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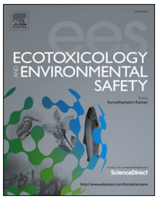

# Bacterial community shaped by heavy metals and contributing to health risks in cornfields

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ABSTRACT

Scientists are increasingly aware that heavy metal contamination in soils, especially in farmland ecosystems, can negatively affect human health and alter the bacterial community that plays a critical role in plant growth and heavy metal accumulation. The goal of the present paper was to uncover how various heavy metals and non-metallic elements affect human health and bacterial diversity in cornfields and to explore the contribution of soil bacteria to heavy metal accumulation in crops. Soil samples were collected from five counties in Shandong Province, China, where abnormally high levels of heavy metals and metalloids were caused by mining and heavy industry. We calculated a hazard quotient (HQ) to evaluate the health risk these heavy metals cause and analyzed the soil bacterial community using 16S rRNA gene sequencing. The HQ results showed that As posed the greatest threat to human health followed by Pb although concentrations of all metals did not reach the health risk threshold. Meanwhile, principal component analysis (PCA) and redundancy analysis (RDA) revealed soil bacterial richness was significantly influenced by As, Ni, and Cr as well as pH and phosphorus, but not by the species diversity of aboveground weeds. The most abundant bacteria in our study region were heavy metal tolerant groups, specifically *Actinobacteria* and *Proteobacteria*. Moreover, correlation analysis suggested that *Actinobacteria* might reduce the phytoaccumulation of Cr, Cu, Zn, and Hg in corn, while *Proteobacteria* might weaken phytoaccumulation of Pb, Ni, As, and Cd. Our results verified that heavy metals play an important role in shaping the soil bacterial community. Using native bacteria in farmland provides a potential biological strategy for reducing the health risk posed by heavy metals related to food consumption.

## 1. Introduction

Mineral resources provide major economic benefits to local people, while creating major problems related to environmental pollution from metals and metalloids. With the development of industry, more and more mining areas have been exploited. The heavy metals from mining and smelting factories can migrate into farmland soil through soil waste disposal, sewage discharge (Wang et al., 2018), and atmospheric deposition (Türtscher et al., 2017). Meanwhile, the excessive use of pesticides and fertilizers has also contributed to high levels of heavy metals in large areas of farmland. The most common toxic heavy metals include mercury (Hg), lead (Pb), cadmium (Cd), copper (Cu), chromium (Cr), manganese (Mn), zinc (Zn), and arsenic (As, a metalloid) (Duruibe et al., 2007) among which Zn, Mn, and Cu are micronutrients in plants, while Hg, Pb, Cd, Cr, and As have no useful biological functions. Heavy metals exist in various forms such as free metal ions, interchangeable metal ions, soluble metal complexes, and metals bound in other compounds. These forms result in different levels of bioavailability and toxicity in agricultural soils and their mobility is influenced by different factors including soil properties, climatic conditions, plant characteristics, and farm management (Dube et al., 2001; Fayiga, 2017). The concentrations and forms of heavy metals in farmland can affect crop yield and many contaminate the food chain. Therefore, the presence of these heavy metals pose health risks to both humans and the ecosystem (Iqbal, 2012; Liu et al., 2016; Yi et al., 2011).

Heavy metals can enter the human body through food consumption and exposure to soil. Soil exposure mainly occurs via ingestion, dermal contact, and inhalation (Wuana et al., 2011). Exposure to heavy metals can cause cancer, cardiovascular disease, chronic anemia, cognitive impairment, reproductive health problems, and other damage to body

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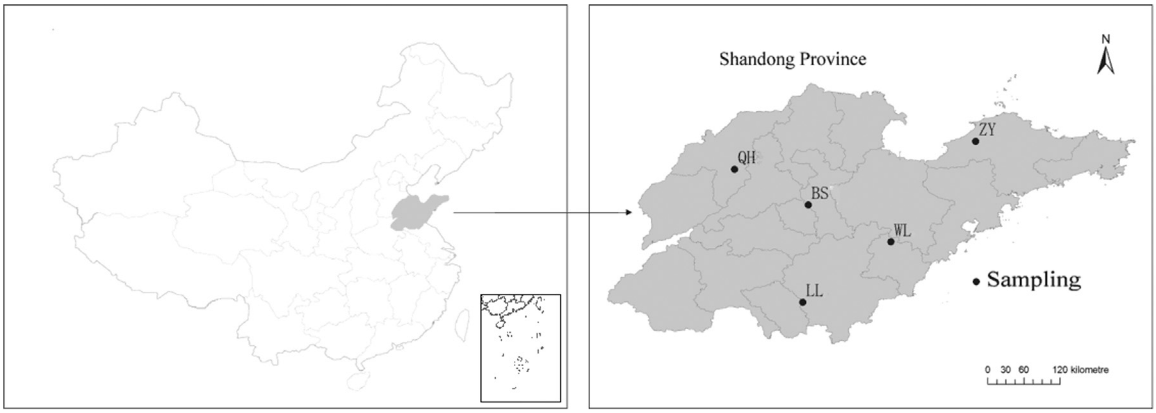

**Fig. 1.** Locations of sampling points in Shandong province, China.

organs of humans (Fayiga, 2017; Ullah et al., 2015). Hence, assessing and mitigating the health risks caused by heavy metals in farmland soil is important. Assessment criteria for human health risks can be used to evaluate the risks caused by soil metal contamination, which is the basis for determining the safe levels of contamination and for evaluating the effectiveness of safety management. The hazard quotient (HQ)serves as one of the major indices used to evaluate the non-carcinogenic risk caused by heavy metal contamination.

The richness and variety of microorganisms can reflect the health status of soil (Fierer et al., 2007) and affect plant fitness (Martin et al., 2017). Although some soil bacteria can cause plant disease, substantial populations of beneficial bacteria can promote plant growth through atmospheric nitrogen fixation, phosphorus solubilisation, phytohormone production, and disease suppression (Gyaneshwar et al., 2002; Hayat et al., 2010; Latz et al., 2012; Singh and Millard, 2004). Heavy metals can impair soil bacteria through protein denaturation, cell membrane disruption, and through the inhibition of cell division or enzyme activity (Abdu et al., 2017). For this reason, in soils with high levels of metal concentration, bacterial communities are generally shaped by various heavy metals (Feris et al., 2003; Gołębiewski et al., 2014; Li et al., 2011; Zhang et al., 2016). However, some tolerant bacteria have the ability to protect themselves and surrounding plants from metal toxicity through volatilization, metal complexation, and enzymatic detoxification (Gadd, 2010; Osman and Cavet, 2008; Pavel et al., 2013). These bacteria can alter the mobility and availability of heavy metals to plants through the release of chelating agents, acidification, and redox changes (Gadd, 2004, 2010; Jing et al., 2007). Therefore, bacteria that can promote plant growth are often used to assist in the phytoremediation of soils contaminated by heavy metals (Ashraf et al., 2017; Ullah et al., 2015). In this situation, bacteria can transform insoluble metals into bioavailable forms and promote the accumulation of heavy metals in plant tissues. However, in cropland, humans need bacteria that will stabilize heavy metals in the soil, lessen the bioavailability of these metals, and reduce the uptake of heavy metals in crops while decreasing the human health risk. The use of heavy metal resistant bacteria is one of the major techniques available for reducing the bioavailability of heavy metals in farmland especially for mildly contaminated soil (Fayiga, 2017). Numerous studies have demonstrated the effects of heavy metals on bacterial diversity and community structure in soil and identified the dominant bacterial taxa in heavy metal contaminated soil. Bacterial groups play an important role in the absorption and inhibition of heavy metals by plants; nevertheless, the importance of the bacterial community in the accumulation of heavy metals in grain crops has not been fully explored.

Goals of the present study included revealing the relationships among heavy metal concentrations, soil bacterial communities, and heavy metal accumulation in crops, and determining whether heavy metal elements that have the largest negative effects on human health also have strong negative effects on bacterial abundance. The present paper estimated the health risk with a HQ based on the heavy metal concentrations in soil from five cornfields, and investigated the bacterial richness using the 16S rRNA gene with high-throughput Illumina MiSeq sequencing. We assessed and compared the effects of each heavy metal on the risk to human health and the alteration of soil bacterial populations. Moreover, we determined the dominant bacteria that influenced the process of heavy metal accumulation in maize. Our results can help researchers to understand the risk heavy metals in farmland soil pose to the human health, the ecological services provided by bacteria, and practically researchers to develop bioremediation strategies using native bacteria with the goal of decreasing the health risk of heavy metals in farmland soils.

## 2. Materials and methods

### 2.1. Sampling design

The investigation was performed in farmlands of Shandong Province in North China. The region has rich mineral resources and high levels of grain yield. Five counties were selected for the present study, specifically Boshan (BS, 36°27′41″N, 117°49′37″E), Lanling (LL, 35°00′03″N, 117°44′39″E), Qihe (QH, 36°59′00″N, 116°43′59.35″E), Wulian (WL, 35°54′27″N, 119°03′58″E), and Zhaoyuan (ZY, 37°24′07″N, 120°19′44″E; Fig. 1). The soil type at sampling sites in QH was Solonchaks, whose formation was conditioned by the arid and semi-arid climate of the region. The soil of the LL and ZY's sampling were Luvisols, in an area with a (sub-)humid temperate climate. The soil type of sampling sites in BS was Calcic cinnamon soil, which are Cambisols; this kind of soil is conditioned by their limited age that is not confined to any particular climatic region. The soil types collected in WL were Regosols with their formation conditioned by the topography and physiography of the terrain; soils in lowlands (wetlands) have level topography while soils in elevated regions have non-level topography (Driessen et al., 2001). With the development of mining and heavy industry, the content of heavy metals in farmland in these areas were found to be elevated when compared with other places (Dai et al., 2012). The five regions belong to the monsoon region, with mean annual rainfall and temperature were 547, 657.8, 671.1, 767.1, and 835.3 mm and 12.9, 12.6, 11.5, 12.5, and 13.5 °C in QH, WL, ZY, BS, and LL, respectively.

The present study focused on cornfields that were tended using conventional agricultural management practices. The selected corn fields were surrounded by factories or mines. Three soil quadrats (1 m × 1 m) in the cornfields were randomly established in each sampling area in November 2015. In each quadrat, surface soil (0–20 cm deep) samples were collected from four subsamples in the corners with one subsample in the centre, which were homogeneously mixed to