

Antibiotics and antibiotic resistance genes in agricultural soils: A systematic analysis

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4 **A systematic analysis of antibiotics and antibiotic resistance genes in**
5 **global agricultural soils**

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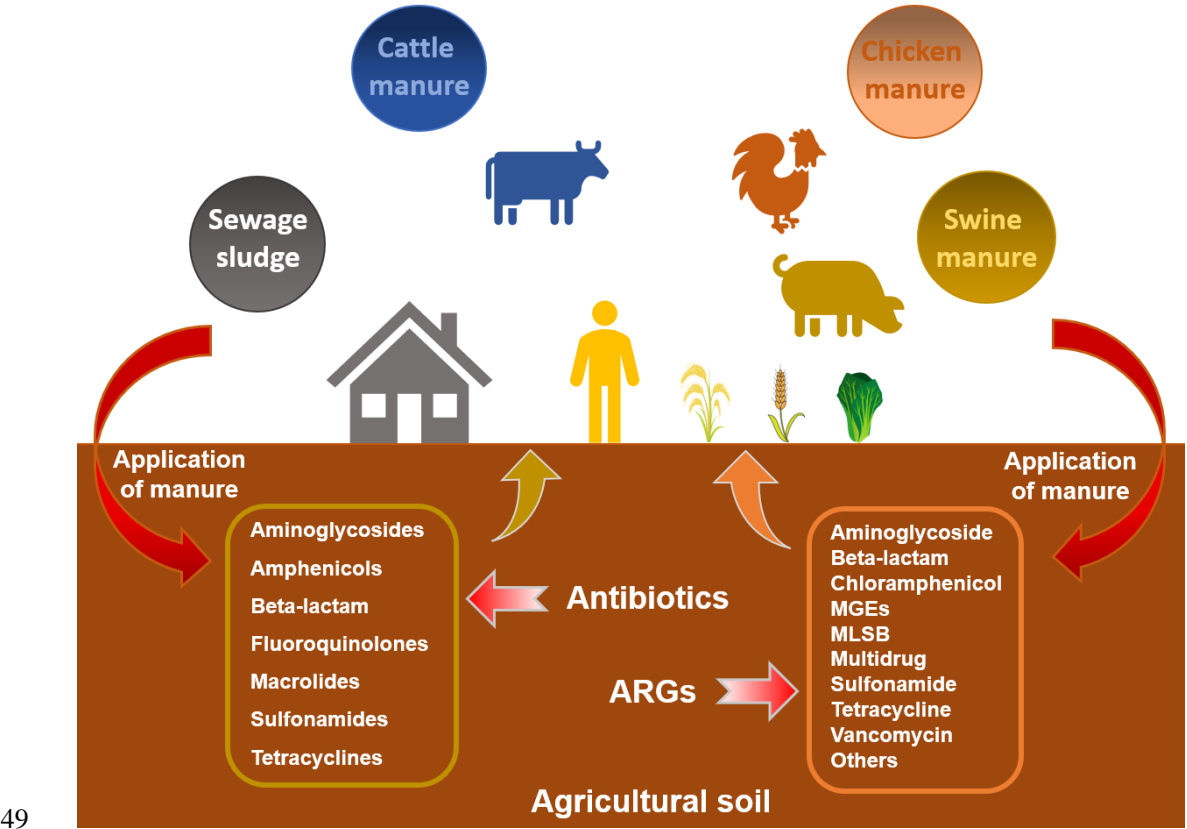
Abstract

Antibiotics and antibiotic resistance genes (ARGs) reach the agricultural soils through fertilization in the form of livestock manure are the main environmental pollutants at present. This study discussed the sources and distribution characteristics of antibiotics and ARGs in global agricultural soils, revealed the main driving factors affecting antibiotic concentration and ARGs abundance, analyzed the environmental risk, and environmental impact of typical antibiotics, and summarized the common elimination methods in current research. We retrieved the papers related to antibiotics and ARGs, and extracted 2093 observations from 135 papers. The results showed that cattle manure, chicken manure, swine manure, and sewage sludge were the primary pollution sources of antibiotics and ARGs in agricultural soils, and sulfonamides, tetracycline, fluoroquinolones, and their corresponding ARGs were the main pollution types. The distribution of the antibiotic concentration and the ARGs abundance was affected by land-use types and soil physicochemical parameters. The mobile genetic elements (MGEs) play a vital role in promoting the dissemination of ARGs, and *sul1* genes strongly correlate with *int1*. The occurrence, fate, and ecotoxicity of antibiotics and ARGs in agricultural ecosystems have become the most pressing environmental issues. Our results provide a systematic analysis of the environmental behavior of antibiotics and ARGs in agricultural soils.

Keywords: veterinary antibiotic; antibiotic resistance genes; agricultural soil; environmental factors; ecological risk; horizontal transfer

Synopsis statement: A systematical description for the distribution of antibiotics and antibiotic resistance genes in agricultural soils worldwide.

Graphical Abstract



1 Introduction

Antibiotics are widely used in many countries worldwide to treat human diseases and protect the health of animals in agricultural fields ^{1,2}. Antibiotics cannot be fully absorbed in animals, and large amounts of antibiotics and their degradation products were introduced into agricultural ecosystems in the form of fertilization and irrigation through antibiotic-contaminated manures and sewage sludge ³⁻⁵. More importantly, accounting for selective pressure on the environmental resistome, antibiotics are

considered to be the driver of increasing antibiotic resistance genes (ARGs) ⁶.

The increasing use of fertilizers in agriculture poses a rapidly increasing threat of antibiotic and ARGs contamination ⁵. Distribution of antibiotic and ARGs abundance varied greatly under different land-use patterns in soils. It has been documented that the concentration of antibiotics differs significantly, typically in the μg antibiotic per kg soil ⁷. ARGs can spread from animal farms to surrounding agricultural soils, posing a potential high risk to environmental ecology ⁸. Remarkably, antibiotics and ARGs contamination status in vegetable, paddy, and upland soil vary greatly. Continuously manure fertilization significantly increased the ARGs abundances in typical greenhouse vegetable production bases, and antibiotic types and vegetable species affected the distribution of ARGs in soil ^{9,10}. The occurrence and abundance of most ARGs increased significantly in wheat soils irrigated with wastewater ¹¹. Organic fertilizer partially replaced mineral fertilizer significantly increased the relative abundance of *sul* genes in north China under wheat-maize rotation ¹². Long-term application of organic fertilizer usually increases antibiotic concentration and ARGs abundance in paddy soil, and paddy soil showed higher ARG accumulation than dryland soil, corresponding to higher microbial biomass ^{13,14}.

Land-use of livestock and poultry manure is an essential agricultural measure. Intensive animal husbandry is the main cause of the increasing environmental contamination of antibiotics and ARGs in soil ¹⁵. Antibiotic-resistant bacteria (ARB) reproduction was induced in soils modified by cow manure ¹⁶. The long-term

application of chicken manure significantly increased the abundance and diversity of ARGs and various classes of antibiotics in soil ¹⁷. The application of untreated swine manure resulted in antibiotic and ARGs pollution, and changed the composition of soil microbial community ^{18,19}. Application of antibiotic-contaminated biogas slurry and residue in the field increased the occurrence of ARB and ARGs ²⁰. Irrigated with tap water spiked with different concentrations of antibiotics significantly altered microbial populations in pot experiments ²¹. Therefore, the pollution of antibiotics and ARGs caused by manure application is worth pondering.

Soil properties, including pH, soil organic carbon (SOC), total nitrogen (TN), soil texture, and heavy metals, are the key factors determining the change and distribution of antibiotics and ARGs. The texture of soil with manure application determines the persistence of target ARGs, thus influencing the fate of antimicrobial resistance in the soil ²². The adsorption potential of sulfonamides increases with decreasing pH because of the abundance of positively charged species capable of electrostatic binding to soil mineral surfaces under acidic conditions ²³. Reducing the manure pH promotes degradation of sulfonamides and inhibits *sul* gene-level transfer, which may be a simple and effective management measure to reduce antibiotic resistance ²⁴. Pot experiments comparing antibiotic resistance in three typical soils found that soil with the highest SOC was least affected by swine slurry amendment ²⁵. In general, SOC strongly influenced the retention of sulfonamides; the higher the SOC content, the higher the adsorption capacity, and the lower the desorption capacity due to competition for adsorption sites ^{26,27}. Clay improver greatly influenced the change of original ARB in

chicken manure compost, and the diversity and relative abundance of ARB in 4% clay was greater than that in high dose clay²⁸. The soil TN was positively correlated with *sul1* in the agricultural soils of the Yangtze River Delta, China²⁹. The soluble organic nitrogen was a critical explanatory variable for the model to determine the swine manure-water distribution coefficients of typical veterinary antibiotics³⁰. Metal contamination plays an essential selective role in spreading antibiotic resistance through the co-selection mechanisms^{31,32}. However, many studies fail to adequately address the role of soil properties as a factor affecting the distribution of antibiotics and ARGs in agricultural soils.

Horizontal gene transfer of ARGs is considered a significant contributor to the spread of antibiotic resistance^{33,34}. The class I integron-integrase gene (*intI1*) is a good proxy for ARGs pollution³⁵. There is an excellent positive co-occurrence pattern of ARGs and mobile genetic elements (MGEs) in manure-amended greenhouse soils³⁶. The abundance of *intI1* increased significantly in the soil exposed to pig manure, and was positively correlated with ARGs in a long-time field experiment³⁷. Another study revealed sub-inhibitory heavy metals widely present in various environments could promote resistance by promoting the horizontal transfer of ARGs³⁸. This means that the analysis of MGEs contamination status is imperative to better understand ARGs dissemination in the soil.

Until now, numerous reports demonstrate the sources, spatial patterns, driving factors, and environmental implications of antibiotics and ARGs in global estuarine and

coastal environments^{39,40}, no comprehensive study of antibiotics and ARGs in agricultural soils has been conducted. In this study, we collated the occurrence, distribution, and risk of antibiotics and ARGs in agricultural soil. The objectives were to (1) analyze the pollution characteristics of antibiotics and the occurrence of ARGs in agricultural soil under different land-use modes, and determine the potential ecological risk of typical antibiotics in soil; (2) analyze the relationship between soil properties and antibiotic concentration and ARGs abundance, and reveal the key factors affecting its pollution characteristics; (3) discuss the coexistence mode of ARGs and antibiotics, and the horizontal transfer of ARGs in the soil, and summarize the common elimination methods and effects in current researches.

2 Methodology

2.1 Data collection

To survey antibiotics and ARGs in agricultural soil, we performed a systematic search using the ISI Web of Science (Thomson Reuters, New York, NY, USA), Google Scholar (Google, Mountain View, CA, USA), and China National Knowledge Infrastructure (CNKI, Beijing, China). Our search terms included (“soil*” OR “agricultural soil*” AND (“antibiotic*” OR “antibiotic resistance gene*” OR “ARG*” OR “resistome*”). Studies included in this study had to meet the following criteria: (a) both laboratory field and incubation studies were selected; (b) only studies that had an adequate number of replicates; (c) the means, standard errors, and replication of the variables could be extracted directly from the text, tables, or digitized graphs. The following information

was documented for each study: the type of experiment, site, planting type, contaminated type, soil physiochemical parameter, type of antibiotic and ARGs, the concentration of antibiotics, and abundance of ARGs. A total of 2039 observations were taken from 135 papers (Text S1). The distribution location of observations is shown in Fig. 1. China accounted for about 85.87% of observations, and other countries, including Israel, Brazil, Korea, Pakistan, Spain, and the USA accounted for 14.13%.



Fig. 1 The distribution of data collected from different countries. The point sizes represent the number of replications, and different colors denote antibiotics and ARGs.

2.2 Ecological risk assessment of antibiotics

A risk quotient (RQ) was used to evaluate the potential risk and calculated as follows

^{41,42}:

$$RQ = \frac{MEC}{PNEC_{soil}} \quad (1)$$

$$PNEC_{soil} = PNEC_{water} \times K_d \quad (2)$$

$$\text{PNEC}_{\text{water}} = \text{EC}_{50} / \text{AF} \quad (3)$$

where MEC is the actual measured concentration in the environment ($\mu\text{g kg}^{-1}$), $\text{PNEC}_{\text{soil}}$ is the predicted no-effect concentration in soil ($\mu\text{g kg}^{-1}$), and $\text{PNEC}_{\text{water}}$ is the predicted no-effect concentration in water, and which represents the maximum drug concentration that will not have an adverse effect on the microorganisms or the ecosystem in the environment under the existing cognition ($\mu\text{g L}^{-1}$). K_d is the soil-water partition coefficient (L kg^{-1}). EC_{50} is the half-maximum effect concentration (mg L^{-1}). EC_{50} is all obtained from the literature. AF is the evaluation factor, which is the recommended value of the European Water Framework Directive (1,000). The ecological risk parameters of antibiotics are summarized in Table S1.

RQ was used to assess the ecological risk of the target antibiotic. The RQ values were mainly divided into four levels: $\text{RQ} \geq 1$ high risk; $0.1 \leq \text{RQ} < 1$ medium risk; $0.01 \leq \text{RQ} < 0.1$ low risk; $\text{RQ} \leq 0.01$ no risk⁴³.

2.3 Statistical analysis

The mean concentrations of antibiotics and ARGs abundance were either provided in the publications or calculated using the original data. Concentrations that were “not detected” or “below detection limit” were entered as zero values. To minimize the differences in background bacterial abundances, the absolute number of all detected genes was further normalized to that of the ambient 16S rRNA gene. We used linear mixed-effects models to test the fixed effects of planting type, contaminated type, type

of antibiotic, and ARGs on the concentration of antibiotics and the ARGs abundance, respectively. Study ID was included as a random effect. The “nlme” package was used to conduct linear mixed-effects models in R version 4.0.3 (R Core Team, 2020). Multiple comparisons were made using the Tukey HSD test. The network analysis was visualized on the Gephi platform (version 0.9.2).

3 Results

3.1 Antibiotics in agricultural soils

3.1.1 Distribution characteristics of antibiotics in agricultural soils

The concentrations of antibiotics in soil samples are shown in Fig. 2. In this survey, beta-lactam, sulfonamides, tetracyclines, and fluoroquinolones classes were the dominant antibiotics with a concentration of 364.16, 241.17, 176.98, and 112.02 $\mu\text{g kg}^{-1}$, respectively, much higher than others (Fig. 2A). Simultaneously, tetracycline, chlortetracycline, oxytetracycline, sulfamethoxazole, norfloxacin, and ciprofloxacin have higher concentrations, and their concentrations differed in several orders of magnitude (Fig. S1). Applications of different manure could make a difference in the contamination status of antibiotics. Soil applied with cattle, chicken, and swine manure contains higher concentrations of antibiotics, followed by sewage sludge and mix manure ($P < 0.05$; Fig. 2B). Concentrations of antibiotics in vegetable and paddy soil were significantly higher than other planting types ($P < 0.01$; Fig. 2C).

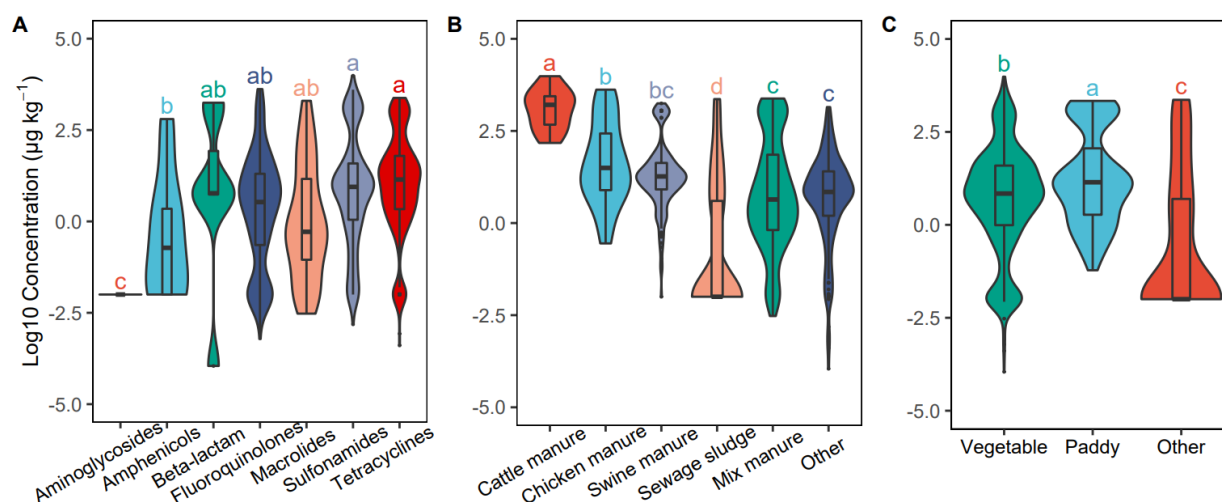


Fig. 2 Concentrations of antibiotics in global agricultural soils. Violin plots show the concentration of antibiotics (A) for each type of antibiotics, (B) in the soil with different manure applied, and (C) from different land-use types. Different lowercase letters in each panel indicate significant differences between groups. The solid line inside the box represents the median.

3.1.2 Correlation between antibiotics and soil physicochemical parameters

The correlations between the major environmental variables and the concentration of antibiotics were analyzed for all studied data. A significant positive correlation was observed between antibiotics and SOC ($P < 0.05$) and several heavy metals (e.g., Pb, Hg, and Na) ($P < 0.01$; Fig. 3), respectively. In contrast above factors, the concentration of antibiotics indicated negative correlations with clay, TN, Cd, and As ($P < 0.01$).

In detail, sulfonamides and tetracyclines showed strong negative correlations with TN and pH ($P < 0.01$), respectively. We also observed that most heavy metal contents were significantly correlated to the two antibiotics, but they show opposite correlations with most heavy metals (e.g., Cu, Cd, Na, Ca, and Mg). Beta-lactam and

fluoroquinolones antibiotic showed a significant positive correlation with pH ($P < 0.05$), while showed a significant negative correlation with TN ($P < 0.05$). Moreover, the strongly negative relationship between macrolides and clay, and several heavy metals (e.g., Cu, Zn, and Pb), the significant correlation between aminoglycosides, amphenicols, and environmental factors existed in the soils in this study.

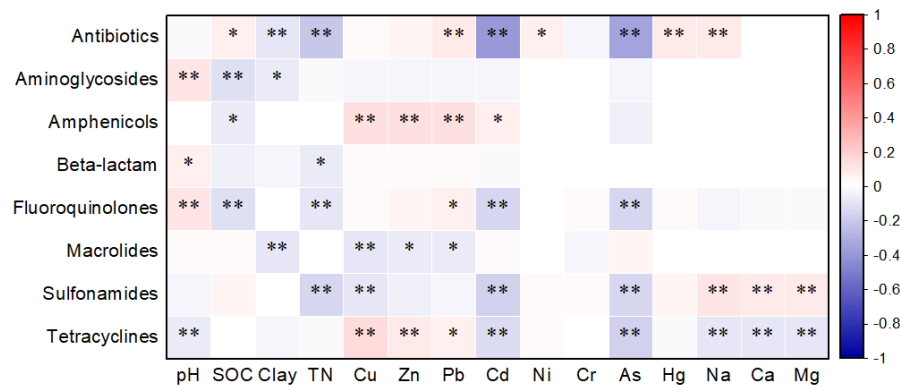


Fig. 3 Spearman correlations between antibiotics and environmental factors in soil.

Pairwise comparisons are shown, with a color gradient denoting spearman's correlation coefficient. *** $P < 0.001$, ** $P < 0.01$, and * $P < 0.05$. Cu, copper; Zn, zinc; Pb, lead; Cd, cadmium; Ni, nickel; Cr, chromium; As, arsenic; Hg, mercury; Na, sodium; Ca, calcium; Mg, magnesium.

3.1.3 Potential ecological risk of antibiotics

To better elucidate the ecological risk of antibiotics in agricultural soils, the RQ values of concentrations of sulfonamides, tetracyclines, and fluoroquinolones classes in soil were analyzed (Fig. 4). In this study, the RQ of the three antibiotic classes detected were all less than one. The RQ of norfloxacin is significantly higher than the other antibiotics

of all target compounds and may present a relatively higher ecological risk, followed by the sulfadiazine. Contrarily, the RQ values of other antibiotics examined here were below 0.1 poses a low or no environmental risk. We can speculate that the soil was seriously polluted with antibiotics, leading to ARGs contamination and potential ecological risks.

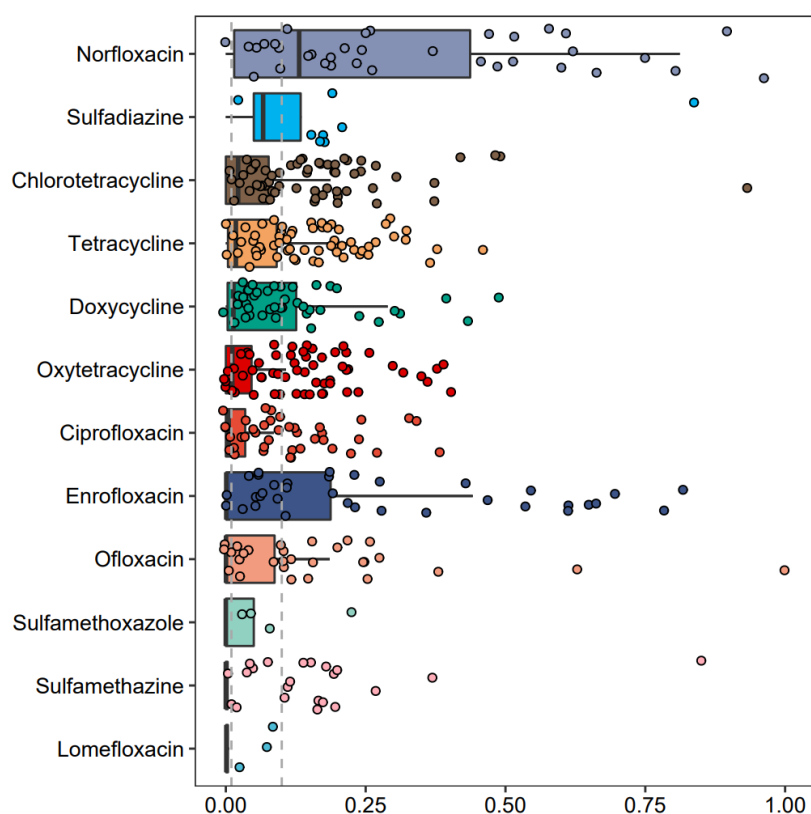


Fig. 4 Risk quotient values of various antibiotics in agricultural soil. The RQ values could be divided into four levels according to the European Commission: $RQ \geq 1$ high risk; $0.1 \leq RQ < 1$ medium risk; $0.01 \leq RQ < 0.1$ low risk; $RQ \leq 0.01$ no risk. The dotted line in the figure represents the values of 0.01 and 0.01, respectively. RQ values greater than one is not shown in the figure for clearer display (6% of total observations).

3.2 ARGs in agricultural soils

3.2.1 The abundance of ARGs in agricultural soils

The abundance of ARGs varied greatly across soils. ARGs conferred resistance to multidrug, sulfonamide, and tetracycline classes were the most abundant ARG subtypes in samples, while other ARGs were found at much lower levels (Fig. 5A). Additionally, the abundance of the *sul1* and *sul2* genes was higher than that of other *sul* genes, the *tetM*, *tetW*, *tetO*, and *tetX* were the most abundant across *tet* genes, and *intI1* was the most abundant integron genes (Fig. S2). The abundance of ARGs was found at much higher levels in soil applied with cattle, chicken, swine manure, and mixed manure ($P < 0.05$; Fig. 5B). It is noteworthy that a similar distribution pattern was observed for ARGs among planting types compared with antibiotics (Fig. 5C).

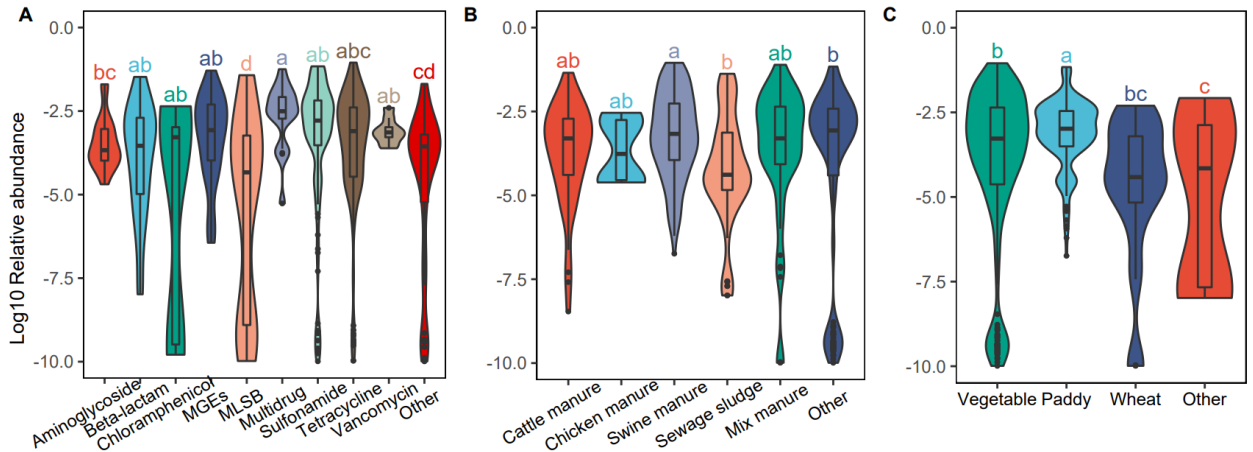


Fig. 5 The abundance of ARGs in global agricultural soils. The abundance of ARGs in soil (A) for each type of AGRs, (B) in the soil with different manure applied, and (C) from different land-use types. Different lowercase letters in each panel indicate significant differences between groups. The solid line inside the box represents the median.

3.2.2 Correlation between ARGs and soil physicochemical parameters

Correlation analysis showed that the abundance of target ARGs in soil was positively correlated with the clay content generally but negatively correlated with the SOC, TN, and metallic elements (e.g., Cd, Ni, Cr, As, and Hg) ($P < 0.01$; Fig. 6).

Specifically, sulfonamide and tetracycline classes exhibited significant negative correlations with pH ($P < 0.05$). Almost no correlations were found between most heavy metals and sulfonamide ARGs except for Na, Ca, and Mg ($P < 0.05$). Cu, Zn, and Pb are correlated positively with tetracycline ARGs ($P < 0.01$). However, we found an opposite trend between other various heavy metals and tetracycline ARGs. Moreover, none of the ARGs detected above in samples showed a significant correlation with SOC, TN, and clay.

Unlike the above two ARGs, the analysis revealed that beta-lactam and multidrug ARGs were significantly and positively correlated with pH. A stronger correlation was found observed between beta-lactam and pH ($P < 0.01$) than between multidrug and pH ($P < 0.05$). Our results did not show statistically significant correlations between the beta-lactam and multidrug ARGs with SOC and clay. Beta-lactam ARGs showed a significantly negative correlation with TN but a significantly positive correlation with various heavy metals ($P < 0.01$). Furthermore, the MGEs were positively correlated with Ni ($P < 0.05$), while other soil physicochemical parameters were less intensely or even not associated.

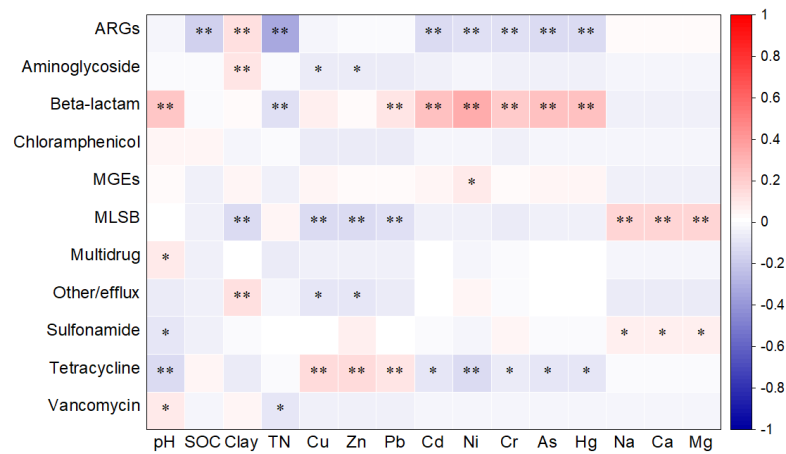


Fig. 6 Spearman correlations between abundance of ARGs and environmental factors in soil. Pairwise comparisons are shown, with a color gradient denoting spearman's correlation coefficient. *** $P < 0.001$, ** $P < 0.01$, and * $P < 0.05$. Cu, copper; Zn, zinc; Pb, lead; Cd, cadmium; Ni, nickel; Cr, chromium; As, arsenic; Hg, mercury; Na, sodium; Ca, calcium; Mg, magnesium.

3.2.3 Correlations between ARGs and *intI1*

Based on the correlation analysis between the most prevalent mobile genetic elements (*intI1*) and ARGs (Fig. 7), there were strong positive correlations between *intI1* and the abundance of sulfonamide ARGs (*sul1*) in soils ($P < 0.01$) and weaker correlations with the rest of ARGs. This might indicate that *sul1* is generally located on the more readily disseminated class I integron and a lower frequency of horizontal gene transfer caused by *intI1* for other types of ARGs. This is not surprising given that these correlations do not entirely reflect variations in ARGs dissemination based on existing data.

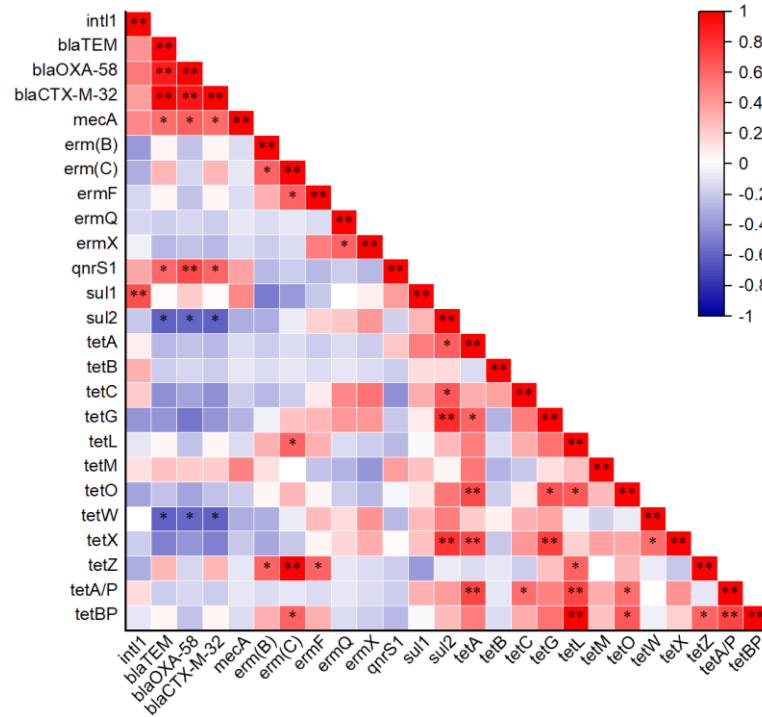


Fig. 7 Spearman correlations between the abundance of *intI1* and ARGs in soil. Pairwise comparisons are shown, with a color gradient denoting spearman's correlation coefficient. *** $P < 0.001$, ** $P < 0.01$, and * $P < 0.05$.

4 Discussion

4.1 Effects of land-use on the distribution of antibiotics and ARGs

The distribution characteristics of antibiotics and ARGs were significantly affected by land-use patterns. Our finding provides evidence that both the concentration of antibiotics and the abundance of ARGs were higher in the paddy fields than that in uplands, which may be attributed to differences in the degradation of antibiotics and the migration capacity of ARGs in soil under different planting types. The rate of transformation and degradation of antibiotics in the soil largely depends on many

abiotic factors, and anaerobic conditions significantly prolonged the half-life of antibiotics⁴⁴. Paddy soil generally showed a higher accumulation of ARGs due to higher microbial biomass than host populations of ARGs under anaerobic conditions¹⁴. Increasing the amount of manure in a paddy-upland rotation system resulted in a sharp increase in soil ARGs; however, the high moisture content may block the path of contact from the manure-improved soil to the phyllosphere, thereby affecting the dissemination of ARGs^{45,46}. Therefore, it should be fully considered that multiple soil factors may drive the distribution of antibiotics and ARGs to confirm the different fates of them in the soil introduced by manure under aerobic and anaerobic conditions.

Prior studies that have noted the behavior of antibiotics and ARGs proved to be governed by the combination of soil and plants under the real planting conditions⁴⁷. Further analysis showed that vegetable species had a significant effect on the content of antibiotics and ARGs in the soil, and the soil planting pakchoi accumulates more than other vegetables (Fig. 8). Antibiotics and ARGs absorbed by plants are further harmful to human health as they travel up the food chain. Therefore, it may represent higher health risks than other vegetables when consumed pakchoi by humans. Plant root exudates indirectly affect the fate of antibiotics by changing soil properties during vegetable growth¹⁰. Antibiotics accumulate in plants through passive absorption by water⁴⁸, which is the main reason for dissipation and removal of antibiotics effects differed in the soil of different vegetable growing types. The growth of plant roots alters the soil microbial composition, and the selectivity of plants to environmental bacteria may be an important factor leading to the difference of distribution of ARGs^{49,50}.

Additionally, different types of antibiotics are absorbed and accumulated in different parts of plants^{51,52}. Although this aspect is not involved in this study, the environmental behavior of complex types of antibiotics in agricultural soils warrants more exhaustive investigation.

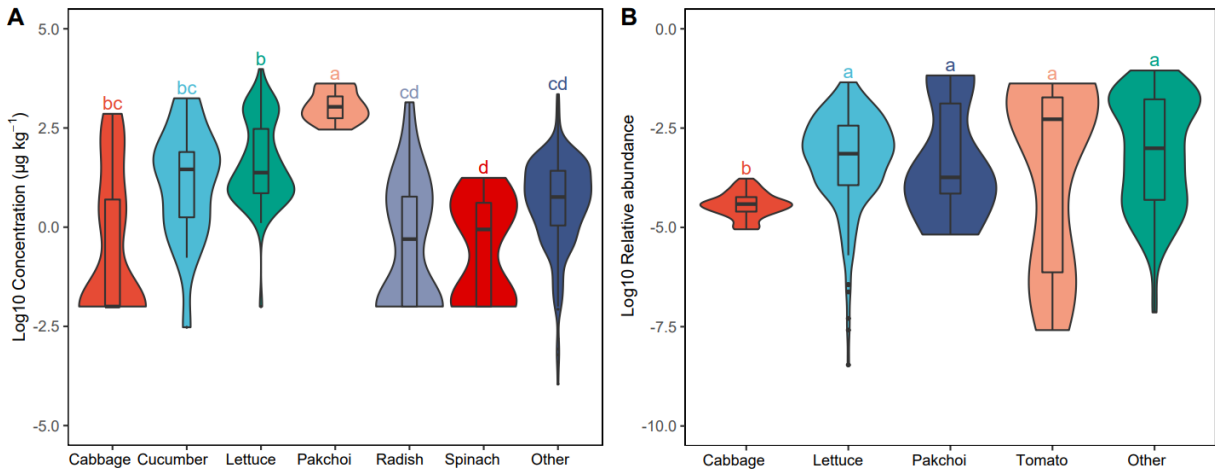


Fig. 8 Concentrations of antibiotics and abundance of ARGs in vegetable soils. Violin plots show the concentration of antibiotics (A) and abundance of ARGs (B) for each type of vegetable. Different lowercase letters in each panel indicate significant differences between groups. The solid line inside the box represents the median.

4.2 Effects of fertilization on the distribution of antibiotics and ARGs

The contamination of antibiotics and ARGs in the soil is related to fertilization methods and local farming practices. The amount of antibiotics taken by farm animals varies by livestock species and region, and the abundance of ARGs is related to the amount of antibiotics used by these animal species⁵³. Previous studies found significant differences in the abundance of ARGs in different types of farm soils, with the highest

concentrations in pig farm soils ⁵⁴. This also accords with our observations above that swine manures generally had higher levels of ARGs, followed by soils applied cattle and chicken manure, and the ARGs contamination caused by the mixed-use of manure should not be ignored. Pork production is the main pillar industry of animal husbandry, and antibiotics are widely used in pig breeding due to the disease types and incidence rate of pigs being relatively high compared with other animals ⁵⁵. Metagenomic sequencing indicated that the long-term application of swine manure could increase microbial diversity and reshape ARGs in agricultural soils ⁵⁶. Cattle manure is widely used as fertilizer because it is an excellent source of nutrients, they exhibit more abundant ARGs than poultry manure, and the application of fresh cattle manure may lead to a greater transmission potential of antimicrobial resistance genes in the soil ⁵⁷. The current study found that a lower bacterial diversity in poultry litter might be linked to the higher total solids ⁵⁸, which in turn affects the abundance of ARGs in the soil. Taken together, it is essential to reasonably control the abuse of antibiotics in livestock and poultry breeding.

Applied sewage sludge increased soil microbial activity, thereby contributing to the risk of the spread of ARGs and MGEs ^{59,60}. Peri-urban agricultural plots were irrigated with treated wastewater and groundwater from rural organic farms suggested the agricultural practices and water quality for irrigation influenced microbial communities and associated resistances ⁶¹. However, in reviewing the literature, the study did not reveal the risk of sludge improvement associated with the development of antibiotic resistance in an agricultural field trial ⁶². Our study demonstrated that

antibiotics and ARGs pollution caused by sewage sludge were not significant compared with other fertilization methods. Hence, it should be inferred carefully due to the different sludge quality and application practices in various agricultural regions.

4.3 Effects of soil environmental factors on the distribution of antibiotics and ARGs

The sorption ability is stronger at a lower pH value and higher clay fraction in the soil⁶³. Although the effects of pH on the occurrence of soil antibiotics and ARGs have been studied extensively, no unified results have been formed. Our results indicated that pH was not strongly correlated with antibiotics and ARGs, and the effects of pH on them were not consistent. Only beta-lactam and tetracyclines and their corresponding ARGs were significantly positively and negatively correlated with pH, respectively. The adsorption mechanism and degradation performance of antibiotics were significantly different in soils with different pH values. Hydrophobic interaction and cation exchange might be the primary mechanism of tetracyclines adsorption in acidic and alkaline soil, respectively⁶⁴. Studies on the effects of pH on the adsorption capacity of sulfamethoxazole in pasture soils showed that the adsorption capacity decreased when pH increased⁶⁵. pH value is an essential parameter of the hydrolysis process of antibiotics; the hydrolysis rate of base catalysis was significantly higher than that of acid catalysis and neutral pH and altered the fate of antibiotics⁶⁶. Moreover, pH also strongly affected the bacterial diversity and shaped the ARGs profiles, given that bacteria were the dominant carriers of ARGs⁶⁷. Positive correlations between sorption

coefficients and clay content were found for several antibiotics ⁶⁸. Contrary to the results, this study did not detect evidence for the phenomenon, which may relate to the type of antibiotics we analyzed. The findings observed a significant positive correlation between ARGs and clay content in this study mirror those of the previous studies that soil texture was the top edaphic factor that correlated with the abundance of ARGs ⁶⁹.

Nutrient factors in fertilized soil affected the distribution of ARGs, with SOC and TN being contributed significantly to the distribution of ARGs ⁷⁰⁻⁷². In general, soils with higher SOC showed higher adsorption for antibiotics, dissolved organic matter released from farmland straw may reduce the ecological risk of sulfamethoxazole by inhibiting the migration of antibiotic pollutants ⁷³. The removal of SOC by calcination results in a sharp decrease in the adsorption capacity of antibiotics ⁷⁴. This finding supports the result that there was a significant positive correlation between SOC content and antibiotics. However, the risk is serious in soils with lower SOC because antibiotics adsorption is limited ⁷⁵. TN was significantly positively correlated with the abundance of target ARGs in wastewater contamination of coastal waters ⁷⁶. Our study suggested that TN showed a significant negative correlation with ARGs. This somewhat contradictory result may be due to promoting soil microbial degradation of antibiotics under high TN content ⁷⁷. More research on this topic needs to be undertaken before the association between soil physical and chemical properties, and distribution of antibiotics and ARGs is more clearly understood.

Large amounts of heavy metals are used in large intensive livestock and poultry

farms worldwide to promote growth and control disease ⁷⁸. As mentioned in the literature review, an obvious co-selection mechanism between heavy metals and ARGs includes co-resistance, cross-resistance, co-regulation, and biofilm phenotypes ³¹. In the first instance, a positive correlation between ARGs and high metal concentrations, some antibiotic-resistant bacterial strains also showed tolerance to high concentrations of heavy metals ^{79,80}. In the next place, most ARGs and heavy metal resistance genes are carried on MGEs, which contributes to their spread and reproduction in contaminated soils. Heavy metal resistance genes are thought to play an essential role in the proliferation of ARGs ⁸¹. Furthermore, heavy metals can trigger the proliferation of ARGs by increasing the abundance of MGEs or altering the bacterial community structure ⁸².

Specifically, there was a significant positive correlation between tetracycline ARGs and Cu, Zn, and Pb, which was confirmed by related studies ^{83,84}. As and Cd as common heavy metals pollution types had a relatively high accumulation proportion in the soil, the strong positive correlation between Cd, As, and beta-lactam ARGs in this study were found. Cd stress in sludge compost increased the relative abundance of target ARGs in rhizosphere soil, which exacerbated the risk of ARGs transferring from the underground to the aboveground part of plants ⁸⁵. As (III) may coexist with ARGs on the mobile genome as a co-selection driver in paddy soils ⁸⁶. Notably, long-term exposure to far below the minimal inhibitory concentration of certain metals also increases bacterial resistance to antibiotics, demonstrating that even low concentrations of heavy metals may pose a considerable risk to the transmission of ARGs ⁸⁷.

Additionally, the occurrence of co-selection is also related to soil characteristics; Hg-driven ARGs were observed in alkaline soils but not in neutral pH soils⁸⁸. More seriously, multiple antibiotics form complexes with metal cations (e.g., Cu and Zn) that impede (or enhance) antibiotic activity⁸⁹. In this study, the significant positive correlation between amphenicols, tetracyclines, and heavy metals may be due to the complexes promoting the adsorption of antibiotics in the soil^{90,91}. Therefore, controlling heavy metal pollution is crucial to reducing the adsorption, transfer, and diffusion of antibiotics and ARGs in agricultural soils.

4.4 Coexistence of antibiotics, MGEs, and ARGs in agricultural soils

The abundance of ARGs is associated with the overuse and misuse of antibiotics in the soil. To determine the association between antibiotics and ARGs in agricultural soils, we performed network analysis on the antibiotics and ARGs data of the same treatment (Fig. 9). Specifically, tetracyclines are closely related to *tetB*, *tetX*, *tetM*, *tetL*, *tetW* and *tetO*, but the correlation between sulfonamides and *sul1* and *sul2* is weak, although they are the main ARGs. Pot experiment showed that the abundance of *sul1* in sulfamethoxazole-contaminated soil increased significantly⁹². The emergence of tetracycline and quinolone resistance genes was consistent with the results of antibiotic risk assessments in farm soils fertilized with animal manure⁹³. It is interesting to note that an antibiotic can be closely associated with the generation of different types of ARGs, which may cause more significant risks to the farmland environment and is worth our vigilance. Assessment of the relative contributions of antibiotics to ARGs in

a natural farming environment suggests that sulfonamides are significantly associated with multiple genes and dominate the spread of ARGs⁹⁴. However, studies have questioned there was no clear correlation between the presence of targeted antibiotics and the relative abundance of ARGs in the soil irrigated with treated wastewater⁹⁵. Hence, the distribution factors of ARGs in the soil should be considered comprehensively to assess its environmental impact more accurately.

The integrase gene *intI1* that can be horizontal transferred between bacteria is usually closely associated with ARGs^{37,96}. The positive correlation between ARGs and integrase genes confirmed that the application of manure fertilizer increased the possibility of ARGs transmission in agricultural soils⁹⁷⁻⁹⁹. Once ARGs are integrated into successful gene delivery elements, they can persist and spread even without antibiotics². The positive correlation between *intI1* and *sul1* in estuarine sediments suggested that *intI1* may be involved in the occurrence and reproduction of sulfonamides resistance genes¹⁰⁰, which is consistent with our results (Fig. 7). In addition, the correlation between *intI1* and beta-lactam ARGs (*bla*_{TEM}, *bla*_{OXA-58}, and *bla*_{CTX-M-32}) was also strong, although these results were not statistically significant (Fig. 7). Simultaneously, this study provides evidence that *intI1* was significantly positively correlated with a variety of ARGs (e.g., *tetB*, *cmlA*, and *qnrA*), which further confirmed the role of *intI1* in promoting the transmission of ARGs (Fig. 9).

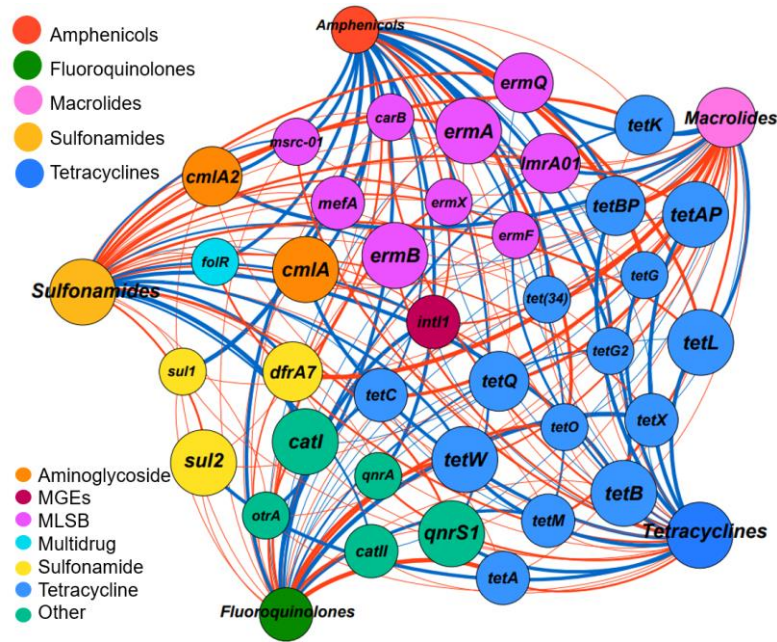


Fig. 9 Network visualizes the antibiotics and ARGs in soils. A connection represents a strong Spearman's correlation coefficient $\rho > 0.7$ with statistical significance ($P < 0.05$). The nodes were colored according to antibiotics and ARGs types. The blue and red lines represent positive and negative correlations, respectively. Node size is proportional to the number of connections, and edge thickness is proportional to the correlation coefficient.

4.5 Environmental implications and mitigation strategies of antibiotics and ARGs

The antibiotics and ARGs burden have serious implications for human health and the agricultural environment. In most cases, agricultural management adopts the repeated use of manure-containing antibiotics, whereas little is known about the long-term effects of this measure on natural microbial communities in the soil, which plays a crucial role in ecological processes such as the nitrogen cycle in the soil^{3,101,102}. A more sophisticated analysis of the fate and effects of antibiotics accounting for spatiotemporal variability of microorganisms in agricultural soils is necessary. In

addition, The obvious consequence of antibiotic release is the emergence of resistant bacteria with the increasing use of antibiotics in the natural environment, and the high mortality caused by multidrug-resistant bacterial infections is of particular concern^{103–105}. The presence of antibiotics in agricultural soils can adversely affect plant growth and productivity, their absorption, and accumulation in crops, and ultimately affect human health through the food chain^{106–108}. Antibiotic compounds were ubiquitous in soil samples from organic vegetable farms. The norfloxacin is a typical quinolone antibiotic which could pose a high risk in vegetable soils^{109,110}. This study further confirms the ecological risk of norfloxacin. Sulfanilamide and tetracycline are most common in farms, wastewater treatment plants, and soil, and the highest concentrations were above 10 mg kg⁻¹ in agricultural soils from multiple feedlots^{111,112}. These findings seem to be consistent with our research, and this may be due to unique structure of them affecting the degradation and migration processes in the soil. Hence, it is necessary to understand the fate and transmission mechanisms of antibiotics and ARGs and develop effective technologies to minimize the risk of antibiotic resistance.

In general, anaerobic and aerobic digestions of compost, manure, or sludge are reasonable disposal measures to reduce antibiotic resistance in agricultural soils at source¹¹³. The removal effect of ARGs, ARB, and transposon genes was pronounced after aerobic composting¹¹⁴. Anaerobic digestion can reduce the relative abundance of macrolide-lincosamide-streptogramin and tetracycline resistance genes in biogas slurry²⁰. Adsorbent materials such as biochar used as a soil amendment can effectively remove antibiotic residues in feces and reduce the ecological risk of their release into

the environment ^{115–117}. In addition, constructed wetlands are considered an economical and efficient ecological restoration technology are emerging in recent years. Their performance in removing antibiotics is superior to many traditional wastewater treatment technologies under certain conditions ¹¹⁸. With the increasing pollution of antibiotics and ARGs in agricultural soils, it is necessary to grasp the mechanisms of their adsorption, migration, and degradation. It is imperative to understand the expression and inhibition of ARGs for the study of their removal mechanisms.

Regulated management in the treatment of antibiotics and ARGs prior to exposure to the soil is lacking. The Ministry of Agriculture and Rural Affairs of the People's Republic of China formulated the National Action Plan for the Reduction of Veterinary Antimicrobial Use (2021-2025). This plan aims to effectively improve the ability and level of the safe, standardized, and scientific use of veterinary antimicrobials in livestock and poultry breeding and establish a sound and strict implementation of a management system for the safe use of veterinary drugs. China faces the most severe challenge of any country in dealing with antimicrobial resistance due to the large production and usage of antibiotics. In fact, the long-term effects of antibiotics and ARGs on natural microbial communities and agricultural ecosystems are still largely unknown. The transmission of antibiotic-resistant bacterial might occur on a worldwide scale ¹¹⁹. The widespread nature of antibiotics and ARGs in agricultural soils is well documented in countries. Nevertheless, limited researches from low-income countries hinder a comprehensive understanding of antibiotics and ARGs, and this knowledge gap limits our ability to assess the risk of antibiotic resistance outbreaks in agricultural

ecosystems³⁹. As such, antibiotics and ARGs should be treated as a global health issue that all countries strive to solve.

5 Conclusions

In summary, this study systematically described the distribution of antibiotics and ARGs in agricultural soil. Cattle manure, chicken manure, swine manure, and sewage sludge are the primary sources of antibiotic pollution in agricultural soil. For ARGs, the pollution caused by the application of mixed manure cannot be underestimated. Sulfanilamide, tetracyclines, fluoroquinolones, and corresponding ARGs were the main pollution types, and the abundance of multidrug resistance was also high in agricultural soil. Land-use types and environmental variables regulated the distributions of antibiotics and ARGs. There were significant differences in antibiotic concentration and ARGs abundance under different planting types and soil physicochemical parameters (e.g., pH, SOC, clay content, TN, and heavy metals). It is worth emphasizing that the *intI1* plays a vital role in promoting the dissemination of ARGs. In addition, it is necessary to find appropriate conditions and scientific management methods for the removal of antibiotics and ARGs to play a greater efficiency. Hence, a more sophisticated analysis of the occurrence and fate of different antibiotic and ARGs classes to combat this type of increasingly serious pollution.

References

- (1) Sarmah, A. K.; Meyer, M. T.; Boxall, A. B. A. A Global Perspective on the Use, Sales, Exposure Pathways, Occurrence, Fate and Effects of Veterinary Antibiotics (VAs) in the Environment. *Chemosphere* **2006**, *65* (5), 725–759.
<https://doi.org/10.1016/j.chemosphere.2006.03.026>.
- (2) Martinez, J. L. Environmental Pollution by Antibiotics and by Antibiotic Resistance Determinants. *Environ. Pollut.* **2009**, *157* (11), 2893–2902.
<https://doi.org/10.1016/j.envpol.2009.05.051>.
- (3) Jechalke, S.; Heuer, H.; Siemens, J.; Amelung, W.; Smalla, K. Fate and Effects of Veterinary Antibiotics in Soil. *Trends Microbiol.* **2014**, *22* (9), 536–545.
<https://doi.org/10.1016/j.tim.2014.05.005>.
- (4) Du, L.; Liu, W. Occurrence, Fate, and Ecotoxicity of Antibiotics in Agro-Ecosystems. A Review. *Agron. Sustain. Dev.* **2012**, *32* (2), 309–327. <https://doi.org/10.1007/s13593-011-0062-9>.
- (5) Kuppusamy, S.; Kakarla, D.; Venkateswarlu, K.; Megharaj, M.; Yoon, Y. E.; Lee, Y. B. Veterinary Antibiotics (VAs) Contamination as a Global Agro-Ecological Issue: A Critical View. *Agric. Ecosyst. Environ.* **2018**, *257*, 47–59. <https://doi.org/10.1016/j.agee.2018.01.026>.
- (6) Pruden, A.; Pei, R.; Storteboom, H.; Carlson, K. H. Antibiotic Resistance Genes as Emerging Contaminants: Studies in Northern Colorado. *Environ. Sci. Technol.* **2006**, *40* (23), 7445–7450.

- 557 <https://doi.org/10.1021/es060413l>.
- 558 (7) Qiao, M.; Ying, G. G.; Singer, A. C.; Zhu, Y. G. Review of Antibiotic Resistance in China and
 559 Its Environment. *Environ. Int.* **2018**, *110*, 160–172.
 560 <https://doi.org/10.1016/j.envint.2017.10.016>.
- 561 (8) Fang, H.; Han, L.; Zhang, H.; Long, Z.; Cai, L.; Yu, Y. Dissemination of Antibiotic Resistance
 562 Genes and Human Pathogenic Bacteria from a Pig Feedlot to the Surrounding Stream and
 563 Agricultural Soils. *J. Hazard. Mater.* **2018**, *357*, 53–62.
 564 <https://doi.org/10.1016/j.jhazmat.2018.05.066>.
- 565 (9) Pu, Q.; Zhao, L. X.; Li, Y. T.; Su, J. Q. Manure Fertilization Increase Antibiotic Resistance in
 566 Soils from Typical Greenhouse Vegetable Production Bases, China. *J. Hazard. Mater.* **2020**,
 567 *391*, 122267. <https://doi.org/10.1016/j.jhazmat.2020.122267>.
- 568 (10) Sun, Y.; Guo, Y.; Shi, M.; Qiu, T.; Gao, M.; Tian, S.; Wang, X. Effect of Antibiotic Type and
 569 Vegetable Species on Antibiotic Accumulation in Soil-Vegetable System, Soil Microbiota, and
 570 Resistance Genes. *Chemosphere* **2021**, *263*, 128099.
 571 <https://doi.org/10.1016/j.chemosphere.2020.128099>.
- 572 (11) Dungan, R. S.; McKinney, C. W.; Leytem, A. B. Tracking Antibiotic Resistance Genes in Soil
 573 Irrigated with Dairy Wastewater. *Sci. Total Environ.* **2018**, *635*, 1477–1483.
 574 <https://doi.org/10.1016/j.scitotenv.2018.04.020>.
- 575 (12) Xu, F.; Liu, Y.; Du, W.; Li, C.; Xu, M.; Xie, T.; Yin, Y.; Guo, H. Response of Soil Bacterial

- 576 Communities, Antibiotic Residuals, and Crop Yields to Organic Fertilizer Substitution in North
577 China under Wheat–Maize Rotation. *Sci. Total Environ.* **2021**, 785, 147248.
578 <https://doi.org/10.1016/j.scitotenv.2021.147248>.
- 579 (13) Tang, X.; Lou, C.; Wang, S.; Lu, Y.; Liu, M.; Hashmi, M. Z.; Liang, X.; Li, Z.; Liao, Y.; Qin,
580 W.; Fan, F.; Xu, J.; Brookes, P. C. Effects of Long-Term Manure Applications on the
581 Occurrence of Antibiotics and Antibiotic Resistance Genes (ARGs) in Paddy Soils: Evidence
582 from Four Field Experiments in South of China. *Soil Biol. Biochem.* **2015**, 90, 179–187.
583 <https://doi.org/10.1016/j.soilbio.2015.07.027>.
- 584 (14) Wang, F.; Xu, M.; Stedtfeld, R. D.; Sheng, H.; Fan, J.; Liu, M.; Chai, B.; Soares De Carvalho,
585 T.; Li, H.; Li, Z.; Hashsham, S. A.; Tiedje, J. M. Long-Term Effect of Different Fertilization
586 and Cropping Systems on the Soil Antibiotic Resistome. *Environ. Sci. Technol.* **2018**, 52 (22),
587 13037–13046. <https://doi.org/10.1021/acs.est.8b04330>.
- 588 (15) Zhu, Y. G.; Johnson, T. A.; Su, J. Q.; Qiao, M.; Guo, G. X.; Stedtfeld, R. D.; Hashsham, S. A.;
589 Tiedje, J. M. Diverse and Abundant Antibiotic Resistance Genes in Chinese Swine Farms.
590 *Proc. Natl. Acad. Sci. U. S. A.* **2013**, 110 (9), 3435–3440.
591 <https://doi.org/10.1073/pnas.1222743110>.
- 592 (16) Udikovic-Kolic, N.; Wichmann, F.; Broderick, N. A.; Handelsman, J. Bloom of Resident
593 Antibiotic-Resistant Bacteria in Soil Following Manure Fertilization. *Proc. Natl. Acad. Sci. U.*
594 *S. A.* **2014**, 111 (42), 15202–15207. <https://doi.org/10.1073/pnas.1409836111>.
- 595 (17) Chen, Q.; An, X.; Li, H.; Su, J.; Ma, Y.; Zhu, Y. G. Long-Term Field Application of Sewage

- 596 Sludge Increases the Abundance of Antibiotic Resistance Genes in Soil. *Environ. Int.* **2016**,
597 92–93, 1–10. <https://doi.org/10.1016/j.envint.2016.03.026>.
- 598 (18) Gao, F. Z.; He, L. Y.; He, L. X.; Zou, H. Y.; Zhang, M.; Wu, D. L.; Liu, Y. S.; Shi, Y. J.; Bai,
599 H.; Ying, G. G. Untreated Swine Wastes Changed Antibiotic Resistance and Microbial
600 Community in the Soils and Impacted Abundances of Antibiotic Resistance Genes in the
601 Vegetables. *Sci. Total Environ.* **2020**, 741, 140482.
602 <https://doi.org/10.1016/j.scitotenv.2020.140482>.
- 603 (19) Cheng, W.; Li, J.; Wu, Y.; Xu, L.; Su, C.; Qian, Y.; Zhu, Y. G.; Chen, H. Behavior of
604 Antibiotics and Antibiotic Resistance Genes in Eco-Agricultural System: A Case Study. *J.*
605 *Hazard. Mater.* **2016**, 304, 18–25. <https://doi.org/10.1016/j.jhazmat.2015.10.037>.
- 606 (20) Pu, C.; Liu, H.; Ding, G.; Sun, Y.; Yu, X.; Chen, J.; Ren, J.; Gong, X. Impact of Direct
607 Application of Biogas Slurry and Residue in Fields: In Situ Analysis of Antibiotic Resistance
608 Genes from Pig Manure to Fields. *J. Hazard. Mater.* **2018**, 344, 441–449.
609 <https://doi.org/10.1016/j.jhazmat.2017.10.031>.
- 610 (21) Cerqueira, F.; Christou, A.; Fatta-Kassinos, D.; Vila-Costa, M.; Bayona, J. M.; Piña, B. Effects
611 of Prescription Antibiotics on Soil- and Root-Associated Microbiomes and Resistomes in an
612 Agricultural Context. *J. Hazard. Mater.* **2020**, 400, 123208.
613 <https://doi.org/10.1016/j.jhazmat.2020.123208>.
- 614 (22) Macedo, G.; Hernandez-Leal, L.; van der Maas, P.; Heederik, D.; Mevius, D.; Schmitt, H. The
615 Impact of Manure and Soil Texture on Antimicrobial Resistance Gene Levels in Farmlands and

- 616 Adjacent Ditches. *Sci. Total Environ.* **2020**, *737*, 139563.
- 617 <https://doi.org/10.1016/j.scitotenv.2020.139563>.
- 618 (23) Park, J. Y.; Huwe, B. Effect of PH and Soil Structure on Transport of Sulfonamide Antibiotics
619 in Agricultural Soils. *Environ. Pollut.* **2016**, *213*, 561–570.
- 620 <https://doi.org/10.1016/j.envpol.2016.01.089>.
- 621 (24) Lin, H.; Sun, W.; Yu, Q.; Ma, J. Acidic Conditions Enhance the Removal of Sulfonamide
622 Antibiotics and Antibiotic Resistance Determinants in Swine Manure. *Environ. Pollut.* **2020**,
623 *263*, 114439. <https://doi.org/10.1016/j.envpol.2020.114439>.
- 624 (25) Sui, Q.; Zhang, J.; Chen, M.; Wang, R.; Wang, Y.; Wei, Y. Fate of Microbial Pollutants and
625 Evolution of Antibiotic Resistance in Three Types of Soil Amended with Swine Slurry.
626 *Environ. Pollut.* **2019**, *245*, 353–362. <https://doi.org/10.1016/j.envpol.2018.11.003>.
- 627 (26) Conde-Cid, M.; Ferreira-Coelho, G.; Fernández-Calviño, D.; Núñez-Delgado, A.; Fernández-
628 Sanjurjo, M. J.; Arias-Estévez, M.; Álvarez-Rodríguez, E. Single and Simultaneous Adsorption
629 of Three Sulfonamides in Agricultural Soils: Effects of PH and Organic Matter Content. *Sci.*
630 *Total Environ.* **2020**, *744*, 140872. <https://doi.org/10.1016/j.scitotenv.2020.140872>.
- 631 (27) Conde-Cid, M.; Ferreira-Coelho, G.; Núñez-Delgado, A.; Fernández-Calviño, D.; Arias-
632 Estévez, M.; Álvarez-Rodríguez, E.; Fernández-Sanjurjo, M. J. Competitive Adsorption of
633 Tetracycline, Oxytetracycline and Chlortetracycline on Soils with Different PH Value and
634 Organic Matter Content. *Environ. Res.* **2019**, *178*, 108669.
635 <https://doi.org/10.1016/j.envres.2019.108669>.

- 636 (28) Awasthi, M. K.; Chen, H.; Liu, T.; Awasthi, S. K.; Wang, Q.; Ren, X.; Duan, Y.; Zhang, Z.
637 Respond of Clay Amendment in Chicken Manure Composts to Understand the Antibiotic
638 Resistant Bacterial Diversity and Its Correlation with Physicochemical Parameters. *J. Clean.*
639 *Prod.* **2019**, 236, 117715. <https://doi.org/10.1016/j.jclepro.2019.117715>.
- 640 (29) Sun, J.; Zeng, Q.; Tsang, D. C. W.; Zhu, L. Z.; Li, X. D. Antibiotics in the Agricultural Soils
641 from the Yangtze River Delta, China. *Chemosphere* **2017**, 189, 301–308.
642 <https://doi.org/10.1016/j.chemosphere.2017.09.040>.
- 643 (30) Cheng, D.; Feng, Y.; Liu, Y.; Li, J.; Xue, J.; Li, Z. Quantitative Models for Predicting
644 Adsorption of Oxytetracycline, Ciprofloxacin and Sulfamerazine to Swine Manures with
645 Contrasting Properties. *Sci. Total Environ.* **2018**, 634, 1148–1156.
646 <https://doi.org/10.1016/j.scitotenv.2018.04.114>.
- 647 (31) Baker-Austin, C.; Wright, M. S.; Stepanauskas, R.; McArthur, J. V. Co-Selection of Antibiotic
648 and Metal Resistance. *Trends Microbiol.* **2006**, 14 (4), 176–182.
649 <https://doi.org/10.1016/j.tim.2006.02.006>.
- 650 (32) Li, L. G.; Xia, Y.; Zhang, T. Co-Occurrence of Antibiotic and Metal Resistance Genes
651 Revealed in Complete Genome Collection. *ISME J.* **2017**, 11 (3), 651–662.
652 <https://doi.org/10.1038/ismej.2016.155>.
- 653 (33) Lu, J.; Wang, Y.; Jin, M.; Yuan, Z.; Bond, P.; Guo, J. Both Silver Ions and Silver
654 Nanoparticles Facilitate the Horizontal Transfer of Plasmid-Mediated Antibiotic Resistance
655 Genes. *Water Res.* **2020**, 169, 115229. <https://doi.org/10.1016/j.watres.2019.115229>.

- 656 (34) Ashbolt, N. J.; Amézquita, A.; Backhaus, T.; Borriello, P.; Brandt, K. K.; Collignon, P.; Coors,
657 A.; Finley, R.; Gaze, W. H.; Heberer, T.; Lawrence, J. R.; Larsson, D. G. J.; McEwen, S. A.;
658 Ryan, J. J.; Schönfeld, J.; Silley, P.; Snape, J. R.; Van den Eede, C.; Topp, E. Human Health
659 Risk Assessment (HHRA) for Environmental Development and Transfer of Antibiotic
660 Resistance. *Environ. Health Perspect.* **2013**, *121* (9), 993–1001.
661 <https://doi.org/10.1289/ehp.1206316>.
- 662 (35) Gillings, M. R.; Gaze, W. H.; Pruden, A.; Smalla, K.; Tiedje, J. M.; Zhu, Y. G. Using the Class
663 1 Integron-Integrase Gene as a Proxy for Anthropogenic Pollution. *ISME J.* **2015**, *9* (6), 1269–
664 1279. <https://doi.org/10.1038/ismej.2014.226>.
- 665 (36) Zhang, H.; Zhang, Q.; Song, J.; Zhang, Z.; Chen, S.; Long, Z.; Wang, M.; Yu, Y.; Fang, H.
666 Tracking Resistomes, Virulence Genes, and Bacterial Pathogens in Long-Term Manure-
667 Amended Greenhouse Soils. *J. Hazard. Mater.* **2020**, *396*, 122618.
668 <https://doi.org/10.1016/j.jhazmat.2020.122618>.
- 669 (37) Peng, S.; Feng, Y.; Wang, Y.; Guo, X.; Chu, H.; Lin, X. Prevalence of Antibiotic Resistance
670 Genes in Soils after Continually Applied with Different Manure for 30 Years. *J. Hazard.*
671 *Mater.* **2017**, *340*, 16–25. <https://doi.org/10.1016/j.jhazmat.2017.06.059>.
- 672 (38) Zhang, Y.; Gu, A. Z.; Cen, T.; Li, X.; He, M.; Li, D.; Chen, J. Sub-Inhibitory Concentrations of
673 Heavy Metals Facilitate the Horizontal Transfer of Plasmid-Mediated Antibiotic Resistance
674 Genes in Water Environment. *Environ. Pollut.* **2018**, *237*, 74–82.
675 <https://doi.org/10.1016/j.envpol.2018.01.032>.

- 676 (39) Zheng, D.; Yin, G.; Liu, M.; Chen, C.; Jiang, Y.; Hou, L.; Zheng, Y. A Systematic Review of
677 Antibiotics and Antibiotic Resistance Genes in Estuarine and Coastal Environments. *Sci. Total*
678 *Environ.* **2021**, 777, 146009. <https://doi.org/10.1016/j.scitotenv.2021.146009>.
- 679 (40) Yang, Y.; Song, W.; Lin, H.; Wang, W.; Du, L.; Xing, W. Antibiotics and Antibiotic
680 Resistance Genes in Global Lakes: A Review and Meta-Analysis. *Environ. Int.* **2018**, 116, 60–
681 73. <https://doi.org/10.1016/j.envint.2018.04.011>.
- 682 (41) Huang, L.; Mo, Y.; Wu, Z.; Rad, S.; Song, X.; Zeng, H.; Bashir, S.; Kang, B.; Chen, Z.
683 Occurrence, Distribution, and Health Risk Assessment of Quinolone Antibiotics in Water,
684 Sediment, and Fish Species of Qingshitan Reservoir, South China. *Sci. Rep.* **2020**, 10 (1),
685 15777. <https://doi.org/10.1038/s41598-020-72324-9>.
- 686 (42) Tang, J.; Sun, J.; Wang, W.; Yang, L.; Xu, Y. Pharmaceuticals in Two Watersheds in Eastern
687 China and Their Ecological Risks. *Environ. Pollut.* **2021**, 277, 116773.
688 <https://doi.org/10.1016/j.envpol.2021.116773>.
- 689 (43) Han, Q. F.; Zhao, S.; Zhang, X. R.; Wang, X. L.; Song, C.; Wang, S. G. Distribution,
690 Combined Pollution and Risk Assessment of Antibiotics in Typical Marine Aquaculture Farms
691 Surrounding the Yellow Sea, North China. *Environ. Int.* **2020**, 138, 105551.
692 <https://doi.org/10.1016/j.envint.2020.105551>.
- 693 (44) Cycoń, M.; Mroziński, A.; Piotrowska-Seget, Z. Antibiotics in the Soil Environment-Degradation
694 and Their Impact on Microbial Activity and Diversity. *Front. Microbiol.* **2019**, 10.
695 <https://doi.org/10.3389/fmicb.2019.00338>.

- 696 (45) Lin, H.; Sun, W.; Zhang, Z.; Chapman, S. J.; Freitag, T. E.; Fu, J.; Zhang, X.; Ma, J. Effects of
697 Manure and Mineral Fertilization Strategies on Soil Antibiotic Resistance Gene Levels and
698 Microbial Community in a Paddy-Upland Rotation System. *Environ. Pollut.* **2016**, *211*, 332–
699 337. <https://doi.org/10.1016/j.envpol.2016.01.007>.
- 700 (46) Zhou, S. Y. D.; Zhu, D.; Giles, M.; Yang, X. R.; Daniell, T.; Neilson, R.; Zhu, Y. G.
701 Phyllosphere of Staple Crops under Pig Manure Fertilization, a Reservoir of Antibiotic
702 Resistance Genes. *Environ. Pollut.* **2019**, *252*, 227–235.
703 <https://doi.org/10.1016/j.envpol.2019.05.098>.
- 704 (47) Wang, J.; Lin, H.; Sun, W.; Xia, Y.; Ma, J.; Fu, J.; Zhang, Z.; Wu, H.; Qian, M. Variations in
705 the Fate and Biological Effects of Sulfamethoxazole, Norfloxacin and Doxycycline in Different
706 Vegetable-Soil Systems Following Manure Application. *J. Hazard. Mater.* **2016**, *304*, 49–57.
707 <https://doi.org/10.1016/j.jhazmat.2015.10.038>.
- 708 (48) Ahmed, M. B. M.; Rajapaksha, A. U.; Lim, J. E.; Vu, N. T.; Kim, I. S.; Kang, H. M.; Lee, S.
709 S.; Ok, Y. S. Distribution and Accumulative Pattern of Tetracyclines and Sulfonamides in
710 Edible Vegetables of Cucumber, Tomato, and Lettuce. *J. Agric. Food Chem.* **2015**, *63* (2),
711 398–405. <https://doi.org/10.1021/jf5034637>.
- 712 (49) Guo, Y.; Qiu, T.; Gao, M.; Sun, Y.; Cheng, S.; Gao, H.; Wang, X. Diversity and Abundance of
713 Antibiotic Resistance Genes in Rhizosphere Soil and Endophytes of Leafy Vegetables:
714 Focusing on the Effect of the Vegetable Species. *J. Hazard. Mater.* **2021**, *415*, 125595.
715 <https://doi.org/10.1016/j.jhazmat.2021.125595>.

- 716 (50) Cerqueira, F.; Matamoros, V.; Bayona, J. M.; Berendonk, T. U.; Elsinga, G.; Hornstra, L. M.;
717 Piña, B. Antibiotic Resistance Gene Distribution in Agricultural Fields and Crops. A Soil-to-
718 Food Analysis. *Environ. Res.* **2019**, *177*, 18–26. <https://doi.org/10.1016/j.envres.2019.108608>.
- 719 (51) Li, X. W.; Xie, Y. F.; Li, C. L.; Zhao, H. N.; Zhao, H.; Wang, N.; Wang, J. F. Investigation of
720 Residual Fluoroquinolones in a Soil-Vegetable System in an Intensive Vegetable Cultivation
721 Area in Northern China. *Sci. Total Environ.* **2014**, *468–469*, 258–264.
722 <https://doi.org/10.1016/j.scitotenv.2013.08.057>.
- 723 (52) Li, X.; Yu, H.; Xu, S.; Hua, R. Uptake of Three Sulfonamides from Contaminated Soil by
724 Pakchoi Cabbage. *Ecotoxicol. Environ. Saf.* **2013**, *92*, 297–302.
725 <https://doi.org/10.1016/j.ecoenv.2013.03.010>.
- 726 (53) Heuer, H.; Schmitt, H.; Smalla, K. Antibiotic Resistance Gene Spread Due to Manure
727 Application on Agricultural Fields. *Curr. Opin. Microbiol.* **2011**, *14* (3), 236–243.
728 <https://doi.org/10.1016/j.mib.2011.04.009>.
- 729 (54) Duan, M.; Gu, J.; Wang, X.; Li, Y.; Zhang, R.; Hu, T.; Zhou, B. Factors That Affect the
730 Occurrence and Distribution of Antibiotic Resistance Genes in Soils from Livestock and
731 Poultry Farms. *Ecotoxicol. Environ. Saf.* **2019**, *180*, 114–122.
732 <https://doi.org/10.1016/j.ecoenv.2019.05.005>.
- 733 (55) Zhi, S.; Shen, S.; Zhou, J.; Ding, G.; Zhang, K. Systematic Analysis of Occurrence, Density
734 and Ecological Risks of 45 Veterinary Antibiotics: Focused on Family Livestock Farms in
735 Erhai Lake Basin, Yunnan, China. *Environ. Pollut.* **2020**, *267*, 115539.

736 <https://doi.org/10.1016/j.envpol.2020.115539>.

737 (56) He, L. Y.; He, L. K.; Liu, Y. S.; Zhang, M.; Zhao, J. L.; Zhang, Q. Q.; Ying, G. G. Microbial
738 Diversity and Antibiotic Resistome in Swine Farm Environments. *Sci. Total Environ.* **2019**,
739 685, 197–207. <https://doi.org/10.1016/j.scitotenv.2019.05.369>.

740 (57) Gurmessa, B.; Ashworth, A. J.; Yang, Y.; Savin, M.; Moore, P. A.; Ricke, S. C.; Corti, G.;
741 Pedretti, E. F.; Cocco, S. Variations in Bacterial Community Structure and Antimicrobial
742 Resistance Gene Abundance in Cattle Manure and Poultry Litter. *Environ. Res.* **2021**, 197,
743 111011. <https://doi.org/10.1016/j.envres.2021.111011>.

744 (58) Zealand, A. M.; Mei, R.; Papachristodoulou, P.; Roskilly, A. P.; Liu, W. T.; Graham, D. W.
745 Microbial Community Composition and Diversity in Rice Straw Digestion Bioreactors with
746 and without Dairy Manure. *Appl. Microbiol. Biotechnol.* **2018**, 102 (19), 8599–8612.
747 <https://doi.org/10.1007/s00253-018-9243-7>.

748 (59) Urrea, J.; Alkorta, I.; Mijangos, I.; Epelde, L.; Garbisu, C. Application of Sewage Sludge to
749 Agricultural Soil Increases the Abundance of Antibiotic Resistance Genes without Altering the
750 Composition of Prokaryotic Communities. *Sci. Total Environ.* **2019**, 647, 1410–1420.
751 <https://doi.org/10.1016/j.scitotenv.2018.08.092>.

752 (60) Chen, C.; Li, J.; Chen, P.; Ding, R.; Zhang, P.; Li, X. Occurrence of Antibiotics and Antibiotic
753 Resistances in Soils from Wastewater Irrigation Areas in Beijing and Tianjin, China. *Environ.*
754 *Pollut.* **2014**, 193, 94–101. <https://doi.org/10.1016/j.envpol.2014.06.005>.

- 755 (61) Cerqueira, F.; Matamoros, V.; Bayona, J.; Elsinga, G.; Hornstra, L. M.; Piña, B. Distribution of
 756 Antibiotic Resistance Genes in Soils and Crops. A Field Study in Legume Plants (*Vicia Faba*
 757 L.) Grown under Different Watering Regimes. *Environ. Res.* **2019**, *170*, 16–25.
 758 <https://doi.org/10.1016/j.envres.2018.12.007>.
- 759 (62) Rutgersson, C.; Ebmeyer, S.; Lassen, S. B.; Karkman, A.; Fick, J.; Kristiansson, E.; Brandt, K.
 760 K.; Flach, C. F.; Larsson, D. G. J. Long-Term Application of Swedish Sewage Sludge on
 761 Farmland Does Not Cause Clear Changes in the Soil Bacterial Resistome. *Environ. Int.* **2020**,
 762 *137*, 105339. <https://doi.org/10.1016/j.envint.2019.105339>.
- 763 (63) Hu, S.; Zhang, Y.; Shen, G.; Zhang, H.; Yuan, Z.; Zhang, W. Adsorption/Desorption Behavior
 764 and Mechanisms of Sulfadiazine and Sulfamethoxazole in Agricultural Soil Systems. *Soil*
 765 *Tillage Res.* **2019**, *186*, 233–241. <https://doi.org/10.1016/j.still.2018.10.026>.
- 766 (64) Chen, Y.; Hu, C.; Deng, D.; Li, Y.; Luo, L. Factors Affecting Sorption Behaviors of
 767 Tetracycline to Soils: Importance of Soil Organic Carbon, PH and Cd Contamination.
 768 *Ecotoxicol. Environ. Saf.* **2020**, *197*, 110572. <https://doi.org/10.1016/j.ecoenv.2020.110572>.
- 769 (65) Srinivasan, P.; Sarmah, A. K.; Manley-Harris, M. Co-Contaminants and Factors Affecting the
 770 Sorption Behaviour of Two Sulfonamides in Pasture Soils. *Environ. Pollut.* **2013**, *180*, 165–
 771 172. <https://doi.org/10.1016/j.envpol.2013.05.022>.
- 772 (66) Mitchell, S. M.; Ullman, J. L.; Teel, A. L.; Watts, R. J. PH and Temperature Effects on the
 773 Hydrolysis of Three β -Lactam Antibiotics: Ampicillin, Cefalotin and Cefoxitin. *Sci. Total*
 774 *Environ.* **2014**, *466–467*, 547–555. <https://doi.org/10.1016/j.scitotenv.2013.06.027>.

- 775 (67) Zhang, Y.; Cheng, D.; Zhang, Y.; Xie, J.; Xiong, H.; Wan, Y.; Zhang, Y.; Chen, X.; Shi, X.
776 Soil Type Shapes the Antibiotic Resistome Profiles of Long-Term Manured Soil. *Sci. Total*
777 *Environ.* **2021**, *786*, 147361. <https://doi.org/10.1016/j.scitotenv.2021.147361>.
- 778 (68) Kodešová, R.; Grabic, R.; Kočárek, M.; Klement, A.; Golovko, O.; Fér, M.; Nikodem, A.;
779 Jakšík, O. Pharmaceuticals' Sorptions Relative to Properties of Thirteen Different Soils. *Sci.*
780 *Total Environ.* **2015**, *511*, 435–443. <https://doi.org/10.1016/j.scitotenv.2014.12.088>.
- 781 (69) Wang, L.; Wang, J.; Wang, J.; Zhu, L.; Conkle, J. L.; Yang, R. Soil Types Influence the
782 Characteristic of Antibiotic Resistance Genes in Greenhouse Soil with Long-Term Manure
783 Application. *J. Hazard. Mater.* **2020**, *392*, 122334.
784 <https://doi.org/10.1016/j.jhazmat.2020.122334>.
- 785 (70) Sun, Y.; Qiu, T.; Gao, M.; Shi, M.; Zhang, H.; Wang, X. Inorganic and Organic Fertilizers
786 Application Enhanced Antibiotic Resistome in Greenhouse Soils Growing Vegetables.
787 *Ecotoxicol. Environ. Saf.* **2019**, *179*, 24–30. <https://doi.org/10.1016/j.ecoenv.2019.04.039>.
- 788 (71) Li, S.; Shi, W.; Li, H.; Xu, N.; Zhang, R.; Chen, X.; Sun, W.; Wen, D.; He, S.; Pan, J.; He, Z.;
789 Fan, Y. Antibiotics in Water and Sediments of Rivers and Coastal Area of Zhuhai City, Pearl
790 River Estuary, South China. *Sci. Total Environ.* **2018**, *636*, 1009–1019.
791 <https://doi.org/10.1016/j.scitotenv.2018.04.358>.
- 792 (72) Conde-Cid, M.; Fernández-Calviño, D.; Núñez-Delgado, A.; Fernández-Sanjurjo, M. J.; Arias-
793 Estévez, M.; Álvarez-Rodríguez, E. Estimation of Adsorption/Desorption Freundlich's Affinity
794 Coefficients for Oxytetracycline and Chlortetracycline from Soil Properties: Experimental Data

- 795 and Pedotransfer Functions. *Ecotoxicol. Environ. Saf.* **2020**, *196*, 110584.
- 796 <https://doi.org/10.1016/j.ecoenv.2020.110584>.
- 797 (73) Wang, B.; Li, M.; Zhang, H.; Zhu, J.; Chen, S.; Ren, D. Effect of Straw-Derived Dissolved
 798 Organic Matter on the Adsorption of Sulfamethoxazole to Purple Paddy Soils. *Ecotoxicol.*
 799 *Environ. Saf.* **2020**, *203*, 110990. <https://doi.org/10.1016/j.ecoenv.2020.110990>.
- 800 (74) Alvarez-Esmoris, C.; Conde-Cid, M.; Fernández-Sanjurjo, M. J.; Núñez-Delgado, A.; Álvarez-
 801 Rodríguez, E.; Arias-Estévez, M. Environmental Relevance of Adsorption of Doxycycline,
 802 Enrofloxacin, and Sulfamethoxypyridazine before and after the Removal of Organic Matter
 803 from Soils. *J. Environ. Manage.* **2021**, *287*, 112354.
 804 <https://doi.org/10.1016/j.jenvman.2021.112354>.
- 805 (75) Álvarez-Esmoris, C.; Conde-Cid, M.; Ferreira-Coelho, G.; Fernández-Sanjurjo, M. J.; Núñez-
 806 Delgado, A.; Álvarez-Rodríguez, E.; Arias-Estévez, M. Adsorption/Desorption of
 807 Sulfamethoxypyridazine and Enrofloxacin in Agricultural Soils. *Sci. Total Environ.* **2020**, *706*,
 808 136015. <https://doi.org/10.1016/j.scitotenv.2019.136015>.
- 809 (76) Zhang, Y.; Wang, J.; Lu, J.; Wu, J. Antibiotic Resistance Genes Might Serve as New Indicators
 810 for Wastewater Contamination of Coastal Waters: Spatial Distribution and Source
 811 Apportionment of Antibiotic Resistance Genes in a Coastal Bay. *Ecol. Indic.* **2020**, *114*,
 812 106299. <https://doi.org/10.1016/j.ecolind.2020.106299>.
- 813 (77) Shen, G.; Zhang, Y.; Hu, S.; Zhang, H.; Yuan, Z.; Zhang, W. Adsorption and Degradation of
 814 Sulfadiazine and Sulfamethoxazole in an Agricultural Soil System under an Anaerobic

- 815 Condition: Kinetics and Environmental Risks. *Chemosphere* **2018**, *194*, 266–274.
- 816 <https://doi.org/10.1016/j.chemosphere.2017.11.175>.
- 817 (78) Qian, X. Y.; Shen, G. X.; Wang, Z. Q.; Chen, X. H.; Zhao, Q. J.; Bai, Y. J.; Tang, Z. Z.
- 818 Application of Dairy Manure as Fertilizer in Dry Land in East China: Field Monitoring and
- 819 Model Estimation of Heavy Metal Accumulation in Surface Soil. *Environ. Sci. Pollut. Res.*
- 820 **2020**, *27* (29), 36913–36919. <https://doi.org/10.1007/s11356-020-09786-x>.
- 821 (79) Arya, S.; Williams, A.; Reina, S. V.; Knapp, C. W.; Kreft, J. U.; Hobman, J. L.; Stekel, D. J.
- 822 Towards a General Model for Predicting Minimal Metal Concentrations Co-Selecting for
- 823 Antibiotic Resistance Plasmids. *Environ. Pollut.* **2021**, *275*, 116602.
- 824 <https://doi.org/10.1016/j.envpol.2021.116602>.
- 825 (80) Sherpa, M. T.; Najar, I. N.; Das, S.; Thakur, N. Distribution of Antibiotic and Metal Resistance
- 826 Genes in Two Glaciers of North Sikkim, India. *Ecotoxicol. Environ. Saf.* **2020**, *203*, 111037.
- 827 <https://doi.org/10.1016/j.ecoenv.2020.111037>.
- 828 (81) Salam, L. B. Unravelling the Antibiotic and Heavy Metal Resistome of a Chronically Polluted
- 829 Soil. *3 Biotech* **2020**, *10* (6), 238. <https://doi.org/10.1007/s13205-020-02219-z>.
- 830 (82) Wang, X.; Lan, B.; Fei, H.; Wang, S.; Zhu, G. Heavy Metal Could Drive Co-Selection of
- 831 Antibiotic Resistance in Terrestrial Subsurface Soils. *J. Hazard. Mater.* **2021**, *411*, 124848.
- 832 <https://doi.org/10.1016/j.jhazmat.2020.124848>.
- 833 (83) Mazhar, S. H.; Li, X.; Rashid, A.; Su, J. M.; Xu, J.; Brejnrod, A. D.; Su, J. Q.; Wu, Y.; Zhu, Y.

834 G.; Zhou, S. G.; Feng, R.; Rensing, C. Co-Selection of Antibiotic Resistance Genes, and
 835 Mobile Genetic Elements in the Presence of Heavy Metals in Poultry Farm Environments. *Sci.*
 836 *Total Environ.* **2021**, 755, 142702. <https://doi.org/10.1016/j.scitotenv.2020.142702>.

837 (84) Zhao, X.; Shen, J. P.; Zhang, L. M.; Du, S.; Hu, H. W.; He, J. Z. Arsenic and Cadmium as
 838 Predominant Factors Shaping the Distribution Patterns of Antibiotic Resistance Genes in
 839 Polluted Paddy Soils. *J. Hazard. Mater.* **2020**, 389, 121838.
 840 <https://doi.org/10.1016/j.jhazmat.2019.121838>.

841 (85) Wei, H.; Ding, S.; Qiao, Z.; Su, Y.; Xie, B. Insights into Factors Driving the Transmission of
 842 Antibiotic Resistance from Sludge Compost-Amended Soil to Vegetables under Cadmium
 843 Stress. *Sci. Total Environ.* **2020**, 729, 138990. <https://doi.org/10.1016/j.scitotenv.2020.138990>.

844 (86) Cao, J.; Yang, G.; Mai, Q.; Zhuang, Z.; Zhuang, L. Co-Selection of Antibiotic-Resistant
 845 Bacteria in a Paddy Soil Exposed to As (III) Contamination with an Emphasis on Potential
 846 Pathogens. *Sci. Total Environ.* **2020**, 725, 138367.
 847 <https://doi.org/10.1016/j.scitotenv.2020.138367>.

848 (87) Li, X.; Gu, A. Z.; Zhang, Y.; Xie, B.; Li, D.; Chen, J. Sub-Lethal Concentrations of Heavy
 849 Metals Induce Antibiotic Resistance via Mutagenesis. *J. Hazard. Mater.* **2019**, 369, 9–16.
 850 <https://doi.org/10.1016/j.jhazmat.2019.02.006>.

851 (88) Mahbub, K. R.; King, W. L.; Siboni, N.; Nguyen, V. K.; Rahman, M. M.; Megharaj, M.;
 852 Seymour, J. R.; Franks, A. E.; Labbate, M. Long-Lasting Effect of Mercury Contamination on
 853 the Soil Microbiota and Its Co-Selection of Antibiotic Resistance. *Environ. Pollut.* **2020**, 265,

- 854 115057. <https://doi.org/10.1016/j.envpol.2020.115057>.
- 855 (89) Poole, K. At the Nexus of Antibiotics and Metals: The Impact of Cu and Zn on Antibiotic
856 Activity and Resistance. *Trends Microbiol.* **2017**, 25 (10), 820–832.
857 <https://doi.org/10.1016/j.tim.2017.04.010>.
- 858 (90) Ma, J.; Xiong, Y.; Dai, X.; Yu, F. Coadsorption Behavior and Mechanism of Ciprofloxacin and
859 Cu(II) on Graphene Hydrogel Wetted Surface. *Chem. Eng. J.* **2020**, 380, 122387.
860 <https://doi.org/10.1016/j.cej.2019.122387>.
- 861 (91) Zhang, Z.; Sun, K.; Gao, B.; Zhang, G.; Liu, X.; Zhao, Y. Adsorption of Tetracycline on Soil
862 and Sediment: Effects of PH and the Presence of Cu(II). *J. Hazard. Mater.* **2011**, 190, 856–
863 862. <https://doi.org/10.1016/j.jhazmat.2011.04.017>.
- 864 (92) Cheng, S.; Shi, M.; Xing, L.; Wang, X.; Gao, H.; Sun, Y. Sulfamethoxazole Affects the
865 Microbial Composition and Antibiotic Resistance Gene Abundance in Soil and Accumulates in
866 Lettuce. *Environ. Sci. Pollut. Res.* **2020**, 27 (23), 29257–29265.
867 <https://doi.org/10.1007/s11356-020-08902-1>.
- 868 (93) Wei, R.; He, T.; Zhang, S.; Zhu, L.; Shang, B.; Li, Z.; Wang, R. Occurrence of Seventeen
869 Veterinary Antibiotics and Resistant Bacterias in Manure-Fertilized Vegetable Farm Soil in
870 Four Provinces of China. *Chemosphere* **2019**, 215, 234–240.
871 <https://doi.org/10.1016/j.chemosphere.2018.09.152>.
- 872 (94) Yue, Z.; Zhang, J.; Zhou, Z.; Ding, C.; Wan, L.; Liu, J.; Chen, L.; Wang, X. Pollution

- 873 Characteristics of Livestock Faeces and the Key Driver of the Spread of Antibiotic Resistance
874 Genes. *J. Hazard. Mater.* **2021**, *409*, 124957. <https://doi.org/10.1016/j.jhazmat.2020.124957>.
- 875 (95) Seyoum, M. M.; Obayomi, O.; Bernstein, N.; Williams, C. F.; Gillor, O. Occurrence and
876 Distribution of Antibiotics and Corresponding Antibiotic Resistance Genes in Different Soil
877 Types Irrigated with Treated Wastewater. *Sci. Total Environ.* **2021**, *782*, 146835.
878 <https://doi.org/10.1016/j.scitotenv.2021.146835>.
- 879 (96) Blair, J. M. A.; Webber, M. A.; Baylay, A. J.; Ogbolu, D. O.; Piddock, L. J. V. Molecular
880 Mechanisms of Antibiotic Resistance. *Nat. Rev. Microbiol.* **2015**, *13*, 42–51.
881 <https://doi.org/10.1038/nrmicro3380>.
- 882 (97) Liu, C.; Li, X.; Zheng, S.; Kai, Z.; Jin, T.; Shi, R.; Huang, H.; Zheng, X. Effects of Wastewater
883 Treatment and Manure Application on the Dissemination of Antimicrobial Resistance around
884 Swine Feedlots. *J. Clean. Prod.* **2021**, *280*, 123794.
885 <https://doi.org/10.1016/j.jclepro.2020.123794>.
- 886 (98) Buta, M.; Korzeniewska, E.; Harnisz, M.; Hubeny, J.; Zieliński, W.; Rolbiecki, D.; Bajkacz, S.;
887 Felis, E.; Kokoszka, K. Microbial and Chemical Pollutants on the Manure-Crops Pathway in
888 the Perspective of “One Health” Holistic Approach. *Sci. Total Environ.* **2021**, *785*.
889 <https://doi.org/10.1016/j.scitotenv.2021.147411>.
- 890 (99) Wang, L.; Wang, J.; Wang, J.; Zhu, L.; Yang, L.; Yang, R. Distribution Characteristics of
891 Antibiotic Resistant Bacteria and Genes in Fresh and Composted Manures of Livestock Farms.
892 *Sci. Total Environ.* **2019**, *695*, 133781. <https://doi.org/10.1016/j.scitotenv.2019.133781>.

- 893 (100) Guo, X. pan; Liu, X.; Niu, Z. shun; Lu, D. pei; Zhao, S.; Sun, X. li; Wu, J. yuan; Chen, Y. ru;
894 Tou, F. yun; Hou, L.; Liu, M.; Yang, Y. Seasonal and Spatial Distribution of Antibiotic
895 Resistance Genes in the Sediments along the Yangtze Estuary, China. *Environ. Pollut.* **2018**,
896 242, 576–584. <https://doi.org/10.1016/j.envpol.2018.06.099>.
- 897 (101) Grenni, P.; Ancona, V.; Barra Caracciolo, A. Ecological Effects of Antibiotics on Natural
898 Ecosystems: A Review. *Microchem. J.* **2018**, 136, 25–39.
899 <https://doi.org/10.1016/j.microc.2017.02.006>.
- 900 (102) DeVries, S. L.; Zhang, P. Antibiotics and the Terrestrial Nitrogen Cycle: A Review. *Curr.*
901 *Pollut. Reports* **2016**, 2 (1), 51–67. <https://doi.org/10.1007/s40726-016-0027-3>.
- 902 (103) Aminov, R. I. A Brief History of the Antibiotic Era: Lessons Learned and Challenges for the
903 Future. *Front. Microbiol.* **2010**, 1, 1–7. <https://doi.org/10.3389/fmicb.2010.00134>.
- 904 (104) Ben, Y.; Fu, C.; Hu, M.; Liu, L.; Wong, M. H.; Zheng, C. Human Health Risk Assessment of
905 Antibiotic Resistance Associated with Antibiotic Residues in the Environment: A Review.
906 *Environ. Res.* **2019**, 169, 483–493. <https://doi.org/10.1016/j.envres.2018.11.040>.
- 907 (105) Alekshun, M. N.; Levy, S. B. Molecular Mechanisms of Antibacterial Multidrug Resistance.
908 *Cell* **2007**, 128 (6), 1037–1050. <https://doi.org/10.1016/j.cell.2007.03.004>.
- 909 (106) Pan, M.; Chu, L. M. Fate of Antibiotics in Soil and Their Uptake by Edible Crops. *Sci. Total*
910 *Environ.* **2017**, 599–600, 500–512. <https://doi.org/10.1016/j.scitotenv.2017.04.214>.
- 911 (107) Hu, X.; Zhou, Q.; Luo, Y. Occurrence and Source Analysis of Typical Veterinary Antibiotics

912 in Manure, Soil, Vegetables and Groundwater from Organic Vegetable Bases, Northern China.
 913 *Environ. Pollut.* **2010**, 158 (9), 2992–2998. <https://doi.org/10.1016/j.envpol.2010.05.023>.

914 (108) Zhu, B.; Chen, Q.; Chen, S.; Zhu, Y. G. Does Organically Produced Lettuce Harbor Higher
 915 Abundance of Antibiotic Resistance Genes than Conventionally Produced? *Environ. Int.* **2017**,
 916 98, 152–159. <https://doi.org/10.1016/j.envint.2016.11.001>.

917 (109) Wu, X. L.; Xiang, L.; Yan, Q. Y.; Jiang, Y. N.; Li, Y. W.; Huang, X. P.; Li, H.; Cai, Q. Y.; Mo,
 918 C. H. Distribution and Risk Assessment of Quinolone Antibiotics in the Soils from Organic
 919 Vegetable Farms of a Subtropical City, Southern China. *Sci. Total Environ.* **2014**, 487 (1),
 920 399–406. <https://doi.org/10.1016/j.scitotenv.2014.04.015>.

921 (110) Li, C.; Chen, J.; Wang, J.; Ma, Z.; Han, P.; Luan, Y.; Lu, A. Occurrence of Antibiotics in Soils
 922 and Manures from Greenhouse Vegetable Production Bases of Beijing, China and an
 923 Associated Risk Assessment. *Sci. Total Environ.* **2015**, 521–522, 101–107.
 924 <https://doi.org/10.1016/j.scitotenv.2015.03.070>.

925 (111) Ji, X.; Shen, Q.; Liu, F.; Ma, J.; Xu, G.; Wang, Y.; Wu, M. Antibiotic Resistance Gene
 926 Abundances Associated with Antibiotics and Heavy Metals in Animal Manures and
 927 Agricultural Soils Adjacent to Feedlots in Shanghai, China. *J. Hazard. Mater.* **2012**, 235–236,
 928 178–185. <https://doi.org/10.1016/j.jhazmat.2012.07.040>.

929 (112) Zhuang, M.; Achmon, Y.; Cao, Y.; Liang, X.; Chen, L.; Wang, H.; Siame, B. A.; Leung, K. Y.
 930 Distribution of Antibiotic Resistance Genes in the Environment. *Environ. Pollut.* **2021**, 285,
 931 117402. <https://doi.org/10.1016/j.envpol.2021.117402>.

- 932 (113) Wang, J.; Wang, L.; Zhu, L.; Wang, J.; Xing, B. Antibiotic Resistance in Agricultural Soils:
933 Source, Fate, Mechanism and Attenuation Strategy. *Crit. Rev. Environ. Sci. Technol.* **2020**, *4*,
934 1–43. <https://doi.org/10.1080/10643389.2020.1835438>.
- 935 (114) Pu, C.; Yu, Y.; Diao, J.; Gong, X.; Li, J.; Sun, Y. Exploring the Persistence and Spreading of
936 Antibiotic Resistance from Manure to Biocompost, Soils and Vegetables. *Sci. Total Environ.*
937 **2019**, *688*, 262–269. <https://doi.org/10.1016/j.scitotenv.2019.06.081>.
- 938 (115) Qiu, X.; Zhou, G.; Wang, H.; Wu, X. The Behavior of Antibiotic-Resistance Genes and Their
939 Relationships with the Bacterial Community and Heavy Metals during Sewage Sludge
940 Composting. *Ecotoxicol. Environ. Saf.* **2021**, *216*, 112190.
941 <https://doi.org/10.1016/j.ecoenv.2021.112190>.
- 942 (116) Lin, H.; Sun, W.; Yu, Y.; Ding, Y.; Yang, Y.; Zhang, Z.; Ma, J. Simultaneous Reductions in
943 Antibiotics and Heavy Metal Pollution during Manure Composting. *Sci. Total Environ.* **2021**,
944 *788*, 147830. <https://doi.org/10.1016/j.scitotenv.2021.147830>.
- 945 (117) Jiao, W.; Du, R.; Ye, M.; Sun, M.; Feng, Y.; Wan, J.; Zhao, Y.; Zhang, Z.; Huang, D.; Du, D.;
946 Jiang, X. ‘Agricultural Waste to Treasure’ – Biochar and Eggshell to Impede Soil
947 Antibiotics/Antibiotic Resistant Bacteria (Genes) from Accumulating in *Solanum Tuberosum*
948 *L. Environ. Pollut.* **2018**, *242*, 2088–2095. <https://doi.org/10.1016/j.envpol.2018.06.059>.
- 949 (118) Liang, Y.; Zhu, H.; Bañuelos, G.; Shutes, B.; Yan, B.; Cheng, X. Removal of
950 Sulfamethoxazole from Salt-Laden Wastewater in Constructed Wetlands Affected by Plant
951 Species, Salinity Levels and Co-Existing Contaminants. *Chem. Eng. J.* **2018**, *341*, 462–470.

952 <https://doi.org/10.1016/j.cej.2018.02.059>.

953 (119) Hernando-Amado, S.; Coque, T. M.; Baquero, F.; Martínez, J. L. Defining and Combating
954 Antibiotic Resistance from One Health and Global Health Perspectives. *Nat. Microbiol.* **2019**,
955 *4* (9), 1432–1442. <https://doi.org/10.1038/s41564-019-0503-9>.

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957 **Supplementary Materials Captions**

958 **Figure S1** Concentrations of different types of antibiotics in global agricultural soils.

959 **Figure S2** Abundances of different types of ARGs in global agricultural soils.

960 **Text S1** Studies included in this analysis exploring the antibiotics and ARGs in global
961 agricultural soils.

962 **Table S1** Antibiotics ecological risk parameters.

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