# Endophytic bacterial community reveals antimicrobial resistance in response to poultry manure application

Animesh Tripathi [a] , Anjali Jaiswal [a] , Dinesh Kumar [b] , Priyank Chavda [b] , Ramesh Pandit [b] , Madhvi Joshi [b] , Damer P. Blake [c] , Fiona M. Tomley [c] , Chaitanya G Joshi [b] , and Suresh Kumar Dubey [a] *

a Department of Botany, Institute of Science, Banaras Hindu University (BHU), Varanasi, Uttar Pradesh, India, 221005,
b Gujarat Biotechnology Research Centre (GBRC), Department of Science and Technology (DST), Govt. of Gujarat, Gandhinagar, Gujarat, India, 382011,
c Pathobiology and Population Sciences, The Royal Veterinary College, Hawkshead Lane, North Mymms, Hertfordshire, UK.

## HYPOTHESIS & OBJECTIVES

Poultry manure serve as reservoir of antimicrobial resistant microbes their exposure to agriculture may induce plant endo-microbiome to acquire Antimicrobial resistance genes. In this study, we examine the proposition that exposure to poultry manure in the agro-ecosystem amplifies the prevalence of antimicrobial resistance (AMR) in plant endophytes.

## INTRODUCTION

- Poultry manure, a widely used fertilizer in agriculture, raises concerns due to the continuous use of antibiotics in poultry farming, potentially creating reservoirs for antimicrobial resistance genes (ARGs).

- Previous studies reveal that adding poultry manure to soil increases the prevalence of resistance genes against various antibiotics in both soil and plants.

- It is crucial to comprehend the microbial communities hosting these antimicrobial resistance genes (ARGs) in order to identify microbes that may potentially enter the human food chain.

- Our Previous published data show poultry litter (broiler/layer) enriching plant root/stem ARGs via next-gen sequencing, validated with dPCR in pot/field experiments (Tripathi, et al., 2023).

## METHODS AND MATERIALS

Experimental sampling, Plant pre-treatment and isolation of endophytic bacteria

⬇

Screening of Antibiotic Resistant endophytic bacteria using Antibiotic Susceptibility Test (AST)
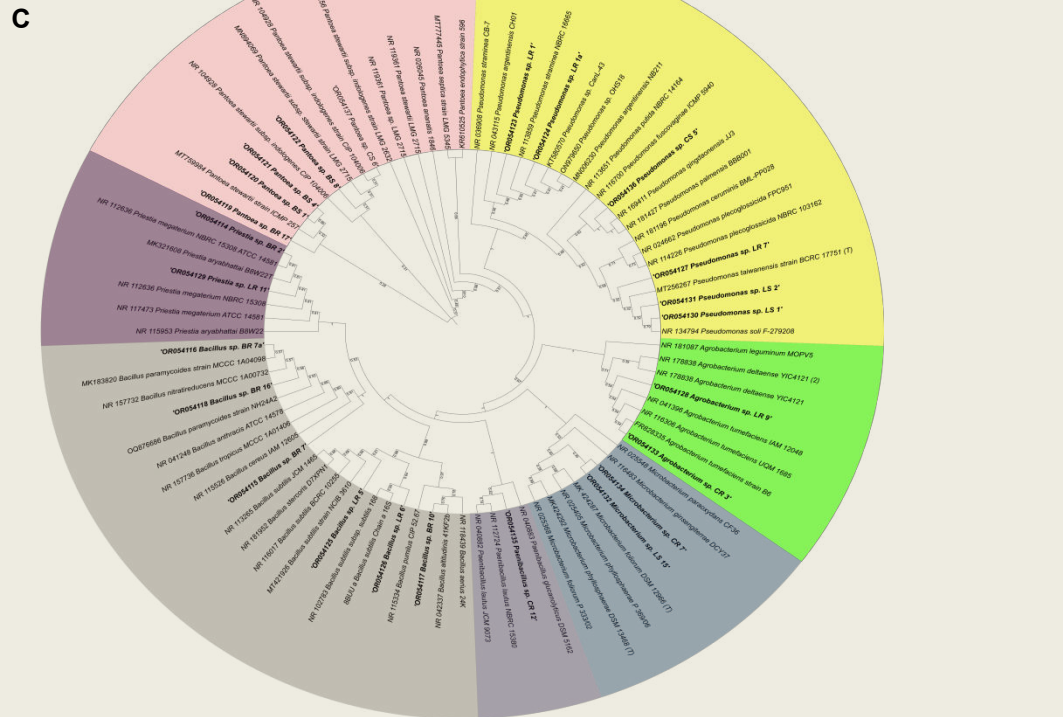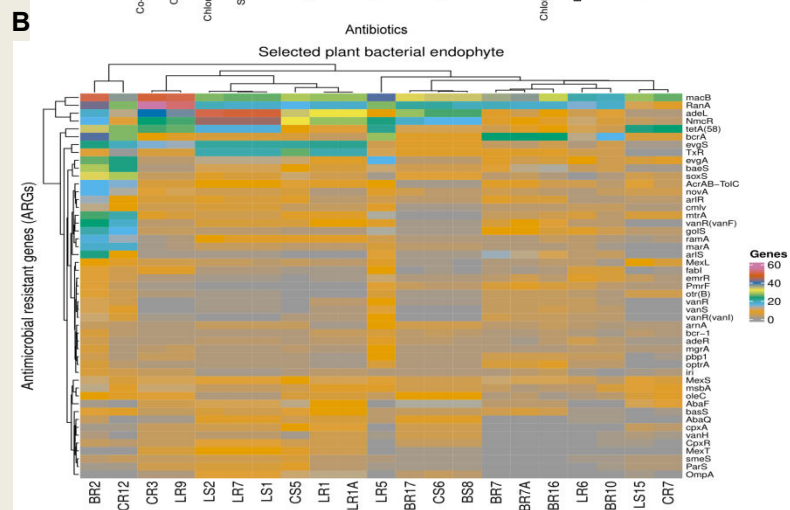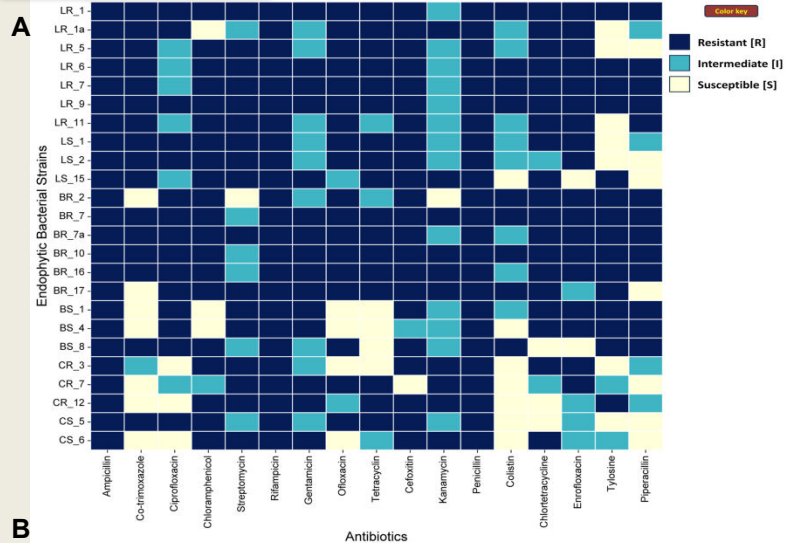
⬇

Molecular identification, phylogenetic analysis and Biotypic characterization of MDR endophytic isolates using MALDI-TOF

⬇

Whole genome sequencing (WGS) of bacterial isolates and Bioinformatics data analysis

A. Heatmap plot of twenty-four selected MDR strains showing resistance patterns against different antibiotics.
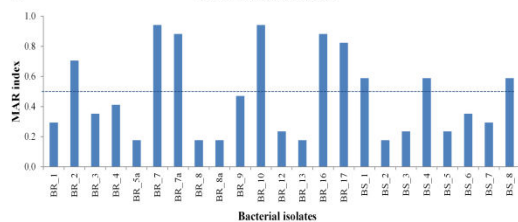B. Phylogenetic tree for twenty-four MDR strains based on 16S rRNA nucleotide sequences constructed by the neighbor-joining approach. Numerical values at the node represent bootstrap values. GenBank accession numbers of nucleotide sequences are shown along with the name of the bacterial strains.
C. Heatmap plot depicting the ARGs detected in different plant endophyte bacterial strains isolated from poultry litter treated or control plant samples (Roots and Stem).
D. Summary of antibiotic resistance profiles for endophytes isolated from plants grown with broiler, layer or no litter supplementation. MAR indexes for broiler (a), layer (b), and no litter treatment (c) groups where BR: Broiler Root, BS: Broiler Stem, LR: Layer Root, LS: Layer Stem, CR: Control Root and CS: Control Stem. The percentage of isolates resistant, intermediate or susceptible to each of 17 antibiotics is shown for broiler (e), layer (f), and no (g) litter treated endophytes.
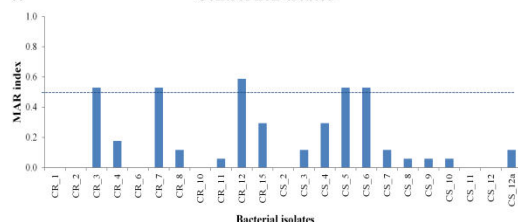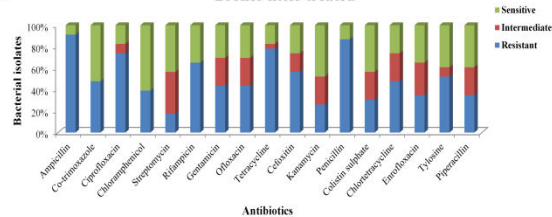
**D**

Broiler litter treated (a)
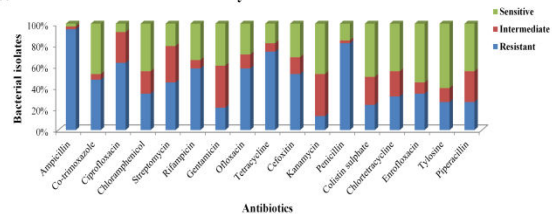
Layer litter treated (b)

Control non-treated (c)

Broiler litter treated (d)

Layer litter treated (e)

Control non-treated (f)

## REFERENCE

Tripathi, A., Kumar, D., Chavda, P., Rathore, D.S., Pandit, R., Blake, D., Tomley, F., Joshi, M., Joshi, C.G. and Dubey, S.K., 2023. Resistome profiling reveals transmission dynamics of antimicrobial resistance genes from poultry litter to soil and plant. *Environmental Pollution*, **327, p.121517**.