Impact of biosolids, ZnO, ZnO/biosolids on bacterial community and enantioselective transformation of racemic–quizalofop–ethyl in agricultural soil

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ABSTRACT

The effects of biosolids, ZnO, and ZnO/biosolids on soil microorganism and the environmental fate of coexisting racemic–quizalofop–ethyl (rac-QE) were investigated. Microbial biomass carbon in native soil, soil/biosolids decreased by 62% and 52% in the presence of ZnO (2‰, weight ratio). The soil bacterial community structure differed significantly among native soil, soil/biosolids, soil/ZnO, and soil/biosolids/ZnO based on a principal co-ordinate analysis (PCoA) of OTUs and one-way ANOVA test of bacterial genera. Chemical transformation caused by ZnO only contributed 4% and 3% of the overall transformation of R-quizalofop-ethyl (R-QE) and S-quizalofop-ethyl (S-QE) in soil/ZnO. The inhibition effect of ZnO on the initial transformation rate of R-QE (rR-QE) and S-QE (rS-QE) in soil only observed when enantiomer concentration was larger than 10 mg/kg. Biosolids embedded with ZnO (biosolids/ZnO) caused a 17%–42% and 22%–38% decrease of rR-QE and rS-QE, although rR-QE and rS-QE increased by 0%–17% and 22%–58% by the addition of biosolids. The results also demonstrated that the effects of biosolids on agricultural soil microorganism and enantioselective transformation of chiral pesticide was altered by the embedded nanoparticles.

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Introduction

Engineered nanoparticles (ENPs) have been widely applied in many realms because of their unique optical and electrical properties (Singh, 2016). ENPs could directly enter the terrestrial system through its application in modern agriculture or soil remediation (Chen et al., 2014; Peters et al., 2016). Moreover, ENPs has a high affinity with activated sludge (Kiser et al., 2012; Tou et al., 2017), and therefore, fertilization with biosolids containing ENPs would also increase the opportunity for ENPs to enter agricultural soil. Despite the unknown concentration of most ENPs in ecosystems, it has been suggested that the concentration of ENPs is higher in soil than in water or air (Giese et al., 2018; Meesters et al., 2016). Once has entered agricultural soil, ENPs underwent a complicated transportation/transformation process (Batley et al., 2013; Larue et al., 2018). Accompanied by these processes, ENPs could directly or indirectly influence soil microbe structure, and the impacts on microbial composition and activity were highly dependent on ENPs and soil characteristics (Ge et al., 2011; Simonin and Richaume, 2015; Xu et al., 2015). ENPs exhibited both stimulatory and inhibitory influences on the terrestrial plants, and the practical effects were mostly determined by ENPs' characteristics and experimental conditions (Rizwan et al., 2017; Zuverza-Mena et al., 2017).

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Compared with the wide investigation of interactions between ENPs and soil microbes, ENPs and terrestrial plants, the study of the influence of ENPs on the environmental fate of coexisting pollutant is quite limited (Gardea-Torresdey et al., 2014). It has been reported that ENPs influenced the uptake and transportation of organic pollutant and heavy metal ions by crops. For example, the bioaccumulation of Cd, Cu, and Pb under co-contamination scenario differed significantly from under single contaminant (Li et al., 2018). It has been reported that the transport of rac-metalaxyl in soil decreased in the presence of nano-SiO₂ (Huang et al., 2017). The chemical transformation process of rac-metalaxyl in agricultural soils was highly promoted in the presence of TiO₂ because of the generation of reactive oxygen species (ROS) (Huang et al., 2018; Liang et al., 2016). Conversely, the biotransformation of rac-metalaxyl was not affected by TiO₂, which was because TiO₂ exhibited no significant influence on bacterial structure (Huang et al., 2018). Compared to TiO₂, some other ENPs like ZnO, CuO, etc. showed a more profound influence on soil microbes or enzyme activities (Rajput et al., 2018; Xu et al., 2015), and because microorganisms play a crucial role in biogeochemical cycling (Nannipieri et al., 2017; Schulz et al., 2013), biotransformation of organic pollutants in soils could be subsequently changed by these ENPs. However, their effects on the biotransformation of coexisting pollutants, and to what extent the difference would be still unknown.

It was well-known that ENPs' photoactivity and their influence on soil microorganisms were highly dependent on their characteristics. Huang et al. (2018) found that difference in photoactivity of four TiO₂ in soil was not as much as that in aqueous solution. Citrate-coated gold nanoparticles (nAu) showed a higher stimulation effect on soil enzyme activity than polyvinylpyrrolidone (PVP)-coated nAu, and bacterial structure in soil exposed to citrate-coated nAu differed significantly from soil spiked with PVP-nAu (Asadishad et al., 2017). It also observed that the nAu morphology exhibited a stronger force in shaping the microbial community structure than does the surface coating (Metch et al., 2018). Introduction of ENPs that formulated with biosolids could also affect soil enzyme activities and shift soil bacterial community (Colman et al., 2013; Judy et al., 2015). Biosolids are rich in organic matter with various functional groups, and therefore ENPs' properties could change to some extent when it is embedded in biosolids and subsequently shift its effect on soil microorganisms (Judy et al., 2015). The investigations of ENPs' affection on soil microorganism structure and transformation of coexisting pollutant are still limited.

We have suggested that investigation the change of enantiomeric degradation of chiral compounds helps in revealing the effects of ENPs on the transformation of coexisting pollutants in agricultural soils (Huang et al., 2018), which was because the change of biotransformation could lead to the change of enantiomer fraction (EF). Quizalofop-ethyl (QE) is a widely applied herbicide to post-emergence control of annual and perennial grass weeds in various field crops, and its enantiomeric transformation in soils has been investigated that could provide background information for comparison (Li et al., 2012; Ma et al., 2016). Nano-ZnO was selected as an ENP proxy because it was one of the most widely used ENPs, and exhibited significant impacts on soil microorganisms (Rajput et al., 2018; Xu et al., 2015). This study systematically investigated the effects of ZnO, biosolids, ZnO/biosolids on soil bacterial structure, and the chemical and biological transformations of rac-QE. The results helped in understanding the potential effects of ENPs on the terrestrial system in the absence and the presence of biosolids.

1. Materials and methods

1.1. Chemicals

Rac-QE agent (96%) and nano-ZnO (30 nm) was purchased from Aladdin (Shanghai, China). HPLC grade methanol and acetonitrile were purchased from Fisher Scientific (Hampton, NH, USA). Biosolids was a commercial product (Lvang biological recycling Co., Ltd., Wuhan, China) and was used as received. The ultrapure water was used through the experiments.

1.2. Soil samples

Native soil: An agricultural soil (47.7% sand, 43.1% slit, and 9.2% clay) was collected from a farmland in Qianjiang (30°33′46.9″N 112°39′19.6″E, Hubei Province, China), and its cation exchange capacity and clay mineralogy was reported in a previous study (Huang et al., 2017). The soil was air-dried and sieved (2 mm) and stored at 4°C. Before incubation of rac-QE, the soil samples were pre-incubated in the dark at 25°C for 2 weeks with a moisture content of 15%.

Sterilized soil: Native soil samples were added to a conical flask and sealed with parafilm, and then were sterilized by autoclaving at 121°C for 90 min. The flask was taken out and placed in a dark incubator at 25°C for 2 days before the next sterilization. The autoclave sterilization was repeated three times.

Soil amended with ZnO (soil/ZnO): ZnO powder (2 mg ZnO per kilogram native soil) was directly added to the pre-incubated native soil, and then the mixture was shaken for 2 days to get a homogeneous ZnO amended soil (soil/ZnO).

Soil amended with biosolids (soil/biosolids): Biosolids powder (50 g biosolids per kg native soil) was directly added to the pre-incubated native soil, and then the mixture was shaken for 2 days to get a homogeneous biosolids amended soil (soil/biosolids).

Soil amended with biosolids containing ZnO (soil/biosolids/ZnO): ZnO and biosolids (2 mg ZnO per 50 g biosolids) was first thoroughly mixed, and then added to the pre-incubated native soil, and then soil/biosolids/ZnO was shaken for 2 days to get a homogeneous mixture.

Soil/ZnO, soil/biosolids, and soil/biosolids/ZnO were pre-incubated in the dark at 25°C for 2 weeks with the moisture content of 15% before they were used for rac-QE incubation.

1.3. Soil characterization

Elements for native soil, soil/biosolids, soil/ZnO and soil/biosolids/ZnO were determined by a Vario MACRO cube element analyzer (Elementar, Germany). Soil pH was
determined according to ISO 10390: 2005. Microbial biomass carbon (MBC) was quantified by chloroform fumigation.

1.4. Incubation of rac-QE in soil samples

Different volumes of rac-QE stock solution were added to a glass beaker containing 30 g of soil samples, and the resulting initial rac-QE ranged from 5.0 to 50 mg/kg. rac-QE spiked soil was then thoroughly mixed to achieve a homogeneous sample, and then the beaker (with sample inside) was sealed with fine-meshed transparent film to decrease evaporation. The incubation was conducted either under light irradiation or under dark at 25°C with a soil moisture content of 15%. The spectral irradiance for light incubation was shown in Appendix A Fig. S1. Transformation of rac-QE was also investigated in artificial soil (composed of silica sand and kaolinite) with biosolids (5%, weight ratio) to assess the contribution of biosolids.

1.5. DNA extraction of soil sample and bacterial community analysis

Detailed procedure for DNA extraction and sequences analysis was reported in a previous study (Huang et al., 2018). The operational taxonomic units (OTUs) were clustered with a standard of similarity of 97%. A taxonomic comparison of the OTUs was conducted in the Silva database to obtain the species classification information for each OUT. Bacterial community analyses were performed on the I-sanger platform (http://www.i-sanger.com).

1.6. Extraction and enantioselective analysis of rac-QE

During the transformation of rac-QE, 2 g of subsample was withdrawn at different time intervals. These samples were immediately transferred into a glass tube, and 4 mL of a water:acetonitrile mixture (1:4, V:V) was added to the soil samples. Tubes were vortexed for 1 min, then vigorously shaken for 2 hr at 160 r/min in the dark at 25°C. Aliquots (2 mL) were pipetted from sealed glass tubes and centrifuged at 12,000 r/min for 5 min. The supernatant was then collected and then the beaker (with sample inside) was sealed with fine-meshed transparent film to decrease evaporation. The incubation was conducted either under light irradiation or under dark at 25°C with a soil moisture content of 15%. The spectral irradiance for light incubation was shown in Appendix A Fig. S1. Transformation of rac-QE was also investigated in artificial soil (composed of silica sand and kaolinite) with biosolids (5%, weight ratio) to assess the contribution of biosolids.

1.7. Data analysis

The transformation rate (r, mg/(kg·hr)) of S-QE and R-QE were determined from the linear regression of the slope of a plot of S-QE and R-QE concentration over time. The Michaelis-Menten equation was used to fit correlation between r and the initial concentration of quizalofop-ethyl (CQE, mg/kg).

\[ r = \frac{-dC_{QE}}{dt} = \frac{r_{max} C_{QE}}{K_{m} + C_{QE}} \]  

(1)

In Eq. (1), \( r_{max} \) is the maximum transformation rate of quizalofop-ethyl enantiomer, t (hr) is the reaction time and \( K_{m} \) (mg/kg) is the concentration of quizalofop-ethyl enantiomer at which the reaction rate is half of \( r_{max} \).

EF was calculated by Eq. (2):

\[ EF = \frac{c_{R}}{c_{S} + c_{R}} \]  

(2)

where, \( c_{S} \) (mg/kg) and \( c_{R} \) (mg/kg) are the concentrations of S-QE and R-QE, respectively.

The difference in bacterial communities between native and restored soils was assessed by a one-way analysis of variance (ANOVA) followed by a Tukey test. All the statistical analysis was conducted using the Statistical Packages for the Social Sciences 20 (SPSS, Chicago, USA).

2. Results and discussion

2.1. Soil properties and bacterial diversity

Native soil used in the study is a sandy loam soil that has a pH of 8.3, and the organic carbon (OC) content is 0.92% (Table 1). With the addition of biosolids (mass ratio, 5%), OC increased to 1.62% and content of N, S, H also increased to different extents, but soil pH decreased to 7.7. The presence of ZnO (mass ratio, 0.2%) showed no influence on pH, OC and other element content either in soil/ZnO or soil/biosolids/ZnO. On the other side, MBC increased from 71.5 to 104.9 mg/kg in the presence of 5% biosolids. Conversely, MBC in soil/ZnO decreased to 27.2 mg/kg with the addition of 0.2% ZnO. MBC also decreased to 50.4 mg/kg in soil/biosolids/ZnO, which was 48% of that in soil/biosolids.

Bacterial diversity was also greatly affected by the addition of biosolids and ZnO (Fig. 1). The overall results suggested that

| Table 1 – Properties of soil, soil/biosolids, soil/ZnO and soil/biosolids/ZnO. |
|-----------------------------|-----------------------------|-----------------------------|-----------------------------|
| Properties                  | Soil                        | Soil/biosolids              | Soil/ZnO                    | Soil/biosolids/ZnO           |
| MBC (mg/kg)                 | Dark incubation             | Light irradiation*          |                             |                             |
| 71.5                        | 104.9                       | 27.2                        | 50.4                        |
| 94.6                        | 114.6                       | 32.6                        | 54.7                        |
| Element C (%)               |                             |                             |                             |                             |
| H                           | 0.29                        | 0.41                        | 0.31                        | 0.41                        |
| N                           | 0.09                        | 0.21                        | 0.10                        | 0.23                        |
| S                           | 0.03                        | 0.07                        | 0.03                        | 0.07                        |
| pH                          | 8.3                         | 7.7                         | 8.5                         | 7.5                         |

* Light irradiation means an 3 hr exposure to the irradiation sources (Appendix A Fig. S1).
the bacterial compositions differed between native soil and biosolids or ZnO-amended soil. An average of 43,971 quality sequences (n = 12, range: 37,631–50,372) were obtained, and the obtained OTUs were 1533 (soil), 1454 (soil/biosolids), 1413 (soil/ZnO) and 1483 (soil/biosolids/ZnO), respectively. ZnO caused a substantial decrease of OTUs in native soil, but OTUs number was comparable between soil/biosolids and soil/biosolids/ZnO. The presence of ZnO caused a significant decrease of MBC in soil with or without biosolids. Besides bacterial richness indexed by OTUs, a sharper decline in the rank-abundance curve for soil/ZnO further indicates that the bacterial evenness was lower in soil/ZnO than native soil (Fig. 1a). Conversely, the presence of ZnO did not influence bacterial evenness in soil/biosolids.

As shown in Fig. 1b, most OTUs overlapped among soil, soil/biosolids, soil/ZnO and soil/biosolids/ZnO; 96.7%, 93.2% and 92.0% of OTUs in soil/ZnO, soil/biosolids and soil/biosolids/ZnO were found in native soil, respectively. Principal co-ordinate analysis (PCoA) by a weighted UniFrac method indicated a dense cluster of bacterial communities in these soil samples, and bacteria were distributed separately from each other among the four samples (Fig. 1c). One-way ANOVA test indicated that 9/10 of the top 10 bacterial genera were significantly different among soil, soil/biosolids, soil/ZnO and soil/biosolids/ZnO (Fig. 1d). The major bacteria phyla were the same in native soil, soil/ZnO, soil/biosolids, and soil/biosolids/ZnO, but with a significant difference in their relative proportions. Except for Acidobacteria, Planctomycetes, and Verrucomicrobia (Appendix A Fig. S2),

Fig. 1 – (a) Rank-abundance curve of OTUs, (b) Venn diagram, (c) principal co-ordinate analysis (PCoA), (d) relative proportions of bacteria genera from the bacterial communities in native soil, soil/ZnO, soil/biosolids, soil/biosolids/ZnO. Different letters (x, y, and z in Fig. 1d) indicate significant differences (p < 0.05).
one-way ANOVA analysis showed that the relative proportion of the other seven major bacteria phyla was significantly different \((p < 0.05)\). The results clearly demonstrated that bacterial community was shifted by the addition of biosolids, ZnO or both of them, which could lead to the change of enantioselective transformation of chiral pesticides.

### 2.2. Enantioselective transformation of rac-QE in soil samples

Transformation of rac-QE was rapid in native soil and showed slight enantioselectivity (Fig. 2). Control experiment showed that less than 5% of the initial added rac-QE decomposed after a 72-hr incubation in autoclaved soil (data not shown), indicating the transformation of rac-QE in native soil occurred dominantly through a biotransformation process. On the other side, the result of the transformation of enantiopure R-QE demonstrated that no enantiomerization occurred in the conducted soils. Transformation process could be well fitted with the first-order kinetic model, and the obtained transformation kinetic constant \((k_{\text{obs}} \text{ day}^{-1})\) was 8.02 and 9.36 for R-QE and S-QE for an initial concentration of 2.5 mg/kg, respectively. \(k_{\text{obs}}\) decreased to 6.29 and 7.22 day\(^{-1}\) with a spiked concentration of approximately 5 mg/kg for each enantiomer. These values for \(k_{\text{obs}}\) are 3.7 and 3.9 times that of \(k_{\text{obs}}\) reported in a previous study (Li et al., 2012). The main reason might be the vast diversity in microbe’s abundance and structure between these two soils (Huang et al., 2018). Conversely, Ma et al. (2016) found that elimination of R-QE was faster than S-QE in the other two soils. The different enantioselectivity may result from diverse properties of soil samples because both microbes and soil physicochemical property could affect the enantioselective transformation of chiral compounds in the terrestrial system. It also found that transformation of rac-QE was faster in Chinese soils than in soils from other countries (López-Ruiz et al., 2017; Li et al., 2012; Ma et al., 2016; Mantzos et al., 2016). Metabolism of rac-QE in soil usually yields quizalofop-acid as the primary products in soils (López-Ruiz et al., 2017).

Transformation of R-QE and S-QE enantiomer was comparable in soil/ZnO, and \(k_{\text{obs}}\) was 8.69 and 8.88 day\(^{-1}\) for R-QE and S-QE. Although the change was not statistically significant \((p > 0.05)\), the values indicated a slight increase in \(k_{\text{obs}}\) for R-QE and a slight decrease for S-QE compared with corresponding values in the native soil. Therefore, the change of EF was smaller in soil/ZnO than in native soil (Fig. 2b). ZnO is a well-known photocatalyst, and both chemical and biological transformation contributed to the overall transformation of rac-QE in soil/ZnO. \(k_{\text{obs}}\) was both 0.29 day\(^{-1}\) for R-QE and S-QE in autoclaved soil with ZnO (Fig. 3), indicating chemical transformation made a minimal contribution in the transformation of rac-QE in native soil/ZnO. Conversely, TiO\(_2\) induced chemical transformation contributed predominantly to the overall transformation of rac-metalaxyl (Huang et al., 2018; Liang et al., 2016), which is because biotransformation of rac-QE was much faster than that rac-metalaxyl; half-life \((t_{1/2}, \text{day})\) for R-QE and R-metalaxyl was 0.07 and 5.92 days in native soil, respectively.

Moreover, as shown in Table 1 and Fig. 1d, microbes abundance decreased and bacterial structure changed in the presence of ZnO, which could lead to a decrease in biotransformation’s contribution in the overall transformation of rac-QE. Therefore, compared to the transformation of rac-QE in native soil, overall \(k_{\text{obs}}\) enantioselectivity for the transformation of rac-QE slightly decreased in soil/ZnO. Different from the evident effect of ZnO on EF for rac-QE, TiO\(_2\) showed no marked effect on EF for rac-metalaxyl, although TiO\(_2\) promoted the overall transformation of rac-metalaxyl (Huang et al., 2018; Liang et al., 2016). The main reason is TiO\(_2\) showed no significant influence on soil bacterial diversity (Huang et al., 2018). The result was in line with the reference report that ZnO exhibited a more significant impact on soil microorganisms and enzyme activities than TiO\(_2\) or the other metal oxide nanoparticles (Rajput et al., 2018).

The addition of 5% biosolids promoted the overall transformation of rac-QE, but the influence of biosolid on two enantiomers was different. \(k_{\text{obs}}\) was 7.30 and 12.31 day\(^{-1}\) for R-QE and S-QE, respectively, demonstrating an increase of \(k_{\text{obs}}\) for R-QE and a decrease for S-QE yielded a more significant EF in soil/biosolids (Fig. 2b). The change in enantioselectivity

![Fig. 2](image-url) - (a) Transformation of R-QE and S-QE in native soil, soil/ZnO, soil/biosolids, soil/biosolids/ZnO, solid and dashed lines were the fit of experimental data with pseudo-first-order kinetic equation, (b) change of EF with incubation time.
mainly resulted from the additional transformation induced by biosolids, which was proved by the transformation of rac-QE in autoclaved soil with biosolids, where $k_{obs}$ was 1.06 and 1.92 day$^{-1}$ for R-QE and S-QE, respectively (Fig. 3a). Assuming the biotransformation of rac-QE in soil/biosolids was a simple sum of the individual contribution from native soil and biosolids, the theoretical $k_{obs}$ was 9.08 and 11.28 day$^{-1}$ for R-QE and S-QE, which was 24% larger and 8% smaller than the observed values, suggesting the existence of a mutual influence between soil and biosolids in biotransformation of rac-QE in soil/biosolids. This conclusion was supported by the transformation of rac-QE in artificial soil with the addition of equal biosolids as that in native soil (Fig. 3b). $k_{obs}$ for R-QE was comparable, but $k_{obs}$ for S-QE in biosolids spiked artificial soil was 3.2 times higher than that in native soil. Therefore, the transformation of S-QE was more favorable in soil/biosolids relative to R-QE because of the added biosolid, and enantioselectivity for the transformation of rac-QE was between native soil and biosolids. With the addition of biosolids to the native soil, organic carbon content and carbon bioavailability, as well as other nutrients in the mixed phase, could differ significantly from each individual phase that subsequently influenced the metabolism of rac-QE. On the other side, the change of enantioselectivity may also result from the change of soil pH that played a critical role in the enantioselective transformation of chiral pesticide in soil (Buerge et al., 2006).

Compared with the single addition of either ZnO or biosolids, the transformation of rac-QE was the slowest in soil with the co-addition of ZnO and biosolids. $k_{obs}$ for the transformation of R-QE and S-QE in soil/biosolids/ZnO decreased to 4.94 and 5.02 day$^{-1}$, respectively, which showed that two enantiomers had a comparable transformation rate yielded a bare change of EF with incubation time (Fig. 2b). As shown in Fig. 3a, the transformation of rac-QE was completely inhibited in autoclaved “soil/biosolids”/ZnO. The results showed that autoclaved biosolids inhibited the chemical transformation of rac-QE induced by ZnO, which was because of the competition for reactive species between organic matters in biosolids and rac-QE. It has been reported that biosolids embedded with ENPs could shift soil microbial community composition, as well as pristine ENPs, but the influence amplitude was different between ENPs alone and ENPs with biosolids (Colman et al., 2013; Judy et al., 2015). A similar result was also found in this study that was demonstrated in Fig. 1 and Table 1. Therefore, both chemical transformation of rac-QE induced by ZnO and biotransformation caused by biosolids and native soil were suppressed, which led to a slow and nonenantioselective transformation of rac-QE in soil/biosolids/ZnO.

2.3. Effect of irradiation on enantioselective transformation of rac-QE

Effect of irradiation on the enantioselective transformation of rac-QE with an initial concentration of approximately 20 mg/kg was demonstrated in Fig. 4. Because of the small contribution of chemical transformation and the limit transmission of light in soil media, the transformation of rac-QE was not remarkably influenced by a 3-hr simulated solar irradiation. The deviation of $k_{obs}$ between dark incubation and light irradiation was smaller than 10%, except for the transformation of R-QE in soil/biosolids (17%) and soil/biosolids/ZnO (29%), the results were in line with the small change of MBC with light irradiation (Table 1).

Compared to the value of EF obtained under dark incubation (Fig. 4b), the value of EF that obtained under light irradiation was relatively smaller because of a more noticeable increase of $k_{obs}$ for R-QE relative to S-QE (Fig. 4d). The presence of ZnO decreased the EF value in native soil and soil/biosolids regardless of light irradiation. It has been reported that soil microorganism showed a speciation-dependent response to external disturbance, and a higher functional redundancy might act as a buffer against the change of biodiversity (Griffiths and Laurent, 2013). Because of the faster transformation of S-QE relative to R-QE in particular in the presence of biosolids, change of S-QE-degrading and R-QE-degrading
microorganism might to different extents followed by the shift of EF.

2.4. Kinetics for enantioselective transformation of rac-QE

Transformation of rac-QE was conducted from 5 to 40 mg/kg (2.5–20 mg/kg for each enantiomer) to reveal the impact of the additive on the enantioselective transformation kinetics. As shown in Fig. 5a, the initial transformation rate of R-QE (rR-QE) gradually increased and reached a plateau with increase its initial concentration. rR-QE value was the smallest in soil/biosolids/ZnO. rR-QE value in soil/biosolids was comparable with that in native soil. rR-QE value in soil/ZnO was the highest when R-QE initial dosage was lower than 5 mg/kg, after which the highest value of rR-QE was found in soil/biosolids. The slowest transformation of S-QE was also found in soil/biosolids/ZnO (Fig. 5b), while rS-QE value was the largest in soil/biosolids. rS-QE value in soil/ZnO was 10%–25% smaller than that in native soil for S-QE’s initial concentration ranged from 10 to 18 mg/kg.

Because of the small portion of chemical transformation, the transformation of two enantiomers could be well fitted by the Michaelis–Menten kinetic model. The maximum transformation rate (rmax) for R-QE obtained in soil/ZnO was 56% of that obtained in native soil, and rmax for R-QE obtained in soil/biosolids/ZnO was 53% of that obtained in native soil/biosolids. In the case of rmax for S-QE, the value obtained in soil/ZnO and soil/biosolids/ZnO accounted for 67% and 79% of the corresponding values in native soil and soil/biosolids. The different influence extents of ZnO on rR-QE and rS-QE suggested that its effects mainly resulted from a biotransformation process. Otherwise, the change of rR-QE and rS-QE were supposed to be the same. The influence of ZnO on rR-QE was smaller than that on rS-QE, which was mainly because the abundance for S-QE degrading microorganism was much larger than that of R-QE, particularly in the presence of biosolids, and therefore S-QE degrading microorganism would possess a higher resistance to ZnO.

2.5. Relation of rac-QE transformation with bacterial genera and soil properties

rR-QE and rS-QE has a negative correlation with soil organic carbon and nitrogen, but not to a significant extent (data not shown). Conversely, rR-QE and rS-QE has a significant positive relationship (p < 0.05) with two bacterial genera Bacillus and Fictibacillus (Table 2). Although rR-QE and rS-QE were also positively correlated with the other eight bacterial genera, the correlation was not significant. Enantioselectivity indexed by
the ratio of $r_{S-QE}$ to $r_{R-QE}$ was not found to be significantly correlated with these top ten bacterial genera. Only the relative proportion for Pontibacter genus was highly correlated with soil pH (negative), OC and TN (positive). Several QE-degrading bacterial strains including Acinetobacter sp. strain DL-2 (Dong et al., 2015), Bacillus subtilis H (Hou et al., 2018), Pseudomonas sp. J-2 (Zhang et al., 2017b), Rhodococcus ruber JPL-2 (Liu et al., 2015), Rhodococcus sp. JT-3 and Breundimonas sp. JT-9 have been isolated (Zhang et al., 2017a), but none of them was classified as Fictibacillus. These bacterial genera were found in the conducted soils (Appendix A Fig. S3), and their relative proportions ranged from 0.01% to 0.07% that was much smaller than the relative proportion of Bacillus (6.59%–11.47%). Notably, Acinetobacter was only been found in soil/ZnO and soil/biosolids/ZnO with a relative proportion of 0.017%. Therefore, Bacillus might be the major bacterial genus that corresponded for QE transformation in soils conducted in this study.

3. Conclusions

This study investigated the effect of ZnO on soil bacterial community composition and enantioselective transformation of co-existed rac-QE. It was found that ZnO exhibited an adverse effect on soil microorganism abundance and caused bacterial community composition shift, regardless of ZnO directly entered the soil or with biosolids. Although the sole addition of biosolids promoted the transformation of rac-QE and broadened the enantioselectivity, an inverse effect was observed when biosolids collaborated with ZnO. ZnO could enhance the non-enantioselective chemical transformation of rac-QE in native soil. However, chemical transformation contributed little to the overall transformation because of the short half-life of rac-QE in native soil. ZnO induced chemical transformation was suppressed when it was introduced with biosolids together. The overall finding indicated that the pathway through which ENPs enter soil could cause different influences on soil microorganism and the environmental fate of coexisting pollutants. It was also noticed that biosolids/ZnO used in this study was prepared by directly physical mixing, the influence of biosolids containing ENPs produced from waste-water treatment plant needs further investigation because of the possible change of speciation of ENPs.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jes.2019.06.012.

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<th>Table 2 – Correlation between the top 10 bacterial genera and transformation of rac-QE, and soil properties.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bact1a</td>
</tr>
<tr>
<td>$r_{R-QE}$</td>
</tr>
<tr>
<td>$r_{S-QE}$</td>
</tr>
<tr>
<td>pH</td>
</tr>
<tr>
<td>MBC</td>
</tr>
<tr>
<td>OC</td>
</tr>
<tr>
<td>N</td>
</tr>
</tbody>
</table>

Bact1a to Bact10a stands for bacterial genera listed in Fig. 1d.

* denotes a significant difference at 0.01 < $p$ < 0.05.
REFERENCES


Zhang, H., Li, M., Li, J., Wang, G., Li, F., Xu, D., et al., 2017a. A key esterase required for the mineralization of quinalofop-p-ethyl...