

# Metagenomic Characterization of Antibiotic Resistance Genes in Bacterial Communities from a Wastewater Treatment Plant in Lima, Peru

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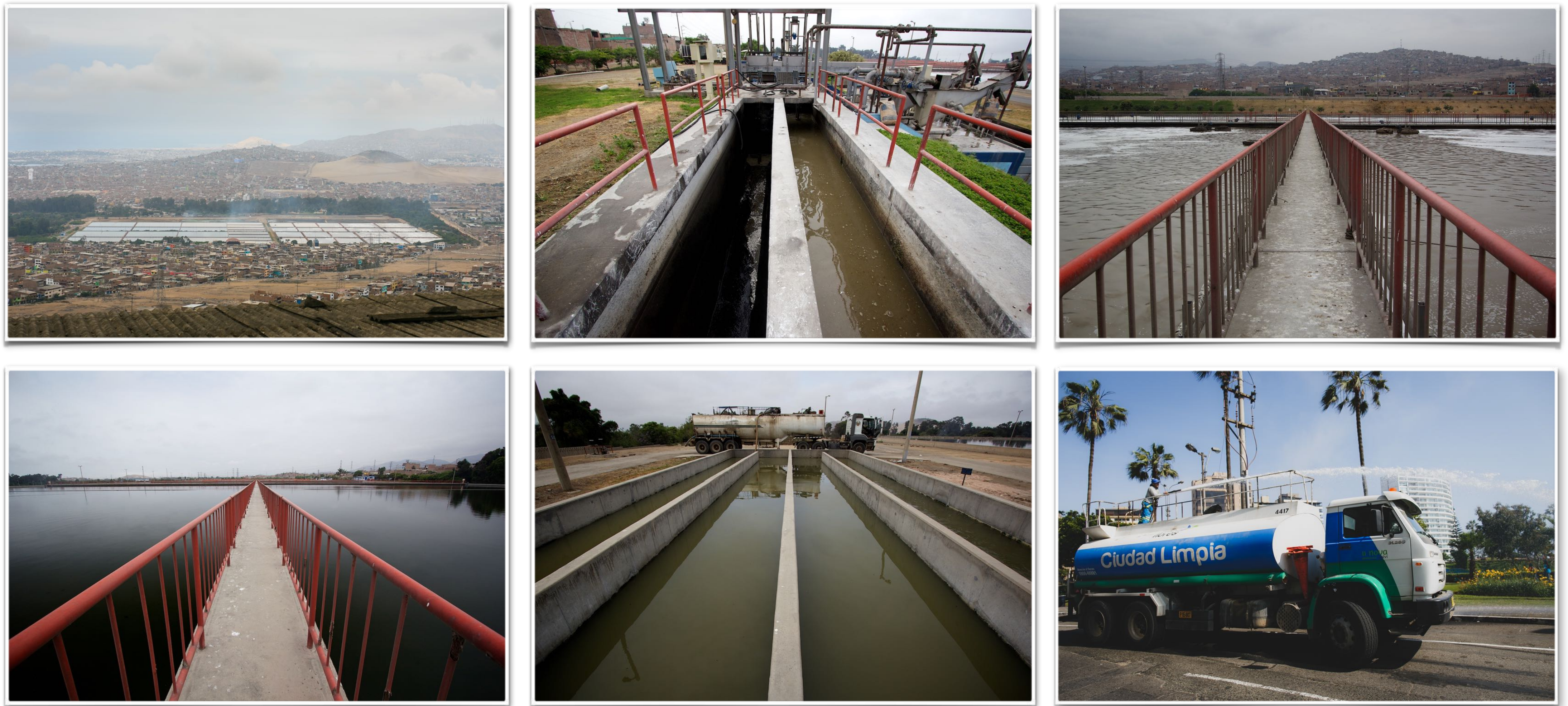
## ABSTRACT

Wastewater treatment plants (WWTPs) have long been postulated as optimal environments for the recombination of antibiotic resistance genes (ARGs) among bacteria: they collect excreta from thousands of individuals and mix them under conditions that promote bacterial growth and genetic recombination, often in the presence of trace amounts of antibiotics from clinical and agricultural use. Treated sewage effluent is usually discharged into oceans and rivers or utilized for the irrigation of agricultural fields and parks, raising concerns of re-introduction and enrichment of resistant bacteria and ARGs in the environment and the community. Sewage microbial communities are potential reservoirs of ARGs available to human pathogens.

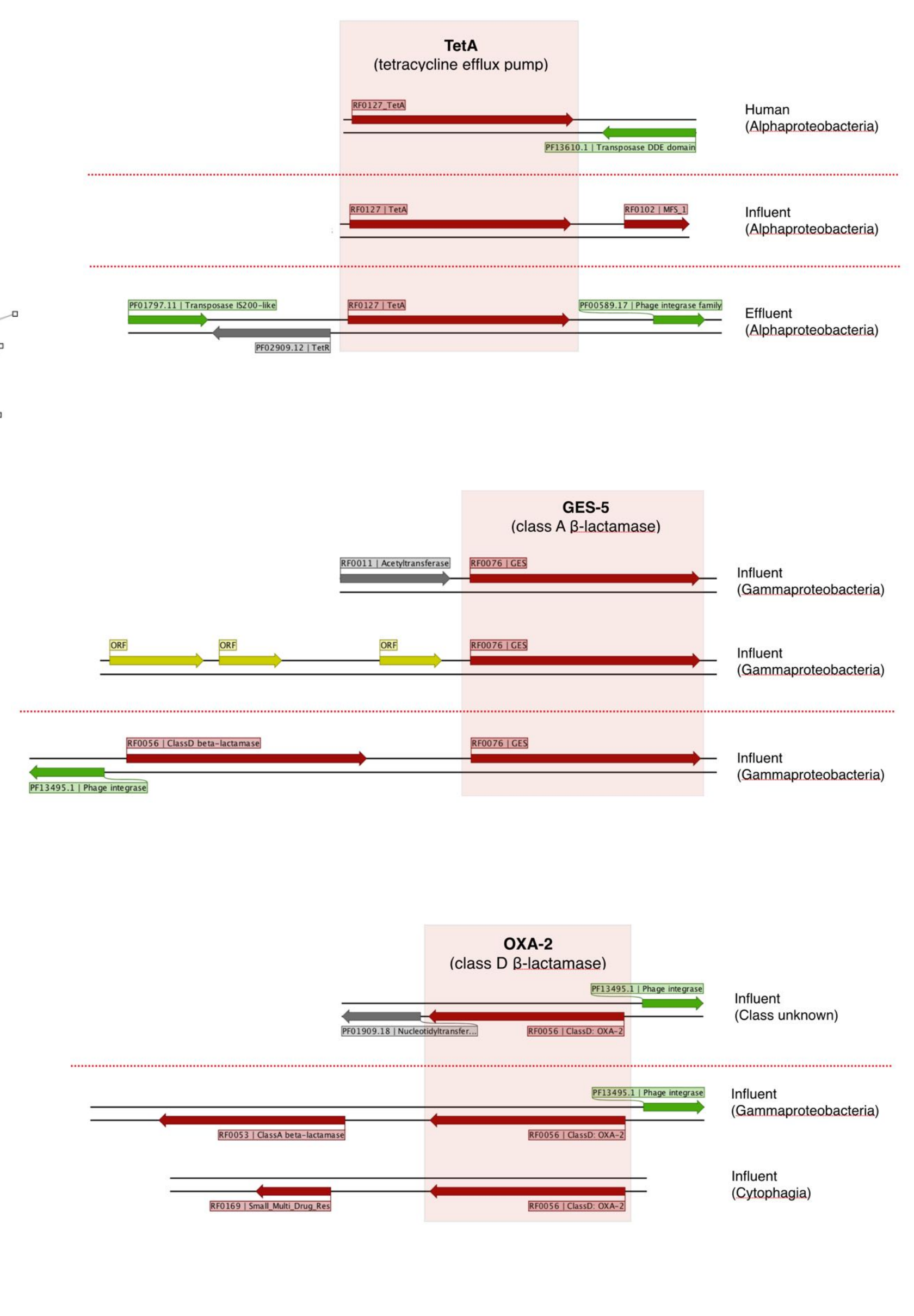
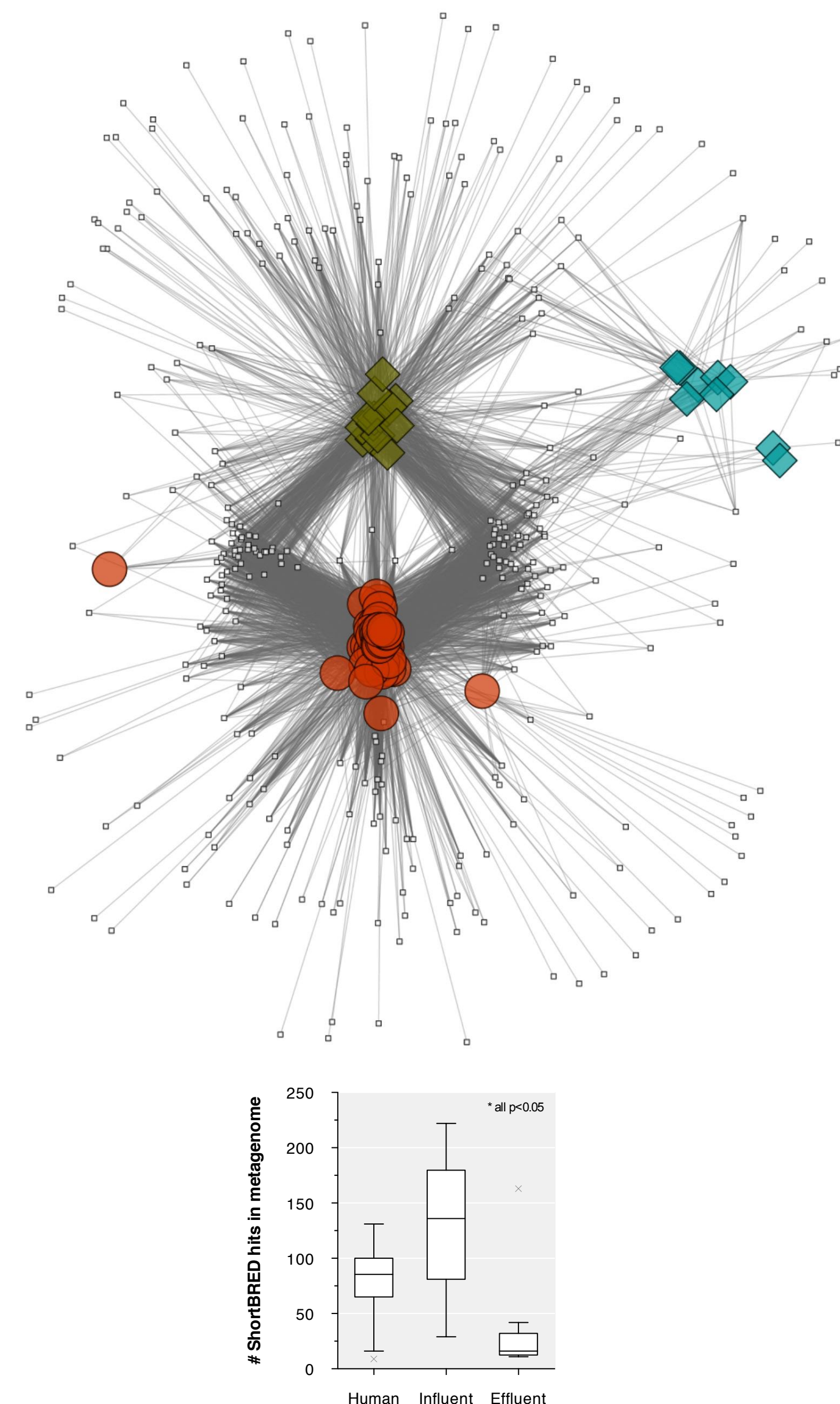
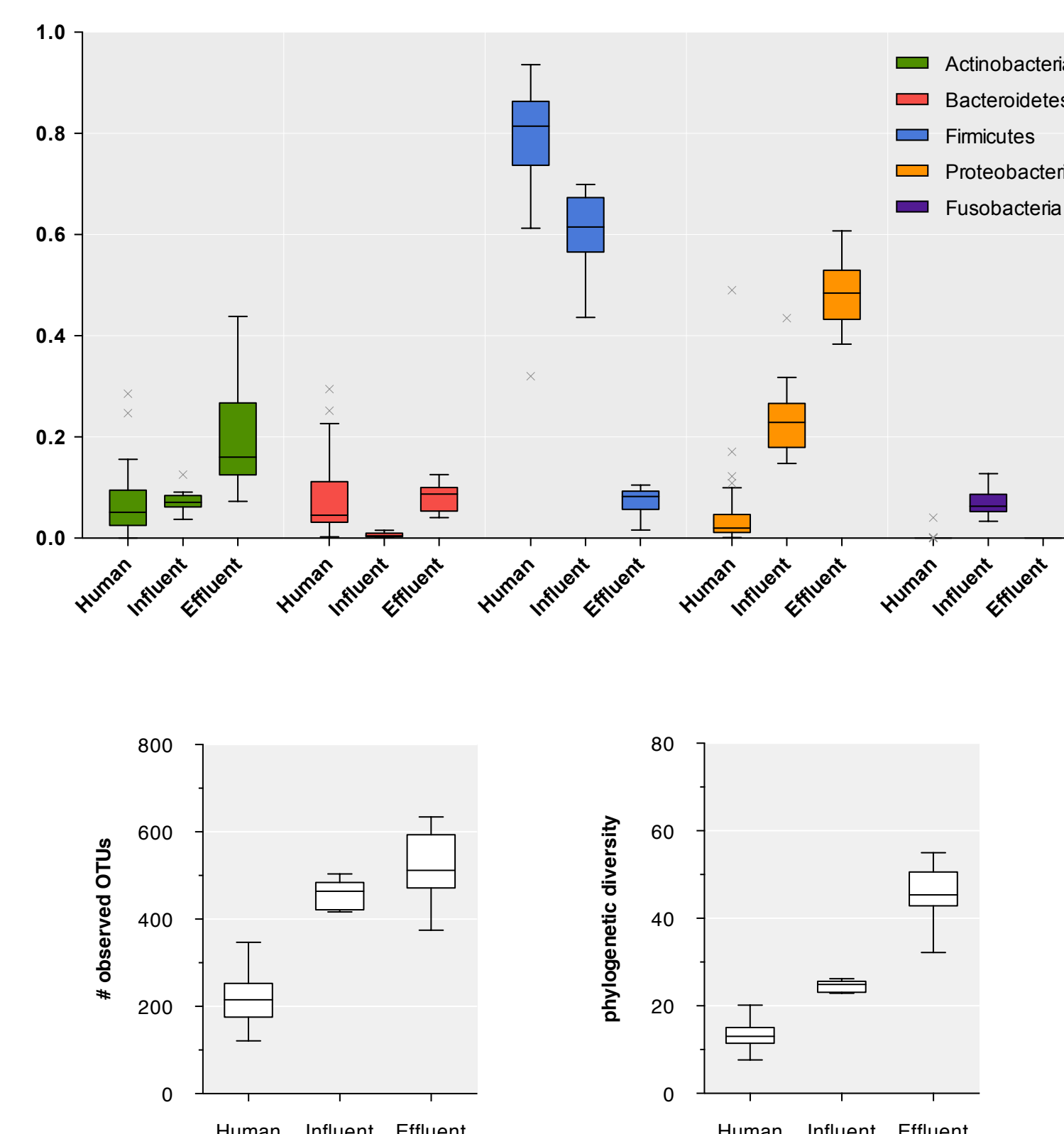
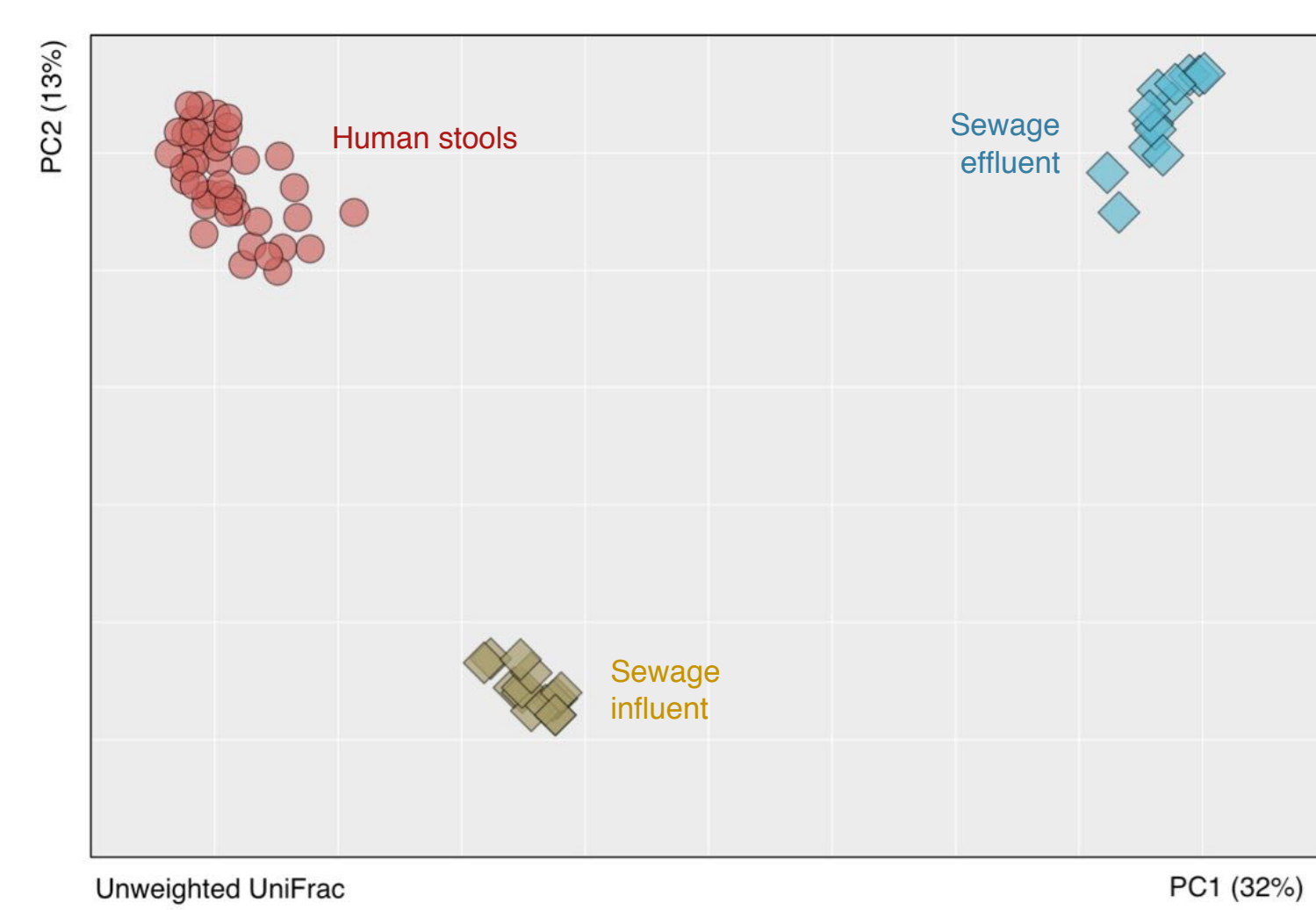
Between 2014 and 2016, we collected monthly sewage influent and effluent samples from a WWTP in Lima, Peru that serves a population of ~1.5 million inhabitants. We determined total coliform counts and cultured members of the Enterobacteriaceae family to assess the efficiency of the WWTP in reducing fecal bacteria in treated effluent. Sewage samples were characterized by 16s rRNA amplicon sequencing and whole metagenomic sequencing to assess changes in microbial composition, antibiotic resistomes, and metabolic potential of these microbial communities as they undergo treatment. Genomes of enterobacterial isolates will be sequenced to assess their phylogenetic relationships and track the movement of ARGs across bacterial genomes. We observed high rates of phenotypic resistance in bacteria isolated from influent and effluent samples. Metagenomic methods revealed that bacterial communities in sewage influent were of 30%-50% human fecal origin and that they were converted into vastly different communities as they progress through treatment. However, many resistant bacteria and ARGs remained in the effluent, and they show evidence of recent recombination across environments.

Our results indicate that potentially pathogenic, multi-drug resistant bacteria are continually introduced into the community via treated effluent, and that metagenomics are a useful tool to assess the efficiency of WWTPs in low- and middle-income country (LMIC) settings.

## Samples: Human stools / Influent + effluent from 'San Juan' WWTP in San Juan de Miraflores, Lima



## Analysis: 16S rRNA sequencing / Functional metagenomic selections / Shotgun sequencing + ShortBRED + CARD



## ARTICLE

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## Interconnected microbiomes and resistomes in low-income human habitats

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Antibiotic-resistant infections annually claim hundreds of thousands of lives worldwide. This problem is exacerbated by exchange of resistance genes between pathogens and benign microbes from diverse habitats. Mapping resistance gene dissemination between humans and their environment is a public health priority. Here we characterized the bacterial community structure and resistance exchange networks of hundreds of interconnected human faecal and environmental samples from two low-income Latin American communities. We found that resistomes across habitats are generally structured by bacterial phylogeny along ecological gradients, but identified key resistance genes that cross habitat boundaries and determined their association with mobile genetic elements. We also assessed the effectiveness of widely used excreta management strategies in reducing faecal bacteria and resistance genes in these settings representative of low- and middle-income countries. Our results lay the foundation for quantitative risk assessment and surveillance of resistance gene dissemination across interconnected habitats in settings representing over two-thirds of the world's population.

More about this work: Nature 533: 212-216 (2016)

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