**Application**: Antimicrobial resistance (AMR) poses a global health and economic challenge, with predictions of up to 10 million annual deaths by 2050 (O’Neill, 2014). It complicates disease management, increases healthcare costs, and threatens sustainable agricultural practices. This study focuses on AMR risks associated with livestock farming in Brazil, a key agricultural producer among BRICS nations. A One Health approach was applied to investigate the presence of AMR genes across dairy cow microbiomes, human nasal and hand microbiomes, and farm environments. Findings contribute to scientific understanding and provide actionable insights to inform policy changes, enhance antimicrobial stewardship, and support sustainable agricultural practices to mitigate resistance risks and preserve public health.

**Introduction**: AMR emerges when bacteria, viruses, or fungi no longer respond to antimicrobials, making infections harder to treat (WHO, 2024). In livestock farming, unregulated antimicrobial use and/or difficulties in implementing regulations exacerbate resistance development. Within the UK, sales patterns for veterinary antibiotics have remained stable over the last few years (UK-VARSS report, 2024). Nonetheless, prescription of antimicrobials for livestock animals in the BRICS countries (Brazil, Russia, India, China, and South Africa) is increasing, and set to double between 2010 and 2030 as a consequence of increased demand for animal protein (O’Neill, 2014). The scale of antibiotic use in agriculture and the large numbers of livestock involved has provided a significant route for dissemination of AMR bacteria to carcasses, to the environment, and subsequently to the human population (Van Boeckel et al., 2015). Brazil's prominence in global livestock and food production and antimicrobial practices make it an important focus for investigating AMR development and dissemination. This study aimed to map AMR trends across human, animal, and environmental interfaces on Brazilian farms and assess the relationship between farming practices and AMR burden in major food-producing regions.

**Materials and Methods**: A total of 186 samples, including ruminant faeces (36), soil (35), udder swabs (32), and human nasal swabs (20), were collected from farms across four major food-producing states: Minas Gerais, Paraná, Santa Catarina, and São Paulo, with varying practices. Illumina sequencing generated 40 GB of metagenomic data, with an average of 2.37 million reads per sample. Quality filtering and assembly using MetaSPADES resulted in 554 million high-quality reads with read counts per sample ranging from 37,008 to 4,066,265 and a mean read count of 1,491,834 reads per sample. The reads were assembled into contigs using MetaSPADES, resulting in contigs with a median N50 value of 250 bp. Contigs with a length of less than 500 bp were filtered from the dataset for downstream analysis.

Microbiome profiling was conducted with MetaPhlan4, and antimicrobial resistance genes (ARGs) were identified using KMA against the PanRes database (Martiny et al., 2023). Community composition and resistance distribution were analyzed using Principal Coordinate Analysis (PCoA) and Permanova tests. OH relevance was categorized into two levels: Level 1 (OH-L1) if ARGs were detected in at least one sample type across all three OH groups (Animal: Faeces and Udder; Environment: Soil and Bedding; and Human: Hand, Nasal, and Saliva), and Level 2 (OH-L2) if ARGs were present in at least one sample type across two OH groups.

**Results**: Across all samples in our dataset, 886 genera and 1,786 prokaryotic species were detected. The relative abundance of dominant taxa associated with human hand samples was observed to vary greatly between states. For instance, at the genus level, hand samples from Minas Gerais were dominated by Kocuria, Corynebacterium, and Acinetobacter. Hand samples from Paraná and Santa Catarina were overwhelmingly dominated by Rothia and Cutibacterium, respectively, while in São Paulo, hand samples were dominated by Ornithinimicobium and Deinococcus. Human nasal samples were dominated, by varying amounts across states, by Cutibacterium, Corynebacterium, and Staphylococcus. Human saliva samples were largely dominated by Staphylococcus, Prevotella, and Actinomyces.

In relation to animal samples, the abundance of Bifidobacterium and an unclassified genus (GGB25583) dominated the microbiota of faecal samples. In addition, Methanobrevibacter was found to largely dominate faecal samples from São Paulo. Udder samples were largely dominated by Corynebacterium and an unclassified genus (GGB44037). Environmental samples revealed that Corynebacterium and GGB44037 were prevalent in samples from Paraná and Santa Catarina, while Brachybacterium, Corynebacterium, and Dietzia dominated bedding samples from São Paulo.

Across all samples, 1,686 ARGs conferring resistance to 25 antibiotic classes were identified, with dominant classes including glycopeptides (284 genes), beta-lactams (279), and aminoglycosides (158). Resistance genes were abundant in human samples, ruminant faeces, udder swabs, and soil samples. Human hand samples were dominated by Tetracyclines and Aminoglycosides (34-55% across samples), human saliva samples by Glycopeptides, Antifolates, and Elfamycins (43-64%), and human nasal samples by Multi-compound, MLS, and Elfamycins (39-57%). Faecal resistomes were dominated by ARGs conferring resistance to Tetracycline, MLS, and Elfamycin (55-74%), while udder samples were dominated by ARGs classified as conferring resistance to Elfamycins, Tetracycline, and MLS (43-59%). Soil ARGs predominantly conferred resistance to Elfamycins, Glycopeptides, and Rifamycin (53-66%), while bedding samples were dominated by Elfamycin, Aminoglycoside, and Rifamycin resistance (41-63%).

PCoA revealed clustering by sample type, with significant differences in microbial and resistome compositions influenced by farming practices and state (P < 0.001). Although self-reported antimicrobial usage did not correspond to observed AMR patterns, AMR genes associated with mobile genetic elements (MGEs) were identified and shared across human, animal, and environmental interfaces. Resistance genes for vancomycin, colistin, and quinolones—last-resort antibiotics—were detected, highlighting the critical need for regulation. Cross-interface analysis indicated the presence of ARGs relevant to One Health (OH), with 434 genes detected across human, animal, and environmental samples (OH-L1 level).

**Conclusions:** This study underscores the widespread dissemination of AMR genes in Brazilian livestock farming systems, which may be driven by unregulated antimicrobial use. Findings highlight the urgent need for regulatory interventions to limit antimicrobial use in livestock farming to reduce resistance risks, protect critical antimicrobials, and ensure sustainable agricultural production. Insights from this work provide a foundation for evidence-based policy changes to inform AMR strategies in Brazil and other BRICS nations.

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