Supplementary Materials

The Effect of Manure Application Rates on the Vertical Distribution of Antibiotic Resistance Genes in Farmland Soil

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Contents: 10 Pages, 7 Figures, 1 Table

Supplementary information contains the following:

Figure S1. The composition of the detected ARGs drug resistance mechanism in different soil layers. A, B, and C represent the 0–20 cm, 20–40 cm, and 40–60 cm soil layers, respectively. The numbers 0, 1, 2, 3, and 4 correspond to the application of 0, 25, 50, 75, and 100% manure in the soil, respectively. JF: organic fertilizer (chicken manure after high-temperature composting).

Figure S2. Relative abundance of ARGs in different soil layers. A: 0－20 cm soil layer, B: 20－40 cm soil layer.

Figure S3. Heatmap of relative abundance of ARGs in different soil layers. A: 0－20 cm soil layer, B: 20－40 cm soil layer.

Figure S4. Changes in pH (A), CEC (B), heavy metals (C), antibiotics (D), carbon (E), and nutrients (F) in different soil layers. A, B, and C represent the 0–20 cm, 20–40 cm, and 40–60 cm soil layers, respectively. The numbers 0, 1, 2, 3, and 4 correspond to the application of 0, 25, 50, 75, and 100% manure in the soil, respectively. JF: organic fertilizer (chicken manure after high-temperature composting).

Figure S5. The relative abundance of microbial phylum in different soil layers (others represent all phyla with relative abundance less than 1%). A: 0–20 cm soil layer, B: 20–40 cm soil layer.

Figure S6. Heatmap of microbial phylum in different soil layers. A: 0–20 cm soil layer, B: 20–40 cm soil layer.

Figure S7. KEGG cluster analysis and functional analysis of microbial community genomes in chicken manure and different soil layers: Enzyme activity annotation of microbial communities in chicken manure and different soil layers (A); KEGG clustering of microbial community genomes in chicken manure and different soil layers (B). A, B, and C represent the 0–20 cm, 20–40 cm, and 40–60 cm soil layers, respectively. The numbers 0, 1, 2, 3, and 4 correspond to the application of 0, 25, 50, 75, and 100% manure in the soil, respectively. JF: organic fertilizer (chicken manure after high-temperature composting).

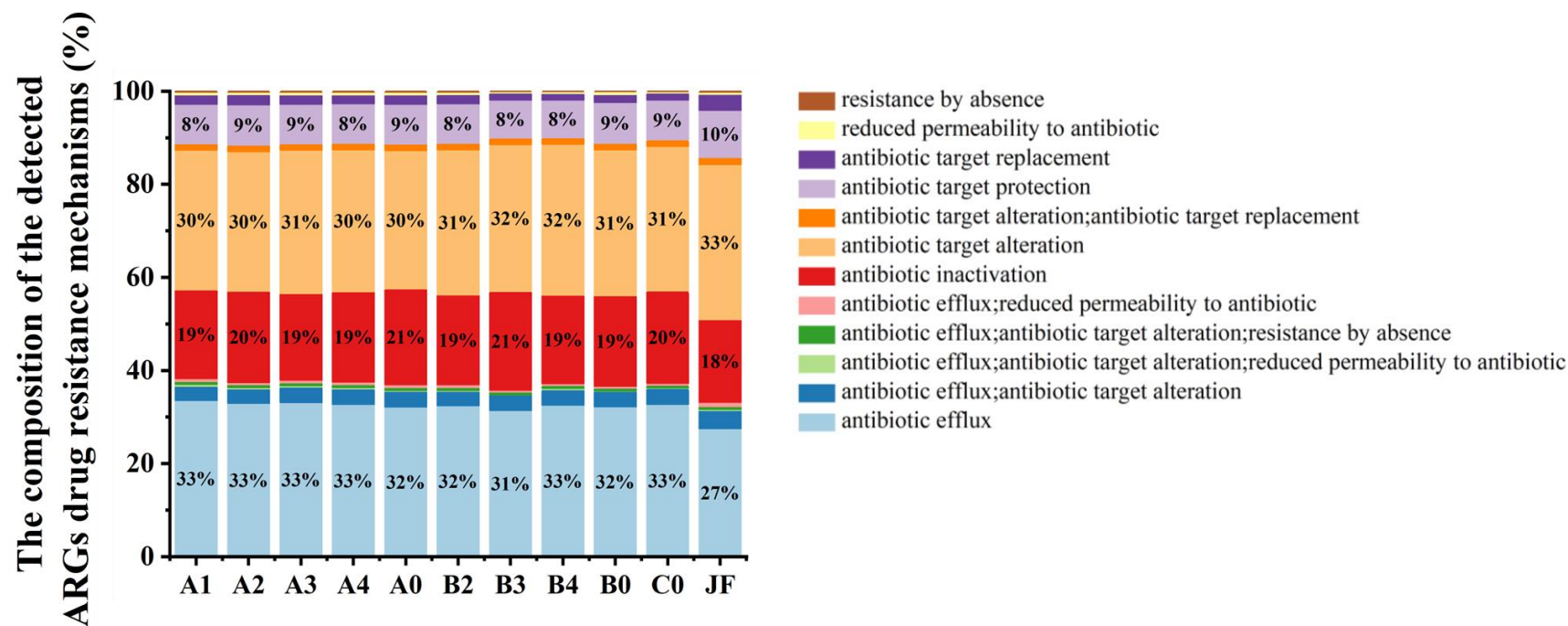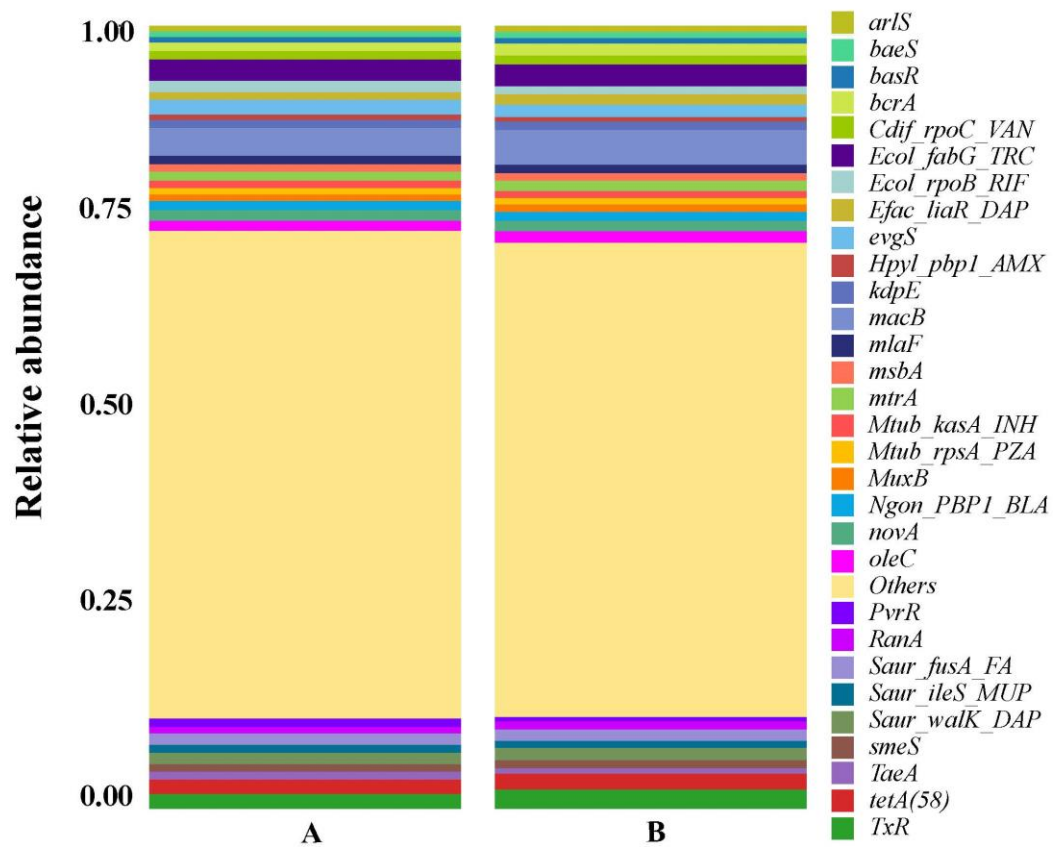**Table S1.** Pathogenic bacteria with ARGs and resistance mechanisms in soil.

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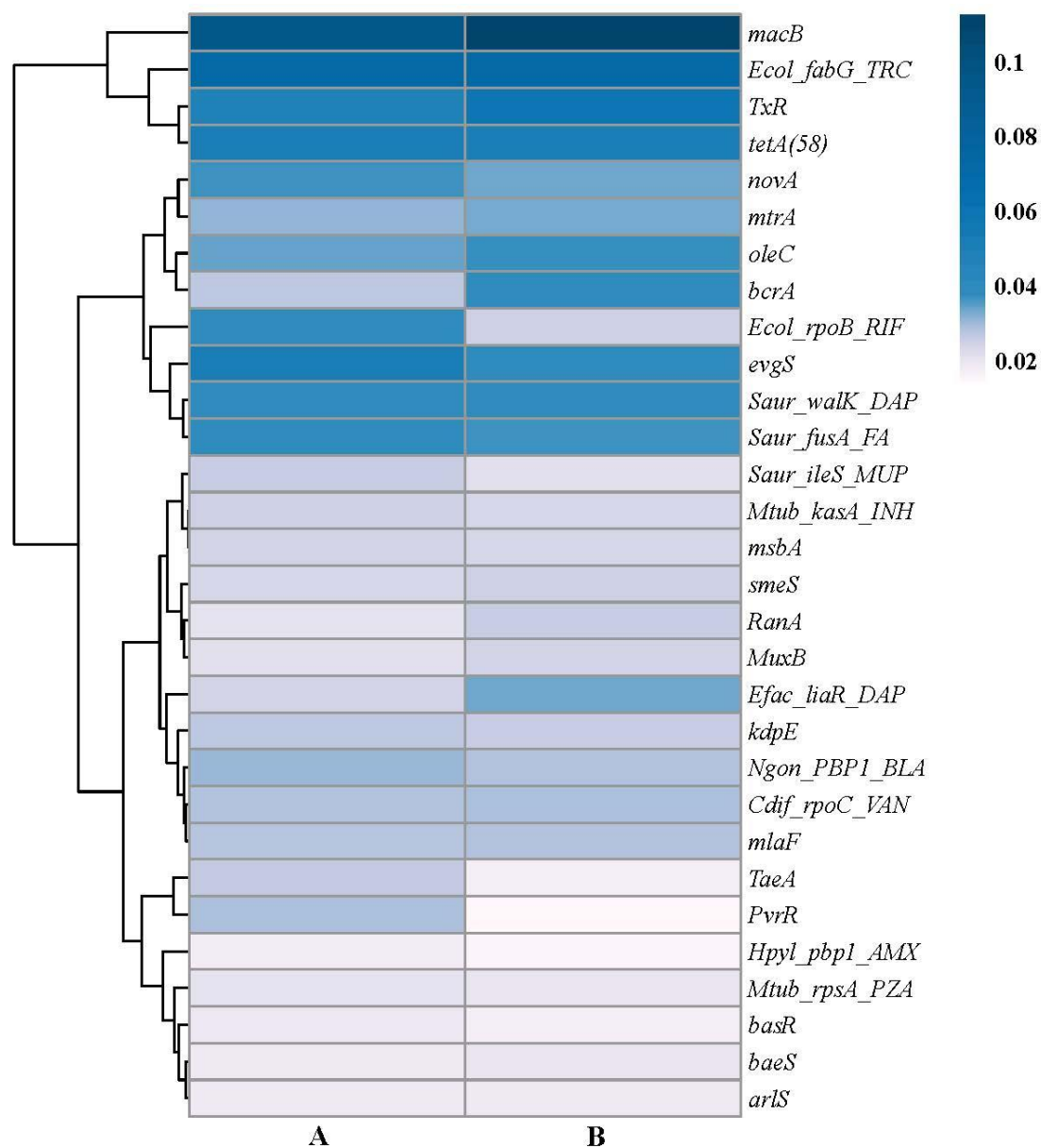
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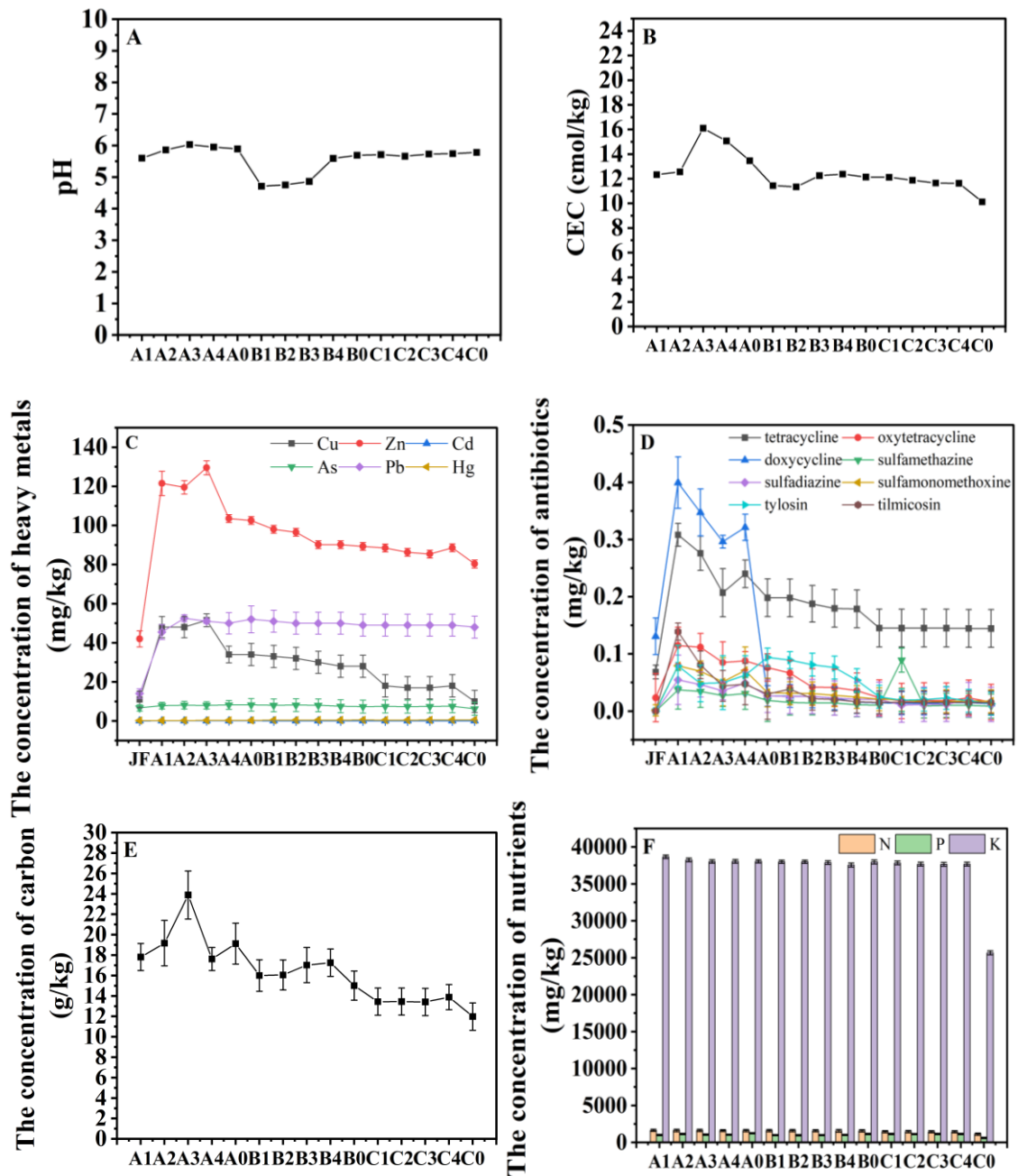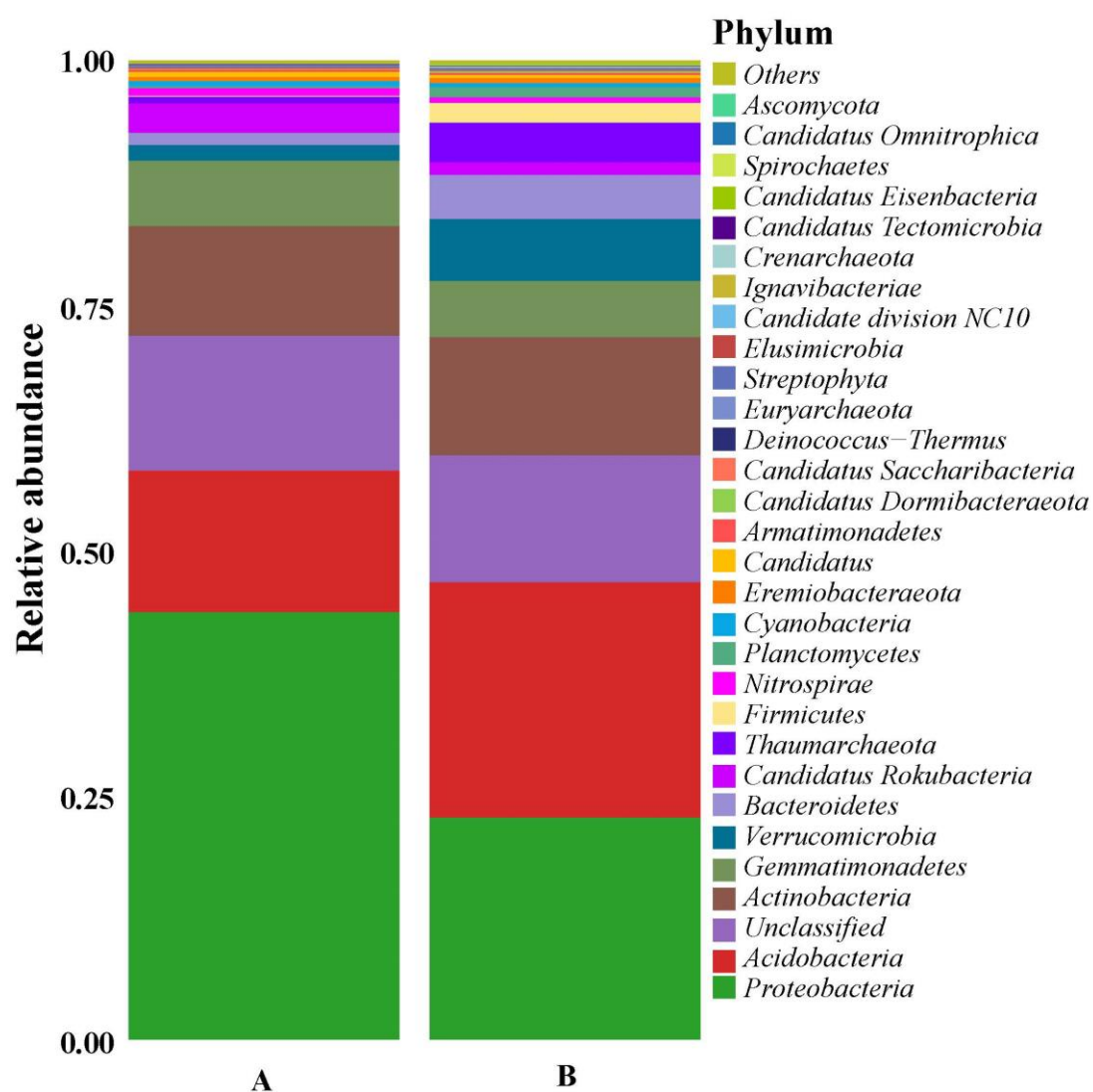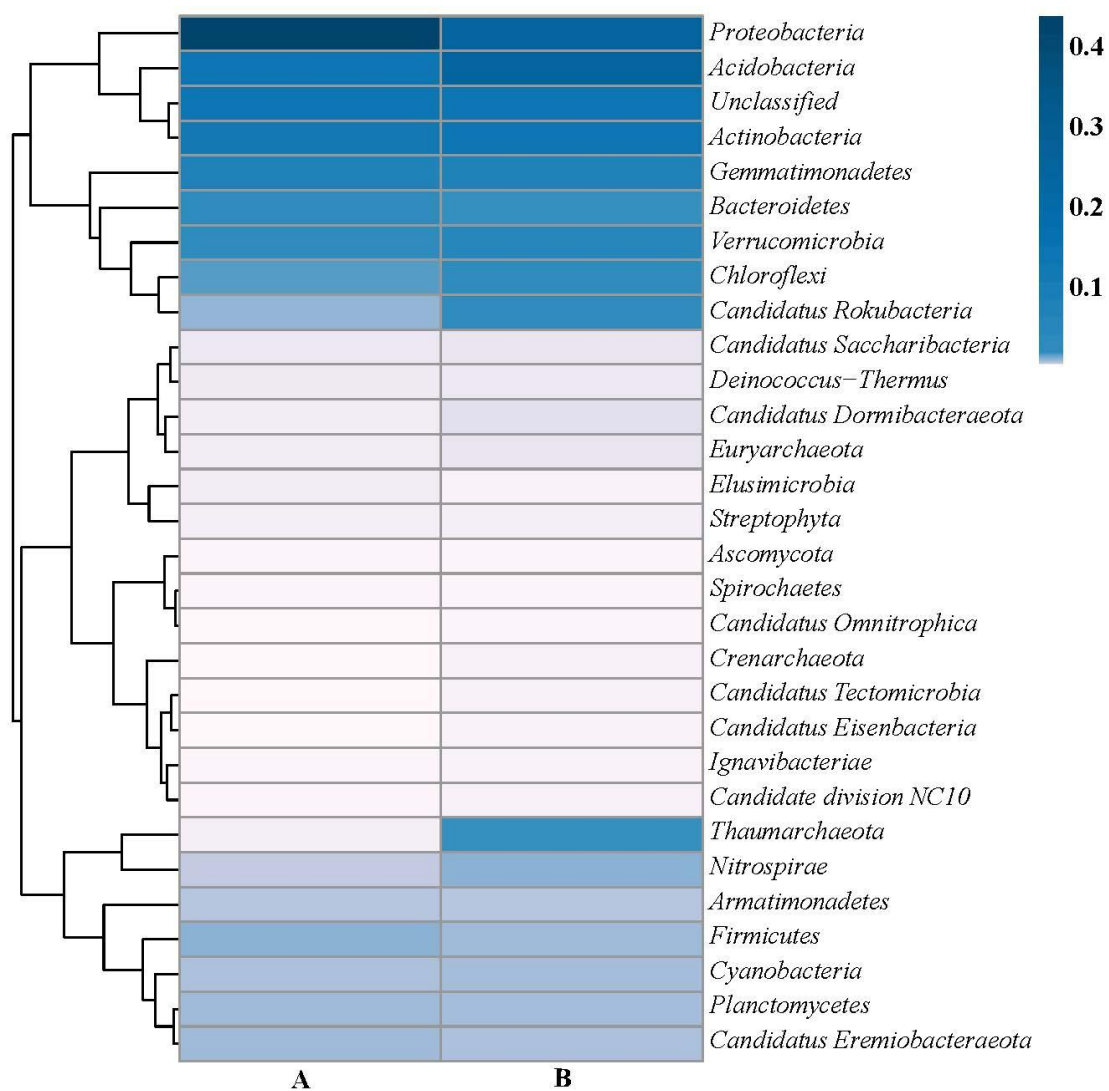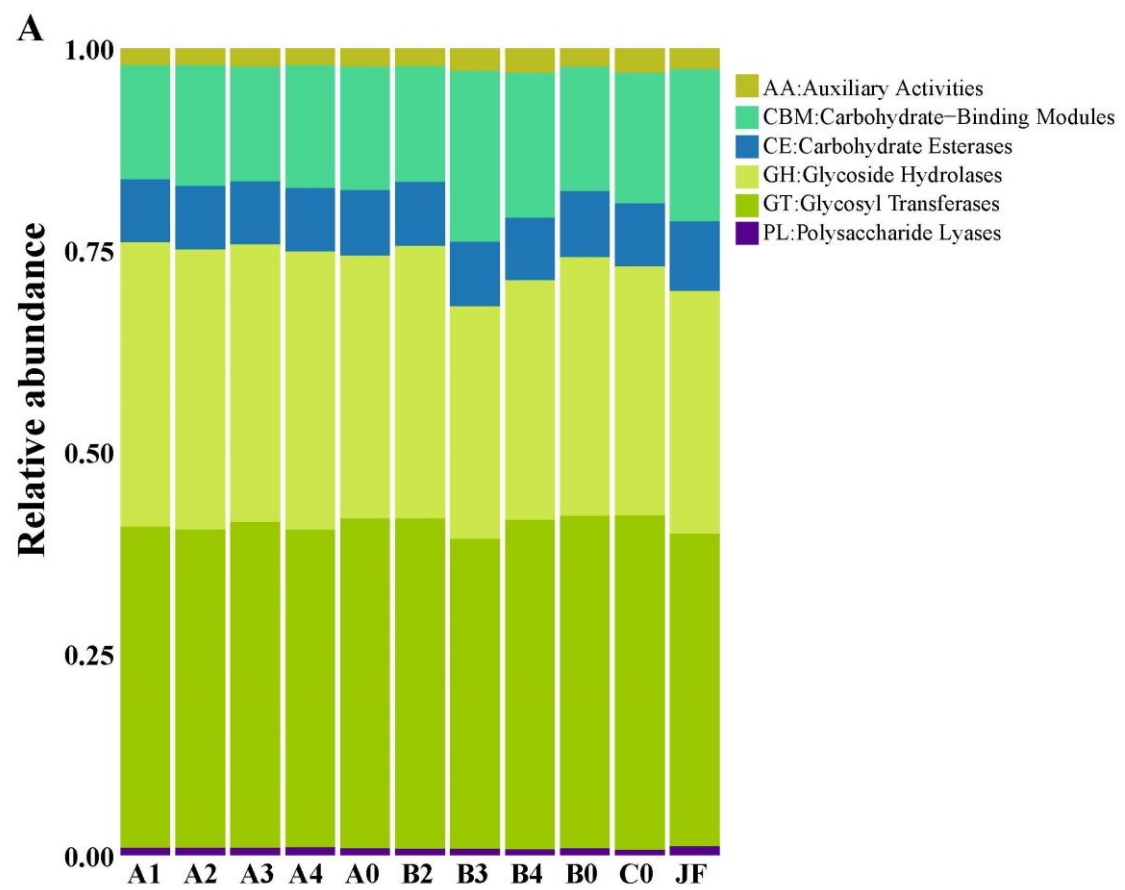
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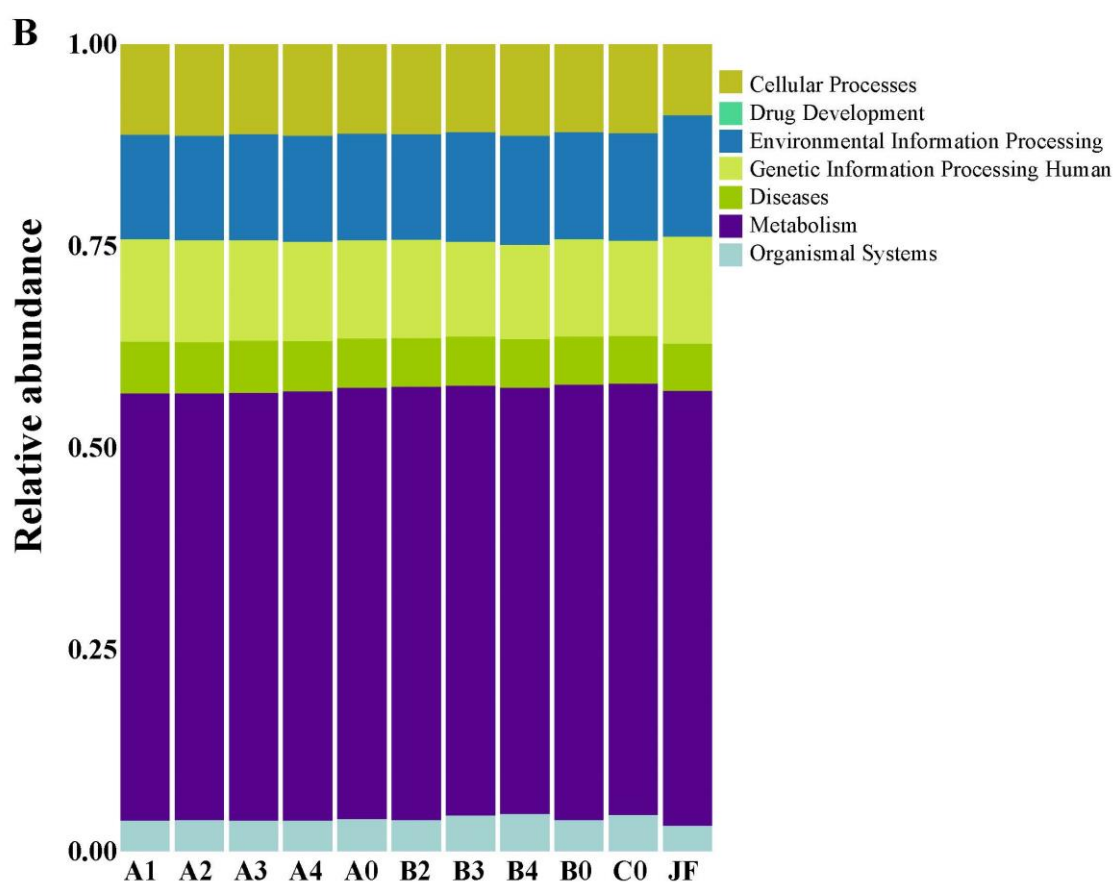
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Table S1 Pathogenic bacteria with ARGs and resistance mechanisms in soil.

Pathogenic Bacteria	Resistance Class	Resistance Gene	Mechanism of Resistance
Pseudomonas aeruginosa LESB58、 *Pseudomonas aeruginosa PAO1*	Macrolide	*macB*、*macA*	Antibiotic efflux
	Diaminopyrimidine Antibiotics; Fluoroquinolones; Phenolic Antibiotics	*MexF*	Antibiotic efflux
	Fluoroquinolones; Macrolide Antibiotics; Penems; Tetracyclines	*evgS*	Antibiotic efflux
	Peptide Class	*PmrF*	Antibiotic efflux、antibiotic target alteration
	Carbapenems; Cephalosporins; Monobactams; Aminoglycosides	*mdsB*	Antibiotic efflux
Listeria monocytogenes EGD-e	Lincosamides	*lin*	Antibiotic Inactivation