

Effects of cucumber root exudates components on *Fusarium* spp. community structure and abundance

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(Received in revised form: February 18, 2019)

ABSTRACT

In a microcosm experiment, soils were treated with selected cucumber root exudates components and the soil *Fusarium* spp. community structure and abundance were estimated by PCR-denaturing gradient gel electrophoresis and quantitative PCR, respectively. Results showed that all added organic compounds (Glucose, succinic acid, *p*-hydroxybenzoic acid, *p*-coumaric acid and glutamic acid) changed the soil *Fusarium* spp. community structure. The *p*-Coumaric acid, glucose, succinic acid and glutamic acid decreased the Shannon-Wiener and Evenness indices. The *p*-Hydroxybenzoic acid, *p*-coumaric acid, succinic acid and glutamic acid also decreased the soil *Fusarium* spp. community abundance. Treatment with *p*-hydroxybenzoic acid and *p*-coumaric acid had higher diversity indices but lower abundance of *Fusarium* spp. community than treatment with succinic acid and glutamic acid.

Key words: Amino acid, community structure, cucumber, *Fusarium* spp., organic acid, PCR, *p*-coumaric acid, *p*-hydroxybenzoic, root exudates, sugar.

INTRODUCTION

Plant roots through root exudates release variety of low molecular weight organic compounds (organic acids, amino acids and sugars) into the rhizosphere (2). These root exudates play important roles in plant-plant and plant-microbe interactions (2,19-21,28). For example, one plant species can inhibit the growth of another species through its root exudates, a phenomenon known as allelopathy (11). There are numerous microorganisms in soil and plant-microbial interactions can have strong influences on plant performance (10,17,33,47,55). Root exudates can also act as specific substrates or signaling molecules for a large group of microbial species in soil (1). Moreover through root exudation, plants can select microorganisms in the rhizosphere to their own benefit, such as deterring the pathogenic microorganisms, while attracting the mutualistic ones (8).

Fungi show the greatest eukaryotic diversity on the planet (3,25) and behave as mutualists, pathogens of plants and have profound influences on the growth, nutrition and health of plants in agroecosystems (4,7,25,32,44). *Fusarium* (Sordariomycetes: Hypocreales: Nectriaceae) genus of filamentous ascomycete fungus contains many species of agricultural importance (16,22-24,35). Besides playing important roles in organic matter decomposition, many *Fusarium* spp. are phytopathogenic and cause wilt disease in many

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crop plants (36,39). But, how plant root exudate components affect the *Fusarium* spp. in soil is still not known.

Cucumber vegetable is commonly continuously cropped in the greenhouse (14,30,52). Previous studies showed that root exudates from cucumber (*Cucumis sativus* L.) contain sugars (glucose and fructose), organic acids (citric, malic and succinic acids and phenolic acids) and amino acids (serine, glutamic acid aspartic acid) (15,27,34). Phenolic acids, such as *p*-hydroxybenzoic and *p*-coumaric acids are phytotoxic to cucumber growth (27,41). Previously, we reported that phenolic acids were able to change the cucumber rhizosphere soil bacterial, fungal and *Fusarium* spp. communities (12,37,42,50,51,55). In this study, we have further evaluated the effects of selected main components of cucumber root exudates on soil *Fusarium* spp. communities.

MATERIALS AND METHODS

Microcosm experiment

The soil used was collected from the undisturbed upper soil layer (0-15 cm) of an open field in our Experimental Station, Northeast Agricultural University, Harbin, China (45°41'N, 126°37'E, mean height above sea level: 127.95 m, annual precipitation: 524.5 mm, maximum and minimum temperature: 36.7°C, -37.7°C). The soil was sandy loam, contained organic matter: 3.67%, available N: 89.02 mg/kg, available P: 63.36 mg/kg, available K: 119.15 mg/kg, EC (1:2.5, w/v): 0.33 mS/cm and pH (1:2.5, w/v): 7.78 (43).

Microcosm experiment was done in flasks containing 60 g fresh soils. To stabilize the soil microbial communities, these soils were pre-incubated at 20°C in dark for 5-days with soil water content maintained at about 50% of its water holding capacity. Then NH₄NO₃ solution was added at 450 µg N/g soil to avoid potential microbial growth limitation by nitrogen (29). Organic compounds were added into the soil periodically as described before (29). Glucose (G), succinic acid (SA), *p*-hydroxybenzoic acid (pHA) *p*-coumaric acid (pCA) and glutamic acid (GA), were purchased from Solarbio Life Science Company, Beijing, China. These were added 5-times into soil at 20 µg C/g soil every two days. Soils treated with distilled water (W) served as control. Added solutions were uniformly mixed with the soil after each addition to avoid concentration gradients. Each treatment had five flasks and replicated thrice. The solution pH was adjusted to 7.0 with 0.1 M NaOH because the soil pH is dominant factor that regulates soil microbial communities (9). Flasks containing these treated soils were sealed with Parafilm (Bemis Company, Inc., Wisconsin, USA) and incubated at 20°C in dark. Soil water content was maintained at about 50% of its water holding capacity.

Soil sampling and DNA extraction

One day after the fifth application of organic compounds, 10 g fresh soils were sampled from each flask with a sterilized lab spoon and soils from five flasks in each replicate were mixed to make a composite sample. Total soil DNA was extracted with the PowerSoil DNA Isolation Kit (MO BIO Laboratories, Carlsbad, USA) as per the manufacturer's instructions.

PCR-DGGE analysis

Fusarium spp. community structure was analyzed by PCR-DGGE method. Nested PCR was used to amplify the *Fusarium* spp. *Eflα* genes with primer sets of EF-1/EF-2 (26) and Alfie1-GC/Alfie2 (38) in the first and second round of PCR amplifications, respectively, as described before (46,54). DGGE analysis was performed on a 6% (w/v) acrylamide gel with 40-60% denaturant gradient. The gel was run, stained and photographed as described before (46,54).

Quantitative PCR assay

Fusarium spp. community abundance was estimated by SYBR Green qPCR assays with an IQ5 real-time PCR system (Bio-Rad Lab, LA, USA) as described before (46,54). The *Eflα* gene was nested and amplified with EF-1/EF-2 (26) and Alfie1/Alfie2 (38) in the first and second round of PCR amplifications, respectively. Care was taken to ensure that first-round PCR products were all in exponential amplification phase of the PCR (36). The relative *Fusarium* spp. community abundance was calculated as describes by Wakelin *et al.* (36) and then, all treatments were compared with the control soil and expressed as the percentage of the abundance in the control soil.

Statistical analysis

Banding patterns of the DGGE profiles were analyzed using Quantity One V4.5 as described before (52). Principal component analysis (PCA) was used to compare the banding patterns between samples with normalized data using Canoco for Windows 4.5 software (56). Analysis of similarities (ANOSIM) was used to test for the overall effect of treatment on microbial community structures using the Vegan package in 'R' (Version 3.3.1). The microbial community diversity indices, including number of bands, Shannon-Wiener index and evenness index, were calculated as described before (56). Data were analyzed following analysis of variance (ANOVA) and mean comparison between treatments was performed based on the Tukey's honestly significant difference (HSD) test at 0.05 probability level.

RESULTS AND DISCUSSION

Fusarium spp. community structure

Visual inspection of *Fusarium* spp. community DGGE profiles showed that banding patterns of triplicate samples per treatment were similar (Fig 1a). However, there were differences among the treatments, both in terms of presence/absence of individual DGGE bands and the intensity of co-migrating DGGE bands.

PCA analysis of *Fusarium* spp. community DGGE profiles clearly separated all treatments from each other (Fig 1b). ANOSIM analysis also confirmed that *Fusarium* spp. community structure significantly differed among treatments ($R=0.999$, $P=0.001$). The PC1 and PC2 components together accounted for 56.4% of the variation. On the PCA plot, *p*-hydroxybenzoic acid, *p*-coumaric acid and succinic acid treatments were close to each other, indicating that compared with glucose and glutamic acid, these three organic acids had similar effects on *Fusarium* spp. community structure. Compared with organic acids

and glucose, the glutamic acid was more close to control, indicating that glutamic acid had weaker influence on *Fusarium* spp. community structure.

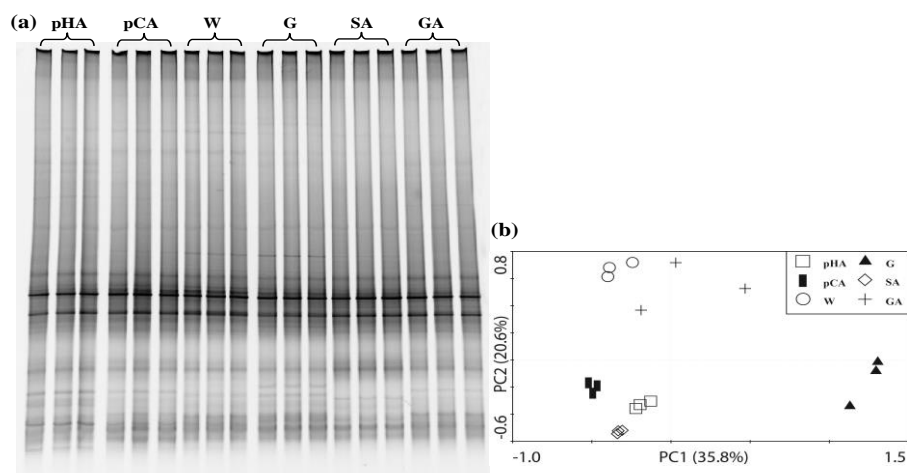


Figure 1. PCR-DGGE profile (a) and PCA analysis (b) of soil *Fusarium* spp. community. pHA: *p*-hydroxybenzoic acid, CA: *p*-coumaric acid, G: glucose, SA: succinic acid, GA: glutamic acid, W: Water

Compared with control, treatments with *p*-coumaric acid, glucose, succinic acid and glutamic acid had lower number of bands and treatments with *p*-coumaric acid, succinic acid and glutamic acid had lower Shannon-Wiener and Evenness indices ($P < 0.05$) (Table 1). Treatment with *p*-hydroxybenzoic acid had higher diversity indices than treatments with succinic acid and *p*-coumaric acid ($P < 0.05$). Treatments with glucose, *p*-hydroxybenzoic acid and *p*-coumaric acid had higher diversity indices than treatments with succinic acid and glutamic acid ($P < 0.05$).

Table 1. Effects of root exudates components on *Fusarium* spp. community diversity

Treatment	Number of bands	Shannon-Wiener	Evenness
Water	2.95±0.05 a	21.33±1.15 ab	0.88±0.01 b
<i>p</i> -Hydroxybenzoic acid	3.00±0.01 a	22.33±0.58 a	0.89±0.00 a
<i>p</i> -Coumaric acid	2.83±0.01 c	19.00±0.00 c	0.84±0.00 c
Glucose	2.89±0.01 b	20.00±0.00 bc	0.86±0.00 b
Succinic acid	2.68±0.01 d	16.00±0.00 d	0.80±0.00 d
Glutamic acid	2.59±0.00 e	15.00±0.00 d	0.77±0.00 e

Fusarium spp. community abundance

Quantitative PCR showed that *p*-hydroxybenzoic acid, *p*-coumaric acid, succinic acid and glutamic acid significantly decreased soil *Fusarium* spp. community abundance (Fig 2). Moreover, treatment with *p*-hydroxybenzoic acid and *p*-coumaric acid, had lower *Fusarium* spp. community abundance than treatments with succinic acid or glutamic acid ($P < 0.05$).

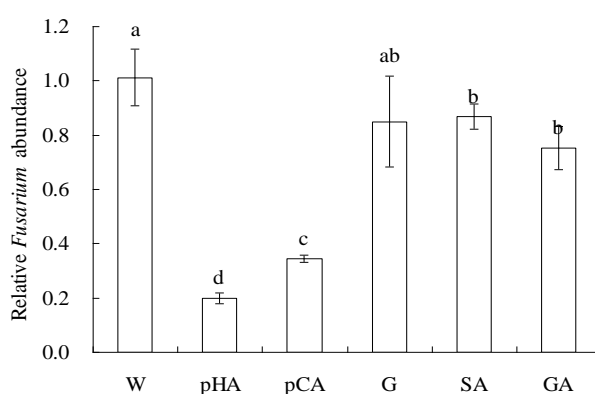


Figure 2. Effects of selected root exudates components on *Fusarium* spp. community abundance. pHA : *p*-hydroxybenzoic acid ,CA : *p*-coumaric acid, G : glucose, SA : succinic acid, GA : glutamic acid, W : Water

In this study, we found that different components of root exudates had variable influences on the *Fusarium* spp. community structure and abundance, suggesting that different components of root exudates play different roles in plant-microbial interactions. The structure and function of a soil microbial community are tightly linked and changes in microbial community's structure can alter its function (6,18,48). For example, *p*-coumaric acid could change the cucumber rhizosphere microbial community, which in turn exerted a negative effects on the cucumber growth (55). Further studies are needed to illustrate the functional differences of changes in soil microbial communities induced by different components of root exudates.

Our observation that *p*-coumaric acid, succinic acid and glutamic acid decreased the *Fusarium* spp. community diversity and abundance, indicates that these compounds inhibited some taxa of *Fusarium* spp. However, the *Fusarium* spp. community structures differed among the treatments. Therefore, these three chemicals may target different taxa of *Fusarium* spp. In agricultural ecosystems, continuous monocropping of the same crop in the same land, usually negatively affects the crop growth, a phenomenon called 'soil sickness' (6,45,47,49). Accumulation of allelochemicals and build-up of soil-borne pathogens are among the principle possible factors contributing to soil sickness (12,13,31,53). Phenolic acids are one of important autotoxins of cucumber (45,50). *Fusarium* spp. contain species beneficial to plants (those involved in organic matter decomposition) and also phytopathogenic species (36,39). *In vitro* studies to evaluate the effects of different components of root exudates on various *Fusarium* spp. species should be performed and to understand the relationships among the root exudates, soil pathogens and soil sickness.

The observation that *p*-hydroxybenzoic and *p*-coumaric acids changed the soil *Fusarium* spp. community structure is consistent with our previous studies (42,46)

showing that phenolic compounds affects the cucumber rhizosphere *Fusarium* spp. community structure. However, contrasting results about *Fusarium* spp. community abundance were observed. In this study, *p*-hydroxybenzoic and *p*-coumaric acids decreased the *Fusarium* spp. community abundance. However, vanillin and syringic acid increased in the cucumber rhizosphere *Fusarium* spp. community abundance (42,46). One possible explanation for this inconsistency is that different phenolic compounds differ in their influences on *Fusarium* spp. community abundance. This inconsistency may also be due to different kinds of soil samples that were analysed: bare soils without plants in this study and cucumber rhizosphere soils in our previous studies (42,46). Phenolic compounds disrupts the plant root cell membrane integrity and increase the ion leakage (5,40) and these changes could also affect the rhizosphere microbial communities. Thus, the effects of phenolic compounds on the rhizosphere communities were likely to be a combination of phenolic compounds' including both direct effects and indirect effects through regulating plant's physiological status.

CONCLUSIONS

The exogenously added glucose, succinic acid, *p*-hydroxybenzoic acid, *p*-coumaric acid and glutamic acid changed the soil *Fusarium* spp. community structure. *p*-Coumaric acid, glucose, succinic acid and glutamic acid decreased the number of bands, while the *p*-coumaric acid, succinic acid and glutamic acid decreased the Shannon-Wiener and Evenness indices. Besides, the *p*-Hydroxybenzoic acid, *p*-coumaric acid, succinic acid and glutamic acid decreased the soil *Fusarium* spp. community abundance.

ACKNOWLEDGMENTS

This work was supported by the National Natural Science Foundation of China (31772361), 'Academic Backbone' Project of Northeast Agricultural University (17XG05) and China Agricultural Research System (CARS-23-B-10).

REFERENCES

1. Badri, D.V., Chaparro, J.M., Zhang, R., Shen, Q. and Vivanco, J.M. (2013). Application of natural blends of phytochemicals derived from the root exudates of *Arabidopsis* to the soil reveal that phenolic-related compounds predominantly modulate the soil microbiome. *Journal of Biological Chemistry* **288**: 4502-4512.
2. Bais, H.P., Weir, T.L., Perry, L.G., Gilroy, S. and Vivanco, J.M. (2006). The role of root exudates in rhizosphere interactions with plants and other organisms. *Annual Review of Plant Biology* **57**: 233-266.
3. Bamisile, B.S., Dash, C.K., Akutse, K.S., Ravindran, K. and Liande, W. (2018). Fungal endophytes: beyond herbivore management. *Frontiers in Microbiology* **9**: 544.
4. Bardgett, R.D. and van der Putten, W.H. (2014). Belowground biodiversity and ecosystem functioning. *Nature* **515**: 505-511.
5. Baziramakenga, R., Leroux, G. and Simard, R. (1995). Effects of benzoic and cinnamic acids on membrane permeability of soybean roots. *Journal of Chemical Ecology* **21**: 1271-1285.
6. Bever, J.D., Platt, T.G. and Morton, E.R. (2012). Microbial population and community dynamics on plant roots and their feedbacks on plant communities. *Annual Review of Microbiology* **66**: 265-283.

7. Bini, D., dos Santos, C.A., da Silva, M.C.P., Bonfim, J.A. and Cardoso, E.J.B.N. (2018). Intercropping *Acacia mangium* stimulates AMF colonization and soil phosphatase activity in *Eucalyptus grandis*. *Scientia Agricola* **75**: 102-110.
8. Chaparro, J.M., Badri, D.V. and Vivanco, J.M. (2014). Rhizosphere microbiome assemblage is affected by plant development. *The ISME Journal* **8**: 790-803.
9. Fierer, N. and Jackson, R.B. (2006). The diversity and biogeography of soil bacterial communities. *Proceedings of the National Academy of Sciences of the United States of America* **103**: 626-631.
10. Hazard, C., Kruitbos, L., Davidson, H., Mbow, F.T., Taylor, A.F.S. and Johnson, D. (2017). Strain identity of the ectomycorrhizal fungus *Laccaria bicolor* is more important than richness in regulating plant and fungal performance under nutrient rich conditions. *Frontiers in Microbiology* **8**: 1874.
11. Inderjit, Wardle, D.A., Karban, R. and Callaway, R.M. (2011). The ecosystem and evolutionary contexts of allelopathy. *Trends in Ecology & Evolution* **26**: 655-662.
12. Jia, H.T., Chen, S.C., Yang, S.Y., Shen, Y.H., Qiao, P.L., Wu, F.Z. and Zhou, X.G. (2018). Effects of vanillin on cucumber rhizosphere bacterial community. *Allelopathy Journal* **44**: 191-200.
13. Jia, X.L., Wang, H.B., Ye, J.H., Wang, F.Q., Lu, L., Hu, Y.L., Zheng, M.Z., Zhang, Q. and Wu, C.Z. (2018). Identification of allelochemicals responsible for soil degradation in continuously cropped Tea plantations. *Allelopathy Journal* **45**: 1-12.
14. Jin, X., Wang, J., Li, D., Wu, F. and Zhou, X. (2019). Rotations with Indian mustard and wild rocket suppressed cucumber *Fusarium* wilt disease and changed rhizosphere bacterial communities. *Microorganisms* **7**: 57.
15. Kamilova, F., Kravchenko, L.V., Shaposhnikov, A.I., Azarova, T., Makarova, N. and Lugtenberg, B. (2006). Organic acids, sugars and L-tryptophane in exudates of vegetables growing on stonewool and their effects on activities of rhizosphere bacteria. *Molecular Plant-Microbe Interactions* **19**: 250-256.
16. Khan, N., Maymon, M. and Hirsch, A.M. (2017). Combating *Fusarium* infection using *Bacillus*-based antimicrobials. *Microorganisms* **5**: 75.
17. Kirker, G.T., Bishell, A.B., Jusino, M.A., Palmer, J.M., Hickey, W.J. and Lindner, D.L. (2017). Amplicon-based sequencing of soil fungi from wood preservative test sites. *Frontiers in Microbiology* **8**: 1997.
18. Kulmatiski, A., Beard, K.H., Stevens, J.R. and Cobbold, S.M. (2008). Plant-soil feedbacks: a meta-analytical review. *Ecology Letters* **11**: 980-992.
19. Li, H.Y., Zhou, X.G. and Wu, F.Z. (2018). Effects of root exudates from potato onion on *Verticillium dahliae*. *Allelopathy Journal* **43**: 217-222.
20. Li, S., Xu, C., Wang, J., Guo, B., Yang, L., Chen, J. and Ding, W. (2017). Cinnamic, myristic and fumaric acids in tobacco root exudates induce the infection of plants *Ralstonia solanacearum*. *Plant and Soil* **412**: 381-395.
21. Liang, Z.Y., Li, X.Y., Zhang, H., Li, J., Bian, X.J. and Xu, J.C. (2018). Allelopathic effects of Bermuda grass (*Cynodon dactylon* L.) root exudates on seed germination and seedling growth of Tall fescue (*Festuca arundinacea* Schreb). *Allelopathy Journal* **44**: 25-34.
22. Ma, L.J., Geiser, D.M., Proctor, R.H., Rooney, A.P., O'Donnell, K., Trail, F., Gardiner, D.M., Manners, J.M. and Kazan, K. (2013). *Fusarium* pathogenomics. *Annual Review of Microbiology* **67**: 399-416.
23. Manici, L. M., Caputo, F. and Sacca M.L. (2017). Secondary metabolites released into the rhizosphere by *Fusarium oxysporum* and *Fusarium* spp. as underestimated component of nonspecific replant disease. *Plant and Soil* **415**: 85-98.
24. Morales-Corts, M.R., Perez-Sanchez, R. and Gomez-Sanchez, M.A. (2018). Efficiency of garden waste compost teas on tomato growth and its suppressiveness against soilborne pathogens. *Scientia Agricola* **75**: 400-409.
25. Nagati, M., Roy, M., Manzi, S., Richard, F., Desrochers, A., Gardes, M. and Bergeron, Y. (2018). Impact of local forest composition on soil fungal communities in a mixed boreal forest. *Plant and Soil* **432**: 345-357.
26. O'Donnell, K., Kistler, H.C., Cigelnik, E. and Ploetz, R.C. (1998). Multiple evolutionary origins of the fungus causing Panama-disease of banana: concordant evidence from nuclear and mitochondrial gene genealogies. *Proceedings of the National Academy of Sciences of the United States of America* **95**: 2044-2049.

27. Pramanik, M.H.R., Nagai, M., Asao, T. and Matsui, Y. (2000). Effects of temperature and photoperiod on phytotoxic root exudates of cucumber (*Cucumis sativus*) in hydroponic culture. *Journal of Chemical Ecology* **26**: 1953-1967.
28. Premathilake, U., Wathugala, D.L. and Dharmadasa, R.M. (2018). Phytotoxic potential of lemongrass (*Cymbopogon citratus* (DC.) Stapf). *Allelopathy Journal* **44**: 261-268.
29. Shi, S., Richardson, A.E., O'Callaghan, M., Deangelis, K.M., Jones, E.E., Stewart, A., Firestone, M.K. and Condron, L.M. (2011). Effects of selected root exudate components on soil bacterial communities. *FEMS Microbiology Ecology* **77**: 600-610.
30. Shi, Y.J., Wang, J., Jin, X., Wang, Z.L., Pan, D.D., Zhuang, Y., Wu, F.Z. and Zhou, X.G. (2019). Effects of intercropping of wheat on cucumber seedling rhizosphere fungal community composition. *Allelopathy Journal* **46**: 241-250.
31. Singh, H.P., Batish, D.R. and Kohli, R.K. (1999). Autotoxicity: Concept, organisms and ecological significance. *Critical Reviews in Plant Sciences* **18**: 757-772.
32. Smith, M.E., Facelli, J.M. and Cavagnaro, T.R. (2018). Interactions between soil properties, soil microbes and plants in remnant-grassland and old-field areas: a reciprocal transplant approach. *Plant and Soil* **433**: 127-145.
33. van der Heijden, M.G., Bardgett, R.D. and Van Straalen, N.M. (2008). The unseen majority: soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems. *Ecology Letters* **11**: 296-310.
34. Vančura, V. and Hovadik, A. (1965). Root exudates of plants: II. Composition of root exudates of some vegetables. *Plant and Soil* **22**: 21-32.
35. Verma, S.K., Kingsley, K.L., Bergen, M.S., Kowalski, K.P. and White, J.F. (2018). Fungal disease prevention in seedlings of rice (*Oryza sativa*) and other grasses by growth-promoting seed-associated endophytic bacteria from invasive *Phragmites australis*. *Microorganisms* **6**: 21.
36. Wakelin, S.A., Warren, R.A., Kong, L. and Harvey, P.R. (2008). Management factors affecting size and structure of soil *Fusarium* communities under irrigated maize in Australia. *Applied Soil Ecology* **39**: 201-209.
37. Wang, Z., Zhang, J., Wu, F. and Zhou, X. (2018). Changes in rhizosphere microbial communities in potted cucumber seedlings treated with syringic acid. *PLoS One* **13**: e0200007.
38. Yergeau, E., Filion, M., Vujanovic, V. and St-Arnaud, M. (2005). A PCRdenaturing gradient gel electrophoresis approach to assess *Fusarium* diversity in asparagus. *Journal of Microbiological Methods* **60**: 143-154.
39. Yergeau, E., Labour, K., Hamel, C., Vujanovic, V., Nakano-Hylander, A., Jeannotte, R. and St-Arnaud, M. (2010). Patterns of *Fusarium* community structure and abundance in relation to spatial, abiotic and biotic factors in soil. *FEMS Microbiology Ecology* **71**: 34-42.
40. Yu, J.Q. and Matsui, Y. (1997). Effects of root exudates of cucumber (*Cucumis sativus*) and allelochemicals on ion uptake by cucumber seedlings. *Journal of Chemical Ecology* **23**: 817-827.
41. Yu, J.Q. and Matsui, Y. (1994). Phytotoxic substances in the root exudates of *Cucumis sativus* L. *Journal of Chemical Ecology* **20**: 21-31.
42. Zhang, J.H., Pan, D.D., Ge, X., Shen, Y.H., Qiao, P.L., Yang, S.Y., Wu, F.Z. and Zhou, X.G. (2018). Effects of syringic acid on *Fusarium* and *Trichoderma* communities in cucumber (*Cucumis sativus* L.) seedling rhizosphere. *Allelopathy Journal* **44**: 181-190.
43. Zhang, J.H., Yu, H.J., Ge, X., Pan, D.D., Shen, Y.H., Qiao, P.L., Wu, F.Z. and Zhou, X.G. (2018). Effects of vanillin on cucumber (*Cucumis sativus* L.) seedling rhizosphere fungal community composition. *Allelopathy Journal* **44**: 169-180.
44. Zhang, K.R., Cheng, X.L., Shu, X., Liu, Y. and Zhang, Q.F. (2018). Linking soil bacterial and fungal communities to vegetation succession following agricultural abandonment. *Plant and Soil* **431**: 19-36.
45. Zhou, X.G., Gao, D., Zhao, M., Zhang, J., Li, L. and Wu, F. (2016). Dynamics of soil bacterial communities in Jerusalem artichoke monocropping system. *Allelopathy Journal* **39**: 167-178.
46. Zhou, X.G., Jia, H., Ge, X. and Wu, F. (2017). Effects of vanillin on the community structures and abundances of *Fusarium* and *Trichoderma* spp. in cucumber seedling rhizosphere. *Journal of Plant Interactions* **13**: 45-50.
47. Zhou, X.G., Liu, J. and Wu, F. (2017). Soil microbial communities in cucumber monoculture and rotation systems and their feedback effects on cucumber seedling growth. *Plant and Soil* **415**: 507-520.

48. Zhou, X.G., Shen, Y., Fu, X. and Wu, F. (2018). Application of sodium silicate enhances cucumber resistance to *Fusarium* wilt and alters soil microbial communities. *Frontiers in Plant Science* **9**: 624.
49. Zhou, X.G., Wang, Z., Jia, H., Li, L. and Wu, F. (2018). Continuously monocropped Jerusalem artichoke changed soil bacterial community composition and ammonia-oxidizing and denitrifying bacteria abundances. *Frontiers in Microbiology* **9**: 705.
50. Zhou, X.G. and Wu, F. (2012). *p*-Coumaric acid influenced cucumber rhizosphere soil microbial communities and the growth of *Fusarium oxysporum* f. sp. *cucumerinum* Owen. *PLoS One* **7**: e48288.
51. Zhou, X.G. and Wu, F. (2018). Vanillic acid changed cucumber (*Cucumis sativus* L.) seedling rhizosphere total bacterial, *Pseudomonas* and *Bacillus* spp. communities. *Scientific Reports* **8**: 4929.
52. Zhou, X.G., Yu, G. and Wu, F. (2011). Effects of intercropping cucumber with onion or garlic on soil enzyme activities, microbial communities and cucumber yield. *European Journal of Soil Biology* **47**: 279-287.
53. Zhou, X.G., Yu, G. and Wu, F. (2012). Soil phenolics in a continuously mono-cropped cucumber (*Cucumis sativus* L.) system and their effects on cucumber seedling growth and soil microbial communities. *European Journal of Soil Science* **63**: 332-340.
54. Zhou, X.G., Zhang, J., Gao, D., Gao, H., Guo, M., Li, L., Zhao, M. and Wu, F. (2017). Conversion from long-term cultivated wheat field to Jerusalem artichoke plantation changed soil fungal communities. *Scientific Reports* **7**: 41502.
55. Zhou, X.G., Zhang, J., Pan, D., Ge, X., Jin, X., Chen, S. and Wu, F. (2018). *p*-Coumaric can alter the composition of cucumber rhizosphere microbial communities and induce negative plant-microbial interactions. *Biology and Fertility of Soils* **54**: 363-372.
56. Zhou, X.G., Wang, Z.L., Pan, D.D. and Wu, F.Z. (2018). Effects of vanillin on cucumber (*Cucumis sativus* L.) seedling rhizosphere *Bacillus* and *Pseudomonas* spp. community structures. *Allelopathy Journal* **43**: 255-264.