopenskem okolju
Vodja projekta	8314 Radmila Milačič
Tip projekta	J Temeljni projekt
Obseg raziskovalnih ur	8430
Cenovni razred	B
Trajanje projekta	07.2011 - 06.2014
Nosilna raziskovalna organizacija	106 Institut "Jožef Stefan"
Raziskovalne organizacije - soizvajalke	105 Nacionalni inštitut za biologijo
Raziskovalno področje po šifrantu ARRS	1 NARAVOSLOVJE 1.08 Varstvo okolja
Družbeno-ekonomski cilj	02. Okolje
Raziskovalno področje po šifrantu FOS	1 Naravoslovne vede 1.04 Kemija

B. REZULTATI IN DOSEŽKI RAZISKOVALNEGA PROJEKTA

2.Povzetek raziskovalnega projekta[1]

SLO

Med odlaganjem in razgradnji odpadkov kot so odpadna blata čistilnih naprav ali komunalni odpadki, se z biološkimi in kemijskimi procesi tvorijo in sproščajo številne strupene snovi. Pomembno skupino strupenih spojin v odpadnih blatih in izcednih vodah predstavljajo kovinske in organokovinske spojine. Da bi lahko sledili tvorbam,

pretvorbam in mobilnosti v okolju ter toksikološkim vplivom organokovinskih spojin in preprečili njihovo sproščanje v okolje, moramo razviti ustrezne analizne postopke.
Za celotne koncentracije kovin v odpadnih blatih in izcednih vodah odlagališč odpadkov je na voljo veliko podatkov, medtem ko podatkov o posameznih kemijskih zvrsteh kovin, vključno s strupenimi organokovinskimi spojinami, praktično ni. Malo je znanega tudi o mikrobioloških pretvorbah kovinskih spojin v omenjenih materialih. Pozanvanje porazdelitve kemijskih oblik elementov med trdno fazo in koloidnimi delci, ki so večinoma velikosti nanodelcev, je ključno za oceno njihove mobilnosti in preučevanje usode strupenih kovinskih in organokovinskih spojin v okolju.
V raziskovalnem projektu smo izvedli interdisciplinarno, kompleksno raziskavo prisotnosti, strupenosti in mobilnosti izbarnih kovin in njihovih kemijskih spojin v odpadnem blatu čistilnih naprav in izcednih vod iz odlagališč odpadkov. Preučevali smo mobilnost organokovinskih spojin kositra. Razvili smo zanesljive, selektivne in občutljive analizne postopke za njihov speciacijo v vzorcih z zelo kompleksno matrico. Preučevali smo inhibicijo nitrifikacije mikroorganizmov v aktivnem blatu s spojinami trivalentnega in šestvalentnega kroma. Pri preiskavah smo v laboratorijskih poskusih z uporabo stabilnih izotopov preučevali transformacije kemijskih zvrsti elementov ter procese metilacije. Delce, na katere so vezane kovine smo ločili po velikosti s sekvenčnim filtriranjem vzorcev in ocenili porazdelitev različnih zvrsti kovin med trdno ter raztopljeno in koloidno vodno frakcijo v preučevanih vzorcih izcednih vod iz odlagališč odpadkov. Škodljive učinke kemijskih zvrsti trivalenthega in šestvalentnega kroma smo ovrednotili z uporabo izbranih testov strupenosti. Novo znanje o usodi različnih kemijskih zvrsti preučevanih kovinskih elementov v odpadnem blatu in izcednih vodah smo uporabili za študije načinov preprečitve njihovih izpustov v terestično in vodno okolje in za remediacijo izcednih vod uporabili nanodelce železa.

ANG

During the disposal and decomposition of different waste materials such as sewage sludge and municipal wastes, several toxic compounds may be released or formed either by microbiological or chemical pathways. In order to evaluate their environmental and toxicological impacts, and to follow the pathways of organometallic compounds formation, their species transformations and their mobility in different environmental compartments and to prevent their release into the environemnt, it is necessary to develop reliable analytical procedures.
Presently there is a lot of data available on total metal concentrations in sewage sludge and landfill leachates. However, there is a lack of information on elemental chemical speciation, in particular regarding their toxic organometallic compounds. There is also insufficient information on transformation of metal species by microorganisms or chemicals present in sewage sludge and landfill leachates. Distribution of chemical species of elements between particulate matter and colloids, including nano particles play a key role in mobility of toxic metal species.
In the present project, a comprehensive investigation was performed on the occurrence, toxicity and mobility of selected metals in sewage sludge and landfill leachates. Transformation and

mobility of organotin compounds was studied. The inhibition of nitrification process by trivalent and hexavalent chromium was investigated in activated sludge. To follow the species transformation and methylation processes in landfill leachates and sewage sludge enriched stable isotopes were used. The size distribution and concentration of metal species between dissolved, colloidal and particulate matter between different compartments of landill leachate samples was evaluated by sequential filtration of samples. Adverse effects of trivalent and hexavalent chromium on the environment was studied with suitable toxicity testings. Based on the knowledge on the fate of organometallic compounds in sewage sludge and landfill leachates measures were proposed to prevent their potential release to the terrestrial and aquatic environment, while for remediation of landfill leachates, iron nanoparticles were applied.

3.Poročilo o realizaciji predloženega programa dela na raziskovalnem projektu[2]

Ogromne količine odpadkov, ki se zbirajo na odlagališčih se lahko, če z njimi primerno ne upravljamo, sproščajo v okolje. Kovine in organokovnske spojine predstavljajo pomembno skupino onesnažil v odpadnih blatih iz čistilnih naprav in v izcednih vodah iz odlagališč. Zaradi kompleksne matrice vzorcev je omenjena onesnažila težko analizirati.
Pri raziskavah kroženja organokositrovih spojin v okolju in oceni njihovih okoljskih in toksičnih vplivov, moramo razviti zanesljive in točne metode njihovega določanja. Med kovinami se v odpadnih blatih mnogokrat pojavlja tudi šestvalentni krom, ki lahko neagtivno vpliva na proces nitrifikacije v aktivnem balatu in povzroča še druge škodljive učinke v okolju. Za določitv šestavlentnega kroma v kompleksni matrici odpadnega blata, je potrebno vpeljati zanesljive analizne postopke.

RAZVOJ ANALIZNIH ORODIJ
V prvem delu projekta smo razvili ustrezna analizna orodja za preučevanje prisotnosti kovin in organokovinskih spojin (OTC) v odpadnih blatih in izcednih vodah.
Najprej smo razvili metodo za določanje organokositrovih spojin v vzorcih izcednih vod in delo objavili v eni izmed najbolj uglednih revij s področja analizne kemije:
VAHČIČ, Mitja, MILAČIČ, Radmila, ŠČANČAR, Janez. Development of analytical procedure for the determination of methyltin, butyltin, phenyltin and octyltin compounds in landfill leachates by gas chromatography-inductively coupled plasma mass spectrometry. *Analytica Chimica Acta*, 2011, 694, 21-30 [COBISS.SI-ID 24660775]
IF=4.555, EA - chemistry, analytical; 5/73; A' četrtina: 1

Z uporabo komercialno dostopne izotopsko označene mešanice butilkositrovih spojih ^{119}Sn (tributil kositer (TBT), dibutil kositer (DBT) in monobutil kositer (MBT)), smo razvili analizni postopek za določitev koncentracije TBT, DBT in MBT v vzorcih odpadnih blat in OTC kvantificirali z metodo izotopskega redčenja v masni spektrometriji (ID-ICP-MS). Delo smo objavili v zelo ugledni reviji:
ZULIANI, Tea, MILAČIČ, Radmila, ŠČANČAR, Janez. Preparation of a sewage sludge laboratory quality control material for butyltin compounds and their determination by isotope-dilution mass

spectrometry. *Analytical and Bioanalytical Chemistry*, 2012, 403, 857-865 [COBISS.SI-ID 25672231] IF=3.659, EA - chemistry, analytical; 9/75; A′ četrtina: 1

Obogateni stabilni izotopi elementov predstavljajo eno izmed najmočnejših orodij, ki jih lahko uporabljamo pri sledenju pretvrob kemijskih zvrsti elementov v okoljskih študijah in živih organizmih, omogočajo pa tudi točno kvantifikacijo kemijskih zvrsti elementov v speciacijski analizi.
V okviru projekta smo razvili postopek priprave obogatenih stabilnih izotopov kroma ^{50}Cr(VI) in ^{53}Cr(III) iz obogatenih oblik kromovega trioksida, pri čemer nismo uporabljali oksidantov niti reducentov, ki bi, če bi le v sledovih ostali v izotopski raztopini, povzročali kemijske pretvorbe kroma v vzorcu. Delo smo objavili v ugledni reviji:
NOVOTNIK, Breda, ZULIANI, Tea, MARTINČIČ, Anže, ŠČANČAR, Janez, MILAČIČ, Radmila. Preparation of Cr(VI) and Cr(III) isotopic spike solutions from ^{50}Cr and ^{53}Cr enriched oxides without the use of oxidizing and/or reducing agents. *Talanta*, 2012, 99, 83-90, [COBISS.SI-ID 25807399] IF=3.498, EA - chemistry, analytical; 12/75; A′ četrtina: 1

Da bi sledili kemijskim pretovorbam OTC v izcednih vodah iz odlagališč odpadkov, smo iz obogatenih izotopov kovinskega kositra pripravili ionski Sn^{4+}, Sn^{2+}, ^{117}TBT in ^{119}DBT. Točno koncentracijo obogatenega ^{117}TBT smo določili s plinsko kromatografijo (GC) v povezavi z reverzno ID-ICP-MS in delo objavli v ugledni reviji:
PEETERS, Kelly, ISKRA, Jernej, ZULIANI, Tea, ŠČANČAR, Janez, MILAČIČ, Radmila. The micro-scale synthesis of ^{117}Sn-enriched tributyltin chlorideand its characterization by GC-ICP-MS and NMR techniques. *Chemosphere*, 2014, 107, 386-392 [COBISS.SI-ID 27435559] IF=3.499, 32/216; A′ četrtina: 1

VPLIV Cr(VI) IN Cr(III) NA NITRIFIKACIJO V AKTIVNEM BLATU ČISTILNIH NAPRAV
V aktivnem blatu biološkega dela čistilnih naprav poteka nitrifikacija z mikroorganizmi. Morebitnim škodljivim učinkom Cr(VI) in Cr(III) na nitrifikatorje v aktivnem blatu smo sledili s poskusi, pri katerih smo k aktivnemu blatu dodajali različne koncentracije Cr(VI) in Cr(III). Točne koncentracije Cr(III) in Cr(VI) smo določili z metodo HPLC-ID-ICP-MS. Ugotovili smo, da koncentracije Cr(VI) višje od 1 mg/L in 50 mg/L Cr (III) povzročajo inhibicijo nitrifikacije. Delo smo objavili v ugledni reviji:
NOVOTNIK, Breda, ZULIANI, Tea, ŠČANČAR, Janez, MILAČIČ, Radmila. Inhibition of the nitrification process in activated sludge by trivalent and hexavalent chromium, and partitioning of hexavalent chromium between sludge compartments. *Chemosphere*, 2014, 105, 87-94, [COBISS.SI-ID 27432487], IF= 3.499, JA - environmental sciences; 32/216; A' četrtina: 1

RAZISKAVE PRETVORB ORGANOKOVINSKIH SPOJIN V IZCEDNIH VODAH IZ DEPONIJ ODPADKOV

S pomočjo obogatenih izotopov doma sintetiziranih kositrovih spojin ^{117}TBT (klorida), ^{119}dibutilkositrovega klorida (DBT), $^{117}SnCl_2$, $^{117}SnCl_4$ in komercialno dostopne mešanice ^{119}Sn butilkositrovih spojin smo študirali pretvorbe OKS v vzorcih izcednih vod iz odlagališč odpadkov. OKS smo kvantificirali z GC-ID-ICP-MS. Biotske pretvorbe, ki potekajo z mikroorganizmi smo od abiotskih procesov ločili tako, da smo del vzorca izcedne vode sterilizirali in rezultate primerjali z nesterilnimi vzorci. Rezultati so pokazali, da poteka razgradnja TBT z mikroorganizmi, DBT pa z abiotskimi pretvorbami. Z obogatenimi izotopi Sn^{2+} ali Sn^{4+}, ki smo jih dodali v izcedno vodo smo pokazali, da poteka metilacija kositrovih spojin v izcednih vodah s pomočjo mikroorganizmov. Rezultati študije pomembno prispevajo k poznavanju procesov pretvorb OTC v izcednih vodah in pomagajo načrtovati ustrezne ukrepe, ki preprečujejo sproščanje strupenih metilkositrovih spojin v okolje. Delo smo objavili v najbolj ugledni reviji s področja preučevanja voda:

PEETERS, Kelly, ZULIANI, Tea, ŠČANČAR, Janez, MILAČIČ, Radmila. The use of isotopically enriched tin tracers to follow the transformation of organotin compounds in landfill leachate. *Water Research*, 2014, 53, 297-309 [COBISS.SI-ID 27462695], IF=5.323, ZR - water resources; 1/81; A'' četrtina: 1

PREUČEVANJE MOBILNOSTI ORGANOKOSITROVIH SPOJIN IN KOVIN V IZCEDNIH VODAH IZ DEPONIJ ODPADKOV IN MOŽNOSTI NJIHOVE REMEDIACIJE Z NANODELCI ŽELEZA

Biotska in abiotska razgradnja OTCs v izcednih vodah iz odlagališč odpadkov mnogokrat ni popolna. Da bi našli načine učinkovite odstranitve TBT in trimetil kositra (TMeT) iz izcednih vod, smo preučevali možnosti uporabe različnih nanodelcev železa (FeNPs): nano ničvalentno železo (nZVI), FeONPs in Fe_3O_4NPs. En alikvot vzorca izcedne vode smo obdržali neremediiran. Ta vzorec je imel pH 8. Drugim alikvotom vzorca pa smo pri pH 8 dodali FeNPs, ki smo jih dispergirali s tetrametil amonijevim hidroksidom (TMAH) ali z mešanjem vzorca. Vzorce smo nato stresali pod aerobnimi pogoji 7 dni. Enake eksperimente smo opravili v vzorcih izcednih vod, katerim smo pH nastavili na vrednost 3 s citronsko kislino. S sekvenčno filtracijo vzorca smo vzorec ločili na frakcije z velikostjo delcev > 5000 nm, 450 do 5000 nm, 2.5 do 450 nm in < 2.5 nm ter TBT in TMeT v ločenih frakcijah določili z GC-ICP-MS. Rezultati so pokazali, da se je TBT nahajal pretežno v frakcijah z delci večjimi od 2.5 nm, TMeT pa je bil prisoten predvsem v raztopini, v frakciji z velikostjo delcev pod 2.5 nm. Pri pH 8 je prevladovala adsorpcija TBT na FeNPs, medtem ko se je pri pH 3, TBT razgradil s hidroksilnimi radikali, po mehanizmu Fentonske reakcije. TBT smo najbolj učinkovito odstranili (96 %) tako, da smo vzorcem zaporedoma dodali nZVI (dispergiran z mešanjem) pri pH 8, nato pa smo vodno fazo vzorca nakisali na pH 3 s citronsko kislino in vzorcu še enkrat dodali nZVI. Tak način remediacije z FeNPs je manj učinkovito odstranil TMeT (le okoli 40 %). Dokazali smo, da TMAH povzroči metilacijo ionskega kositra prisotnega v vzorcu, zato je za disperzijo nZVI potrebno uporabiti mešanje.

Iz omenjenega dela izhaja rokopis članka, ki je rezultat sodelovanja med raziskovalno skupino in Univerzo v Pau-ju Francija. Članek z naslovom: Adsorption and degradation processes of tributyltin and trimethyltin in landfill leachates treated with iron nanoparticles, avtorjev:

Kelly Peeters, Gaëtane Lespes, Janez Ščančar in Radmila Milačič, smo poslali v objavo v revijo z visokim faktorjem vpliva: Journal of Hazardous Materials IF=4,331; IM - engineering, civil; 1/106; A'' četrtina: 1. Rokopis pripenjamo k temu poročilu.

Kadar FeNPs uporabljamo za remediacijo vzorcev okoljskih vod, lahko NPs, ki ostanejo v vodni raztopini po končani remediaciji, onesnažijo okolje z nanodelci. Da bi ocenili, ali preostali nanodelci lahko predstavljajo nevarnost za okolje, smo v različnih časovnih razmikih sledili usedanju in velikostni porazdelitvi FeNPs v vzorcih okoljskih vod. Preučevali smo tudi učinkovitost odstranitve izbranih kovin z FeNPs. Koncentracije elementov v vzorcih vod pred in po nanoremediaciji smo določili z ICPMS. Rezultati so pokazali, da je usedanje delcev in s tem odstranitev FeNPs, iz vzorcev vod po remediaciji odvisna od karakteristik posameznega vzorca in načina disperzije. Disperzija nanodelcev s TMAH je bila bolj učinkovita kot z mešanjem. Ko smo za disperzijo nanodelecv uporabili mešanje, so se nZVI, FeONPs in Fe O NPs hitro usedali v Milli Q vodi, izvirski gozdni vodi in izcedni vodi iz odlagališč odpadkov. Pri disperziji s TMAH so se nanodelci usedali bolj počasi. V Milli Q in izvirski vodi, ki smo ji dodali FeONPs, se je okoli nanodelcev železa tvorila dvojna plast oleinske kisline in TMAH, ki je preprečila tvorbo železovih hidroksidov. Zato je nano železo, ki je ostalo v raztopini tudi še 24 h po remediaciji, predstavljalo nevarnost za okolje.

Izkazalo se je, da je učinkovitost odstranitve elementov iz okoljskih vod odvisna od vrste FeNPs in učinkovitosti disperzije nanodelcev. K učinkovitosti remediacije so pomembno prispevali tudi koncentracija elementa v vzorcu, karakteristike vzorca kot so pH, ionska moč, vsebnost organske snovi, prisotnost ligandov, ki tvorijo komplekse s kovinami, interakcije FeNPs z matrico vzorca ter kemijska speciacija elementov. Če je površina FeNPs onesnažena z določeno kovino, se to onesnažilo lahko sprosti v vodo, ki jo remediiramo.

Iz omenjenega dela izhaja rokopis članka, ki je rezultat sodelovanja med raziskovalno skupino in Univerzo v Pau-ju Francija. Članek z naslovom: The fate of iron nanoparticles in environmental waters treated with nanoscale zerovalent iron, FeO and Fe_3O_4, avtorjev: Kelly Peeters, Gaëtane Lespes, Janez Ščančar in Radmila Milačič, smo poslali v objavo v revijo z visokim faktorjem vpliva: Water Reasearch IF=5.323, ZR - water resources; 1/81; A'' četrtina: 1

Rokopis pripenjamo k temu poročilu.

Preučevanje mobilnosti organokositrovih spojin in kovin v izcednih vodah iz deponij odpadkov in možnosti njihove remediacije z nanodelci železa je potekalo v sodelovanju z Univerzo v Pau-ju, Francija. Iz omenjenega dela izhaja doktorat z deljenim mentorstvom »Co-tutelle«, med Univerzo v Ljubljani in Univerzo v Pau-ju, Francija, MR Kelly Peeters. Zagovor bo 20.4.2015.

STRUPENOSTNI TESTI
Genotoksično aktivnost Cr(VI) in različnih oblik Cr(III) smo določali s spremljanjem povzročanja poškodb DNA s testom komet (elektroforeza posamezne celice), ki je nova zelo občutljiva metoda ugotavljanja genotoksičnosti. Rokopis članka je v pripravi.

PRIPOROČILA ZA VAROVANJE KOPENSKEGA IN VODNEGA OKOLJA
Rezultate projekta so prispevali k uporabnim znanje o usodi in obnašanju preučevanih strupenih kovin ter OTC v odpadnih blatih in izcednih vodah iz odlagališč odpadkov.Na osnovi študije lahko upravljalci čistilnih naprav ali odlagališč odpadkov preprečijo izpuste onesnažil v kopensko in vodno okolje. Pokazali smo tudi možnosti odstranitve onesnažol z uporabo nanodelcev železa. Pomen rezultatov projekta je lokalen in tudi splošno globalen.

4.Ocena stopnje realizacije programa dela na raziskovalnem projektu in zastavljenih raziskovalnih ciljev[3]

Ocenjujemo, da so bili vsi cilji projekta izpolnjeni. Ker smo tekom dela ugotovili, da predstavljajo organokositrove spojine (OTC) izjemno pomembno skupino organokovinskih onesnažil v izcednih vodah in odpadnih blatih iz čistilnih naprav, smo delo intenzivno usmerili predvesm v njihovo preučevanje. Prav tako se je izkazalo, da inhibicijo nitrifikacije v odpadnih blatih povzroča predvsem šestavlentni krom. Ker so imeli na Centralni čistilni napravi Domžale-Kamnik probleme povezane prav z nekontroliranimi izpusti tega onesnažila na čistilno napravo, kar je povzročilo veliko škodo (aktivno blato so morali v celoti zamenjati, naprava ni obratovala 14 dni), smo se v projektu orientirali predvsem na reševanje tega problema. Raziskave so bile zelo uspešne in so dale odlične rezultate, tako z vidika študija bazičnih raziskav kroženja in pretvorb OTC v izcednih vodah iz deponij odpadkov, preučevanja mobilnosti OTC in izbranih kovin v izcednih vodah iz deponij, preučevanja procesov inhibicije nitrifikacije v aktivnem blatu s Cr(VI) in Cr(III) in preučevanja možnosti uporabe nanodelcev železa za remediacijo odpadnih vod, kakor tudi uporabne rezultate za upravljalce čistilnih naprav.
O kvaliteti opravljenih raziskav pričajo sledeča dejstva:
Iz projekta izhaja 6 člankov (vsi z IF iz 1 kvartila, pet člankov z A' in eden z A''), dva poslana rokopisa člankov v objavo, ter eden v pripravi. V zbornikih konferenc smo objavili 6 samostojnih sestavkov, z vabljenim predavanjem smo poročali na ugledni mednarodni konferenci Nordic Plasma Conference 2012, v Loenu na Norveškem, objavili smo 8 prispevkov na konferencah in eno samostojno znanstveno poglavje v monografski publikaciji. Iz projekat izhajata dva doktorata, en MR bo zaključil doktorat (»Co-tutelle«, delo je potekalo med raziskovalnima skupinana v iz Univerze v Pau-ju, Francija in Odsekom za zanosti o okolju, IJS) v aprilu 2105.

5.Utemeljitev morebitnih sprememb programa raziskovalnega projekta oziroma sprememb, povečanja ali zmanjšanja sestave projektne skupine[4]

V projektu ni bilo bistvenih odstopanj od predvidenega programa. V teku projekta se raziskovalna skupina se ni povečala, niti se ni zmanjšala.

6.Najpomembnejši znanstveni rezultati projektne skupine[5]

	Znanstveni dosežek			
1.	COBISS ID		25672231	Vir: COBISS.SI
	Naslov	*SLO*	Priprava laboratorijskega kontrolnega materiala butilkositrovih spojin v odpadnem baltu iz čistilnih naprav in določitev njihovih koncentracij z ID-ICP-MS	
		ANG	Preparation of a sewage sludge laboratory quality control material for butyltin compounds and their determination by isotope-dilution mass spectrometry	
	Opis	*SLO*	V prispevku opisujemo karakterizacijo laboratorijskega kontrolnega materiala (QCM) za določitev koncentracij dibutilkositra (DBT) in tributilkositra (TBT) v odpadnem blatu iz čistilnih naprav. V ta namen smo optimizirali postopek ekstrakcije (mikrovalovno, ultrazvočno in mehansko stresanje) ter koncentracije organokositrovih spojin (OTC) določili z metodo GC-ID-ICP-MS. QCM bomo uporabili v interlaboratorijskih primerjalnih testih in dnevno v laboratoriju za določitev točnosti analiznih postopkov pri študiju pretvorb OTC v odpadnih blatih.	
		ANG	The characterisation of a laboratory quality control material (QCM) for dibutyltin (DBT) and tributyltin (TBT) in sewage sludge is described. For this purpose the extraction procedure was optimized by the use of different extraction methods were tested (microwave- and ultrasound-assisted extraction and mechanical stirring). The reference values were determined by the use GC-ID-ICP-MS. The QCM will be used for an intercomparison study on DBT and TBT in sewage sludge, and as a day-to-day QCM during studies of the transformation of OTC in sewage sludge.	
	Objavljeno v		Springer; Analytical and bioanalytical chemistry; 2012; Vol. 403, no. 3; str. 857-865; Impact Factor: 3.659;Srednja vrednost revije / Medium Category Impact Factor: 2.187; A': 1; WoS: CO, EA; Avtorji / Authors: Zuliani Tea, Milačič Radmila, Ščančar Janez	
	Tipologija		1.01 Izvirni znanstveni članek	
2.	COBISS ID		27432487	Vir: COBISS.SI
	Naslov	*SLO*	Inhibicija nitrifikacije v aktivnem blatu s trivalentnim in šestavlentnim kromom ter porazdelitev šestavlentnega kroma med kompartmente blata	
		ANG	Inhibition of the nitrification process in activated sludge by trivalent and hexavalent chromium, and partitioning of hexavalent chromium between sludge compartments	
			Večina čistilnih naprav sprejema odplake iz gospodinjstev in industrije. Industrijske odplake lahko vsebujejo povišane koncentracije Cr(III) in/ali Cr (VI), ki v aktivnem blatu negativno vplivata na mikrobiološke procese, kot je nitrifikacija. Zato je bil namen našega dela preučiti vpliv različnih koncentracij Cr(III) in Cr(VI) na nitrifikacijo v aktivnem blatu in, za boljše razumevanje strupenosti Cr(VI), slediti njegovi redukciji, adsorpciji in prevzemu v posamezne predele aktivnega blata. Cr(VI) smo določili s speciacijsko analizo z uporabo metode izotopskega redčenja v masni spektrometriji z induktivno sklopljeno plazmo (ID-ICP-MS). Poskuse smo	

	Opis	*SLO*	opravili v šaržnih sistemih. Ugotovili smo, da koncentracije Cr(VI) nad 1,0 mg L-1 in koncentracije Cr(III) nad 50,0 mg L-1 negativno vplivajo na nitrifikacijo. Študije speciacijskih analiz po 24 h inkubacije so pokazale, da se Cr(VI) s koncentracijo do 2,5 mg L-1, skoraj v celoti reducira, medtem ko se je pri dodatku 5,0 mg Cr(VI) L-1, reduciralo le 40 % dodanega Cr (VI). Podatki o porazdelitvi kroma med predelkih aktivnega blata kažejo na to, da je večina Cr v medceličnem prostoru, medtem ko intracelični in adsorbirani krom predstavljata manj kot 0,1 % kroma v aktivnem blatu. Cr (VI) se je reduciral v vseh predelkih aktivnega blata, najbolj učinkovito v intraceličnem in medceličnem prostoru.	
		ANG	The input of wastewater treatment plants (WWTPs) may contain high concentrations of Cr(III) and Cr(VI), which can affect nitrogen removal. In the present study the influence of different Cr(III) and Cr(VI) concentrations towards activated sludge nitrification was studied. To better understand the mechanisms of Cr(VI) toxicity, its reduction, adsorption and uptake in activated sludge was investigated in a batch growth system. Quantification of Cr(VI) was performed by speciated isotope dilution inductively coupled plasma mass spectrometry. It was found that Cr(VI) concentrations above 1.0 mg L-1 and Cr(III) concentrations higher than 50 mg L-1 negatively affected nitrification. Speciation studies indicated almost complete reduction of Cr(VI) after 24 hours of incubation when Cr(VI) concentrations were lower than 2.5 mg L-1, whereas for Cr(VI) added to 5 mg L-1 around 40 % remained unreduced. The study of the partitioning of Cr in the activated sludge was performed by the addition of Cr(VI) in concentrations of 2.5 and 5.0 mg L-1. Results revealed that Cr was allocated mainly within the intercellular compartments, whereas intracellular and adsorbed Cr represented less than 0.1 % of the Cr sludge concentrations. Cr(VI) was reduced in all compartments, the most efficiently (about 94 %) within the intracellular and intercellular fractions. The extent of reduction of adsorbed Cr was 92 and 80 % for 2.5 and 5.0 mg of Cr(VI) L-1, respectively. The results of present investigation provide a new insight into the toxicity of Cr species towards activated sludge nitrification, which is of significant importance for the management of WWTPs in order to prevent them from inflows containing harmful Cr(VI) concentrations.	
	Objavljeno v		Pergamon Press.; Chemosphere; 2014; Vol. 105; str. 87-94; Impact Factor: 3.499;Srednja vrednost revije / Medium Category Impact Factor: 2.143; A': 1; WoS: JA; Avtorji / Authors: Novotnik Breda, Zuliani Tea, Ščančar Janez, Milačič Radmila	
	Tipologija		1.01 Izvirni znanstveni članek	
3.	COBISS ID		27435559	Vir: COBISS.SI
	Naslov	*SLO*	Mikro sinteza 117Sn-obogatenega tributil kositorbega klorida in njegoav karakterizacija z GC-ICP-MS in NMR tehnikami	
		ANG	The micro-scale synthesis of 117Sn-enriched tributyltin chloride and its characterization by GC-ICP-MS and NMR techniques	
	Opis	*SLO*	Opisan je enostaven postopek sinteze 117Sn obogatenih izotopov tributil kositrovih spojin (TBT). Sinteza prične z brominiranjem kovinskega Sn in butilacijo SnBr4 z butil litijem. Nastali tetrabutil kositer (TeBT) pretvorimo v TBTCl z uporabo koncentrirane klorvodikove kisline pri kontroliranih temperaturnih pogojih. Čistost sintetiziranega 117Sn-obogatenega izotopa TBT smo preverili s speciacijsko analizo z uporabo GC-ICP-MS in NMR tehnik. rezultati so pokazali, da je čistost nastalega 117TBT večja od 93 %, v preostalih 7 % je bil prisoten DBT. Nastale spojine smo kvantificirali z metodo reverznega izotopskega redčenja GC-ICP-MS. Izkoristek sinteze je bil boljši od 60 %, sintetizirana spojina pa je obstojna več kot 6 mesecev.	
			Described is a procedure for synthesis of 117Sn tributil butyltin. Synthesis	

		ANG	starts with bromination of metallic Sn, followed by butylation with butyl lithium. The formed tetrabutyltin (SnBu4) was transformed to tributyltin chloride (TBTCl) using concentrated hydrochloric acid (HCl). The purity of synthesized TBT was verified by the speciation analysis using gas chromatography coupled to inductively coupled plasma mass spectrometry (GC-ICP-MS) and nuclear magnetic resonance (NMR) techniques. Data revealed that TBT has a purity of more than 93 %. The remaining 7 % corresponded to DBT. TBT was quantified by reverse isotope dilution GC-ICP-MS. The synthesis yield was better than 60 % and the stability of product more than 6 months.	
	Objavljeno v		Pergamon Press.; Chemosphere; 2014; Vol. 107; str. 386-392; Impact Factor: 3.499;Srednja vrednost revije / Medium Category Impact Factor: 2.143; A': 1; WoS: JA; Avtorji / Authors: Peeters Kelly, Iskra Jernej, Zuliani Tea, Ščančar Janez, Milačič Radmila	
	Tipologija		1.01 Izvirni znanstveni članek	
4.	COBISS ID		27462695	Vir: COBISS.SI
	Naslov	SLO	Uporaba izotopsko označenih seldilcev kositra v študijah pretvorb organokositrovih spojin v izcednih vodah iz deponij odpadkov	
		ANG	The use of isotopically enriched tin tracers to follow the transformation of organotin compounds in landfill leachate	
	Opis	SLO	Izcedne vode iz odlagališč odpadkov so pomemben vir organokositrovih spojin (OTC). V literaturi zasledimo podatke o koncentracijah OTC in izcednih vodah, malo pa je del, ki bi poročala o razgradnji in biometilacijskih procesih. V predstavljeni študiji smo šest mesecev preučevali pretvorbe OTC v izcednih vodah pri simuliranih pogojih, kot so v izcednih vodah z uporabo obogatenih stabilnih izotopov kositra: 117Sn tributil kositer (TBT), 119Sn dibutil kositer (DBT), 119Sn mešanice butil kositrovih spojin (TBT, DBT in monobutil kositer (MBT)) ter obogatenih izotopov 117SnCl2 in 117SnCl4. Kvantifikacijo OTC smo opravili s plinsko kromatografijo in masno spektrometrijo z induktivno sklopljeno plazmo (GC-ICP-MS). Da bi ločili med biotskimi in abiotskimi pretvorbami OTC in zvrsti anorganskega kositra smo izcedno vodo sterilizirali in rezultate primerjali z ne-sterilnimi vzorci. Poskusi so pokazali, da je razgradnja TBT potekala z bakterijami, DBT pa abiotsko. V izcedni vodi z dodatkom obogatenih izotopov Sn2+ ali Sn4+ je potekala biometilacija. Izkazalo se je, hidroliza Sn2+ in Sn4+ zvrsti zavira proces metilacije in določa obseg nastalih metil kositrovih spojin. Predstavljena raziskava pomembno prispeva k boljšemu razumevanju procesov pretvorb OTC v izcednih vodah in preprečevanju izpustov strupenih metil kositrovih spojin v okolje. Inovativni pristop uporabe stabilnih izotopov kot sledilcev procesov in originalnost dela sta omogočila, da je bil članek objavljen v najuglednejši reviji, PRVI s področja: Water Research (IF=4.655).	
		ANG	Landfill leachates are an important pool of organotin compounds (OTCs). In the present study transformation of OTCs in landfill leachate was investigated under simulated landfill conditions over a time span of six months by the use of isotopically enriched tin tracers, namely 117Sn-enriched tributyltin (TBT), 119Sn-enriched dibutyltin (DBT), 119Sn-enriched butyltin mix containing TBT, DBT and monobutyltin (MBT) and 117Sn-enriched SnCl2 and SnCl4. Quantification of OTCs was performed by gas chromatography – inductively coupled plasma mass spectrometry (GC-ICP-MS). To discriminate the biotic and abiotic transformations of OTCs and inorganic tin species, sterilization of leachate was also performed and data compared to non-sterilized samples. During the course of the experiment the microbial degradation of TBT was clearly manifested in Sn-enriched spiked leachate samples, while abiotic pathway of degradation was observed for DBT. Biomethylation process was also observed in the	

			leachate spiked with Sn-enriched Sn2+ or Sn4+. Hydrolysis of Sn2+ and Sn4+ species was found to be a limiting factor which controlled the extent of methyltin formation. The present study importantly contributes to a better understanding of the processes that OTCs undergo in leachates, providing also useful information for taking measures necessary to prevent the release of toxic methyltin species to the nearby environment. Due to its novel approach by the use of stable isotopes as tracers to follow the fate of OTC and its originality the article was published in the most renowned journal FIRST from the field: Water Research (IF=4.655).	
	Objavljeno v		Pergamon Press.; Water research; 2014; Vol. 53; str. 297-309; Impact Factor: 5.323;Srednja vrednost revije / Medium Category Impact Factor: 1.497; A'': 1;A': 1; WoS: IH, JA, ZR; Avtorji / Authors: Peeters Kelly, Zuliani Tea, Ščančar Janez, Milačič Radmila	
	Tipologija		1.01 Izvirni znanstveni članek	
5.	COBISS ID		25807399	Vir: COBISS.SI
	Naslov	*SLO*	Priprava izotopskih raztopin Cr(VI) in Cr(III) iz obogatenih izotopov oksidov 50Cr in 53Cr brez uporabe oksidantov in reducentov	
		ANG	Preparation of Cr(VI) and Cr(III) isotopic spike solutions from [sup](50)Cr and [sup](53)Cr enriched oxides without the use of oxidizing and/or reducing agents	
	Opis	*SLO*	Razvili smo nov originalen postopek za pravilno pripravo raztopin stabilnih izotopov šestvalentnega kroma temelji na pripravi alkalne taline, ki jo nato raztopimo v klorovodikovi kislini. Šestvalentni krom se kvantitativno okisdira pri visoki temperaturi in visokem pH. Trivalentni krom iz kromovega trioksida pa pripravimo z mikrovalovnim razkrojem z uporabo dušikove (V) kisline. Razkrojeni vzorec nato v platinasti izparilnici odparimo do približno 0,2 mL in nato dodamo 1 mL klorvodikove kisline za stabilizacijo vzorca ter ustrezno razredčimo z vodo. Čistost izotopskih razotpin 50Cr(VI) in 53Cr(III) smo preverili s speciacijsko analizo FPLC-ICP-MS. Omenjeni način priprave raztopin stabilnih izotopov šestvalentnega in trivalentnega kroma iz kromovega trioksida omogoča njihovo pripravo brez dodatka oksidanotv ali reducentov. Dokazali smo, da že sledovi preostalega peroksida v sledilcu lahko v nevtralnem in alkalnem pH območju oksidirajo krom prisoten v preučevanem vzorcu, oziroma ga reducirajo v kislih pH območjih. Sledilce obogatenih izotopov smo uporabili pri študiju pretvorb Cr (VI) v odpadnem baltu in inhibiciji nitrifikacije s šestavlentnimin trivalentnim kromom.	
		ANG	In the present work, the procedure based on alkaline melting of 50Cr enriched oxide for preparation of pure 50Cr(VI) spike solution is reported. Moreover, the microwave assisted digestion procedure of 53Cr enriched oxide for preparation of pure 53Cr(III) spike solution is also presented. The purity of 50Cr(VI) and 53Cr(III) isotopic spike solutions was verified by the speciation analysis applying hyphenation of anion-exchange FPLC to ICP-MS. In addition, the artefacts in Cr speciation, which may be initiated by the traces of oxidising and/or reducing agents present in Cr spike solutions, are also shown. The outcomes of our investigation highlighted the importance of the adequate preparation of spike solutions of Cr isotopes that may be used as reliable tracers in the investigations of the oxidation-reduction processes of Cr in wide range of environmentally relevant pH values. The isotopic tracers were applied in the investigations of the transformations of Cr(VI) in sewage sludge and inhibition of nitrification processes by hexavalent and trivalent chromium.	
	Objavljeno v		Pergamon Press; Talanta; 2012; Vol. 99; str. 83-90; Impact Factor: 3.498;Srednja vrednost revije / Medium Category Impact Factor: 2.187; A': 1; WoS: EA; Avtorji / Authors: Novotnik Breda, Zuliani Tea, Martinčič Anže, Ščančar Janez, Milačič Radmila	

	Tipologija	1.01 Izvirni znanstveni članek

7.Najpomembnejši družbeno-ekonomski rezultati projektne skupine[6]

	Družbeno-ekonomski dosežek			
1.	COBISS ID		24575271	Vir: COBISS.SI
	Naslov	SLO	Varno odlaganje in ponovna uporaba odpadkov bogatih na kromu	
		ANG	Safe disposal and re-use of chromium rich waste materials	
	Opis	SLO	V poglavju v knjigi podajamo pregled stanja v svetu na področju varnega odlaganja odpadkov bogatih na kromu, kot so usnjarski odpadki, odpadki iz železarske industrije in predlagamo možnosti ponovne varne uporabe železarskih odpadkov, pri čemer se razbremenjujejo deponije. Pregled stanja vključuje lastne bogate reference z omenjenega področja in reference drugih raziskovalcev.	
		ANG	In book chaprter alternatives of safe disposal and re-use of chromium rich waste materials are discussed, including tannery waste, steel making waste and by-products. The alternatives of re-use of chromium rich waste materials are proposed. The extensive list of own references as well as references from other researchers are included in book chapter.	
	Šifra		D.11 Drugo	
	Objavljeno v		Nova Science Publishers; Management of hazardous residues containing Cr (VI); 2011; Str. 295-317; A': 1; Avtorji / Authors: Ščančar Janez, Milačič Radmila	
	Tipologija		1.16 Samostojni znanstveni sestavek ali poglavje v monografski publikaciji	
2.	COBISS ID		259188224	Vir: COBISS.SI
	Naslov	SLO	Pogovor z dobitnico Preglove nagrade za leto 2011 prof. dr. Radmilo Milačič	
		ANG	Interwiev	
	Opis	SLO	Za izjemne dosežke an področju speciacije elementov je prof.dr. Radmila Milačič prejela v letu 2011 Preglovo nagrado.	
		ANG	For extraordinary achievements from the filed of speciation analysis, in 2011 prof.dr. Radmila Milačič recieved Pregl's aword.	
	Šifra		E.01 Domače nagrade	
	Objavljeno v		Institut Jožef Stefan; Novice IJS; 2011; Št. 156; str. 3-4; Avtorji / Authors: Milačič Radmila	
	Tipologija		1.22 Intervju	
3.	COBISS ID		25912359	Vir: COBISS.SI
	Naslov	SLO	Priprava izotopskih raztopin Cr(VI) in Cr(III) iz obogatenih izotopov oksidov 50Cr in 53Cr brez uporabe oksidantov in reducentov	
		ANG	Preparation and use of [sup](50)Cr isotopic solutions for Cr speciation in environmental samples by HPLC-ICP-MS	
	Opis	SLO	Prof. dr. Radmila Milačič je imela vabljeno predavanje o pomenu pravilne priprave in uporabe obogatenih izotopskih raztopin Cr(VI) in Cr(III) v okoljskih študijah na 6th Nordic Conference on Plasma Spectrochemistry, June 10-13, 2012, Loen, Norway.	
		ANG	Prof. dr. Radmila Milačič gave an invite lecture on adequate preparation and the use of Cr isotopic tracers in environmental studies on 6th Nordic Conference on Plasma Spectrochemistry, June 10-13, 2012, Loen, Norway.	

	Šifra		B.04 Vabljeno predavanje	
	Objavljeno v		s. n.]; Programme and book of abstracts; 2012; Str. 39; Avtorji / Authors: Milačič Radmila, Zuliani Tea, Ščančar Janez	
	Tipologija		1.10 Objavljeni povzetek znanstvenega prispevka na konferenci (vabljeno predavanje)	
4.	COBISS ID		274296320	Vir: COBISS.SI
	Naslov	SLO	Uporaba stabilnih izotopov kroma v speciacijski analizi in pri študiju pretvorb kromovih zvrsti v okoljskih vzorcih	
		ANG	The use of stable chromium isotopes in speciation analysis and investigation of transformation of chromium species in environmental samples	
	Opis	SLO	Doktorska naloga Brede Novotnik, v okviru katere je pripravila obogatene stabilne izotope kroma in jih uporabila v speciacijski analizi in pri študiju pretvorb kromovih zvrsti v okoljskih vzorcih	
		ANG	Doctoral Dissertation of Breda Novotnik in which she prepared and used stable chromium isotopes in speciation analysis and investigation of transformation of chromium species in environmental samples	
	Šifra		D.09 Mentorstvo doktorandom	
	Objavljeno v		[B. Novotnik]; 2014; XII, 108 str.; Avtorji / Authors: Novotnik Breda	
	Tipologija		2.08 Doktorska disertacija	
5.	COBISS ID		26263335	Vir: COBISS.SI
	Naslov	SLO	Kroženje mikroelementov v okolju	
		ANG	Trace elements cycling in the environment	
	Opis	SLO	Prof.dr. Radmila Milačič je skupaj s prof. Janezom Ščančarjem imela v okviru predmeta Kroženje mikroelementov (Univerza v Novi Gorici, Fakulteta za znanosti o okolju) sklop predavanj, v katerih je predstavila študentom tudi izbrane študije s tematiko iz predstavljenega projekta.	
		ANG	Prof.dr. Radmila Milačič and prof. Janez Ščančar gave lectures on Trace elements cycling in the environment (University of Nova Gorica, Faculty for Environmental Sciences) in which thex presented selected case studies from the present project.	
	Šifra		D.10 Pedagoško delo	
	Objavljeno v		University of Nova Gorica, Faculty for Environmental Sciences; 2012; 1 zv. (loč. pag. ; prosojnice); Avtorji / Authors: Milačič Radmila	
	Tipologija		2.05 Drugo učno gradivo	

8.Drugi pomembni rezultati projetne skupine[7]

V okviru preučevanja mobilnosti OTC in kovin v izcednih vodah iz deponij odpadkov in možnosti njihove remediacije z nanodelci železa smo pripravili dva rokopisa člankov:

Prvi rokopis članka je rezultat sodelovanja med raziskovalno skupino in Univerzo v Pau-ju Francija. Članek z naslovom: Adsorption and degradation processes of tributyltin and trimethyltin in landfill leachates treated with iron nanoparticles, avtorjev:
Kelly Peeters, Gaëtane Lespes, Janez Ščančar in Radmila Milačič, smo poslali v objavo v revijo z visokim faktorjem vpliva: Journal of Hazardous Materials IF=4,331; IM - engineering, civil; 1/106; A'' četrtina: 1. Rokopis pripenjamo k temu poročilu.

Drugi rokopis članka je rezultat sodelovanja med raziskovalno skupino in Univerzo v Pau-ju Francija. Članek z naslovom: The fate of iron nanoparticles in environmental waters treated

with nanoscale zerovalent iron, FeO and Fe3O4, avtorjev:
Kelly Peeters, Gaëtane Lespes, Janez Ščančar in Radmila Milačič, smo poslali v objavo v revijo z visokim faktorjem vpliva: Water Reasearch IF=5.323, ZR - water resources; 1/81; A'' četrtina: 1. Rokopis pripenjamo k temu poročilu.

Preučevanje mobilnosti organokositrovih spojin in kovin v izcednih vodah iz deponij odpadkov in možnosti njihove remediacije z nanodelci železa je potekalo v sodelovanju z Univerzo v Pau-ju, Francija. Iz omenjenega dela izhaja doktorat z deljenim mentorstvom »Co-tutelle«, med Univerzo v Ljubljani in Univerzo v Pau-ju, Francija, mlada raziskovalka Kelly Peeters. Zagovor bo 20.4.2015.
Junija 2011 je vodja projekta Radmila Milačič prejela Preglovo nagrado Za dosežke na področju kemijske speciacije elementov.

9.Pomen raziskovalnih rezultatov projektne skupine[8]

9.1.Pomen za razvoj znanosti[9]

SLO

Predlagana raziskava je aktualna in je znatno prispevala k osnovnim raziskavam na področju kemijske speciacije elementov in velikostne porazdelitve kovin in njihovih kemijskih zvrsti v odpdanih vodah.
Razsikava je reševala prereče probleme odlaganja odpadkov in vplivov odlagališč odpadkov na okolje in je tako sledila svetovnim trendom na področju varovanja okolja.
V projektu smo s kombinacijo novih pristopov, ki vključujejo sintezo in uporabo stabilnih izotopov v speciaicjski analizi sledili procesom tvorbe in pretvorbe kemijskih zvrsti elementov v izcednih vodah. Z raziskavami smo prispevali k novim dognanjem, ki omogočajo upravljalcem odlagališč odpadkov izvajanje ustreznih ukrepov za preprečevanje izpustov strupenih kovin in organokovinskih spojin v v terestrično in vodno okolje.
Rezultati projekta so znatno prispevali k boljšemu razumevanju kroženja toksičnih kovin v kopenskem okolju. Pomembno je poudariti, da smo pri ovrednotenju celovite toksičnosti upoštevali tudi tvorbo izjemno strupenih metilkositrovih spojin, katerih negativni učinki na okolje so bili mnogokrat spregledani.

ANG

The proposed investigation is actual and considerably contributed to the basic research in the field of chemical speciation and sizing of metals and their chemical species in wastewaters.
The investigation followed the world trends of solving the acute environmental problems of waste disposal and trends in the field of environemntal protection.
In the project advanced analytical approaches, which included the synthesis and the use of enriched stable isotopes in combination with speciation analysis were applied to follow the processes of chemical species formation and transformation in landfill leachates. Based on the comprehensive knowledge, gained within the project tasks, on the fate of organometallic compounds in sewage sludge and landfill leachates, measures were recommended in order to prevent the potential release of pollutants investigated into the terrestrial and aquatic environment.
The results significantly contributed towards a better understanding of cycling of hazardous metals in the terrestrial environment, considering also formation of their more toxic organometallic compounds, which can significantly contribute towards overall toxicity, a fact often overlooked.

9.2. Pomen za razvoj Slovenije[10]

SLO

Vpliv in vloga določenega elementa v sledovih na okolje in žive organizme je odvisna tako od njegove celotne koncentracije, kot od kemijske oblike v kateri se element nahaja. Zelo malo je podatkov o onesnaženosti z zelo toksičnimi organokovinskimi spojinami v odpadnih blatih iz čistilnih naprav in v izcednih vodah iz deponij komunalnih odpadkov. Znano je, da prisotnost strupenih kovin in njihovih organokovinskih zvrsti v aktivnemu blatu, lahko bitveno zmanjša učinkovitost čiščenja odpadnih vod na čistilnih napravah. Poleg tega pa za odlaganje

kontaminiranega blata ni veliko možnosti. Prisotnost toksičnih kovin in njihovih organokovinskih zvrsti v izcednih vodah iz odlagališč komunalnih odpadkov prav tako zahteva njihovo učinkovito čiščenje pred izpustom izcednih vod v površinske vode.
Na osnovi obsežnega znanja o usodi organokovinskih spojin v odpadnem blatu in izcednih vodah, ki smo ga pridobili v okviru projekta, smo priporočili ukrepe za preprečitev morebitnih izpustov preiskovanih onesnažil v kopensko in vodno okolje. Pokazali smo, kako lahko z nanodelci železa učinkovito odstranimo strupeno obliko tributilkositra iz izcednih vod odlagališč odpadkov in v veliki meri odstranimo tudi trimetil kositrove spojine.
Rezultati raziskovalnega projekta bodo pomagali upravljalcem čistilnih naprav in odlagališč komunalnih odpadkov doma in po svetu k boljšim načinom ravnanja z odpadki, kar bo imelo pozitivne okoljske in ekonomske učinke.
Raziskave bodo torej pomembno prispevale tudi k varovanju naravne in kulturne dediščine in k izboljšanju kvalitete življenja. Varovanje okolja in zagotavljanje kvalitete življenja tudi za bodoče generacije, je glavna prioriteta ne samo v Sloveniji, temveč tudi v EU in drugih deželah širom po svetu.
Komplementarnost in dobro sodelovanje naše skupine, z vodilnimi mednarodnimi skupinami na področju speciacijske analize, v številnih mednarodnih projektih omogočata hitro medsebojno izmenjavo novih odkritij in njihovo integracijo v kompleksne, mednarodne, interdisciplinarne okoljske in biološke študije. Mednarodne povezave z izmenjavo raziskovalcev in doktorskih študentov so omogočale dostop do tujih znanj, kakor tudi dostop tujih raziskovalcev do novih znanj v Sloveniji. Iz mednarodne povezave izhaja en doktorat z deljenim metorstvom med Slovenijo in Francijo "co-tutelle" doktorantke Kelly Peeters (zagovor doktorata bo 20.4.2015).
Mednarodne povezave, objave znanstvenih člankov, ki izhajajo iz projekta, v uglednih mednarodnih revijah in predstavitev novih dosežkov na mednarodnih konferencah, so vsekakor promovirale slovensko znanost. Projekt je pomembno prispeval tudi k izobraževanju mladih raziskovalcev za dosego doktorata. V okviru projketa sta dva mlada raziskovalca zaključila doktorski študij. Doktorant Anže Martinčič se je zaposlil v slovenski industriji, doktorantka Breda Novotnik pa opravlja post-doktorkso izobraževanje v Kanadi.

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The fate and role of trace elements in the environment and living organisms depend not only on their total concentrations but also on different chemical forms in which they are present. An insufficient data exist on the occurence of highly toxic organometalic compounds in sewage sludge and landfill leachates. There is evidence that the presence of toxic metals and organometallic compounds in activated sludge could contribute to significant loss in treatment efficiency. Moreover, there are limited options for disposal of the contaminated sludge. The presence of toxic metals and organometallic compounds in landfill leachates also requires measures for their effective removal before the release of leachates in the surface waters.
Based on the comprehensive knowledge, gained within the project tasks, on the fate of organometallic compounds in sewage sludge and landfill leachates, measures were recommended in order to prevent the potential release of pollutants investigated into the terrestrial and aquatic environment. We have demonstrated that iron nanoparticles can be used for efficient remediation of tributyltin and in great extent also trimethyltin from landfill leachates.
The results of the proposed research will also help wastewater treatment plants and municipal waste landfills in Slovenia and world-wide in better management of wastes that will have positive environmental and economic impacts.
The proposed investigations will therefore significantly contribute to protection of the Slovenian environment and its cultural heritage and improved also to quality of life. The protection of the environment and guarantee for quality of life for future generations is the main priority not only in Slovenia, but also in EU and other countries worldwide.

10. Samo za aplikativne projekte in podoktorske projekte iz gospodarstva! Označite, katerega od navedenih ciljev ste si zastavili pri projektu, katere konkretne rezultate ste dosegli in v kakšni meri so doseženi rezultati uporabljeni

Cilj	

F.01	**Pridobitev novih praktičnih znanj, informacij in veščin**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.02	**Pridobitev novih znanstvenih spoznanj**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.03	**Večja usposobljenost raziskovalno-razvojnega osebja**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.04	**Dvig tehnološke ravni**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.05	**Sposobnost za začetek novega tehnološkega razvoja**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.06	**Razvoj novega izdelka**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.07	**Izboljšanje obstoječega izdelka**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.08	**Razvoj in izdelava prototipa**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.09	**Razvoj novega tehnološkega procesa oz. tehnologije**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.10	**Izboljšanje obstoječega tehnološkega procesa oz. tehnologije**	
	Zastavljen cilj	○ DA ○ NE

	Rezultat	
	Uporaba rezultatov	
F.11	**Razvoj nove storitve**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.12	**Izboljšanje obstoječe storitve**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.13	**Razvoj novih proizvodnih metod in instrumentov oz. proizvodnih procesov**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.14	**Izboljšanje obstoječih proizvodnih metod in instrumentov oz. proizvodnih procesov**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.15	**Razvoj novega informacijskega sistema/podatkovnih baz**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.16	**Izboljšanje obstoječega informacijskega sistema/podatkovnih baz**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.17	**Prenos obstoječih tehnologij, znanj, metod in postopkov v prakso**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.18	**Posredovanje novih znanj neposrednim uporabnikom (seminarji, forumi, konference)**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.19	**Znanje, ki vodi k ustanovitvi novega podjetja ("spin off")**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	

	Uporaba rezultatov	
F.20	**Ustanovitev novega podjetja ("spin off")**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.21	**Razvoj novih zdravstvenih/diagnostičnih metod/postopkov**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.22	**Izboljšanje obstoječih zdravstvenih/diagnostičnih metod/postopkov**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.23	**Razvoj novih sistemskih, normativnih, programskih in metodoloških rešitev**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.24	**Izboljšanje obstoječih sistemskih, normativnih, programskih in metodoloških rešitev**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.25	**Razvoj novih organizacijskih in upravljavskih rešitev**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.26	**Izboljšanje obstoječih organizacijskih in upravljavskih rešitev**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.27	**Prispevek k ohranjanju/varovanje naravne in kulturne dediščine**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.28	**Priprava/organizacija razstave**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	

F.29	**Prispevek k razvoju nacionalne kulturne identitete**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.30	**Strokovna ocena stanja**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.31	**Razvoj standardov**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.32	**Mednarodni patent**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.33	**Patent v Sloveniji**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.34	**Svetovalna dejavnost**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	
F.35	**Drugo**	
	Zastavljen cilj	○ DA ○ NE
	Rezultat	
	Uporaba rezultatov	

Komentar

11. Samo za aplikativne projekte in podoktorske projekte iz gospodarstva! Označite potencialne vplive oziroma učinke vaših rezultatov na navedena področja

	Vpliv	**Ni vpliva**	**Majhen vpliv**	**Srednji vpliv**	**Velik vpliv**	
G.01	**Razvoj visokošolskega izobraževanja**					
G.01.01.	Razvoj dodiplomskega izobraževanja	○	○	○	○	
G.01.02.	Razvoj podiplomskega izobraževanja	○	○	○	○	
G.01.03.	Drugo:	○	○	○	○	

G.02	**Gospodarski razvoj**					
G.02.01	Razširitev ponudbe novih izdelkov/storitev na trgu	○	○	○	○	
G.02.02.	Širitev obstoječih trgov	○	○	○	○	
G.02.03.	Znižanje stroškov proizvodnje	○	○	○	○	
G.02.04.	Zmanjšanje porabe materialov in energije	○	○	○	○	
G.02.05.	Razširitev področja dejavnosti	○	○	○	○	
G.02.06.	Večja konkurenčna sposobnost	○	○	○	○	
G.02.07.	Večji delež izvoza	○	○	○	○	
G.02.08.	Povečanje dobička	○	○	○	○	
G.02.09.	Nova delovna mesta	○	○	○	○	
G.02.10.	Dvig izobrazbene strukture zaposlenih	○	○	○	○	
G.02.11.	Nov investicijski zagon	○	○	○	○	
G.02.12.	Drugo:	○	○	○	○	
G.03	**Tehnološki razvoj**					
G.03.01.	Tehnološka razširitev/posodobitev dejavnosti	○	○	○	○	
G.03.02.	Tehnološko prestrukturiranje dejavnosti	○	○	○	○	
G.03.03.	Uvajanje novih tehnologij	○	○	○	○	
G.03.04.	Drugo:	○	○	○	○	
G.04	**Družbeni razvoj**					
G.04.01	Dvig kvalitete življenja	○	○	○	○	
G.04.02.	Izboljšanje vodenja in upravljanja	○	○	○	○	
G.04.03.	Izboljšanje delovanja administracije in javne uprave	○	○	○	○	
G.04.04.	Razvoj socialnih dejavnosti	○	○	○	○	
G.04.05.	Razvoj civilne družbe	○	○	○	○	
G.04.06.	Drugo:	○	○	○	○	
G.05.	**Ohranjanje in razvoj nacionalne naravne in kulturne dediščine in identitete**	○	○	○	○	
G.06.	**Varovanje okolja in trajnostni razvoj**	○	○	○	○	
G.07	**Razvoj družbene infrastrukture**					
G.07.01.	Informacijsko-komunikacijska infrastruktura	○	○	○	○	
G.07.02.	Prometna infrastruktura	○	○	○	○	
G.07.03.	Energetska infrastruktura	○	○	○	○	
G.07.04.	Drugo:	○	○	○	○	
G.08.	**Varovanje zdravja in razvoj zdravstvenega varstva**	○	○	○	○	
G.09.	**Drugo:**	○	○	○	○	

Komentar

12.Pomen raziskovanja za sofinancerje[11]

	Sofinancer		
1.	Naziv		
	Naslov		
	Vrednost sofinanciranja za celotno obdobje trajanja projekta je znašala:		EUR
	Odstotek od utemeljenih stroškov projekta:		%
	Najpomembnejši rezultati raziskovanja za sofinancerja		Šifra
	1.		
	2.		
	3.		
	4.		
	5.		
	Komentar		
	Ocena		

13.Izjemni dosežek v letu 2014[12]

13.1. Izjemni znanstveni dosežek

Obogatene izotope kositrovih spojin 117tributilkositrovega klorida (TBT), 119dibutilkositrovega klorida (DBT), 117SnCl2, 117SnCl4 in 119Sn mešanice butilkositrovih spojin smo uporabili pri študiju pretvorb organokositrovih spojin (OKS) v izcednih vodah iz odlagališč odpadkov. Biotske pretvorbe, ki potekajo z mikroorganizmi smo od abiotskih procesov ločili tako, da smo del vzorca izcedne vode sterilizirali in rezultate primerjali z nesterilnimi vzorci. Rezultati so pokazali, da poteka razgradnja TBT z mikroorganizmi, DBT pa z abiotskimi pretvorbami. Z obogatenimi izotopi Sn2+ ali Sn4+, ki smo jih dodali v izcedno vodo smo dokazali, da poteka metilacija kositrovih spojin v izcednih vodah s pomočjo mikroorganizmov. Rezultati študije pomembno prispevajo k poznavanju procesov pretvorb OTC v izcednih vodah in pomagajo načrtovati ustrezne ukrepe za preprečitev sproščanja strupenih metilkositrovih spojin v okolje. Rezulate smo objavili v prestižni reviji Water Research, IF=5.323, 1/81; A''.

13.2. Izjemni družbeno-ekonomski dosežek

C. IZJAVE

Podpisani izjavljam/o, da:

- so vsi podatki, ki jih navajamo v poročilu, resnični in točni
- se strinjamo z obdelavo podatkov v skladu z zakonodajo o varstvu osebnih podatkov za potrebe ocenjevanja ter obdelavo teh podatkov za evidence ARRS
- so vsi podatki v obrazcu v elektronski obliki identični podatkom v obrazcu v pisni obliki
- so z vsebino zaključnega poročila seznanjeni in se strinjajo vsi soizvajalci projekta

Podpisi:

zastopnik oz. pooblaščena oseba raziskovalne organizacije:	in	*vodja raziskovalnega projekta:*
Institut "Jožef Stefan"		Radmila Milačič

ŽIG

Kraj in datum:	Ljubljana,	1.3.2015

Oznaka poročila: ARRS-RPROJ-ZP-2015/91

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[1] Napišite povzetek raziskovalnega projekta (največ 3.000 znakov v slovenskem in angleškem jeziku) Nazaj

[2] Napišite kratko vsebinsko poročilo, kjer boste predstavili raziskovalno hipotezo in opis raziskovanja. Navedite ključne ugotovitve, znanstvena spoznanja, rezultate in učinke raziskovalnega projekta in njihovo uporabo ter sodelovanje s tujimi partnerji. Največ 12.000 znakov vključno s presledki (približno dve strani, velikost pisave 11). Nazaj

[3] Realizacija raziskovalne hipoteze. Največ 3.000 znakov vključno s presledki (približno pol strani, velikost pisave 11) Nazaj

[4] V primeru bistvenih odstopanj in sprememb od predvidenega programa raziskovalnega projekta, kot je bil zapisan v predlogu raziskovalnega projekta oziroma v primeru sprememb, povečanja ali zmanjšanja sestave projektne skupine v zadnjem letu izvajanja projekta, napišite obrazložitev. V primeru, da sprememb ni bilo, to navedite. Največ 6.000 znakov vključno s presledki (približno ena stran, velikost pisave 11). Nazaj

[5] Navedite znanstvene dosežke, ki so nastali v okviru tega projekta. Raziskovalni dosežek iz obdobja izvajanja projekta (do oddaje zaključnega poročila) vpišete tako, da izpolnite COBISS kodo dosežka – sistem nato sam izpolni naslov objave, naziv, IF in srednjo vrednost revije, naziv FOS področja ter podatek, ali je dosežek uvrščen v A'' ali A'. Nazaj

[6] Navedite družbeno-ekonomske dosežke, ki so nastali v okviru tega projekta. Družbeno-ekonomski rezultat iz obdobja izvajanja projekta (do oddaje zaključnega poročila) vpišete tako, da izpolnite COBISS kodo dosežka – sistem nato sam izpolni naslov objave, naziv, IF in srednjo vrednost revije, naziv FOS področja ter podatek, ali je dosežek uvrščen v A'' ali A'.

Družbeno-ekonomski dosežek je po svoji strukturi drugačen kot znanstveni dosežek. Povzetek znanstvenega dosežka je praviloma povzetek bibliografske enote (članka, knjige), v kateri je dosežek objavljen.

Povzetek družbeno-ekonomskega dosežka praviloma ni povzetek bibliografske enote, ki ta dosežek dokumentira, ker je dosežek sklop več rezultatov raziskovanja, ki je lahko dokumentiran v različnih bibliografskih enotah. COBISS ID zato ni enoznačen, izjemoma pa ga lahko tudi ni (npr. prehod mlajših sodelavcev v gospodarstvo na pomembnih raziskovalnih nalogah, ali ustanovitev podjetja kot rezultat projekta … - v obeh primerih ni COBISS ID). Nazaj

[7] Navedite rezultate raziskovalnega projekta iz obdobja izvajanja projekta (do oddaje zaključnega poročila) v primeru, da katerega od rezultatov ni mogoče navesti v točkah 6 in 7 (npr. ni voden v sistemu COBISS). Največ 2.000 znakov, vključno s presledki. Nazaj

[8] Pomen raziskovalnih rezultatov za razvoj znanosti in za razvoj Slovenije bo objavljen na spletni strani: http://sicris.izum.si/ za posamezen projekt, ki je predmet poročanja Nazaj

[9] Največ 4.000 znakov, vključno s presledki Nazaj

[10] Največ 4.000 znakov, vključno s presledki Nazaj

[11] Rubrike izpolnite / prepišite skladno z obrazcem "izjava sofinancerja" http://www.arrs.gov.si/sl/progproj/rproj/gradivo/, ki ga mora izpolniti sofinancer. Podpisan obrazec "Izjava sofinancerja" pridobi in hrani nosilna raziskovalna organizacija – izvajalka projekta. Nazaj

[12] Navedite en izjemni znanstveni dosežek in/ali en izjemni družbeno-ekonomski dosežek raziskovalnega projekta v letu 2014 (največ 1000 znakov, vključno s presledki). Za dosežek pripravite diapozitiv, ki vsebuje sliko ali drugo slikovno gradivo v zvezi z izjemnim dosežkom (velikost pisave najmanj 16, približno pol strani) in opis izjemnega dosežka (velikost pisave 12, približno pol strani). Diapozitiv/-a priložite kot priponko/-i k temu poročilu. Vzorec diapozitiva je objavljen na spletni strani ARRS http://www.arrs.gov.si/sl/gradivo/, predstavitve dosežkov za pretekla leta pa so objavljena na spletni strani http://www.arrs.gov.si/sl/analize/dosez/. Nazaj

Obrazec: ARRS-RPROJ-ZP/2015 v1.00a
77-92-AE-97-69-37-D3-ED-43-D3-4C-6F-C8-D7-7C-48-B7-96-99-22

Priloga 1

Elsevier Editorial System(tm) for Journal of Hazardous Materials
Manuscript Draft

Manuscript Number:

Title: Adsorption and degradation processes of tributyltin and trimethyltin in landfill leachates treated with iron nanoparticles

Article Type: Research Paper

Keywords: Landfil leachates
Tributyltin
Trimethyltin
Iron nanoparticles
Adsorption degradation processes

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Corresponding Author's Institution: Jožef Stefan Institute

First Author: Kelly Peeters, M.Sc.

Order of Authors: Kelly Peeters, M.Sc.; Gaëtane Lespes, Prof.dr.; Janez Ščančar, Prof.dr.; Radmila Milačič, Ph.D.

Abstract: Adsorption and degradation of tributyltin (TBT) and trimethyltin (TMeT) in landfill leachate treated with iron nanoparticles (FeNPs): nZVI, FeONPs and Fe3O4NPs was investigated to find conditions for their efficient removal. One sample aliquot was kept untreated (pH 8), while to the others (pH 8) FeNPs dispersed with tetramethyl ammonium hydroxide (TMAH) or by mixing were added and samples shaken under aerated conditions for 3 to 7 days. The same experiments were done in leachates acidified to pH 3 with citric acid. Size distribution of TBT and TMeT was determined by sequential filtration and their concentrations in a given fraction (> 5000 nm, 450 - 5000 nm, 2.5 - 450 nm, < 2.5 nm) by GC-ICP-MS. Majority of TBT or TMeT was present in fractions with particles > 2.5 or < 2.5 nm, respectively. At pH 8 adsorption of TBT to FeNPs prevailed, while at pH 3, the Fenton reaction provoked degradation of TBT by hydroxyl radicals. TBT was the most effectively removed (96%) by sequential treatment of leachate with nZVI (dispersed by mixing) first at pH 8, followed by nZVI treatment of previously acidified aqueous phase (pH 3) with citric acid. Such treatment less effectively removed TMeT (40%). Since TMAH provoked methylation of tin, mixing was recommended for dispersion of nZVI.

Dear Editor,
Please find enclosed the manuscript entitled "Adsorption and degradation processes of tributyltin and trimethyltin in landfill leachates treated with iron nanoparticles" by Kelly Peeters, Gaëtane Lespes, Janez Ščančar and Radmila Milačič.

The importance of the work:
Biotic and abiotic degradation of toxic organotin compounds (OTCs) in landfill leachates is usually not complete. In this work adsorption and degradation processes of tributyltin (TBT) and trimethyltin (TMeT) in leachate sample treated with different iron nanoparticles (FeNPs): Fe^0 (nZVI), FeO and Fe_3O_4 were investigated to find conditions for their efficient removal. One sample aliquot was kept untreated (pH 8), while to the others (pH 8) FeNPs dispersed with tetramethyl ammonium hydroxide (TMAH) or by mixing were added and samples shaken under aerated conditions for 7 days. The same experiments were done in leachates in which the pH was adjusted to 3 with citric acid. Size distribution of TBT and TMeT between particles > 5 µm, 0.45 - 5 µm, 2.5 nm - 0.45 µm, and < 2.5 nm was determined by sequential filtration and their concentrations in a given fraction by gas chromatography coupled to inductively coupled plasma mass spectrometry (GC-ICP-MS). Results revealed that most of the TBT or TMeT was present in fractions with particles > 2.5 or < 2.5 nm, respectively. At pH 8 adsorption of TBT to FeNPs prevailed, while at pH 3, the Fenton reaction provoked degradation of TBT by hydroxyl radicals. TBT was the most effectively removed (96 %) when sequential treatment of leachate with nZVI (dispersed by mixing) was applied first at pH 8, followed by nZVI treatment of the aqueous phase, previously acidified to pH 3 with citric acid. Such treatment less effectively removed TMeT (about 40 %). It was proven that TMAH provoked methylation of tin, so mixing was recommended for dispersion of nZVI.

The corresponding author Prof.Dr. Radmila Milačič has long lasting scientific experiences (more than 35 years) on the investigations of cycling of elements in the environment and their role in biological systems. Her and her co-workers special concern is development of reliable analytical procedures for speciation of elements (Cr, Al, Sn, Zn, Pt, Ru, Ni, Br) combining chromatographic techniques hyphenated to inductively coupled plasma mass spectrometry and the use of stable isotopes to follow species transformation in environmental compartments as well as for accurate quantification of their concentrations by species specific and species unspecific isotope dilution mass spectrometry. One of her major interests is investigation of the environmental impacts of the use of nanoparticles for the remediation of the environmental waters and soils. She has published more than 130 scientific papers in renowned scientific journals and 7 book chapters. She is also Editor of one book (Springer). Her work has been citied more than1400 times (pure citations), her Hirsch index h = 25.

The manuscript deals with actual topics on the use of nanoparticles for the remediation of polluted waters and provides the following new scientific achievement:

- useful information on the behaviour of OTCs after the treatment of landfill leachate with FeNPs
- novel approach, which considers the chemical speciation of OTCs and their size distribution before and after the FeNPs treatment
- new important finding is, that when TMAH is used for dispersion of nZVI, it provokes methylation of soluble ionic tin in the leachate.

Authors estimate that the manuscript is of great interest for the journal readers, and hope that it will be considered suitable for publication as a scientific article in Journal of Hazardous Materials.

Sincerely yours,
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*Novelty Statement

Adsorption and degradation processes of TBT and TMeT in landfill leachate treated with nZVI, FeO and Fe_3O_4 were investigated. A novel approach, which considers chemical speciation of OTCs and their size distribution before and after the FeNPs treatment was applied. At pH 8 adsorption of TBT to FeNPs prevailed, while at pH 3 (acidified with citric acid), Fenton reaction provoked degradation of TBT. TBT was the most effectively removed (96%) by sequential treatment of leachate with nZVI first at pH 8, followed by nZVI treatment of previously acidified aqueous phase with citric acid. Such treatment less effectively removed TMeT (40%).

Research highlights

- The removal of TBT and TMeT from landfill leachate by the use of FeNPs was studied
- nZVI, FeONPs and Fe_3O_4NPs were applied at pH 8 and pH 3 (adjusted with citric acid)
- Size distribution and speciation of OTCs was studied before and after NPs treatment
- Combined treatment with nZVI at pH 8 and pH 3 effectively removed TBT, but not TMeT
- If TMAH is used for dispersion of nZVI it provokes methylation of Sn in the leachate

Adsorption and degradation processes of tributyltin and trimethyltin in landfill leachates treated with iron nanoparticles

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ABSTRACT

Adsorption and degradation of tributyltin (TBT) and trimethyltin (TMeT) in landfill leachate treated with iron nanoparticles (FeNPs): nZVI, FeONPs and Fe_3O_4NPs was investigated to find conditions for their efficient removal. One sample aliquot was kept untreated (pH 8), while to the others (pH 8) FeNPs dispersed with tetramethyl ammonium hydroxide (TMAH) or by mixing were added and samples shaken under aerated conditions for 3 to 7 days. The same experiments were done in leachates acidified to pH 3 with citric acid. Size distribution of TBT and TMeT was determined by sequential filtration and their concentrations in a given fraction (> 5000 nm, 450 - 5000 nm, 2.5 – 450 nm, < 2.5 nm) by GC-ICP-MS. Majority of TBT or TMeT was present in fractions with particles > 2.5 or < 2.5 nm, respectively. At pH 8 adsorption of TBT to FeNPs prevailed, while at pH 3, the Fenton reaction provoked degradation of TBT by hydroxyl radicals. TBT was the most effectively removed (96%) by sequential treatment of leachate with nZVI (dispersed by mixing) first at pH 8, followed by nZVI treatment of previously acidified aqueous phase (pH 3) with citric acid. Such treatment less effectively removed TMeT (40%). Since TMAH provoked methylation of tin, mixing was recommended for dispersion of nZVI.

Keywords:
Landfil leachates
Tributyltin
Trimethyltin
Iron nanoparticles
Adsorption degradation processes

1. Introduction

Organotin compounds (OTCs) are highly toxic even at ng L^{-1} concentration levels [1]. Due to their wide use in industrial and agricultural applications, they are globally present as pollutants in the environment [2-13]. The municipal waste commonly dumped on landfill sites is an important pool of OTCs that originates from household products, plastic materials and textiles, and inorganic tin from metallic cans [11-13]. Both, organic and inorganic tin species can be mobilised and released to the environment by percolation of water through the waste pile.

Tributilytin (TBT) is one of the most toxic OTCs and is, despite its global ban (International Convention on the control of harmful anti-fouling systems on ships) [14], still present in the marine[8] and terrestrial environment [15], while trimetyltin (TMeT) is neurotoxic [16] and occurs in notable concentrations in landfill leachates [9-13]. Under landfill conditions tin is undergoing chemical and biological transformation[13,17]. In this way, tin species present in waste can be modified by alkylation (i.e. hydridation, methylation,

ethylation), dealkylation or transalkylation reactions, resulting in the generation of new tin compounds in liquid and gaseous phases [11].

Additional management of municipal solid wastes is required to prevent the release of pollutants from their treatment and disposal facilities. In 1999, the European Commission ratified a Council Directive on the ‘Landfill of Waste’, with the aim to reduce the negative effects of pollutants on the environment [18]. To prevent environmental pollution, researchers are looking for solutions to remove OTCs from contaminated waters such as by flocculation and/or the use of various adsorbents. Recently, nanoparticles (NPs) have received significant interest for environmental remediation [19]. Among different metal NPs based remediation treatments, this one involving iron nanoparticles (FeNPs) is an attractive option as iron is inexpensive, non-toxic and environmentally compatible [20-24]. FeNPs are magnetic nanoparticles with large surface area. They have proven to be useful for adsorbing, reductively transforming or degrading numerous types of organic and inorganic environmental contaminants [19, 21-24]. At neutral to alkaline pHs, adsorption of pollutant onto the surface of FeNPs and co-precipitation with iron corrosion products is the main process of pollutant removal [21]. After remediation, iron precipitate can be simply taken away by a magnet. At acidic pHs, in aerated solutions containing dissolved Fe^{2+} and organic ligands like citrate, oxalate or EDTA, Fenton-type reaction takes place through a series of ligand-mediated reactions, resulting in formation of H_2O_2 and highly reactive hydroxyl ($OH^\bullet$) radicals. $OH^\bullet$ radicals are strong oxidants, capable to rapidly degrade many organic and inorganic compounds [25-31]. To initiate Fenton-type reaction with FeNPs, citrate can be used as an environment-friendly alternative for toxic EDTA [32]. Iron forms stable complexes with citrate that generate the superoxide radical $O_2^{\bullet-}$, which by further reaction with Fe-complexes produces H_2O_2. H_2O_2 is a source for the Fenton process that yields $OH^\bullet$ radicals.

The aim of our work was to investigate the processes of adsorption and degradation of TBT and TMeT in landfill leachates treated with different FeNPs: nanozerovalent iron (nZVI), FeONPs and Fe_3O_4NPs. Representative leachate sample was treated at different pH in order to promote either adsorption onto particles that can be removed from the leachate, or degradation by Fenton-type reaction. In order to investigate the association of OTCs with particles of operationally defined size present in the leachate, sequential filtration was applied and the concentration of OTCs in a given fraction determined by gas chromatography-inductively coupled plasma mass spectrometry (GC-ICP-MS).

2. Materials and methods

2.1. Chemicals and samples

Stabilized magnetic nZVI (manufactural size information approximately 25 nm) were obtained from MKNano, (Williamsville, NY USA), while the magnetic FeONPs stabilized in heptane (particle size 3.5 - 9.5 nm), magnetic Fe_3O_4 nanopowder (particle size 50 - 100 nm) and tertramethylammonium hydroxide (TMAH) (25 wt% in methanol) used for dispersing of FeNPs came from Sigma-Aldrich (Steinheim, Germany). Citric acid monohydrate (p.a.) used for adjusting of pH of the leachate was supplied from Merck (Darmstadt, Germany). Tributyltinchloride (TBTCl, 96%) was purchased from Sigma-Aldrich and trimethyltin chloride (TMeTCl, 99%) from Acros Organics (New Jersey, NY, USA). For sequential filtration of leachate samples, the 5 µm cellulose-nitrate, 0.45 µm cellulose-acetate membrane filters and 10 KDa regenerated cellulose filters (2.5 nm) were obtained from Whatman (Vienna, Austria), Ministart (Goettingen, Germany) and Amicon (Zagreb, Croatia), respectively. To estimate the influence of microorganisms on transformation of OTCs in landfill leachate, an aliquot of the leachate sample was sterilized for 2 hours at 121 °C in autoclave Getinge GE6610EM-2 (Getinge, Sweden). The reagents used for the speciation analysis of OTC are described elsewhere [12].

2.2. Speciation analysis

Speciation analysis was carried out under an optimised procedure for the determination of OTCs in landfill leachates previously developed and validated in our laboratory [12]. It was applied in all the size fractions of the leachate samples in order to have a complete mass balance, for showing in particular if OTCs were degraded or adsorbed on iron NPs. In the extraction step, methanol was used as co-extraction reagent and Tris-citrate buffer for adjusting the pH to 6. $NaBEt_4$ was used for derivatization. Ethylated OTCs were extracted into 2 mL of hexane and speciation analysis performed by GC-ICP-MS.

2.3. Preparation of FeNPs dispersions

For efficient dispersing of FeNPs (nZVI, FeONPs or Fe_3O_4NPs), 20 mL of TMAH (25 wt% in methanol) was added to about 0.5 g of FeNPs, the suspension sonicated for 10 min, shaken for 1 h, and FeNPs removed with a magnet [33]. FeNPs treated with TMAH were then added to leachates spiked with OTC. Alternatively, FeNPs were added to the spiked leachates and dispersion obtained by mixing of contents with a mixer for 15 min. The amount of FeNPs added to a spiked leachate corresponded to concentration of 1.5 ± 0.1 g L^{-1} Fe.

2.4. Experimental design

Representative samples of landfill leachates were obtained from non-hazardous Barje municipal waste landfill, Ljubljana, Slovenia. 400 mL aliquots of landfill leachate were transferred to 500 mL amber glass bottles and spiked with TBT or TMeT (1000 ng L^{-1} Sn). Bottles were capped with glass stoppers and samples shaken for 48 hours to allow spiked TBT or TMeT equilibration. 60 mL sample portion of aliquots was used for OTCs analyses, while other 60 mL portions were transferred to 250 mL amber glass bottles and treated with FeNPs at pH 8 (the pH of the spiked leachate) or the pH of the leachate was first adjusted to 3 with citric acid (1.5 g of citric acid per 60 mL of leachate) and then treated with dispersed FeNPs (iron load 1.5 g L^{-1}). Bottles were capped with glass stoppers and samples shaken for 3 to 7 days. In leachates treated at pH 8, the iron precipitate was collected by a magnet, allowing its quantitative removal (more than 99%) by decanting the clear leachate solution into another beaker. Before untreated leachates and leachates treated with FeNPs were analysed, sequential filtration of samples through 5000 nm, 450 nm and 2.5 nm filters was applied. 10 mL leachate fractions with particle sizes > 5000 nm, 450 - 5000 nm, 2.5 nm - 450 nm, < 2.5 nm were transferred to 30 mL amber glass vials and concentrations of OTCs determined by GC-ICP-MS. All experiments were carried out in duplicate.

3. Results and discussion

It was hypothesized that at basic pH the OTCs studied will be removed by adsorption on the surface of FeNPs and co-precipitation with iron corrosion products, while at acidic pH, the degradation of OTCs by $OH^{\bullet}$ radicals, which are formed under aerated conditions in the presence of citric acid and dissolved Fe^{2+}, will occur. It is expected that Fenton type reaction will take place through a series of reactions, which can be recorded by the below equations (1-5) as proposed by Gylienė et al. [26], where L represents citrate ligand.

$$Fe^{0} \rightarrow Fe^{2+} + 2\,e^{-} \qquad (1)$$

$$Fe^{2+} + L \rightarrow Fe^{2+}L \qquad (2)$$

$$Fe^{2+}L + O_2 \rightarrow Fe^{3+}L + O_2^{\bullet-} \qquad (3)$$

$$Fe^{2+}L + O_2^{\bullet-} + 2H^{+} \rightarrow Fe^{3+}L + H_2O_2 \qquad (4)$$

$$Fe^{2+}L + H_2O_2 \rightarrow Fe^{3+}L + OH^{\bullet} + OH^{-} \qquad (5)$$

The pH is a key parameter in Fenton-like processes. As the pH rises, the production of $OH^{\bullet}$ radicals is decreased, due to formation of iron corrosion product precipitates [30].

To confirm these hypotheses adsorption and degradation processes of TBT and TMeT were investigated in landfill leachate samples spiked with TBT and TMeT (1000 ng L^{-1} Sn). Their concentrations were similar to those observed at landfill sites around Europe [9,11]. Spiked samples were treated at pH 8 (the pH of leachate) and pH 3 (adjusted with citric acid) with nZVI, FeONPs and Fe_3O_4NPs. Further inside to adsorption and degradation processes of TBT and TMeT was achieved by size fractionation of samples. For this purpose sequential filtration of untreated leachates and landfill leachates treated with FeNPs was applied and concentrations of OTC determined by GC-ICP-MS.

For investigation of removal of OTCs by adsorption at pH 8, the leachate and the iron precipitate were analysed after the FeNPs treatment and data compared to untreated leachate.

To investigate the degradation of OTCs at pH 3, the leachate treated with citric acid and FeNPs was analysed and data compared to untreated leachate. At pH 3 FeNPs do not form iron corrosion products, while adsorption on NPs is inhibited due to the presence of H^+ ions.

3.1. Adsorption and degradation of TBT and TMeT in spiked landfill leachates treated with nZVI, FeONPs and Fe_3O_4NPs

The potential of these different FeNPs for TBT and TMeT removal from landfill leachate at pH 8 and 3 was studied. The experiments were carried out under the conditions described in paragraph Experimental design. TMAH was used for efficient dispersing of FeNPs, and treated leachate was shaken for 7 days. 7 days was chosen to ensure the efficient treatment.

The results obtained with nZVI for TBT and TMeT are presented on Fig. 1 in left and right column, respectively.

Insert Fig. 1. about here

In untreated leachate at pH 8 (Fig. 1A, left), about 20% of the total TBT (concentration in the leachate 2 days after spiking 812 ng L^{-1} Sn) degraded into dibutyltin (DBT) and monobutyltin (MBT). These data are in accordance with our previous findings on biotic degradation of TBT in leachates [13]. In addition, the TBT is not dissolved but is found mainly distributed among fractions containing particles of 2.5 - 450 nm (about 55%) and > 5000 nm (30%).

After nZVI treatment at pH 8 (Fig. 1.B, left), the total TBT concentration was reduced to 260 ng L^{-1} Sn. Furthermore, the amounts of TBT associated with leachate particles > 5000 nm and in the fraction 2.5 nm - 450 nm significantly decreased compared to the untreated leachate, of about 20 and 2 times, respectively. This decrease was not associated to an increase of DBT and MBT concentrations, the total concentration of DBT being even decreased for about 65% compared to the untreated leachate. The analysis of OTCs in the precipitate confirmed the presence of TBT, DBT and MBT. The sum of concentrations of OTCs, which remained in the solution and OTCs adsorbed onto the iron precipitate after the nZVI treatment, was the same as the concentration of untreated leachate after 7 days. All of this indicates that TBT was removed by the adsorption onto the surface of nZVI and co-precipitated with iron corrosion products.

When the nZVI treatment was performed at pH of 3 (Fig. 1C, left), about 17% of the total TBT initially present in the untreated leachate was found, and only in the fraction 2.5 nm - 450 nm. In addition, the DBT total concentration slightly increased in comparison to the untreated leachate, while the MBT total concentration was slightly decreased. The present observations indicate that TBT mainly degraded by successive debutylations, which is most likely a consequence of formation of $OH^{\bullet}$ radicals via Fenton-type reaction in the presence of nZVI and citric acid. In comparison to adsorption process by nZVI treatment at pH 8, degradation of TBT more effectively removes this contaminant from the leachate.

In the untreated leachate at pH 8, the total concentration of methyltins after spiking with TMeT (1000 ng L^{-1} Sn) was 1475 ng L^{-1} Sn, since TMeT (about 300 ng L^{-1} Sn) and smaller amounts of dimethyltin (DMeT) and monomethyltin (MMeT) (less than 100 ng L^{-1} Sn, each) were originally present in the leachate analysed. After spiking, 90% of TMeT was present in leachate fraction with particles smaller than 2.5 nm and 10% in fraction 2.5 nm - 450 nm (Fig. 1A, right). After the nZVI treatment at pH 8 (Fig. 1B, right), the total TMeT concentration was reduced for about 20% due to the adsorption of TMeT onto the surface of nZVI, which was confirmed by the analysis of methyltins in the precipitate. When spiked leachate was treated with nZVI at pH 3 (Fig. 1C, right), the TMeT originally present in fraction with particles 2.5 nm - 450 nm disappeared by degradation. The remaining TMeT in fraction with particles < 2.5 nm represented about 75% of its total content in spiked leachate. Hence, the degradation of TMeT at pH 3 was less effective than that of TBT. Our experimental data revealed that TMeT was hardly degradable species, as are some other organic compounds [25].

The same experiments as with nZVI were performed to examine the potential of TBT and TMeT removal from landfill leachate by the use of FeONPs and Fe_3O_4NPs at pH 8 and 3. TMAH was used for dispersion of FeNPs. The results presented in Figs. S1 and S2 (Supplementary) demonstrate similar extent of TBT removal at pH 8 and 3 with both FeONPs and Fe_3O_4NPs, as when the leachate was treated with nZVI. Also, the level of TMeT removal governed by adsorption (pH 8), and degradation (pH 3) is similar for all three FeNPs investigated. So, in the following experiments only nZVI was used.

3.2. Sequential removal of TBT and TMeT in landfill leachate treated with nZVI

In order to further improve the removal of TBT and TMeT, a two-step sequential leachate treatment was investigated, combining adsorption and degradation processes with nZVI. nZVI was dispersed with TMAH. The sequential procedure was planned so, that the treatment time did not exceed duration of 7 days. Leachate samples spiked with TBT or TMeT were first treated with nZVI at pH 8 and shaken for 3 days. Iron precipitate was separated from the leachate by a magnet. Then, in one sample aliquot sequential filtration was applied, while in another, the pH was adjusted to pH 3 with citric acid. Sample was again treated with nZVI, shaken for additional 3 days and sequential filtration performed prior to the determination of OTCs in different fractions. Such a procedure allows to estimate the efficiency of OTCs removal in each step.

The results of these experiments are presented in Fig. 2, where graphs A for TBT (left) and TMeT (right) represent the untreated leachate at pH 8.

Insert Fig. 2. about here

The efficiency of TBT removal at pH 8 is similar when leachate is treated with nZVI for 3 days (Fig. 2B, left) or for 7 days (Fig. 1B, left), meaning that also the shorter treatment time of 3 days is adequate. Data of Fig. 2C (left) further indicate that sequential treatment of leachate, combining adsorption-precipitation (pH 8) and degradation (pH 3), more effectively removed TBT (the extent of TBT removal about 91%) than when leachate is treated by nZVI only at pH 3 (the extent of TBT degradation about 83%, Fig. 1C, left). For TMeT (Fig. 2B and 2C, right), it is evident that sequential treatment with nZVI has almost no influence on the efficiency of its removal from the leachate.

3.3. Investigation of the degradation processes

In order to confirm that the degradation of the OTCs analysed at pH 3 is not biotic but primarily abiotic process governed by the Fenton chemistry, additional experiments were carried out. The knowledge on degradation processes significantly contribute to accomplish efficient removal of OTCs from leachate. Leachate samples spiked with TBT or TMeT were first treated with nZVI (dispersed with TMAH) at pH 8. After iron removal one sample aliquot was sterilized, while the other was kept non-sterilized. To sterilized and non-sterilized samples, citric acid was added or sequential treatment (first addition of citric acid then addition of nZVI) was applied. Results are presented in Fig. 3, where graph A represents the untreated leachate spiked with TBT at pH 8, and graph B leachate spiked with TBT, treated with nZVI at pH 8.

Insert Fig. 3. about here

When only citric acid was added (Figs. 3C1 and 3C2) substantial degradation of TBT was observed. It should be pointed out that the amount of iron in the dissolved and nanosized fractions, which remained after the removal of nZVI precipitate in the leachate (about 1.5 mg L^{-1} Fe), was high enough to provoke degradation of TBT in the presence of citric acid and dissolved oxygen. It was also experimentally proven that citric acid alone is not able to degrade TBT in aqueous solution, which does not contain iron (these data are not shown). In sterilized leachate samples (Fig. 3C1) almost 99% of TBT was removed, and in non-sterilized samples (Fig. 3C2) about 88%. The less effective degradation of TBT in non-sterilized samples is probably related to microbial degradation of citric acid [34], thus decreasing its content available for Fenton chemistry. The same extent of TBT removal (98%) as in sterilized leachate samples with the addition of citric acid (Fig. 3C1) was observed in sterilized leachate, when after the addition of citric acid, samples were treated also with nZVI (Fig. 3D1). TBT removal in non-sterilized samples, after the sequential nZVI treatment at pH 8 and pH 3, was slightly more effective (about 92%) (Fig. 3D2) than in samples to which, after the nZVI treatment at pH 8 (Fig. 3C2), only citric acid was added. In sterilized samples not only TBT, but also DBT and MBT were more effectively degraded than in non-sterilized samples.

Data presented in Fig. 4 exhibit the same extent of TMeT removal in sterilized and non-sterilized samples (about 35%) for both, when leachate was, after the nZVI treatment at pH 8, treated only with citric acid or with citric acid and nZVI.

Insert Fig. 4. about here

The experiments described above confirmed that under aerated conditions in the leachate the degradation of TBT and TMeT treated with nZVI in the presence of citric acid (pH 3) is governed primarily by the Fenton process.

To check whether citric acid alone (without addition of nZVI) provokes the degradation of OTCs in leachate, samples were spiked with TBT or TMeT, citric acid added and samples shaken under the aerated conditions for 3 days. The concentration of dissolved and nanosized iron originally present in the leachate was about 1.5 mg L^{-1} Fe. The results presented in Fig. S3 (Supplementary) indicate that about 75% of TBT was degraded after the addition of citric acid. Significant increase in DBT concentration, in the amount equal to the content of TBT degraded, was observed. By comparing these data with the results obtained when leachate was sequentially treated with nZVI (pH 8) followed by treatment with citric acid and nZVI (pH 3) (Fig. 3D2), the nZVI treatment is, regarding the TBT degradation much more effective, and effectively degrades also DBT. The results presented in Fig. S3 (Supplementary) further demonstrated that addition of citric acid to the leachate has no influence on TMeT degradation.

3.4. *The influence of TMAH used for dispersion of FeNPs on methylation processes in the leachate.*

In the experiments where leachate samples were spiked with TMeT and the pH adjusted to 3 with citric acid, followed by treatment with FeNPs using TMAH for dispersion, formation of MMeT was observed (see data of Figs. 1C, 2C, 4D). The same phenomenon of the transformation of methyltin species, originally present in the leachate, was also observed in samples spiked with TBT at pH 3, after the treatment of the leachate with nZVI (Fig. S4C, Supplementary). The concentration of MMeT formed in the nZVI treated leachate was about 110 ng L^{-1} Sn (Compare the data of Figs. S4C and S4A, Supplementary). To find out whether MMeT appeared as a consequence of TMeT degradation or is a species newly formed by abiotic methylation of inorganic tin (tin concentration in leachate was about 80 µg L^{-1}), new sets of experiments were performed. Leachate samples were spiked with TBT or TMeT and the sequential treatment of the leachate with nZVI carried out at pH 8 and 3. nZVI were dispersed by mixing, or TMAH was used as dispersing agent. In these experiments, the efficiency of TBT and TMeT degradation by the use of different modes of dispersion was also tested. The results are presented in Fig. 5.

Insert Fig. 5. about here

From data of Fig. 5, it is evident that degradation of TBT and TMeT is more effective when nZVI is dispersed by mixing. As a result of higher reactivity of nanoparticles, 96% of TBT and more than 90% of DBT and MBT was removed by sequential treatment of leachate with nZVI when mixing was applied for dispersion (Fig. 5B, left). When TMAH was used as dispersing agent about 90%, 50% and 60% of TBT, DBT and MBT, respectively were removed (Fig. 5C, left). Regarding methyltins removal, about 40% of TMeT, 45% of DMeT and 90% of MMeT was removed when mixing was used for dispersion (Fig. 5B, right). Although TMAH more efficiently disperses nZVI, it also modifies the surface of nanoparticles so that they became less reactive. The decrease in nZVI reactivity was observed also in our previous work in silica coated and choline modified nZVI [35].

When nZVI was dispersed by TMAH about 30% of TMeT was removed from leachate, while DMeT and MMeT were not removed, but formed in concentration of about 60 and 100 ng L^{-1} Sn, respectively (Fig. 5C, right). By comparing data of Figs. 5B and 5C (right), it is clearly evident that TMAH in the presence of citric acid provokes abiotic methylation. Such abiotic methylation was also observed by other investigators for methylation of mercury during its extraction procedure [36]. To avoid abiotic formation of highly toxic methylmercury and methyltin species in the leachate, the use of TMAH for dispersing of nZVI in the presence of citric acid should be omitted.

4. Conclusions

Data of the present investigation demonstrated that the efficiency of removal of TBT and TMeT from the leachate by the use of nZVI, FeONPs and Fe_3O_4NPs depends on the conditions, under which the remediation procedure is performed. Results from the determination of OTCs in leachate fractions with particle sizes > 5000 nm, 450 – 5000 nm, 2.5 - 450 nm, and < 2.5 nm revealed that TBT was associated with particles bigger than 2.5 nm, while TMeT was present mainly in fraction with particle sizes less than 2.5 nm. At pH 8 adsorption of TBT to FeNPs prevailed, while at pH 3 (adjusted with citric acid), the Fenton reaction provoked degradation of TBT by hydroxyl radicals. In the leachate spiked with TBT and TMeT (1000 ng L^{-1} Sn), TBT was the most effectively removed (96%) when sequential treatment of leachate with nZVI (dispersed by mixing) was applied first at pH 8, followed by nZVI treatment of the aqueous phase, previously acidified to pH 3 with citric acid. For TMeT, which is a hardly degradable species, the same treatment was less effective and removed only about 40% of TMeT from the leachate. It was proven that TMAH, which is commonly used as dispersing agent, provoked methylation of ionic tin species. Due to this reason, mixing is recommended for dispersion of nZVI.

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Caption to Figures

Fig. 1. Size distribution of OTCs in landfill leachates. (A) Untreated leachates (pH 8) were spiked with TBT (1000 ng L^{-1} Sn) or TMeT (1000 ng L^{-1} Sn). (B) Spiked leachates (pH 8) were treated with nZVI. (C) The pH of spiked leachates was adjusted to 3 with citric and sample treated with nZVI. nZVI was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

Fig. 2. Size distribution of OTCs in landfill leachates. (A) Untreated leachates (pH 8) were spiked with TBT (1000 ng L^{-1} Sn) or TMeT (1000 ng L^{-1} Sn). (B) Spiked leachates (pH 8) were treated with nZVI. (C) Spiked leachates (pH 8) were treated first with nZVI, iron precipitate was removed, the pH adjusted to 3 with citric acid and sample again treated with nZVI. nZVI was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

Fig. 3. Size distribution of butyltins in landfill leachates. (A) Untreated leachates (pH 8) were spiked with TBT (1000 ng L^{-1} Sn). (B) Spiked leachates (pH 8) were treated with nZVI. (C) Spiked leachates (pH 8) were treated first with nZVI, iron precipitate was removed and then the pH of sterilized samples (1) and non-sterilized samples (2) adjusted to 3 with citric acid. (D) Spiked leachates (pH 8) were treated first with nZVI, iron precipitate was removed and then the pH of sterilized samples (1) and non-sterilized samples (2) adjusted to 3 with citric acid and samples again treated with nZVI. nZVI was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

Fig. 4. Size distribution of methyltins in landfill leachates. (A) Untreated leachates (pH 8) were spiked with TMeT (1000 ng L^{-1} Sn). (B) Spiked leachates (pH 8) were treated with nZVI. (C) Spiked leachates (pH 8) were treated first with nZVI, iron precipitate was removed and then the pH of sterilized samples (1) and non-sterilized samples (2) adjusted to 3 with citric acid. (D) Spiked leachates (pH 8) were treated first with nZVI, iron precipitate was removed and then the pH of sterilized samples (1) and non-sterilized samples (2) adjusted to 3 with citric acid and samples again treated with nZVI. nZVI was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

Fig. 5. Size distribution of OTCs in landfill leachates. (A) Untreated leachates (pH 8) were spiked with TBT (1000 ng L^{-1} Sn) or TMeT (1000 ng L^{-1} Sn). (B) Spiked leachates (pH 8) were treated first with nZVI, iron precipitate was removed, the pH of sample was adjusted to 3 with citric acid and samples again treated with nZVI. nZVI was dispersed by mixing. (C) Spiked leachates (pH 8) were treated first with nZVI, iron precipitate was removed, the pH of sample adjusted to 3 with citric acid and samples again treated with nZVI. nZVI was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

References

[1] K. Fent, Organotin compounds in municipal wastewater and sewage sludge: contamination, fate in treatment process and ecotoxicological consequences, Sci. Tot. Environ. 185 (1996) 151-159.

[2] M. Hoch, Organotin compounds in the environment – an overview, Appl. Geochem. 16 (2001) 719-743.

[3] S. Díez, M. Ábalos, J.M. Bayona, Organotin contamination in sediments from the Western Mediterranean enclosures following 10 years of TBT Regulation, Water Res. 36 (2002) 905-918.

[4] P.J. Craig, G. Eng, R.O. Jenkins, Occurrence and pathways of organometallic compounds in the environment – general considerations, In Organometallic Compounds in the Environment. Ed. P.J. Craig, John Wiley & Sons, Ltd, Chichester (2003) 1-55.

[5] E. Rosenberg, Speciation of Tin. In Handbook of Elemental Speciation II – Species in the Environment, Food, Medicine and Occupational Health, Eds. R. Cornelis, J. Caruso, H. Crews, K. Heumann, John Wiley & Sons, Ltd, Chichester (2005) 422-464.

[6] T. Milivojevič Nemanič, R. Milačič, J. Ščančar, A Survey of Organotin Compounds in the Northern Adriatic Sea, Water Air Soil Pollut. 196 (2009) 211-224.

[7] R.C. de Carvalho Oliveira, R.E. Santelli, Occurrence and chemical speciation analysis of organotin compounds in the environment: A review, Talanta 82 (2010) 9-24.

[8] M. Furdek, M. Vahčič, J. Ščančar, R. Milačič, G. Kniewald, N. Mikac, Organotin compounds in seawater and *Mitilus galloprovioncialis* mussels along the Croatian Adriatic Coast, Marine Pollut. Bullet. 64 (2012) 189-199.

[9] I. Merisiowsky, R. Brandsch, J. Ejlertsson, Organic compounds in the environment: screening for organotin compounds in European landfill leachates, J. Environ. Qual. 30 (2001)1604-1611.

[10] B. Michalzik, G. Ilgen, F. Hertel, S. Hantsch, B. Bilitewski, Emissions of organo-metal compounds via the leachate and gas pathway from two differently pre-treated municipal waste materials - A landfill reactor study, Waste Manage. 27 (2007) 497-509.

[11] P. Pinel-Raffaitin, D. Amouroux, I. LeHécho, P. Rodríguez-Gonzalez, M. Potin-Gautier, Occurrence and distribution of organotin compounds in leachates and biogases from municipal landfills, Water Res. 42 (2008) 987-996.

[12] M. Vahčič, R. Milačič, J. Ščančar, Development of analytical procedure for the determination of methyltin, butyltin, phenyltin and octyltin compounds in landfill leachates by gas chromatography-inductively coupled plasma mass spectrometry, Anal. Chim. Acta 694 (2011) 21-30.

[13] K. Peeters, T. Zuliani, J. Ščančar, R.Milačič, The use of isotopically enriched tin tracers to follow the transformation of organotin compounds in landfill leachate, Water Res. 53 (2014) 297-309.

[14] Regulation (EC) No. 782/2003 of the European parliament and of the council of 14 April 2003 on the prohibition of organotin compounds on ships, Official Journal of the European Union 115 (2003) 1-11.

[15] P.V Silva, A.R. Silva, S. Mendo, S. Loureiro, Toxicity of tributyltin (TBT) to terrestrial organisms and its species sensitivity distribution, Sci Tot. Environ. 466-467 (2014) 1037-1046.

[16] N.Q Huong, Y. Nakamura, N. Kuramoto, M. Yoneyama, R. Nagashima, T. Shiba, T. Yamaguchi, S. Hasebe, K. Ogita, Indomethacin ameliorates trimethyltin-induced neuronal damage in vivo by attenuating oxidative stress in the dentate gyrus of mice, Biol. Pharm. Bull. 34 (2011) 1856-63.

[17] A.S. Stasinakis, N.S. Thomaidis, A. Nikolaou, A. Kantifes, Aerobic biodegradation of organotin compounds in activated sludge batch reactors, Environ. Pollut. 134 (2005) 431-438.

[18] Council Directive 1999/31/EC of 26 April 1999 on the landfill of waste, Official Journal of the European Communities, OJ L 182 (1999) 1-19.

[19] S.H. Joo, I.F. Cheng, Nanotechnology for environmental remediation, Springer (2006) 41-96.

[20] S. Krajangpan, B.J. Chisholm, H. Kalita, A.N. Bezbaruah, Challenges in groundwater remediation with iron nanoparticles: enabling colloidal stability, Nanotechnologies for Water Environmnet Applications. In T.C. Zhang, R.Y. Surampalli, K.C.K. Lai, Z. Hu, R.D. Tyagi, I.M.C. Lo, (Eds.) Nanotechnologies for water environment applications, American Society of Civil Engineers, Reston, Virginia, USA (2009) 191-212.

[21] C. Noubactep, Review: The fundamental mechanism of aqueous contaminant removal by metallic iron, Water SA 36 (2010) 663-670.

[22] R. Singh, V. Misra, R. Prata Singh, Removal of hexavalent chromium from contaminated ground water using zero-valent iron nanoparticles, Environ. Monit. Asses. 184 (2012) 3643-3651.

[23] K.L. Palanisamy, V. Devabharathi,; N.M. Sundaram, The utility of magnetic iron oxide nanoparticles stabilized by carrier oils in removal of heavy metals from waste water, Int. J. Res. Appl. Natural Soc. Sci. 1 (2013) 15-22.

[24] J. Němeček, O. Lhotský, T. Cajthaml, Nanoscale zero-valent iron application for in situ reduction of hexavalent chromium and its effects on indigenous microorganism populations, Sci. Tot. Env. 485–486 (2014) 739–747.

[25] Y. Deng, J.D. Englehardt, Treatment of landfill leachate by the Fenton process, Water Res. 40 (2006) 3683-3694.

[26] O. Gylienė, T. Vengris, A. Stončius, O. Nivinskienė, Decontamination of solutions containing EDTA using metallic iron, J. Hazard. Mater. 159 (2008) 446-451.

[27] C.R. Keenan, D.L. Sedlak, Ligand-enhanced reactive oxidant generation by nanoparticulate zero-valent iron and oxygen, Environ. Sci. Technol. 42 (2008) 6936-6941.

[28] T. Zhou, X. Lu, J. Wang, F. Wong, Y. Li, Rapid decolorization and mineralization of simulated textile wastewater in a heterogeneous Fenton like system with/without external energy, J. Hazard. Mater. 165 (2009) 193–199.

[29] T. Zhou, T. Lim, Y. Li, X. Lu, F. Wong, The role and fate of EDTA in ultrasound-enhanced zero-valent iron/air system, Chemosphere 78 (2010) 576-582.

[30] R.C. Martins, D.V. Lopes, M.J. Quina, R.M. Quinta-Ferreir, Treatment improvement of urban landfill leachates by Fenton-like process using ZVI, Chem. Engin. J. 192 (2012) 219–225.

[31] A. Babuponnusami, K. Muthukumar, A review on Fenton and improvements to the Fenton process for wastewater treatment, J. Env. Chem. Engin. 2 (2014) 557-572.

[32] O. Gylienė, O. Nivinskienė, V. Pakštas, Use of metallic iron for decontamination of solution containig Ni(II)citrate, Polish J. of Environ. Stud. 16 (2007) 397-402.

[33] P. Guardia, N. Pérez, A. Labarta, X. Battle, Controlled synthesis of iron oxide nanoparticles over a wide size range, Langmuir 26 (2010) 5843-5847.

[34] SIDS Initial assessment report for 11th SIAM. Citric acid. Orlando, Fla., January 2001.

[35] N. Kržišnik, A. Mladenovič, A. Sever Škapin, L. Škrlep, J. Ščančar, R. Milačič, Nanoscale zero-valent iron for the removal of Zn^{2+}, Zn(II)–EDTA and Zn(II)–citrate from aqueous solutions, Sci. Tot. Environ. 476–477 (2014) 20–28.

[36] J. Qvarnström, W. Frech, Mercury species transformation during sample pre-treatment of biological tissues studied by HPLC-ICP-MS, J. Anal. At. Spectrom. 17 (2002) 1486-1491.

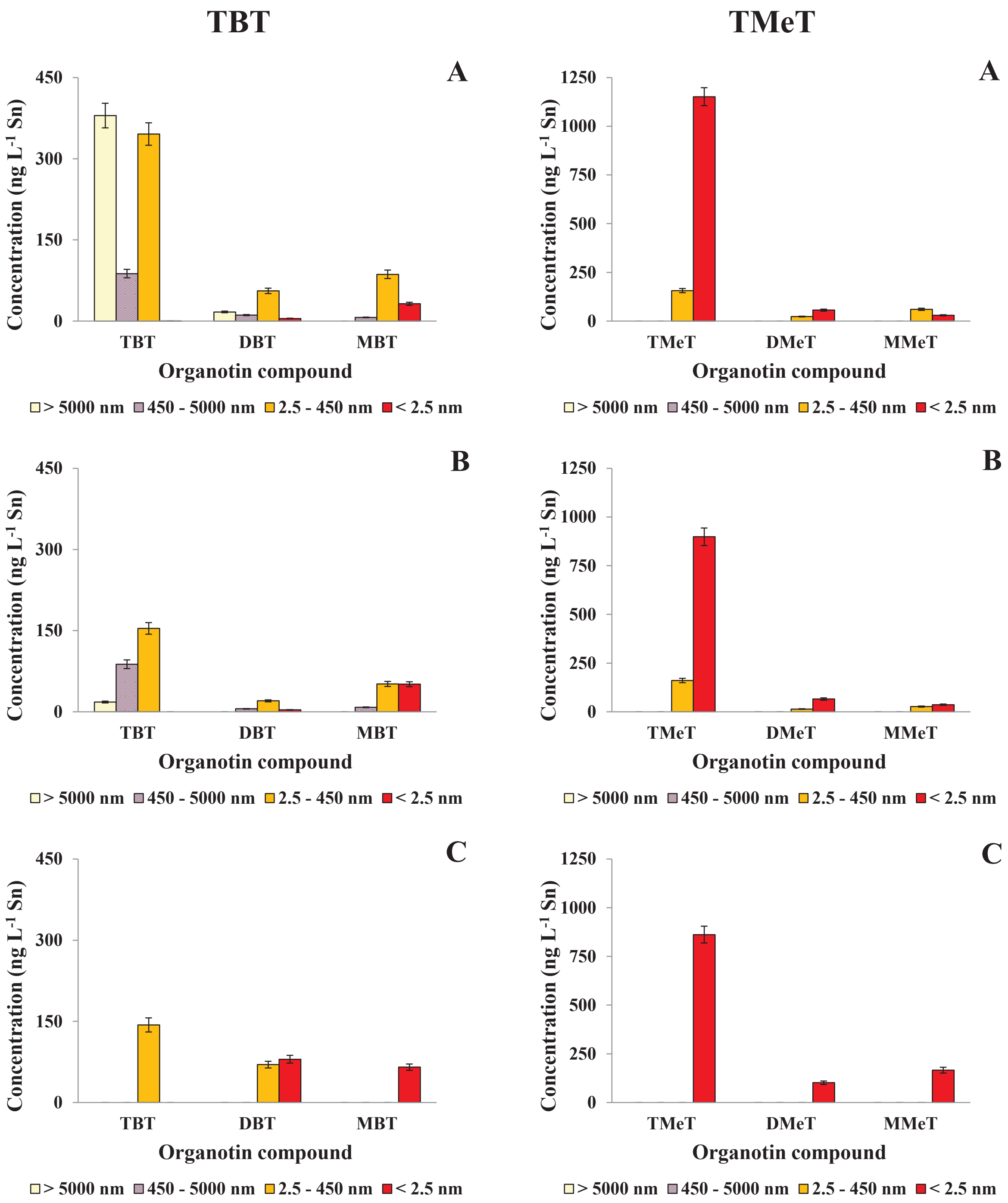
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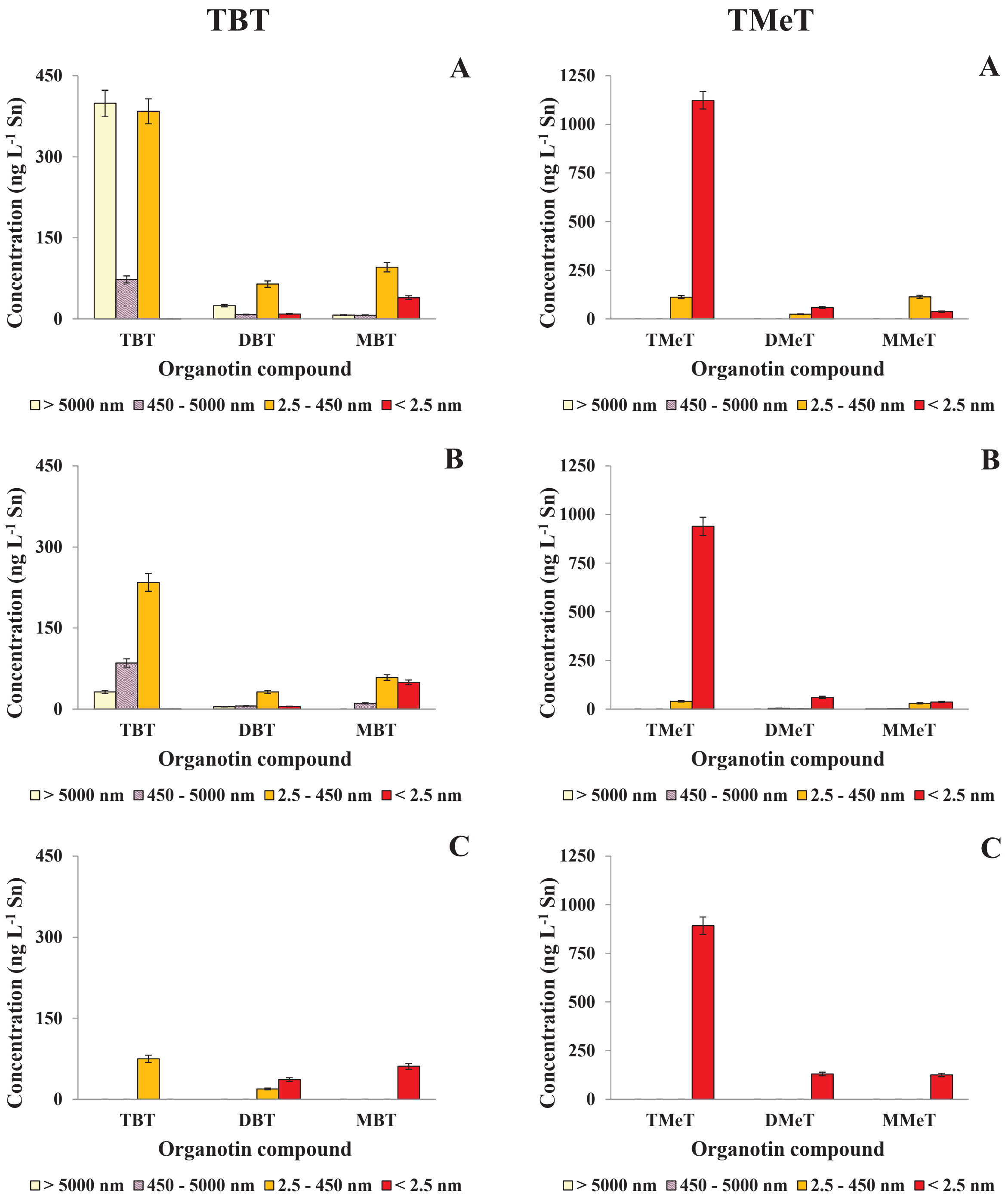
Fig. 1.

Figure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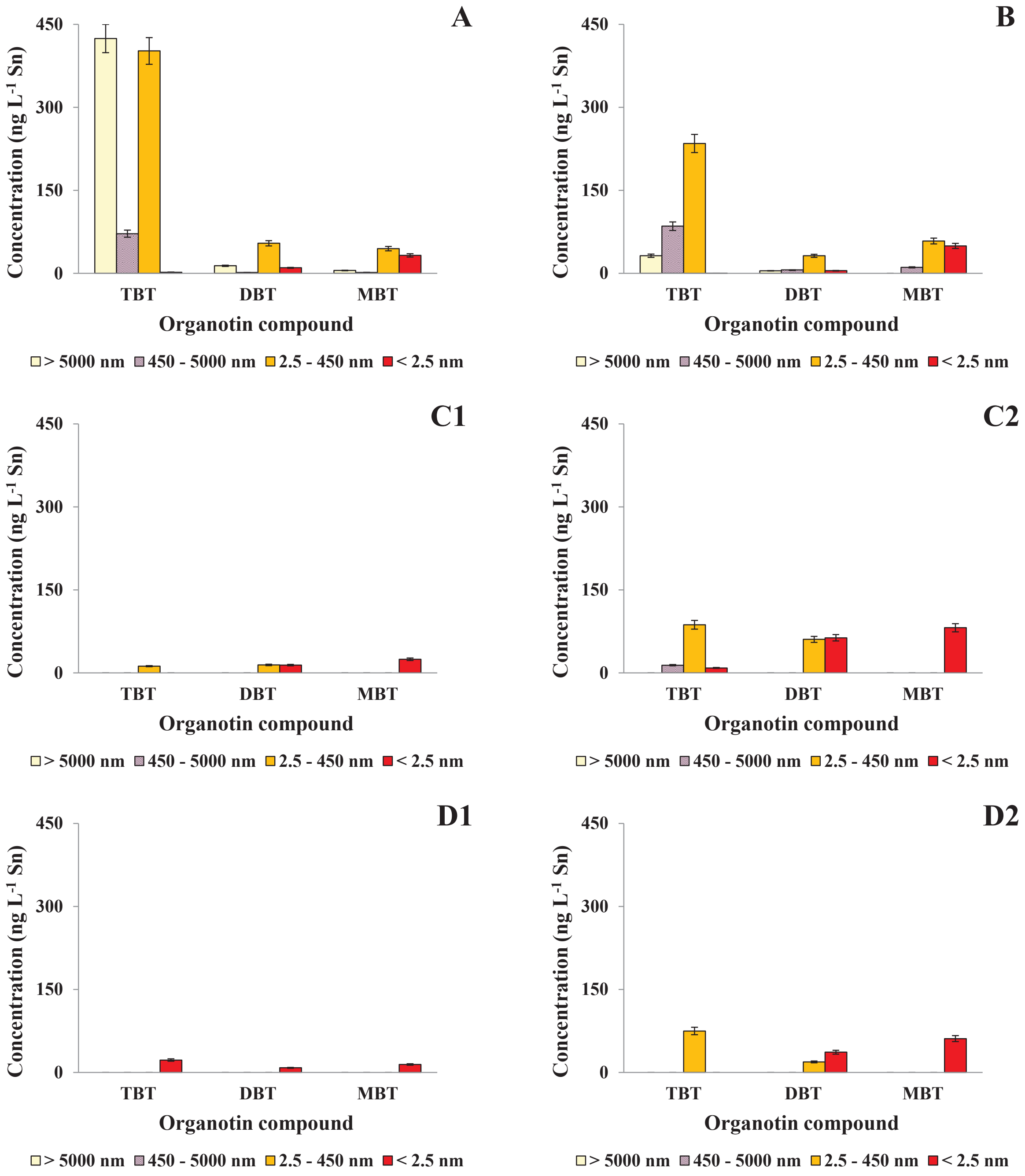
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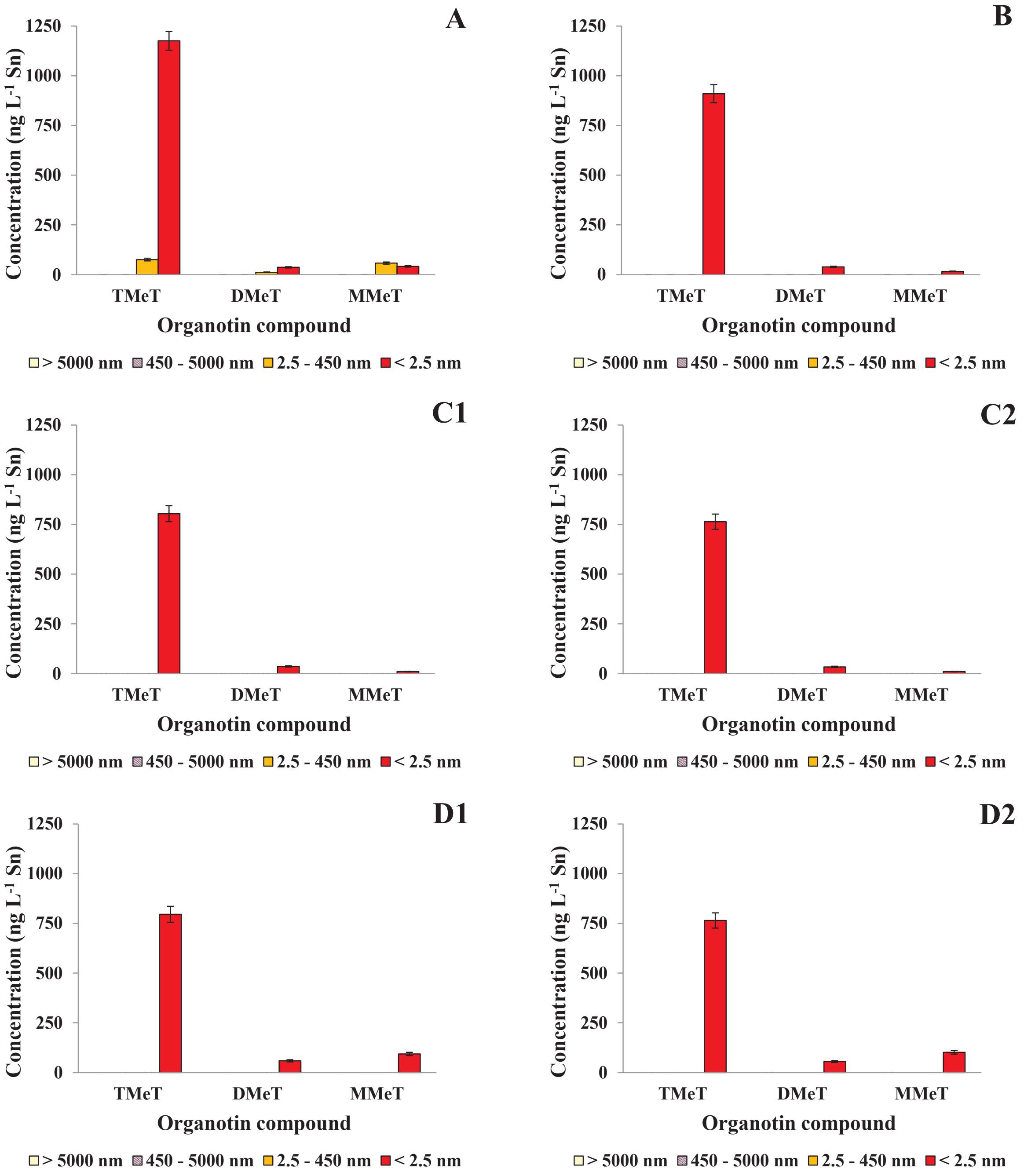
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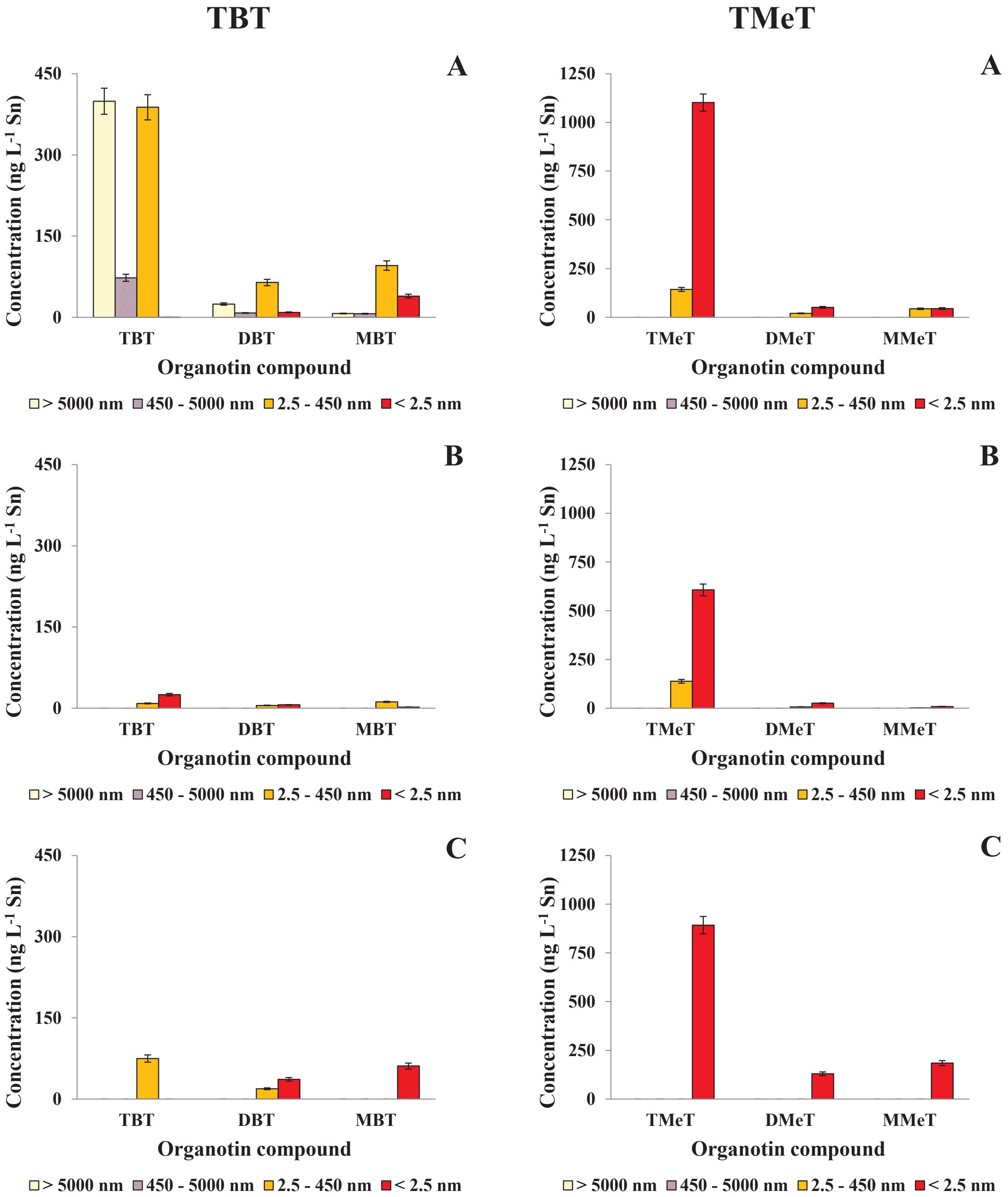
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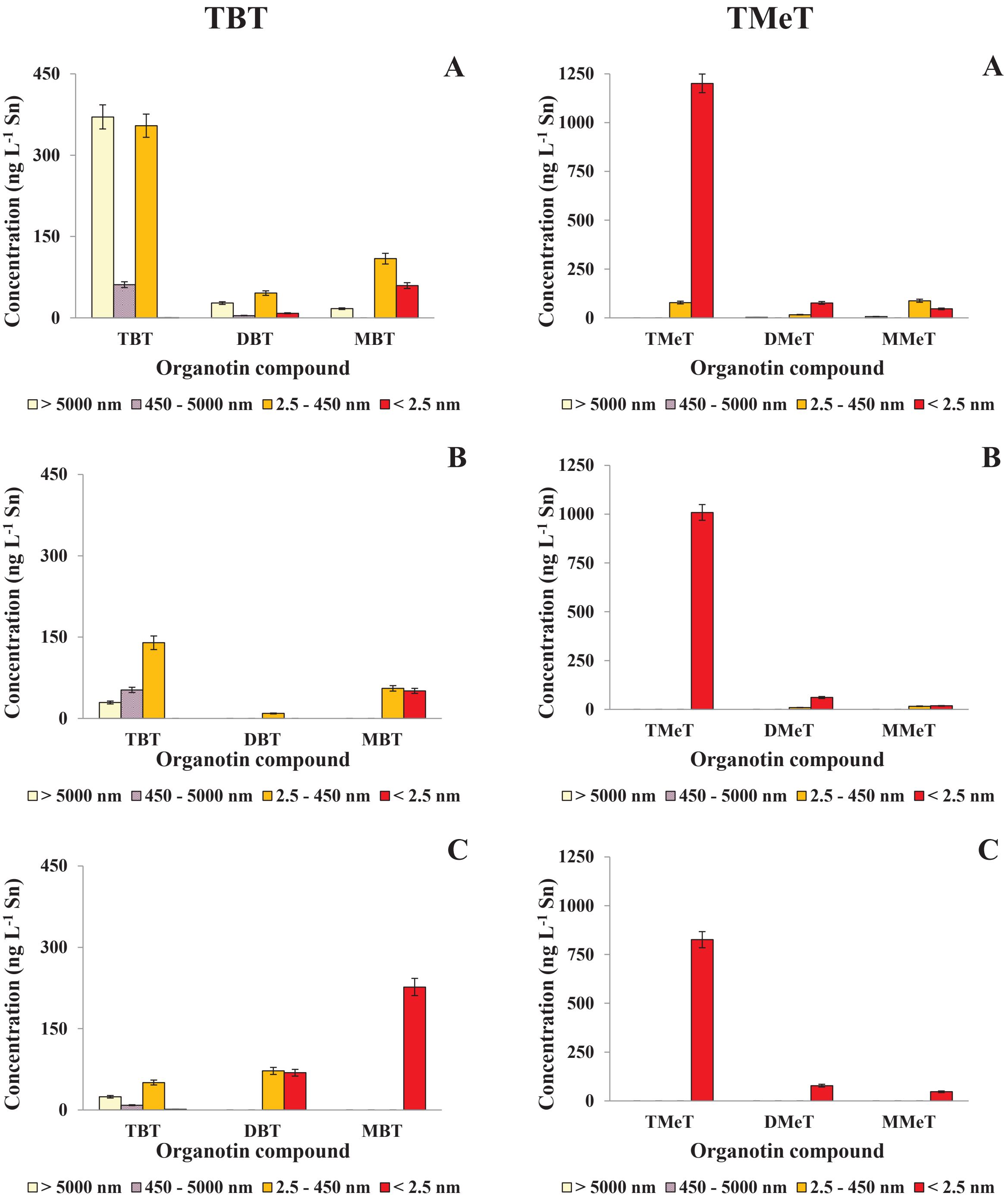

Fig. S1. Size distribution of OTCs in landfill leachates. (A) Untreated leachates (pH 8) were spiked with TBT (1000 ng L^{-1} Sn) or TMeT (1000 ng L^{-1} Sn). (B) Spiked leachates (pH 8) were treated with FeONPs. (C) The pH of spiked leachates was adjusted to 3 with citric acid and samples treated with FeONPs. FeONPs was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

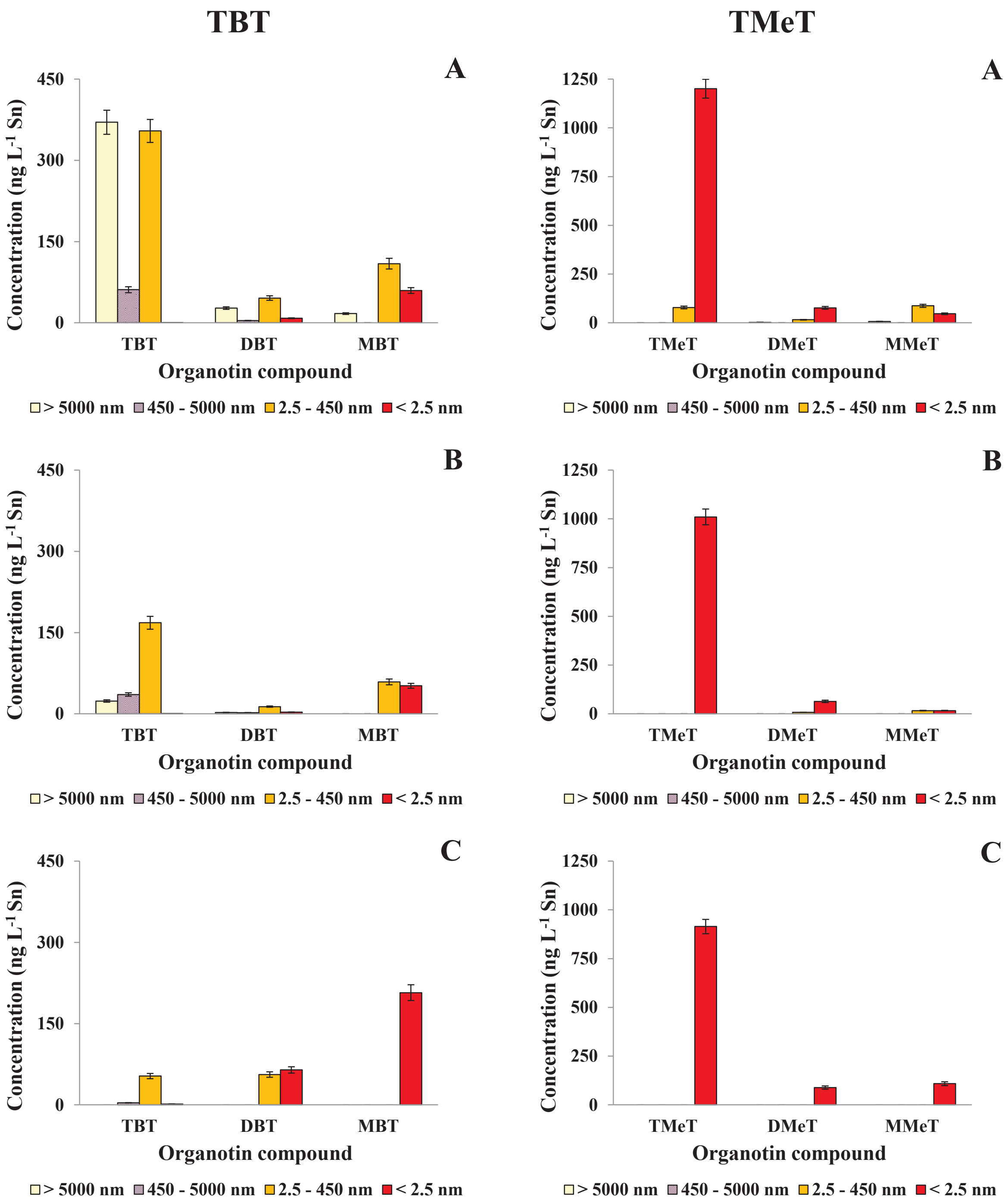

Fig. S2. Size distribution of OTCs in landfill leachates. (A) Untreated leachates (pH 8) were spiked with TBT (1000 ng L^{-1} Sn) or TMeT (1000 ng L^{-1} Sn). (B) Spiked leachates (pH 8) were treated with Fe_3O_4NPs. (C) The pH of spiked leachates was adjusted to 3 with citric acid and sample treated with Fe_3O_4NPs. Fe_3O_4NPs was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

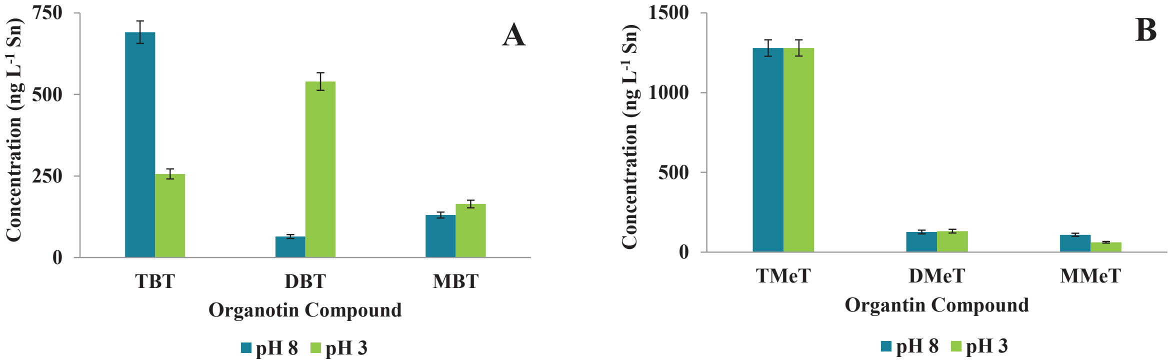

Fig. S3. Speciation of OTCs in landfill leachates at pH 8 or pH 3, spiked with (A) TBT (1000 ng L^{-1} Sn) or (B) TMeT (1000 ng L^{-1} Sn). After spiking samples were shaken for 3 days before being analysed. The pH was adjusted to 3 with citric acid. The error bars represent the experimental and analytical uncertainty.

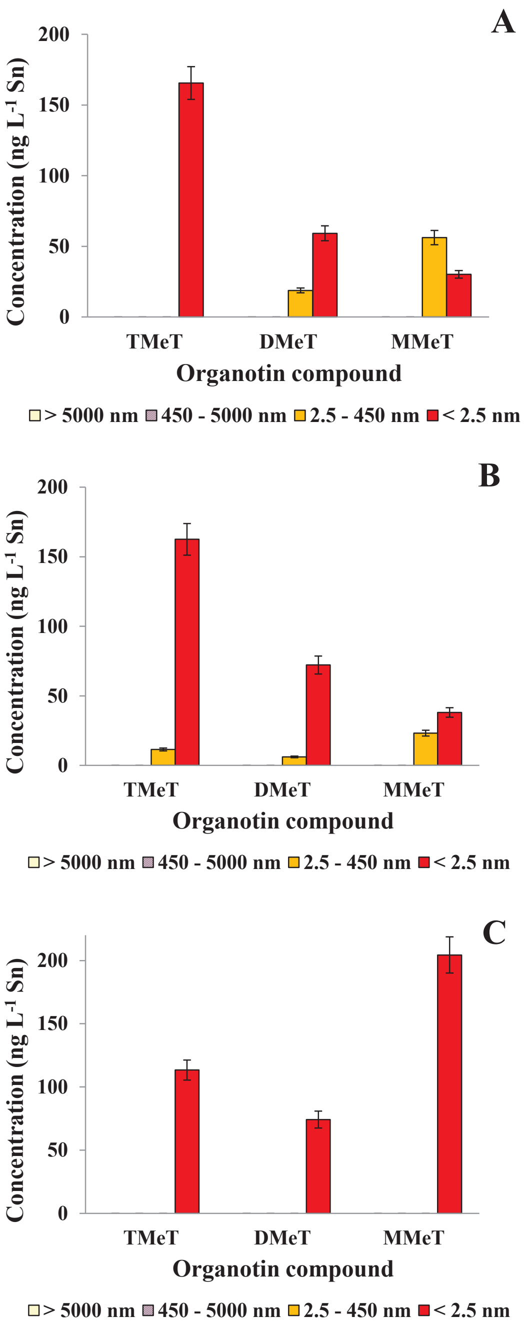

Fig. S4. Size distribution of methyltins in landfill leachates spiked with TBT (1000 ng L^{-1} Sn). (A) Untreated leachates (pH 8). (B) Leachates (pH 8) were treated with nZVI. (C) The pH of spiked leachates was adjusted to 3 with citric acid and sample treated with nZVI. nZVI was dispersed with TMAH. The error bars represent the experimental and analytical uncertainty.

Priloga 2

The fate of iron nanoparticles in environmental waters treated with nanoscale zerovalent iron, FeO and Fe_3O_4

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ABSTRACT

Among the different nanoparticles (NPs), which are used in the remediation of contaminated environmental waters iron nanoparticles (FeNPs) are the most frequently applied. If FeNPs remain in these waters after the treatment, they can cause hazardous effects to the environment. In this work, time dependent size distribution of iron particles was investigated in different aqueous samples after the different FeNPs treatment to evaluate potential environmental threat. The efficiency of metal removal by FeNPs was also examined. Concentrations of metals in aqueous samples were determined before and after the nanoremediation by ICP-MS. The data revealed that settling and removal of FeNPs after the treatment of environmental waters was related to the sample characteristics and the ways of NPs dispersion. Dispersion with TMAH was more effective than by mixing. When mixing was used for the dispersion, nanozerovalent iron (nZVI), FeONPs and Fe_3O_4NPs quickly settled in Milli Q water, forest spring water and in landfill leachate. Dispersion with TMAH resulted in a slower iron particles settling. In Milli Q and forest spring waters treated with FeONPs, a double layer of oleic acid and TMAH was formed around NPs, which prevented the formation of iron corrosion products. Hence, FeNPs, which were detected in solution even24 h after the treatment, may represent a potential environmental threat. Removal of metals from environmental waters depended on the type of FeNPs and the dispersion efficiency. The metals concentration and sample characteristics like the pH, the ionic strength, the content of organic matter, the presence of complexing ligands and the interactions of FeNPs with the sample matrix as well as the chemical speciation of elements, considerably contributed to the remediation efficiency. If the surface of FeNPs is contaminated by a particular metal, this contaminant may be, during the NPs treatment, released into the water, which is being remediated.

Keywords:

Iron nanoparticles

Environmental waters

Dispersion

Aggregation

Metal removal

Inductively coupled plasma mass spectrometry

1. Introduction

Water is one of the most important human resources. Due to the growing human population and related industrial activities the consumption of water extensively grows. On the other hand water is more and more polluted. To reduce the negative impacts of pollution, waters must be remediated. Nanotechnology, among other, provides opportunities for efficient remediation of contaminated soil (Machadoa et al., 2013) and environmental waters (Kamat and Meisel, 2003; Zhang and Surampalli, 2009). It enables *in situ* soil, groundwater and surface water remediation. Such remediation is characterised by the use of highly reactive nanoparticles (NPs), potential cost-effectiveness, access to hard-to-reach sites, shorter clean-up times and the possibility of subsequent NPs injections (Grieger et al., 2010). Application of iron nanoparticles (FeNPs) represents a new generation of environmental remediation technologies that could provide cost-effective solutions to some of the most challenging environmental clean-up problems (Lo et al., 2009). FeNPs possess a large surface area, which is highly reactive. There are different possibilities by which they can be applied for *in situ* remediation. Research has shown that FeNPs very effectively transform and detoxify a wide variety of common environmental contaminants, such as chlorinated, brominated and trihalo-methanes, chlorinated ethenes and benzenes, other polychlorinated hydrocarbons, pesticides and dyes (Zhang, 2003; Tratnyek, 2006; Karn et al., 2009). Among FeNPs the most widely used are magnetic FeNPs, nano zero-valent iron (nZVI), iron oxide (FeONPs) and magnetite (Fe_3O_4NPs). They have all been applied in treatment of polluted water or groundwater (Shen et al., 2009).

The fundamental reaction mechanism behind the removal of contaminant by nZVI is either reductive transformation of the contaminant followed by adsorption and co-precipitation or adsorption and co-precipitation with iron corrosion products. Under environmental conditions, nZVI is oxidised to Fe^{2+}, the stability of which is mostly dependent on the pH value and the redox potential. In the pH ranges relevant for natural waters, Fe^{2+} species are unstable and rapidly reacts with molecular oxygen. The resulting Fe^{3+} species readily hydrolyse and precipitate (Noubactep, 2010). The reactivity of FeONPs and Fe_3O_4NPs is much lower than that of nZVI because on their surface oxides form passivation layer (Jiang et al., 2014; Hotze et al., 2010). The aggregation of FeNPs is highly dependent also on the ionic strength and the content of natural organic matter (Jiang et al., 2009; Hotze et al., 2010).

The fate of NPs in different aqueous media, such as groundwater, rivers, lakes and seawaters, is still not well understood. Despite the concern on their potential environmental or toxicological impacts, there was not enough studies carried out in which the toxicological effects of FeNPs on living organisms were investigated (Auffan et al., 2008; Grieger et al., 2010; Karthikeyeni et al., 2013; Mauer-Jones et al., 2013). Cytotoxic effects of Fe_3O_4NPs and nZVI towards *E. Colli* occurred for doses higher than 700 mg/L and 70 mg/L, respectively (Auffan et al., 2008). For fish species *Oryzias latipes*, nZVI caused a disturbance in the oxidative defence system at concentrations as low as 0.5 mg/L. In its adult stage fish showed histopathological changes and morphological alterations on gills and intestine and antioxidant balance disruption, although they were able to recover afterwards. For the other fish species *Pimephales promelas*, 50 mg/L nZVI cause some changes in gene regulation, including genes that encode proteins involved in tissue repair and antioxidant defences. Acute toxicity to aquatic organisms appears to be relatively low, although some sub-lethal effects have been observed (Grieger et al., 2010).

When FeNPs are used for environmental water remediation, NPs that persist in solution after the treatment may represent an environmental threat. To estimate their potential toxic effects towards biota, it is necessary to study their behaviour after the treatment of polluted waters.

Therefore, the aim of our work was to follow the fate of nZVI, FeONPs and Fe_3O_4NPs in Milli Q water, landfill leachate and forest spring water, which differ in ionic strength and the content of organic matter. Time dependent size distribution of iron in aqueous samples was investigated by the use of sequential filtration. The efficiency of metal removal by FeNPs was also examined. Concentrations of metals in aqueous samples were determined before and after the nanoremediation by ICP-MS.

2. Materials and Methods

2.1. Instruments

Total elemental concentrations were determined by an inductively coupled plasma mass spectrometer (ICP-MS), model 7700x (Agilent Technologies, Tokyo, Japan). ICP-MS operating parameters are presented in Table S1 (Supplementary).

2.2. Reagents and materials

Merck (Darmstadt, Germany) suprapur acids and Milli Q water (18.2 MΩ cm) obtained by Direct-Q 5 Ultrapure water system (Millipore Watertown, MA, USA) were used for the preparation of samples and standard solutions. All the other chemicals used were of analytical reagent grade. Stock Multi Element XVI standard solution (100 mg L^{-1} in 5 % HNO_3) (Merck, Darmstadt, Germany) was used for the preparation of working standard solutions for ICP-MS determinations.

Magnetic nZVI (manufactural size information approximately 25 nm), surface stabilized with a shell layer of FeO around the iron particle core, were obtained from MKNano, (Williamsville, NY, USA). Magnetic FeONPs surface stabilized with oleic acid and kept in heptane (particle size 3.5 - 9.5 nm) and the magnetic Fe_3O_4 nanopowder (particle size 50 - 100 nm) were purchased from Sigma-Aldrich (Steinheim, Germany). Tertramethylammonium hydroxide (TMAH) (25 wt % in methanol) used for the dispersion of FeNPs was obtained also from Sigma-Aldrich.

For sequential filtration of aqueous samples, the 5 μm cellulose-nitrate, 0.45 μm, 0.2 μm and 0.1 μm cellulose-acetate membrane filters and 10 KDa regenerated cellulose filters (2.5 nm) were obtained from Whatman (Vienna, Austria), Ministart (Goettingen, Germany) and Amicon (Zagreb, Croatia), respectively.

2.3. Preparation of FeNPs dispersions

For efficient dispersing of FeNPs (nZVI, FeONPs or Fe_3O_4NPs), 30 mL of TMAH (25 wt % in methanol) was added to 0.75 g of FeNPs, the suspension sonicated for 10 min, shaken for 1 h, and FeNPs removed with a magnet (Guardia et al., 2010). FeNPs treated with TMAH were then added to 500 mL of the aqueous samples. Alternatively, FeNPs were added to samples and dispersed by mixing for 15 min. The amount of FeNPs added to an aqueous sample corresponded to a concentration of 1.5 g L^{-1} Fe.

2.4. Samples

The behaviour of various FeNPs was investigated in the samples of Milli Q water, forest spring water and landfill leachate. Their sample matrices differ in ionic strength and the content of organic matter. Sampling of unpolluted forest spring water was performed at the rivulet Hruševnik close to Ljubljana city, Slovenia, while leachate samples were collected at the non-hazardous Barje municipal waste landfill, Ljubljana, Slovenia. 30 L of sample was collected from a leachate control well of the active landfill basin into a polyethylene bottle and directly transported to the laboratory.

Relevant parameters characterising the samples analysed are presented in Table 1.

Insert Table 1 about here

2.5. Experimental set-up

500 mL aliquots of aqueous samples were transferred to 1 L amber glass bottles. nZVI, FeONPs or Fe_3O_4NPs were added using TMAH or mixing for dispersion of FeNPs, as described under *2.3.* The treatment with FeNPs was performed by shaking of the samples for 24 h. 60 mL sample aliquots were then taken in different time intervals: 15 min, 1 h, 4 h and 24 h after the FeNPs treatment, to follow the particles settlement. Sequential filtration of samples through 5 µm, 0.45 µm, 0.2 µm, 0.1 µm and 2.5 nm filters was applied. Accordingly,

concentrations of Fe and selected elements were determined in 10 mL sample fractions with particle sizes 450 - 5000 nm, 200 - 450 nm, 100 - 200 nm, 2.5 nm - 100 nm and < 2.5 nm by ICP-MS. The same procedure of the sequential filtration and the determination of elements was performed also in untreated aqueous samples.

All experiments were carried out in duplicate.

3. Results and discussion

FeNPs are increasingly used to remove different pollutants from contaminated environmental waters (Krajangpan et al., 2009; Noubactep, 2010; Singh et al., 2012; Palanisamy et al., 2013). At neutral and alkaline pHs, adsorption of pollutant onto the surface of FeNPs and co-precipitation with iron corrosion products is the prevailing process of pollutant removal (Noubactep, 2010). During the remediation, iron corrosion products are formed and precipitated. In this way FeNPs lose their nano character. To prevent the potential toxic effects of the remaining FeNPs towards biota in environmental waters, it is necessary to follow their fate after the treatment. Ionic strength, the content of organic matter and the use of dispersing agents significantly influence the behaviour of NPs (Jiang et al., 2009; Bian et al., 2011; Hu et al., 2010; Hotze et al., 2010). Although it is important to understand the behaviour of NPs after the remediation, the literature data addressing this issue are scarce. The reports are mainly related to toxicological studies (Auffan et al., 2008; Grieger et al., 2008; Karthikeyeni et al., 2013). Therefore, the present investigation was designed to study the size distribution of different FeNPs after the treatment of environmental water samples.

3.1. Size distribution of particles in environmental waters treated with different FeNPs

The fate of FeNPs in Milli Q water, forest spring water and landfill leachate treated with nZVI, FeONPs and Fe_3O_4NPs was investigated over a time span from 15 min to 24 h after the FeNPs treatment. FeNPs were dispersed with TMAH or by mixing. The sample preparation is described in details under paragraphs *2.3.* and *2.5.*

3.1.1. Milli Q water

First, the behaviour of NPs in Milli Q water treated with FeNPs was investigated. As can be seen from the data of Table 1, Milli Q water has an extremely low electrical conductivity (which is related to a low ionic strength) and a very low content of organic matter. In such aqueous sample matrix the interactions that are caused by the presence of organic matter or ionic strength are totally negligible. To follow the size distribution of FeNPs, sequential filtration of samples was applied. The results for the time dependent size distribution of iron in Milli Q water treated with nZVI, FeONPs and Fe_3O_4NPs, are presented in Fig. 1.

Insert Fig. 1. about here

Data from Fig. 1. demonstrate that dispersion of FeNPs with TMAH (left column) is much more effective than by mixing (right column). TMAH acts as a surfactant and creates an electrostatic repulsion layer around nZVI and Fe_3O_4NPs, keeping them dispersed in solution (Andrade et al., 2012; Tang and Lo, 2013). FeONPs coated with a layer of oleic acid possess hydrophobic characteristics and disperses NPs in heptane by steric interactions (Scherer and Neto, 2005). TMAH is frequently used as a surfactant that changes NPs surface properties. It forms a strong electrostatic ligand bilayer that allows the transfer of otherwise hydrophobic NPs to water. The double layer around NPs also inhibits oxidation of FeONPs (Sperling and Park, 2010).

When Milli Q water was treated with nZVI using TMAH for dispersion (Fig. 1A1), iron corrosion products, which were generated by rapid reaction of Fe^{2+} (formed from Fe^0) with dissolved oxygen, aggregated and precipitated in a short time. So, iron precipitate was observed in Milli Q water treated with nZVI immediately after shaking stopped. The coarser iron particles (450 – 5000 nm) were settled mainly within 4 h, while those with sizes 2.5 – 100 nm and 100 – 200 nm persisted in the solution even 24 h after the nZVI treatment. However, iron concentrations in both fractions were low and did not exceed 1 mg L^{-1} Fe.

In Milli Q water treated with FeONPs, using TMAH for dispersion (Fig. 1B1), iron precipitate was not observed. NPs remained dispersed in solution 24 h after the FeONPs treatment. The prevailing iron containing fraction was that with particles 2.5 – 100 nm (about 1300 mg L^{-1} Fe), followed by the fraction with particles of 100 – 200 nm (about 200 mg L^{-1} Fe). FeONPs coated with oleic acid were, after the addition of TMAH, efficiently dispersed in Milli Q water by strong electrosteric interactions (Sperling and Park, 2010; Tang and Lo, 2013). Their oxidation and consequently the formation of iron corrosion products were inhibited by a bilayer of oleic acid and TMAH, which covered NPs. For this reason, the precipitate was not formed and FeONPs remained dispersed in solution.

When Milli Q water was treated with Fe_3O_4NPs (Fig. 1C1) iron precipitate was observed immediately after the shaking was stopped. Since the reactivity of Fe_3O_4NPs is much lower than that of nZVI iron corrosion products were not so effectively formed. Consequently, 24 h after the Fe_3O_4NPs treatment, about 75 mg L^{-1} Fe remained in the nanosized fraction 2.5 – 100 nm, while a very low iron concentration (about 0.2 mg L^{-1} Fe) was determined also in the dissolved fraction (particle sizes < 2.5 nm). However, these iron concentrations were much lower than those found in FeONPs treatment.

Dispersing of FeNPs by mixing was not as efficient as by TMAH. When mixing was applied to disperse nZVI or Fe_3O_4NPs in Milli Q water, iron precipitate was observed as soon as the shaking was stopped. The dispersion by mixing in Milli Q water was more efficient for nZVI with smaller particles (around 25 nm), than for Fe_3O_4 nanopowder with bigger particles (50 - 100 nm). Due to this reason, Fe_3O_4NPs quickly settled and only a negligible fraction of iron (about 0.2 mg L^{-1} Fe) remained dissolved in solution (Fig. 1C2), while slightly higher amount of nZVI (about 1.0 mg L^{-1}) remained dispersed during the course of the experiment (Fig. 1A2).

Oleic acid coated FeONPs are hydrophobic and this characteristic prevented their successful dispersion by mixing in Milli Q water despite their smallest particle sizes (3.5 - 9.5 nm). Instead of being dispersed, FeONPs were stuck on the surface of the glass containers. As evident from Fig. 1B2 only a negligible portion of nanosized iron remained in solution during the course of the experiment (about 0.3 mg L^{-1} Fe).

3.1.2. Forest spring water

Forest spring water is a sample matrix with both a low ionic strength (which is reflected by low electrical conductivity), and low content of dissolved and total organic matter (see data of Table 1). The iron concentration in the samples of forest spring water analysed was below 0.3 mg L^{-1} Fe. The behaviour of NPs in forest spring water treated with different FeNPs using TMAH or mixing for their dispersion is presented in Fig. 2.

Insert Fig. 2. about here

When TMAH was used for the dispersion of nZVI or Fe_3O_4NPs in the forest spring water, the positively charged quaternary ammonium ions reacted with the deprotonated carboxylic groups of organic acids (constituents of organic matter), forming quaternary ammonium salts of the carboxylate anions (Kossa et al., 1979). As a consequence of these reactions, the TMAH dispersion property was lost. nZVI and Fe_3O_4NPs were not efficiently dispersed but agglomerated and precipitated as soon as shaking was stopped (Figs. 2A1and 2C1). On the contrary, in forest spring water treated with FeONPs, using TMAH for dispersion (Fig. 2B1) iron precipitate was not observed since a bilayer of oleic acid and TMAH inhibited the oxidation of FeO and formation of iron corrosion products. NPs remained dispersed in solution for at least 24 h after the FeONPs treatment, exclusively in the fraction with particle sizes 2.5 – 100 nm. This phenomenom can be explained by a strong interaction that occurred between oleic acid and TMAH, which prevented desorption of TMAH from the surface of FeONPs and thereby its reaction with dissolved organic matter. TMAH remained to act as a dispersion agent, while the dissolved organic matter present in samples, additionally enhanced the dispersion efficiency (Maurer-Jones et al., 2013).

When mixing was used for the dispersion of FeNPs, dissolved organic matter has no observable effect on the efficiency of dispersion of nZVI or Fe_3O_4NPs (Figs. 2A2 and 2C2). A slight enhancement in the dispersion efficiency was observed for FeONPs. In this case, dissolved organic matter interacted with oleic acid and, as a result, small amount of iron in particles bigger than 100 nm remained dispersed in samples for at least 24 h after the shaking was stopped.

3.1.3. Landfill leachate

Landfill leachate represents a sample matrix with a very high ionic strength (which is reflected by high electrical conductivity) and a high content of dissolved and total organic matter (see data of Table 1). The iron concentration was about 2 mg L^{-1} Fe, present mainly in the dissolved (< 2.5 nm) fraction. The behaviour of NPs in landfill leachate treated with FeNPs, using TMAH or mixing for dispersion, is presented in Fig. 3.

Insert Fig. 3. about here

Once added to the landfill leachate, FeNPs behaviour depends on the combination of several processes. When TMAH is used to disperse nZVI or Fe_3O_4NPs, it reacts with the dissolved organic matter, forming quaternary ammonium salts of the carboxylate anions. By this, the coating of TMAH is removed from the surface of FeNPs resulting in their significant increased tendency to agglomerate and settle down. Contrary to this, the excess of dissolved organic matter, which did not react with TMAH, forms a coating around FeNPs, which acts as a surfactant that keeps them dispersed in solution. Another influence comes from high ionic strength of the sample matrix. It causes the aggregation of FeNPs by reducing the repulsive forces between NPs and the size of the electrostatic layer around them (Hotze et al., 2010). The results presented on the behaviour of FeNPs in landfill leachate in this work can be explained as a combining influence of all the processes described. As evident from Figs 3A1 and 3C1, the mechanism of settling of NPs in the landfill leachate prevailed. When TMAH was applied for the dispersion of FeONPs (Fig. 3B1), the microorganisms, present in the leachate tested, most likely used oleic acid as a source of carbon and removed it from the surface of FeONPs. Thus, a double layer coating was eliminated. This enabled FeONPs oxidation and further formation of iron corrosion products, causing their settling under the same mechanisms as described for nZVI and Fe_3O_4NPs. From the comparison of the influences of the particular sample matrix on the behaviour of FeONPs dispersed with TMAH (Milli Q water (Fig. 1B1), forest spring water (Fig. 2B1) and landfill leachate (Fig. 3B1)) it can be seen, that the most pronounced effects were observed for the landfill leachate sample matrix.

When mixing was used to disperse FeNPs in the landfill leachate, the effect of the ionic strength prevailed over the influence of dissolved organic matter, leading to the rapid settling of FeNPs (Figs. 3A2, 3B2 and 3C2).

3.2. Removal of metals from forest spring water and landfill leachates by FeNPs

In the same samples in which the fate of FeNPs was investigated, their efficiency of the metal removal was also studied. Samples were analysed 24 h after the treatment. The size distribution of metals in untreated samples and samples treated with different FeNPs, using TMAH or mixing for dispersion, was determined by measuring the total metal concentrations with ICP-MS. Treatment by FeONPs, using mixing for the dispersion, was not carried out as hydrophobicity causes their rapid attachment to the walls of the containers used in experiments. Once attached to the walls FeONPs are not having any influence on the metal removal. Since treatment of water samples with FeNPs may contribute to contamination, the concentrations of selected elements were determined in Milli Q water also after the treatment. These results are presented in Table 2. It was experimentally proven that TMAH was not contaminated with the elements investigated. Their concentrations were below 0.1 µg L^{-1}. In Table 3 the pHs of samples before and after the treatment are given.

Insert Tables 2 and 3 about here

The efficiency of the removal of selected elements from the forest spring water by the use of FeNPs is presented in Fig. 4.

Insert Fig. 4. about here

As can be seen from the data of Fig. 4, the concentration of Ca in untreated forest spring water was 1.5 mg L^{-1}. nZVI and Fe_3O_4NPs dispersed with TMAH were only slightly contaminated with Ca ions (Table 2). A high pH (pH 13) (Table 3) contributes to the co-precipitation of $Ca(OH)_2$ with iron corrosion products and thus to efficient removal of Ca from forest spring water. Despite the high pH, FeONPs dispersed with TMAH did not remove Ca from the forest water, since the double layer of TMAH and oleic acid prevented the formation of iron precipitate. The concentration of Ca after the FeONPs treatment was slightly enhanced due to the contamination of FeONPs particles. Removal of Ca from the forest water was also not effective by nZVI and Fe_3O_4NPs when mixing was used for dispersion. This can be explained by poor formation of iron corrosion products and $Ca(OH)_2$ at pH 6. The concentration of Ca was increased for the amount corresponding to that arising from the contamination of the NPs used.

Ba and Mn concentrations in forest spring water were low (below 1 $\mu g\ L^{-1}$). Treatment with FeONPs or Fe_3O_4NPs particles, which were not contaminated with Ba and Mn did not influence their concentrations in the forest water. Opposite, high contamination of nZVI particles with Ba and Mn (about 170 $\mu g\ L^{-1}$) caused the increase of their concentrations in the forest water (Table 2). As evident from the data of Fig. 4, when mixing is applied for the dispersion of nZVI at pH 6, Ba and Mn were released from the surface of nZVI to the forest water, while when TMAH was used for the dispersion, at pH 13, the released contaminants formed hydroxides that were co-precipitated with iron corrosion products.

In forest spring water, Al was evenly distributed between different particle sizes with a concentration of 60 $\mu g\ L^{-1}$. Al chemistry in aqueous samples depends strongly on the pH. At a pH below 5, $Al(H_2O)_6^{3+}$ species prevails. In less acidic solution, $Al(H_2O)_6^{3+}$ undergoes hydrolysis to yield $Al(H_2O)_5(OH)^{2+}$ and $Al(H_2O)_4(OH)_2^{+}$ species. At pH 6, $Al(H_2O)_5(OH)^{2+}$, $Al(H_2O)_4(OH)_2^{+}$ and sparingly soluble $Al(OH)_3$ species are present. In the neutral pH range, Al is mainly precipitated as $Al(OH)_3$. By increasing the alkalinity of the sample, at pHs higher than 8, the precipitate starts to re-dissolve, resulting in the progressive formation of $Al(OH)_4^-$ species. Between pH 8 and pH 12, the $Al(OH)_3$ and $Al(OH)_4^-$ species exist, while at a pH higher than 12, highly soluble $Al(OH)_4^-$ is the only Al species present (Ščančar and Milačič, 2006). As can be seen from the data of Tables 2 and 3, when nZVI was dispersed by mixing (pH 6), the soluble Al species were released from the surface of nZVI particles into Milli Q water (Al concentration 38 $\mu g\ L^{-1}$). Application of TMAH for the dispersion of nZVI (pH 13) caused the release of Al from their surface due to the formation of highly soluble $Al(OH)_4^-$

species (Al concentration 277 μg L^{-1}). For similar reasons, but in much lower extent, the Fe_3O_4NPs contribute to the contamination of Millli Q water by Al. The behaviour of Al in the forest spring water treated with nZVI or Fe_3O_4NPs that were both dispersed by the addition of TMAH followed the phenomena described above. After the treatment, the pH of the forest spring water samples was increased to pH 13 that solubilized Al originally present in the samples and that one arising from the contamination of the FeNPs particles added.

As evident from Fig. 4, Cu was presented, in the forest spring water in trace amounts (0.5 μg L^{-1}). Consequently, the treatments with FeONPs or Fe_3O_4NPs, which were not contaminated with Cu, have no influence on its concentration in the spring water treated. However, significant increase of its concentration in the forest spring water was observed when water samples were treated with nZVI. The amount of Cu released to forest water was equal to that released from the surface of nZVI particles in Milli Q water (Table 2).

Like Cu, the content of V in the forest spring water was very low (0.1 μg L^{-1}). Treatment of this water with nZVI (dispersed by mixing or TMAH) or Fe_3O_4NPs (dispersed with TMAH) contributed to the contamination of forest water in amounts between 1 to 4 μg L^{-1}. These amounts corresponded to those of V released to the Milli Q water from the surface of nZVI or Fe_3O_4NPs, respectively (Table 2).

The behaviour of the elements determined in landfill leachate after its treatment with FeNPs was different than that observed in the forest spring water. Among all the aqueous samples studied in this work, the leachate samples were characterised by the highest element concentrations, ionic strength, the content of organic matter and a pH (pH 8) (Table 1). The efficiency of the removal of selected elements in landfill leachate by FeNPs is presented in Fig. 5.

Insert Fig. 5. about here

The data from Fig. 5 indicate that, in untreated landfill leachate, Ca was present in the dissolved form. Its concentration was about 100 mg L^{-1}, and was so high that the contamination arising from FeNPs can be considered as negligible. The treatment of leachate with FeNPs dispersed with TMAH increased its pH from 8 to 9. At pH 9, $Ca(OH)_2$ is formed, which is co-precipitated with iron corrosion products. Since the treatment with FeNPs dispersed by mixing, did not change the pH of the leachate (pH 8), Ca was in this case not co-precipitated and thus, not removed from the solution.

In untreated leachate Ba and Mn were present mainly in the dissolved form in concentrations of about 250 µg L^{-1} and 150 µg L^{-1}, respectively. High contamination with Ba and Mn (about 170 µg L^{-1}) arising from nZVI particles (Table 2), is reflected in their remaining concentrations after the leachate treatment. As it is evident from the data of Fig. 4, when mixing was applied to disperse nZVI, only 30% of Ba was removed from the leachate, while the Mn concentration was, due to the contamination of nZVI, increased for about two times. Much more effective removal of these two elements was obtained when nZVI was dispersed with TMAH. Namely, at a pH of 9, Ba and Mn ions are more effectively adsorbed on the surface of nZVI and co-precipitated with iron corrosion products. FeONPs and Fe_3O_4NPs, which are not contaminated with Ba and Mn, effectively removed these two elements from the leachate.

In landfill leachate Al was distributed mainly between the dissolved and nanosized (2.5 – 100 nm) fractions in a concentration of about 185 µg L^{-1}. As can be seen from the data of Fig. 5, none of the treatments applied removed effectively Al from the leachate. The extent of removal depended on the pH, which influences the Al speciation, the dispersion efficiency and the contamination arising from FeNPs.

Cu was presented in the landfill leachate in a concentration of 56 µg L^{-1}. It was bound to coarser particles, distributed between 200 - 450 nm and 450 - 5000 nm particle size fractions.

After leachate was treated with FeONPs or Fe_3O_4NPs, which were not contaminated with Cu, the re-distribution from coarser to smaller particle sizes (2.5 -100 nm and < 2.5 nm) was observed. This indicates that there were interactions of these FeNPs with the particulate matter to which Cu is bound in the leachate. However, the treatment with FeONPs and Fe_3O_4NPs had no influence on the removal of Cu. This phenomenon was most probably related to the strong tendency of Cu to form complexes with available low molecular mass (e.g. citric) and high molecular mass (e.g. fulvic, humic) organic acids present in the leachate. These negatively charged Cu complexes were not adsorbed on the surface of FeNPs due to repulsive forces. Another phenomenon in the behaviour of Cu was observed also after the treatment of leachate with nZVI contaminated with Cu. The Cu concentration released from the surface of nZVI into Milli Q water was 26 $\mu g\ L^{-1}$ or 32 $\mu g\ L^{-1}$, when mixing or TMAH was used for dispersion, respectively (Table 2). After the treatment of landfill leachate with nZVI, strong complexes between organic acids and Cu were formed. This complexation caused the desorption of Cu from the surface of nZVI. As a result, Cu was released into the leachate. Its concentration was increased to about 240 $\mu g\ L^{-1}$, and was much higher than that found in the untreated leachate. The treatment of leachate with nZVI is not efficient for the Cu removal due to the repulsive forces between the formed negatively charged Cu complexes and nZVI particles which kept Cu in solution.

In the untreated leachate and in the leachate treated with FeNPs, V was present in the dissolved form. Its concentration in the leachate was 27 $\mu g\ L^{-1}$. After the treatment with Fe_3O_4NPs, the extent of V removal depended on the pH, which influences the V speciation. The more effective dispersion of FeONPs treated with TMAH in comparison to Fe_3O_4NPs, contributes to a better removal of V from the leachate. When the leachate was treated with nZVI, V present in the sample in oxidation states V^{5+} and V^{4+} was reduced to its stable V^{3+} species. V^{3+} readily reacts with available organic ligands and forms negatively charged complexes in neutral up to slightly alkaline pH ranges (Briucal et al., 2004). Repulsive forces between the negatively charged V(III) complexes and nZVI particles prevented V removal from the leachate investigated.

4. Conclusions

- The settling and removal of FeNPs after the treatment of environmental waters depends strongly on the sample characteristics and the ways of NPs dispersion.
- After the treatment of Milli Q water with nZVI dispersed by mixing or TMAH, a precipitate was formed and nZVI was efficiently removed from the solution within 24 h. FeONPs and Fe_3O_4NPs dispersed by mixing were also effectively removed. However, dispersion with TMAH resulted in the persistence of FeONPs also 24 h after the treatment. Nanosized iron remained dispersed in the same concentration as was added (1500 mg L^{-1} Fe), since a double layer of oleic acid and TMAH was created around the FeONPs and prevented the formation of iron corrosion products. Fe_3O_4NPs dispersed with TMAH persisted in solution still 24 h after the treatment, but in much lower extent (70 mg L^{-1} Fe).
- In forest spring water, the ionic strength of the sample contributes to rapid removal of nZVI, FeONPs and Fe_3O_4NPs (efficient settling of iron corrosion products was achieved after 15 min). The exception was FeONPs dispersed with TMAH, which persisted in the nanosized form also 24 h after the treatment in the same iron concentration that was added (1500 mg L^{-1} Fe).
- In landfill leachate with much higher ionic strength and content of organic matter than in the forest spring water, the efficient removal of nZVI, FeONPs and Fe_3O_4NPs was obtained already 4 h after the FeNPs treatment. The effect of the high ionic strength that favours settling of NPs was more pronounced then the effect of organic matter, which contributes to better dispersion efficiency. The bacteria in the landfill leachate used oleic acid as a source of carbon and removed it from the surface of FeONPs. In

this way, the formation of iron corrosion products and the settling of particles were also possible when TMAH was used for dispersion of FeONPs.

- Despite its efficiency, TMAH is not recommended for being used as a dispersing agent in the treatment of environmental waters by FeNPs, due to its high toxicity.
- The removal of elements from the environmental water samples depends on the contaminant concentration, the type of FeNPs and the dispersion efficiency. An important role in the effectiveness of metal removal by FeNPs play the pH, the ionic strength, the content of organic matter, the presence of complexing ligands and the interactions of FeNPs with the sample matrix. The latter sample characteristics influence the chemical speciation of elements, which also importantly contributes to overall contaminant removal from the environmental waters.
- It should be pointed out that FeNPs may significantly contribute to the appearance of metal contaminants in water, which is remediated.
- The present investigation demonstrates the importance of the knowledge on the behaviour and the fate of FeNPs, when these are implemented for the remediation of environmental waters.

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References

Andrade, A.L., Fabris, J.D., Ardisson, J.D., Valente, M.A., Ferriera, M.F., 2012. Research article: Effect of tetramethylammonium hydroxide on nucleation, surface modification and growth of magnetic nanoparticles. Journal of Nanomaterials 2012, 1-9.

Auffan, M., Achouak, W., Rose, J., Roncato, M., Chanéac, C., Waite, D.T., Masion, A., Woicik, J.C., Wiesner, M.R., Bottero, J., 2008. Relation between redox state of iron-based nanoparticles and theri cytotoxicity toward Escherichia coli. Environmental Science and Technology 42, 6730-6735.

Bian, S., Mudunkotuwa, I.A., Rupasinghe, T., Grassian, V.H., 2011. Aggregation and dissolution of 4 nm ZnO nanoparticles in aqueous environments: influence of pH, ionic strength, size, and adsorption of humic acid. Langmuir, 27, 6059-6068.

Briucal, J., Lubes, V., Araujo, M.L., Brito, F., 2004. Vanadium(III) complexes in aqueous solutions with the dicarboxylic oxalic, malonic and succinic acids. Journal of the Chilean Chemical Society 49, 285-288.

Grieger, K.D., Fjordbøge, A., Hartmann, N.B., Eriksson, E., 2010. Environmental benefits and risks of zero-valent iron nanoparticles (nZVI) for in situ remediation: Risk mitigation or trade-off ? Journal of Contaminant Hydrology 118, 165-183.

Hotze, E.M., Phenrat, T., Lowry, G.V, 2010. Nanoparticle aggregation: Challenges to understanding transport and reactivity in the environment. Journal of Environmental Quality 39, 1909-1924.

Hu, J., Zevi, Y., Kou, X., Xiao, J., Wang, X., Jin, Y., 2010. Effect of dissolved organic matter on the stability of magnetite nanoparticles under different pH and ionic strength conditions. Science of the Total Environment 408 (16), 3477-3489.

Jiang, D., Hu, X., Wang, R., Yin, D., 2014. Oxidation of nanoscale zero-valent iron under sufficient and limited dissolved oxygen: Influences on aggregation behaviors. Chemosphere.

Jiang, J., Oberdöster, G., Biswas, P., 2009. Characterization of size, surface charge, and agglomeration state of nanoparticle dispersions for toxicological studies. Journal of Nanoparticle Research 11, 77-89.

Kamat, P.V., Meisel, D., 2003. Nanoscience opportunities in environmental remediation. Comptes Rendus Chimie 6, 999-1007.

Karn, B., Kuiken, T., Otto, M., 2009. Nanotechnology and in situ remediation: a review of the benefits and potential risks. Environmental Health Perspectives, 117 (12), 1823-1831.

Karthikeyeni, Vijayakumar, T.S., Vasanth, S., Ganesh, A., Manimegalai, M., Subramanian, P., 2013**.** Biosynthesis of iron oxide nanoparticles and its haematological effects on fresh water fish Oreochromis mossambicus. Journal of Academia and Industrial Research 1 (10), 645-649.

Kossa, W.C., MacGee, J., Ramachandran, S., Webber, A.J., 1979. Pyrolytic methylation/gas chromatography: A short review. Journal of Chromatographic Science, 17, 177-187.

Krajangpan, S., Chisholm, B.J., Kalita, H., Bezbaruah, A.N., 2009. Challenges in groundwater remediation with iron nanoparticles: enabling colloidal stability, *In* Nanotechnologies for water environment applications. Edts. Zhang, T.C., Surampalli, R.Y., Lai, K.C.K, Hu, Z., Tyagi, R.D., Lo, I.M.C., American Society of Civil Engineers, Reston, Virginia, USA, 191-212.

Lo, I.M.C., Hu, J., Chen, G., 2009. Iron-based magnetic nanoparticles for removal of heavy metals from electroplating and metal-finishing wastewater, *In* Nanotechnologies for water environment applications. Edts. Zhang, T.C., Surampalli, R.Y., Lai, K.C.K., Hu, Z., Tyagi, R.D., Lo, I.M.C., American Society of Civil Engineers, Reston Virginia, USA, 213-268.

Machadoa, S., Stawińskib, W., Sloninab, P., Pintoa, A.R., Grossoa, J.P., Nouwsa, H.P.A., Albergariaa, J.T., Delerue-Matosa, C., 2013. Application of green zero-valent iron nanoparticles to the remediation of soils contaminated with ibuprofen. Science of the Total Environment 461-462, 323-329.

Mauer-Jones, M.A., Gunsolus, I.L., Murphy, C.J., Haynes, C.L., 2013. Toxicity of engineered nanoparticles in the environment. Analytical Chemistry 85, 3036-3049.

Noubactep, C., 2010. Review: The fundamental mechanism of aqueous contaminant removal by metallic iron. Water SA 36 (5), 663-670.

Palanisamy, K.L., Devabharathi, V., Sundaram, N.M., 2013. The utility of magnetic iron oxide nanoparticles stabilized by carrier oils in removal of heavy metals from waste water. International Journal of Research in Applied, Natural and Social Sciences 1, 15-22.

Scherer, C., Figueiredo Neto, A. M., 2005. Ferrofluids: Properties and applications. Brazilian Journal of Physics 35 (3A), 718-727.

Shen, Y.F., Tang, J., Nie, Z.H., Wang, Y.D., Ren, Y., Zuo, L., 2009. Tailoring size and structural distortion of Fe3O4 nanoparticles for the purification of contaminated water. Bioresource Technology 100, 4139-4146.

Shipley, H.J., Engates, K. E., Guettner, A. M., 2011. Study of iron oxide nanoparticles in soil for remediation of arsenic. Journal of Nanoparticle Research 13, 2387-2397.

Singh, R., Misra, V., Singh, R.P., 2012. Removal of hexavalent chromium from contaminated ground water using zero-valent iron nanoparticles. Environmental Monitoring and Assesment 184, 3643-3651.

Sperling, R.A., Parak, W.J., 2010. Review: Surface modification, functionalization and bioconjugation of colloidal inorganic nanoparticles. Philosophical Transactions of the Royal Society A 368, 1333-1383.

Ščančar, J., Milačič, R., 2006. Aluminium speciation in environmental samples: a review. Analytical and Bioanalytical Chemistry 386, 999-1012.

Tang, S.N.C., Lo, I.M.C., 2013. Magnetic nanoparticles: Essential factors for sustainable environmental applications. Water Research 47, 2613-2632.

Tratnyek, P.G., Johnson, R. L., 2006. Nanotechnologies for environmental cleanup. Nanotoday 1 (2), 44-48.

Zhang, T.C., Surampalli, R.Y., 2009. Introduction, *In* Nanotechnology for Water Environmental Applications. Edts. Zhang, T.C., Surampalli, R.Y., Lai, K.C.K., Hu, Z., Tyagi, R.D., Lo, I.M.C., American Society of Civil Engineers, Reston Virginia, USA, 1-13.

Zhang, W., 2003. Nanoscale iron nanoparticles for environmental remediation: an overview. Journal of Nanoparticle Research 5, 323-332.

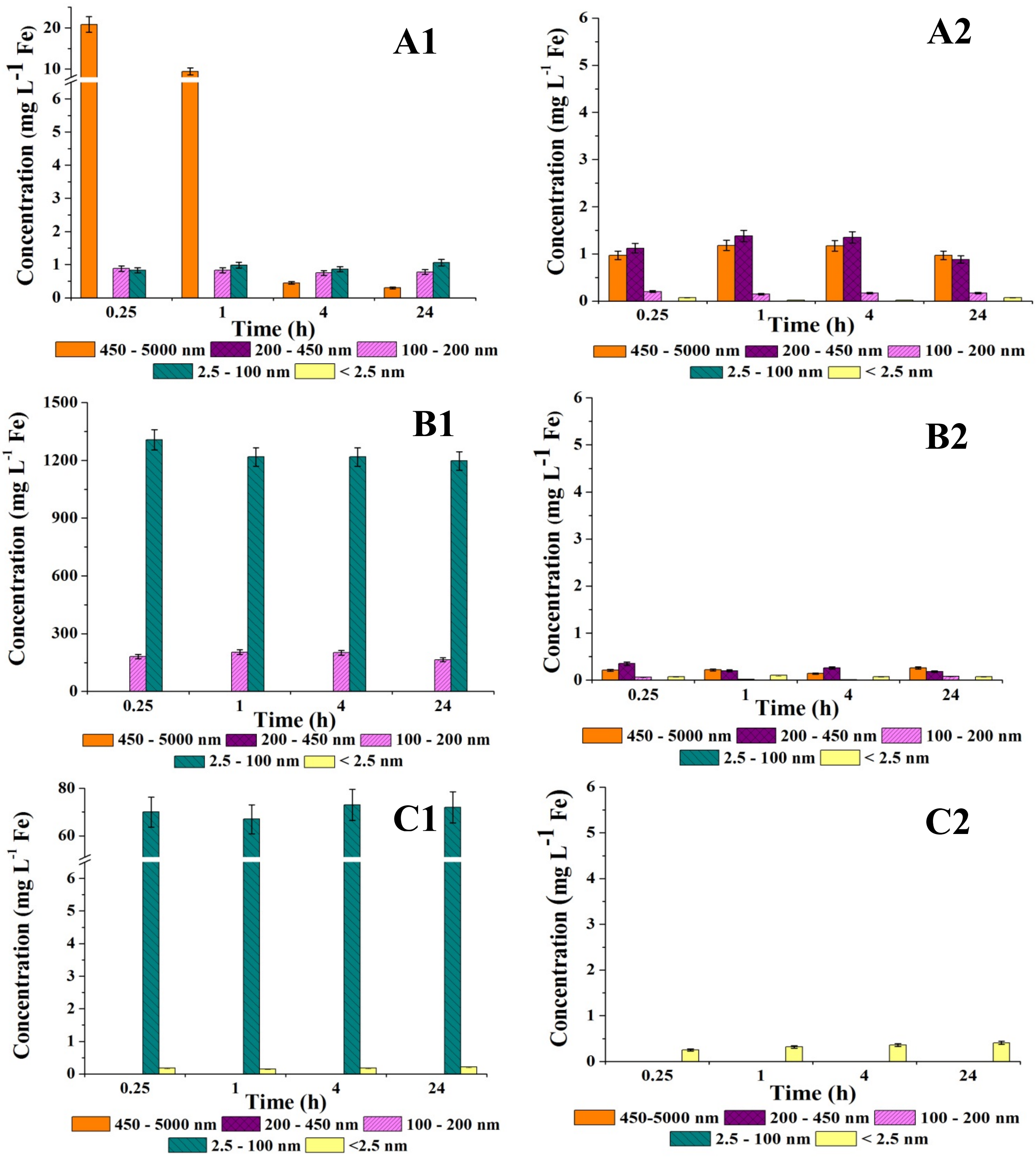


Fig. 1. Time dependent size distribution of iron in Milli Q water treated with nZVI (A), FeONPs (B) and Fe_3O_4NPs (C). FeNPs (iron load 1.5 g L^{-1}) were dispersed with TMAH (1) or by mixing (2). Iron in fractions obtained by the sequential filtration of samples was determined by ICP-MS. The error bars represent the experimental and analytical uncertainty.

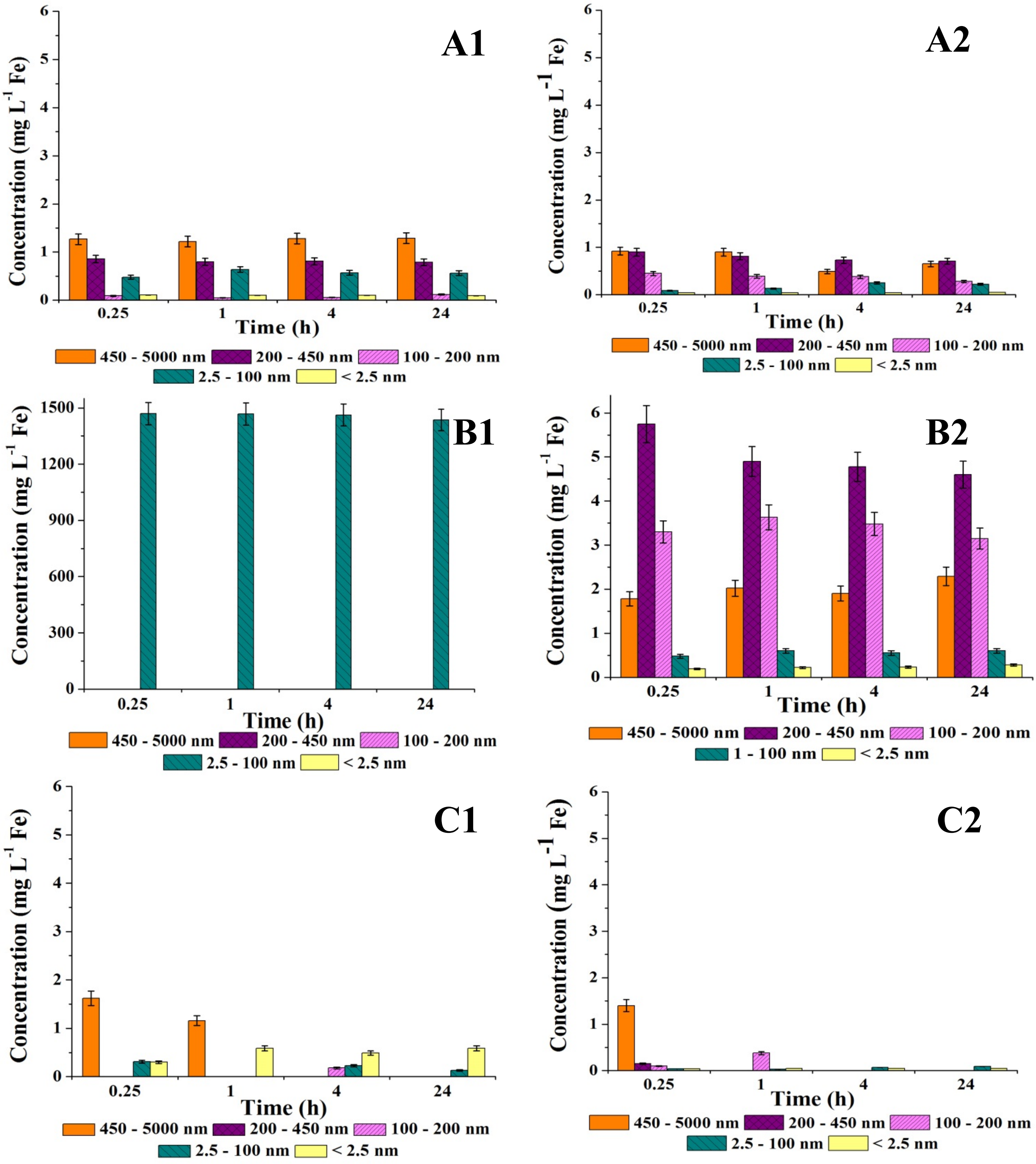


Fig. 2. Time dependent size distribution of iron in forest spring water treated with nZVI (A), FeONPs (B) and Fe_3O_4NPs (C). FeNPs (iron load 1.5 g L^{-1}) were dispersed with TMAH (1) or by mixing (2). Iron in fractions obtained by the sequential filtration of samples was determined by ICP-MS. The error bars represent the experimental and analytical uncertainty.

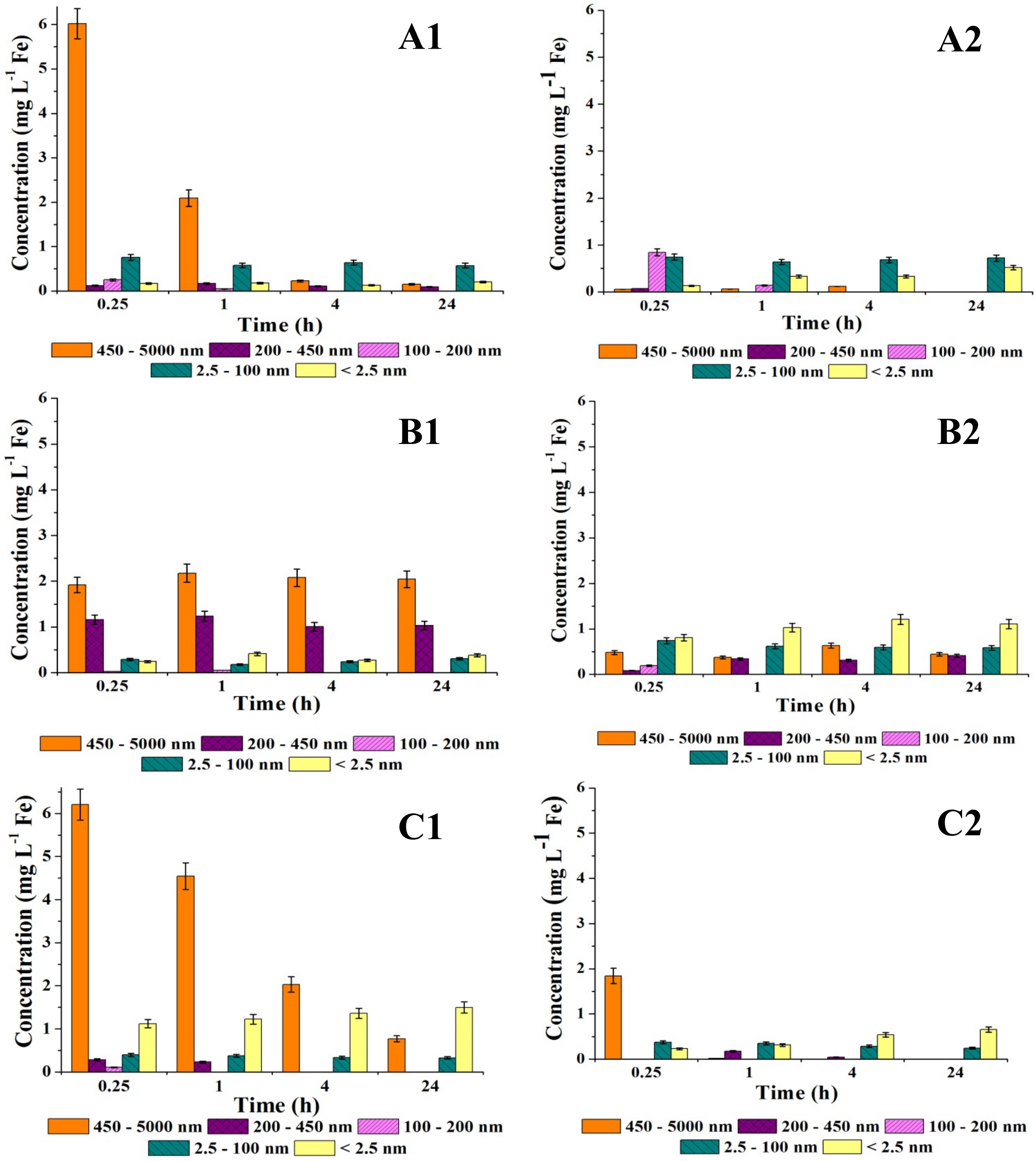


Fig. 3. Time dependent size distribution of iron in landfill leachate treated with nZVI (A), FeONPs (B) and Fe_3O_4NPs (C). FeNPs (iron load 1.5 g L^{-1}) were dispersed with TMAH (1) or by mixing (2). Iron in fractions obtained by the sequential filtration of samples was determined by ICP-MS. The error bars represent the experimental and analytical uncertainty.

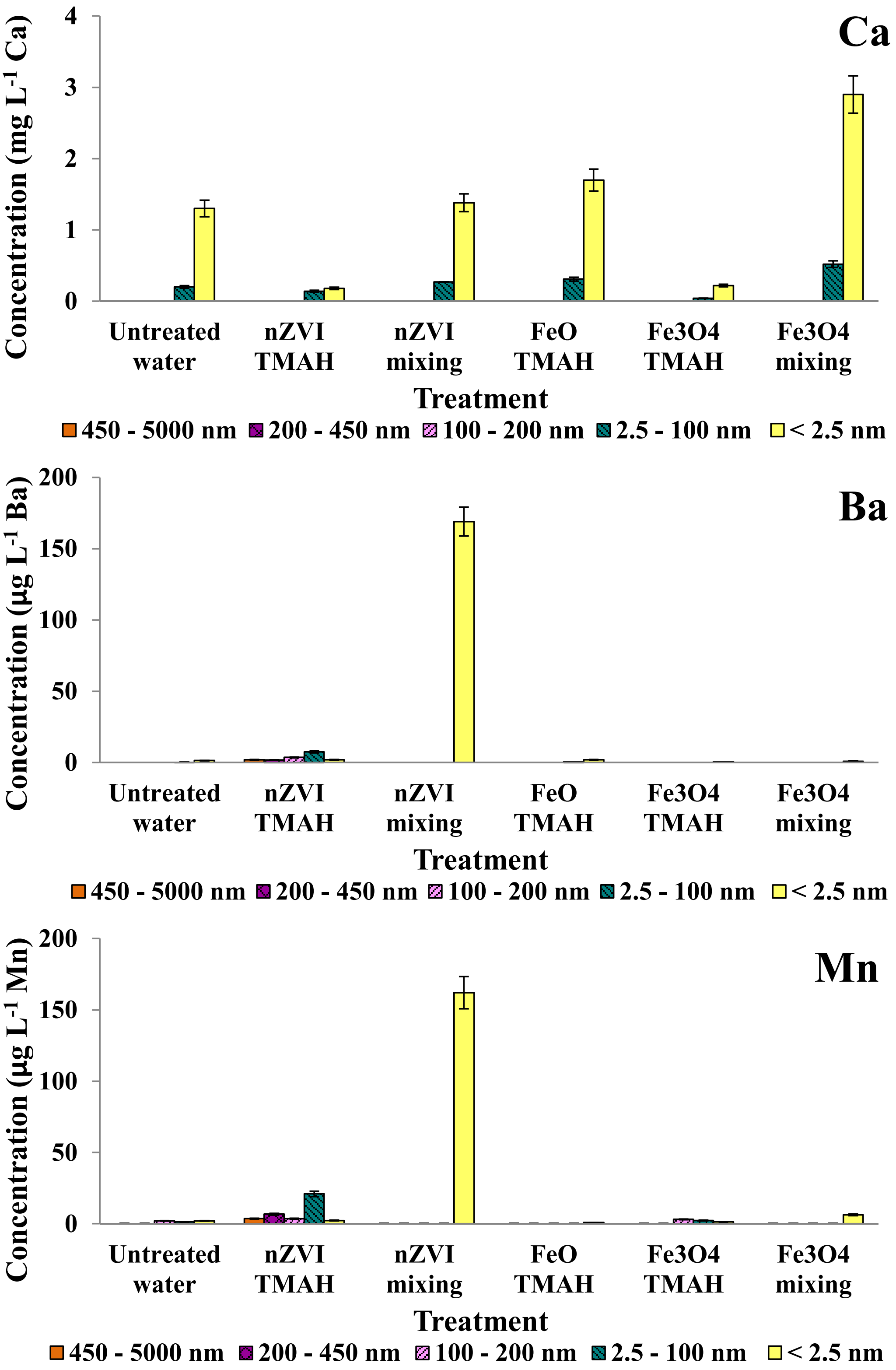


Fig. 4. Size distribution of metals in untreated forest spring water and in spring water 24 h after the treatment with nZVI, FeONPs or Fe_3O_4NPs, using TMAH or mixing for dispersion FeNPs. Elements in fractions obtained by the sequential filtration of samples were determined by ICP-MS. The error bars represent the experimental and analytical uncertainty.

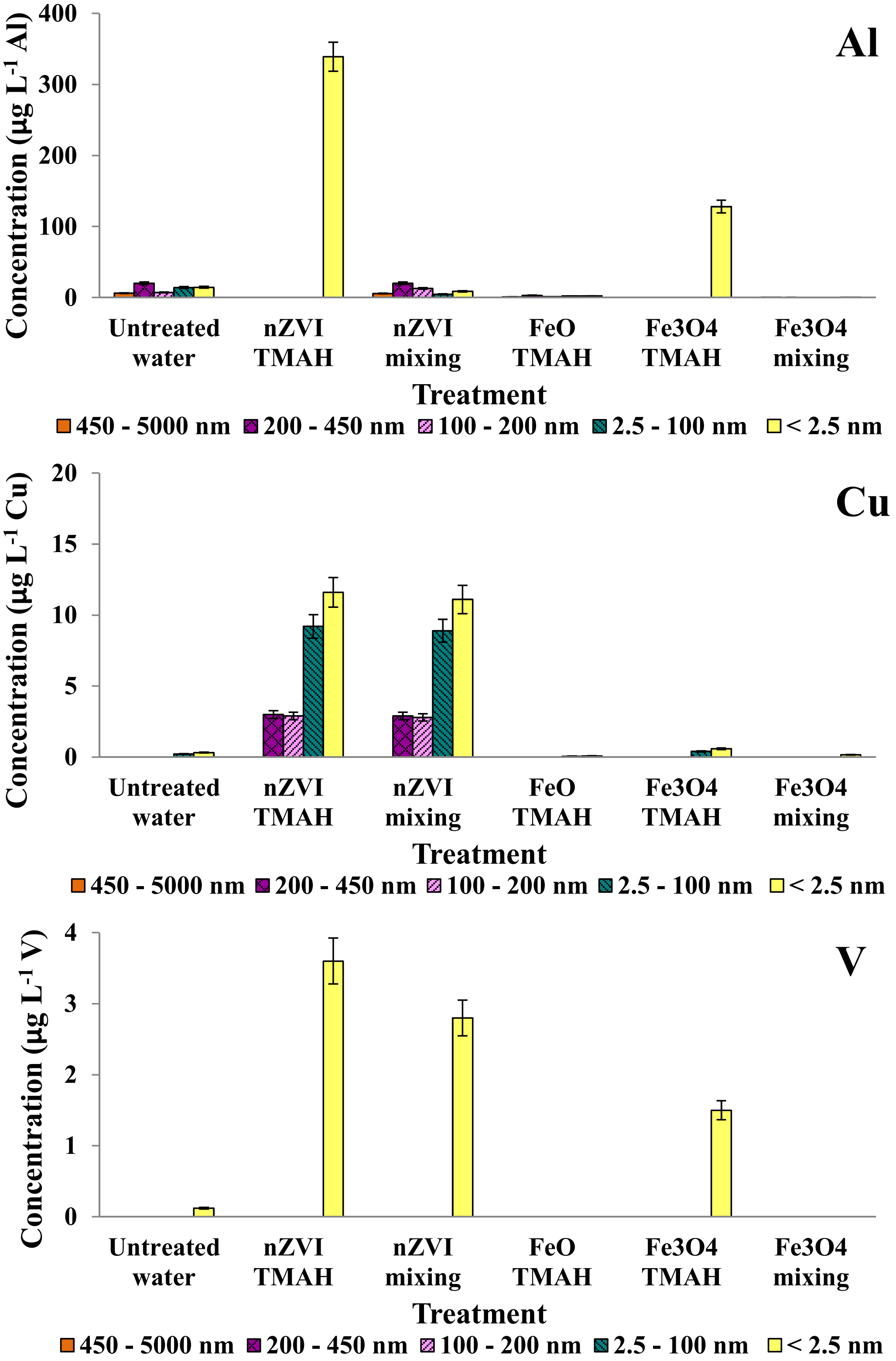


Fig. 4. (continued) Size distribution of metals in untreated forest spring water and in spring water 24 h after the treatment with nZVI, FeONPs or Fe_3O_4NPs, using TMAH or mixing for dispersion FeNPs. Elements in fractions obtained by the sequential filtration of samples were determined by ICP-MS. The error bars represent the experimental and analytical uncertainty.

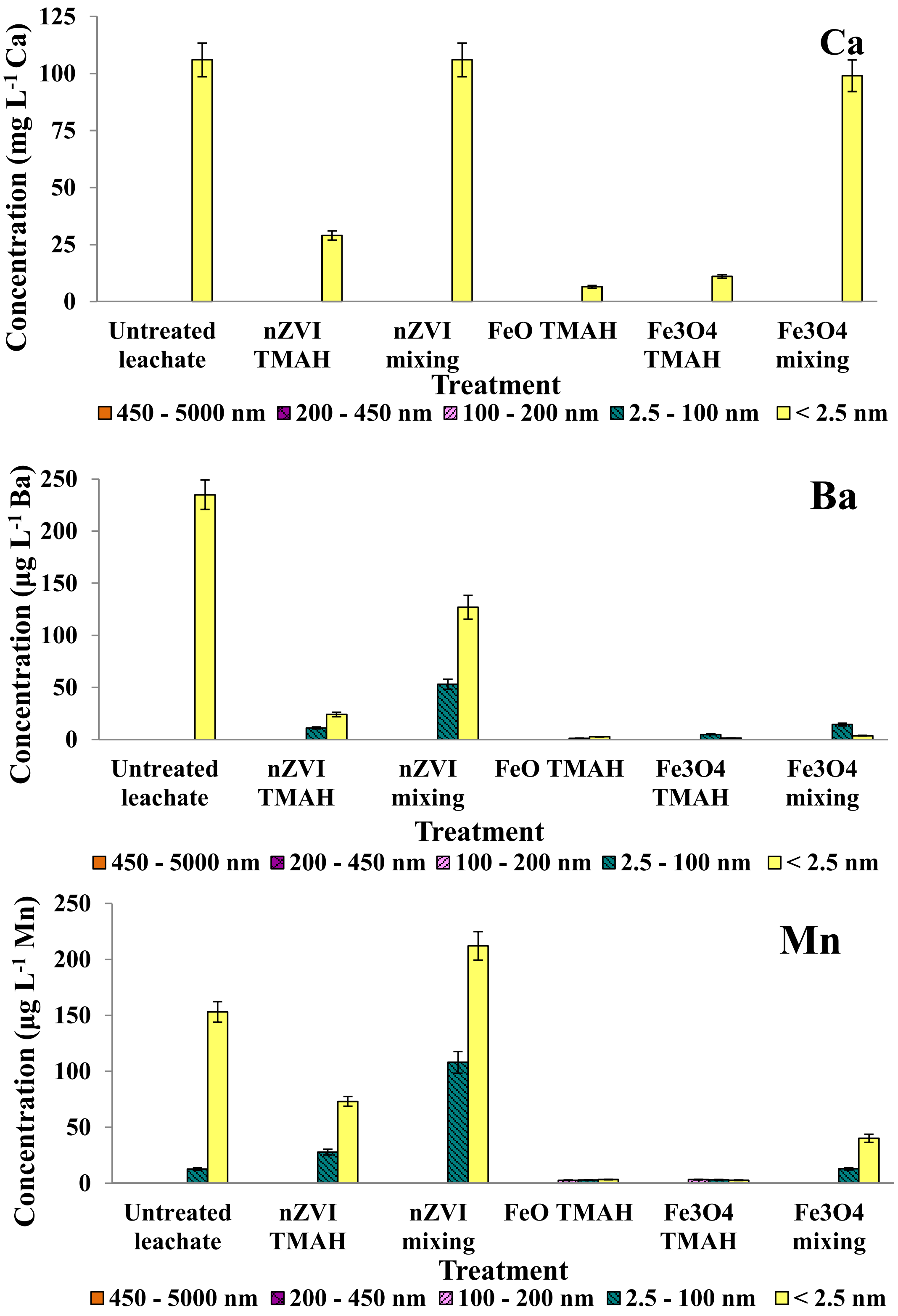


Fig. 5. Size distribution of metals in untreated landfill leachate and in leachate 24 h after the treatment with nZVI, FeONPs or Fe_3O_4NPs, using TMAH or mixing for dispersion FeNPs. Elements in fractions obtained by the sequential filtration of samples were determined by ICP-MS. The error bars represent the experimental and analytical uncertainty.

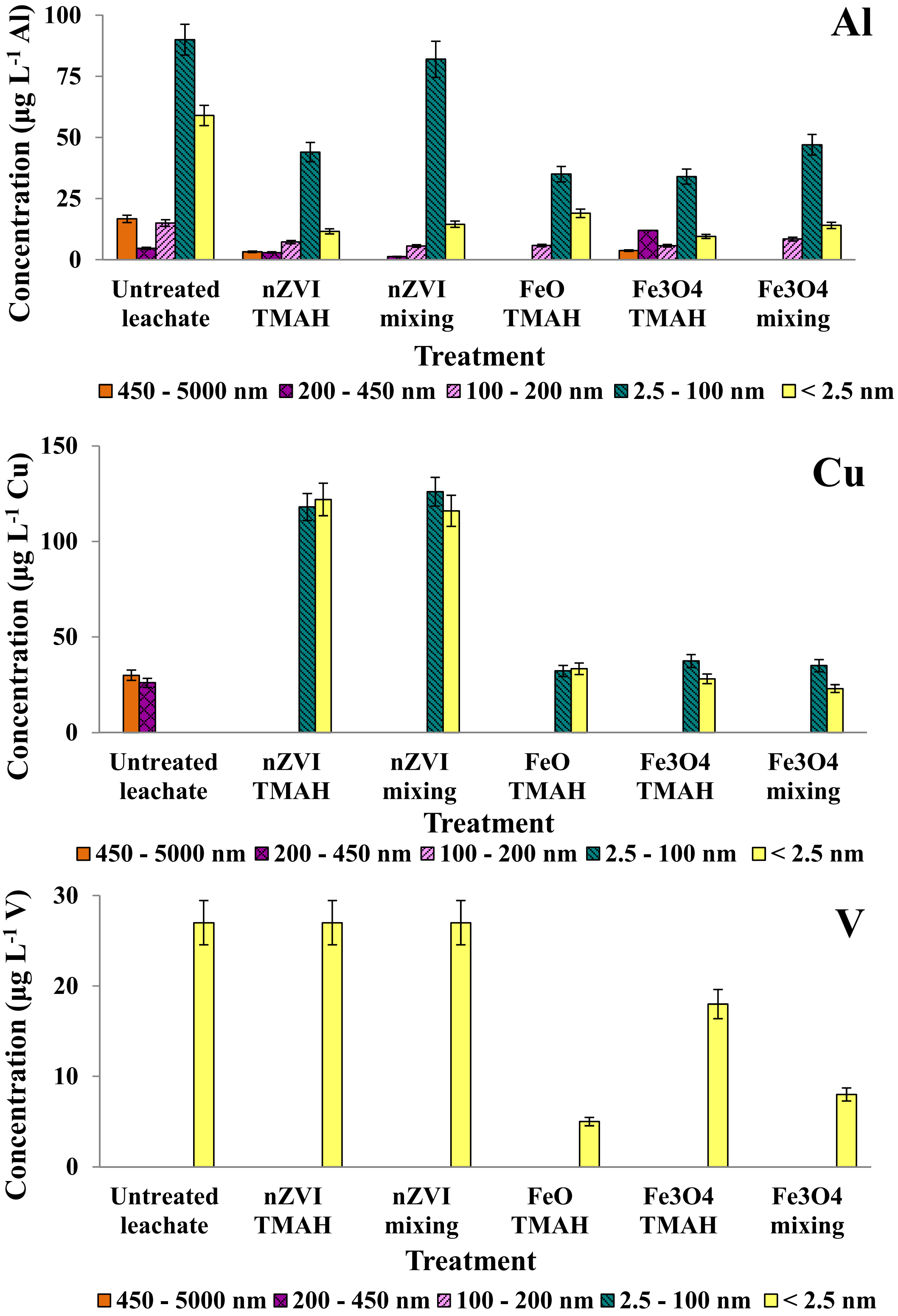


Fig. 5. (continued) Size distribution of metals in untreated landfill leachate and in leachate 24 h after the treatment with nZVI, FeONPs or Fe_3O_4NPs, using TMAH or mixing for dispersion FeNPs. Elements in fractions obtained by the sequential filtration of samples were determined by ICP-MS. The error bars represent the experimental and analytical uncertainty.

Table 1 - Sample characteristics of water samples.

Sample	Electrical conductivity (μS cm^{-1})	Total organic content (mg L^{-1})	Dissolved organic content (mg L^{-1})	pH
Milli Q water	0.055 ± 0.001	< 0.005	< 0.005	5.5 ± 0.1
Forest spring water	58 ± 1	45 ± 1	38 ± 1	5.5± 0.1
Landfill leachate	6800 ± 100	760 ± 15	690 ± 15	8.0 ± 0.1

Table 2 – Concentrations of element in aqueous solution of TMAH in Milli Q water and in Milli Q water treated with nZVI, FeONPs and Fe_3O_4NPs dispersed by mixing 24 h after treatment with FeNPs. Concentrations of elements were determined by ICP-MS.

Parameter	Ca (mg L^{-1})	Ba (µg L^{-1})	Mn (µg L^{-1})	Al (µg L^{-1})	Cu (µg L^{-1})	V (µg L^{-1})
nZVI (Mixing)	0.63 ± 0.06	176 ± 9	181 ± 9	38 ± 3	26 ± 2	2.8 ± 0.3
nZVI (TMAH)	0.30 ± 0.03	8.7 ± 0.8	50 ± 5	277 ± 7	32 ± 3	3.7 ± 0.7
FeONPs (TMAH)	0.53 ± 0.05	1.6 ± 0.1	0.36 ± 0.032	8.1 ± 0.7	0.13 ± 0.01	< 0.1
Fe_3O_4NPs (Mixing)	1.5 ± 0.1	2.7 ± 0.2	25 ± 2	0.25 ± 0.02	< 0.1	< 0.1
Fe_3O_4NPs (TMAH)	0.33 ± 0.03	0.97 ± 0.09	3.1 ± 0.3	65 ± 5	0.54 ± 0.005	1.3 ± 0.1

Table 3 – pH of water samples after the treatment with FeNPS, using TMAH or mixing for dispersion.

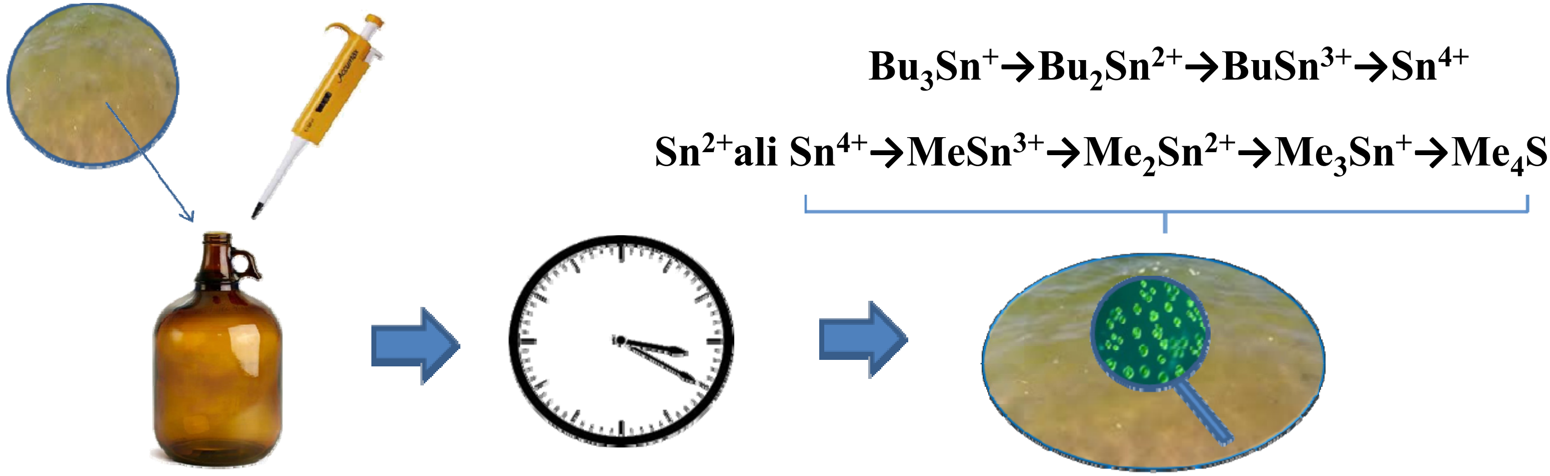
Sample	FeNPs	Treatment TMAH	Treatment mixing
Milli Q water	nZVI	13	6
	FeONPs	12	6
	Fe_3O_4NPs	13	6
Forest spring water	nZVI	13	6
	FeONPs	12	6
	Fe_3O_4NPs	13	6
Landfil leachate	nZVI	9	8
	FeONPs	9	8
	Fe_3O_4NPs	9	8

Table S1 - ICP-MS operating parameters for determination of element concentrations.

Parameter	Type/Value	He mode (4.5 mL He min^{-1})	No gas mode
Sample introduction			
Nebuliser	Miramist		
Spray chamber	Scott		
Skimmer and sampler	Ni		
Plasma conditions			
Forward power	1550 W		
Plasma gas flow	15.0 L min^{-1}		
Carrier gas flow		1.00 L min^{-1}	1.00 L min^{-1}
Make-up gas flow		0.15 L min^{-1}	0.15 L min^{-1}
QP bias		-15.0 V	-3.0 V
Oct bias		-18.0 V	-8.0 V
Cell entrance		-40.0 V	-30.0 V
Cell exit		-60 V	-50 V
Deflect		0 V	12.8 V
Plate bias		-60 V	-40 V
Sample uptake rate	0.3 mL min^{-1}		
Data acquisition parameters			
Isotopes monitored		^{27}Al, ^{51}V, ^{52}Cr, ^{55}Mn, ^{56}Fe, ^{59}Co, ^{60}Ni, ^{63}Cu, ^{66}Zn, ^{75}As, ^{78}Se, ^{88}Sr, ^{95}Mo	^{111}Cd, ^{118}Sn, ^{121}Sb, ^{137}Ba, ^{208}Pb
Isotopes of internal standards		^{45}Sc, ^{72}Ge, ^{103}Rh, ^{115}In	^{45}Sc, ^{72}Ge, ^{103}Rh, ^{115}In

Priloga 3

Študij pretvorb organokositrovih spojin v izcednih vodah iz odlagališč odpadkov



Izcedna voda iz
deponij odpadkov

^{117}Sn, ^{119}Sn obogateni izotopi

$Bu_3Sn^+ \rightarrow Bu_2Sn^{2+} \rightarrow BuSn^{3+} \rightarrow Sn^{4+}$

Sn^{2+} ali $Sn^{4+} \rightarrow MeSn^{3+} \rightarrow Me_2Sn^{2+} \rightarrow Me_3Sn^+ \rightarrow Me_4Sn$ (plin)

Obogatene izotope kositrovih spojin $^{117}Bu_3Sn^+$, $^{119}Bu_2Sn^{2+}$, $^{117}SnCl_2$, $^{117}SnCl_4$ in ^{119}Sn mešanice butilkositrovih spojin smo uporabili pri študiju pretvorb organokositrovih spojin v izcednih vodah iz odlagališč odpadkov. Biotske pretvorbe, ki potekajo z mikroorganizmi smo od abiotskih procesov ločili tako, da smo del vzorca izcedne vode sterilizirali in rezultate primerjali z nesterilnimi vzorci. Rezultati so pokazali, da poteka razgradnja Bu_3Sn^+ z mikroorganizmi, Bu_2Sn^{2+} pa z abiotskimi pretvorbami. Z obogatenimi izotopi Sn^{2+} ali Sn^{4+} smo dokazali, da poteka metilacija kositrovih spojin v izcednih vodah z mikroorganizmi. Rezultati študije pomembno prispevajo k poznavanju procesov pretvorb organokositrovih spojin v izcednih vodah in pomagajo načrtovati ustrezne ukrepe za preprečitev sproščanja strupenih metilkositrovih spojin v okolje.

K. Peeters, T. Zuliani, J. Ščančar, R. Milačič. The use of isotopically enriched tin tracers to follow the transformation of organotin compounds in landfill leachate. *Water Research*, 2014, 53, 297-309. [IF=5.323, ZR - water resources; 1/81, A''']