Soil physico-chemical parameters influence on the level of antibiotic resistance

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Introduction and Methodology

Introduction: The evolution and spread of antibiotic-resistant bacteria (ARB) and antibiotic resistance genes (ARGs) into the environment is a critical global health concern. Tackling AMR requires action aligned with the 'One Health' approach—a holistic framework in which human, veterinary, and environmental settings are interconnected. Up to now multiple environmental dissemination routes for resistant bacteria have been identified as potentially significant for the proliferation of AMR, particularly in the context of intensive agriculture and increasing urbanization. The accumulation of antibiotics in soils depends on their physico-chemical properties so physico-chemical characteristics of the environment should be identified as potential factors influencing the abundance of ARGs.

Methodology: The study was conducted on soil samples collected from areas characterized by high (agricultural fields: manured and non-manured) and low (forests) anthropogenic activity during the autumn of 2022. Collected soil samples were analyzed for their physico-chemical properties. Total DNA was extracted using the DNeasy PowerSoil Pro Kit (Qiagen) according to the manufacturer's instructions.
A total of 27 ARGs and 5 MGEs (mobile genetic elements) were targeted. Determination of the relative abundance of target genes in the samples was performed using a SmartChip Real-Time PCR cycler. Quantification included the 16S rRNA gene. The qPCR cycling conditions and initial data processing were carried out as described by Wang et al., 2014.
Relationships between physico-chemical properties of soils and the relative abundances of ARGs/MGEs were determined using Spearman's correlation coefficient and visualized with GraphPad Prism 9.

Aim of the study

Determination whether there is a quantitative and qualitative relationship between ARGs and the physico-chemical properties of soil

Results

The physico-chemical properties analysis revealed a significant difference between arable and forest soils ($p \leq 0.05$) in terms of soil **water** content, **humus**, **sand** and **silt** proportions, and **mercury** concentration. No significant differences ($p > 0.1$) were found for clay content, carbon-to-nitrogen ratio, or the concentrations of phosphorus, potassium, iron, manganese, zinc, boron, lead, chromium, vanadium, and calcium.

The relative abundance of the analyzed ARGs and MGEs differed between arable and forest soils ($p = 0.0247$). The average abundance of these genes was lower in forest soils compared to arable soils. In total, 5 MGEs (*intl1_1*, *orf37-IS26*, *IncP_oriT*, *IncW_trwAB* and *Tn5*) and 25 ARGs were identified in the analyzed soils (Szadziul et al., 2025).

Correlation analysis between MGEs and ARGs revealed a network consisting of 25 nodes. A total of 47 interactions were associated with MGEs, highlighting their role in network formation. However, no significant interactions were observed for the following ARGs identified in soil samples: *aph3-via*, *ermB_1*, *ermF_1*, *bla*$_{CTX\text{-}M\_2}$ and *tetO_1*. (Figure 1)

Correlation analysis between ARG/MGE and physico-chemical parameters revealed a network consisted of 44 nodes and 275 edges, representing strong and significant correlations ($|Rs| > 0.8$; $p \leq 0.05$) Of these interactions, 26 were negative, while 249 were positive. (Figure 2)

Aluminum (Al.) abundance showed a strong positive correlation with all tested MGE-related genes and 17 out of 20 ARGs (excluding *tetW*, *tetM_2* and *dfrA1_1*) and with 4 out of 5 MGEs (except *Tn5).*
Arsenic (As), chromium (Cr), zinc (Zn) and water content positively correlated with 14 ARGs and with 4 out of 5 MGEs (except *Tn5).*
Calcium (Ca), iron (Fe), nickel (Ni), clay content, and humus content positively correlated with 13 ARGs and with all 5 MGEs.
Magnesium (Mg) and nitrogen (N) positively correlated with 11 ARGs and with 3 out of 5 MGEs (*intl1_1*, *IncW_trwAB* and *Tn5)*
Boron (B) and mercury (Hg) showed positive correlations with 11 ARG and 12 ARG, respectively and with 3 MGEs (*orf37-IS26*, *IncP_oriT*, *IncW_trwAB).*

Figure 1. Gene to gene heatmap

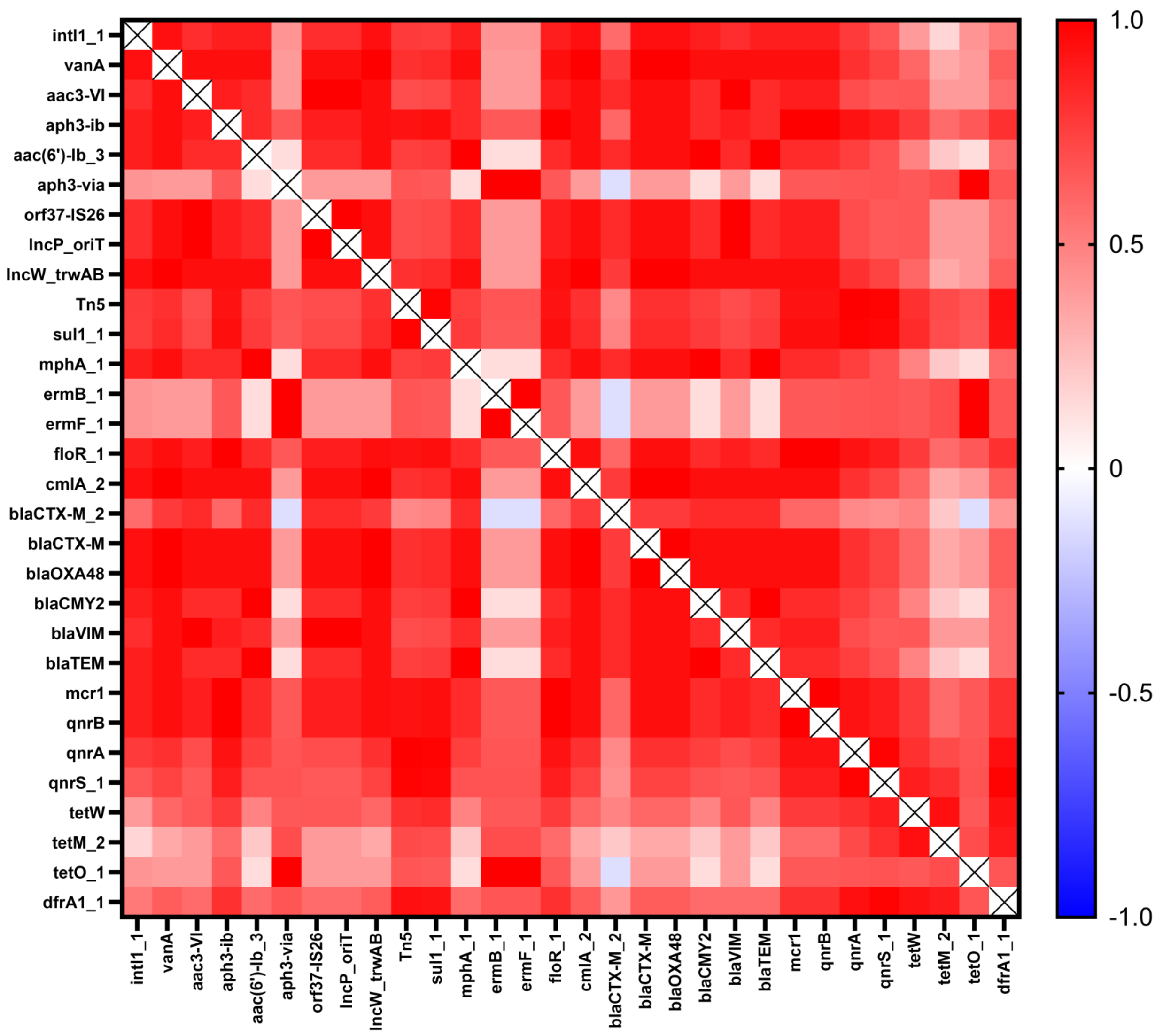
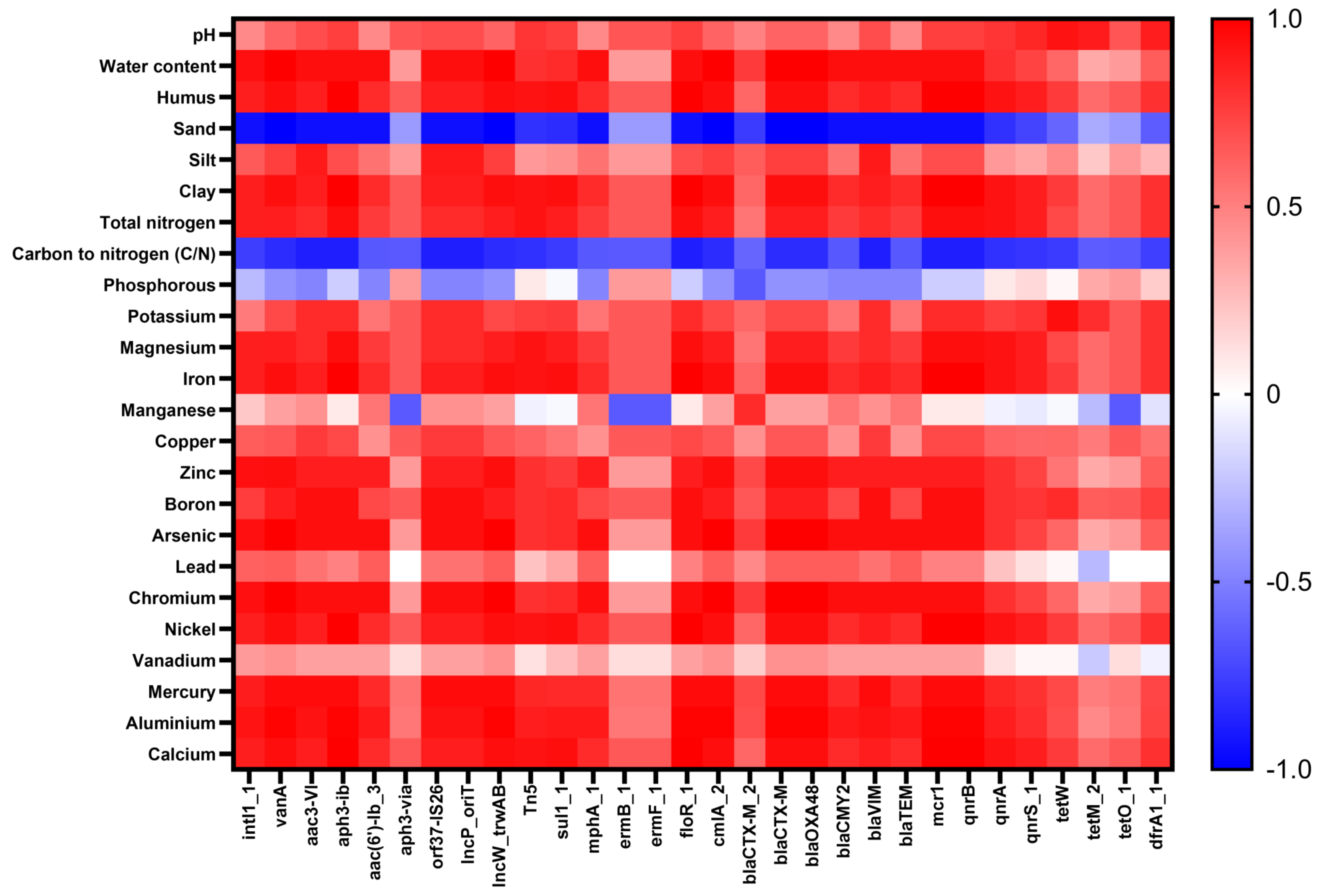


Figure 2. Gene to factor heatmap



Take home message

***S**oil physico-chemical properties influence ARG and MGE abundances*

Acknowledge

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References

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