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Generation and Transmission of Antibiotic Resistance Genes in Livestock and Poultry Breeding Environments

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Abstract: The widespread presence of antibiotic resistance genes (ARGs) in livestock and poultry farming poses a significant threat to the ecological environment and public health. In this paper, the causes, transmission routes and environmental impacts of ARGs in livestock and poultry breeding environments are reviewed. ARGs spread in the environment through horizontal gene transfer (HGT), biological vectors, and airborne transmission, further exacerbating the problem of drug resistance. The presence of ARGs not only has a negative impact on soil and water ecosystems, but also poses a potential threat to human health through the food chain. This article discusses the current methods of detecting and controlling ARGs, and puts forward the focus and direction of future research, aiming to provide theoretical support for the formulation of effective prevention and control strategies to reduce the environmental and health risks caused by ARGs.This review identifies horizontal gene transfer as a critical mechanism in ARG propagation and proposes advanced detection techniques to curb environmental contamination, laying the groundwork for sustainable antibiotic usage in livestock.

Keywords: livestock and poultry farming, ARGs, route of transmission, environmental pollution, public health

1. Introduction

Livestock and poultry farming is an important part of modern agriculture, in which antibiotics are widely used in order to improve production efficiency and prevent diseases. Although antibiotics have played a positive role in the treatment of livestock and poultry diseases and in promoting growth, their overuse and abuse have led to the generation of a large number of antibiotic resistance genes (ARGs), which are transmitted to the natural environment through multiple routes, which has exacerbated the problem of bacterial resistance[1]. Once these drug-resistant bacteria are transmitted to humans through the food chain, they will cause intestinal infections, urinary tract infections, diarrhea, acute gastroenteritis and other diseases, which seriously threaten human health. Globally, over 700,000 deaths annually are attributed to antimicrobial resistance, a figure projected to rise dramatically without effective interventions.

2. The public health hazards of bacterial resistance in livestock and poultry breeding environments

Since Fleming's discovery of penicillin in 1928, antibiotics have gradually begun to be widely used in medical treatment, livestock and poultry breeding and other fields, but with the large use of antimicrobial drugs, drug-resistant bacteria have been screened out, which has increased the resistance rate of bacteria and caused the drug to lose its original efficacy. The problem of drug-resistant bacteria in livestock and poultry breeding environments is becoming more serious, and can be transmitted in a variety of ways, and even through the food chain to humans, posing a serious threat to public health. For instance, a U.S. outbreak of multidrug-resistant Salmonella linked to poultry farming resulted in over 400 cases and raised alarms about livestock-related ARGs. In recent years, there have been many cases of diarrhea and other related diseases caused by drug-resistant bacteria in humans around the world. In China, Nie Li et al. found that 279 strains of Salmonella had a multi-drug resistance rate of 73.48% to 14 antibiotics in 279 strains of Salmonella in Wuhan from 2017 to 2022, which was higher than the national average (69.11%), and the irrational use and abuse of antimicrobial drugs in animal husbandry were important reasons for the development of Salmonella resistance[2]. Qi Haoyu et al. conducted whole-genome sequencing of 101 strains of Escherichia coli isolated from Henan Province from 2017 to 2022, and found that Escherichia coli had multiple drug resistance to ampicillin, tetracycline, naphridic acid, etc., and the drug resistance status was at a high level, and the drug resistance genes and drug resistance mechanisms were diverse[3]. Lv Hua et al. found that Enterobacteriaceae bacteria have multidrug resistance in the analysis of bacterial resistance in 16 large poultry farms in Sichuan Province. In the United States, methicillin-resistant Staphylococcus aureus (MRSA) kills more people each year than AIDS, Parkinson's disease, emphysema, and homicides combined[4]. In 2023, a survey by the Albert Schweitzer Foundation found that 71% of chicken samples from Lidl supermarkets in Germany contained multiple drug-resistant bacteria, most of which are E. coli that can cause diseases such as urinary tract or gastrointestinal infections. The emergence of these drug-resistant E. coli has been linked to poor breeding conditions in chicken farms and the overuse of antibiotics in industrial animal husbandry.

ARGs in livestock and poultry breeding environments increase the risk of zoonotic disease transmission, and public health safety incidents that adversely affect human health are caused by drug-resistant bacteria in livestock and poultry breeding environments, which occur frequently in countries around the world.

3. Mechanism of production and transmission of ARGs

3.1. Genetic mutations

3.1.1. Spontaneous mutations

During the natural growth and reproduction of bacteria, there will be errors in DNA replication. DNA polymerase has a certain error rate during replication, and although the bacteria themselves have some repair mechanisms, there is still a small amount of error accumulation. For example, in Escherichia coli, the DNA replication error rate is approximately 1 error per 10^9 - 10^{10} base pair of replication. These errors can lead to changes in gene sequences, including those associated with antibiotic targets. If there is a mutation in the gene encoding the bacterial ribosomal protein, antibiotics (e.g., aminoglycoside antibiotics acting on the ribosomal 30S subunit) may not be able to effectively bind to the target, resulting in resistance.

3.1.2. Induce mutations

Genetic mutations are induced when bacteria are exposed to antibiotics, especially sublethal doses. The selective pressure created by antibiotics prompts adaptive mutations in bacteria. For example, it has been found that at low concentrations of quinolone antibiotics, the gene for bacterial DNA gyrase, which targets quinolone antibiotics, is mutated. This mutated DNA gyrase has a reduced affinity for quinolone antibiotics, allowing bacteria to resist the effects of quinolone antibiotics to some extent. For example, in intensive farming systems, low-dose antibiotic exposure often triggers these adaptive mutations, contributing to ARG persistence.

3.2. Vertical gene transfer(VGT)

Vertical gene transfer is the process by which bacteria directly pass genes from parents to offspring. In bacteria, reproduction is carried out through cell division, and the mother cell replicates and passes on its complete genome to daughter cells. In this process, if the mother cell contains antibiotic resistance genes, the daughter cells will also inherit these genes, so that the progeny bacteria will also become resistant to antibiotics.

3.3. Horizontal gene transfer(HGT)

3.3.1. Joint

Conjugation is the process by which bacteria transfer plasmids through cell-to-cell contact. Many antibiotic resistance genes are located on plasmids. For example, in Enterobacteriaceae bacteria, the donor bacteria are attached to the recipient bacteria through the sexual piria, passing the plasmid carrying the resistance gene to the recipient bacteria. As in some drug-resistant Escherichia coli, plasmids containing tetracycline resistance genes can be delivered to other susceptible bacteria by conjugation, enabling the recipient bacteria to express resistance proteins (such as efflux pump proteins or enzymes that inactivate antibiotics) and thus acquire resistance to tetracycline.

3.3.2. Transduce

Gene transfer mediated by bacteriophages. When a bacteriophage infects a bacterium, it packages a fragment of the bacteria's genome along with its own DNA. When phages re-infect other bacteria, they pass on DNA fragments from previous bacteria, which may contain antibiotic resistance genes, to the new host bacteria. In Staphylococcus aureus, some phages have been found to carry the mecA gene (a gene resistant to methicillin) that can be transduced to make other susceptible Staphylococcus aureus resistant to the drug.

3.3.3. Invert

Bacteria are able to take up free DNA from their surroundings. For example, in Streptococcus pneumoniae, DNA fragments are released when a bacterium dies and lyses. Other Streptococcus pneumoniae bacteria can absorb this DNA and, if it contains an antibiotic resistance gene, can be recombined into its own genome. Streptococcus pneumoniae has been shown to absorb DNA fragments containing the β-lactamase gene under laboratory conditions, thereby gaining resistance to β-lactam antibiotics.

3.3.4. Vesicle fusion

In addition to the above mechanisms, in recent years, studies have shown that the horizontal transfer of genes between bacteria by secreting antibiotic resistance genes in external vesicles and helping the continuous spread of ARGs in the environment has been identified as a new pathway for the horizontal transfer of ARGs, which is currently mainly found in Gram-negative bacteria.

4. Occurrence characteristics of ARGs among different animal breeds

4.1. Chicken

There is a wide range of subtypes of antibiotic resistance genotypes detected in chicken manure, typically 132-168, mainly aminoglycosides and tetracycline resistance genes. The number of resistance genes was the highest in the breeding stage, and the lowest in the prepartum stage. The total abundance of various resistance genes was the highest at the brooding stage, while the opposite was true at the end of parity. For example, in the study of Ningxia chicken farm, it was found that tetracyclines, aminoglycosides and sulfonamides were the dominant antibiotics in chicken manure, and the corresponding resistance genes were also more common. The number and relative abundance of resistance genes in the soil around the chicken farm gradually decreased with the increase of distance from the farm, but the number and relative abundance of resistance genes in the soil where chicken manure was applied increased significantly, but lower than that of the soil 20 m away from the farm, indicating that the application of chicken manure increased the content of resistance genes in the soil[5][6].

4.2. Pig

The abundance of antibiotic resistance genes and mobile genetic elements in pig manure is generally significantly higher than in chicken manure. Multi-drug resistance genes were dominant in pig manure after intramuscular therapeutic doses of enrofloxacin, and Clostridium and Lactobacillus were the main host bacteria of resistance genes in pig manure after intramuscular enrofloxacin. In the pig lower respiratory tract microbiome, γ-Proteobacteria are the main carriers of resistance genes, and some mobile genetic elements such as tnpA genes are widely distributed and closely related to a variety of resistance genes. The results showed that the increased resistance genes in the soil-plant system after pig manure application were mainly derived from pig manure-derived bacteria, and the community fusion process between pig manure and soil bacteria promoted the spread of resistance genes in the soil-plant system[7].

4.3. Goat

The dominant antibiotics in bovine feces were tetracyclines, quinolones and sulfonamides, and their resistance genes were also dominated by these types, and a total of 79-142 resistance gene subtypes were detected, among which aminoglycosides had the largest number of resistance genes. The resistance genes at the rearing stage were the most abundant and relatively abundant, while the resistance genes coexisting in all rearing feces at the calf stage were tetQ, ermF and tetO-01. Quinolones and tetracyclines were the main antibiotics in the soil around the farm, and the content of quinolones and tetracyclines in the soil applied to cow manure increased significantly, and the number and relative abundance of resistance genes were also significantly higher than those in other soils[8]. Comparatively, chicken farming showed a higher diversity of ARGs, dominated by aminoglycosides and tetracyclines, likely due to widespread use as growth promoters.

4.4. Comparison between different breeds

There were differences in the diversity of antibiotic resistance genes in the manure of different breeds, the diversity of resistance genes in chicken manure was relatively high, the abundance of resistance genes and mobile genetic elements in pig manure was relatively high, while the variety and abundance of resistance genes in cow manure were between chicken and pig, and the variety and abundance of resistance genes in goat's stomach were relatively small, but there was also a significant correlation between age and diet. There is a risk of horizontal movement of β-lactamase, aminoglycosides, and macrolide-lincolnamide-streptomycin B resistance genes in chicken manure; Both sulfonamide and chloramphenicol resistance genes in cow manure are at risk of horizontal movement in both cow manure and soil, suggesting that the transmission potential of resistance genes in manure varies between different breeds in the environment.

5. Occurrence characteristics of ARGs in different environmental media

5.1. ARGs in livestock and poultry manure

Livestock manure is considered to be an important reservoir and source of transmission of ARGs. Most of the antibiotics ingested by animals are excreted in feces and urine in the form of prototypes or metabolites. Feces contain high concentrations of ARGs, which may arise from unabsorbed antibiotics in the gastrointestinal tract, the proliferation of resistant bacteria, and resistance genes in the normal microbial community in animals. ARGs in feces can enter soil and water bodies through fertilization, washing, etc., and then spread widely in the environment. Studies have shown that the concentration of ARGs in pig manure is particularly high, due to the characteristics of the digestive tract of pigs and the types of antibiotics commonly used. The manure of other livestock and poultry such as chickens and cattle is also an important source of ARGs. Once ARGs in feces enter the environment, they can spread through a variety of pathways and have long-term effects on ecosystems[9-12].

5.2. ARGs in the soil environment

Soil is one of the important reservoirs of ARGs, and after livestock and poultry manure is applied as fertilizer, the organic matter rich in ARGs enters the soil ecosystem. Studies have shown that ARGs in soil are mainly derived from animal waste, agricultural antibiotics, and drug-resistant bacteria transmitted through horizontal gene transfer (HGT). Once these genes enter the soil, they can be acquired and transmitted by soil microorganisms such as bacteria and archaea. ARGs in soil have high persistence and complexity, and can migrate and spread in different soil layers. In addition, parameters such as soil properties, such as pH, organic matter content, and humidity, also affect the survival and propagation of ARGs. Because ARGs in the soil can persist for a long time and co-evolve with the soil microorganisms, soil becomes an important source of ARGs pollution, which further affects crop growth and food safety[13].

5.3. ARGs in the water environment

The water environment is another important medium for the spread of ARGs. Manure and wastewater from livestock farms are often discharged directly into nearby water bodies without adequate treatment, causing ARGs to enter rivers, lakes and groundwater systems. These ARGs further diffuse through runoff and osmosis, causing pollution to aquatic ecosystems. The study found that ARGs in the water body are mainly attached to suspended particulate matter and sediments in the water, which allows them to remain active and accumulate in the water body for a long time. In addition, aquatic animals such as fish and crustaceans may also carry and transmit ARGs. The presence of ARGs not only affects water quality safety, but also poses a potential threat to human health through the food chain. Therefore, monitoring and controlling ARGs in the water environment is essential for the conservation of water resources and the maintenance of public health[14].

5.4. Possibility of airborne transmission

Although airborne ARGs have received less attention than soil and water, aerosol transmission remains a potential pathway for ARGs to spread. Livestock farm activities such as spray cooling, ventilation and manure drying processes produce aerosol particles containing ARGs. These particles can be transported over long distances by the action of wind, which can spread ARGs to the surrounding area. In addition, dust storms and agricultural machinery operations can also raise ARGs-containing particles, further expanding their spread. Studies have shown that ARGs in the air can settle back to the ground under certain conditions and re-enter the soil or water body, forming a cross-media propagation cycle. Therefore, the potential for airborne transmission cannot be ignored, especially in areas with intensive agricultural and livestock production. Understanding and controlling ARGs in the air can help to comprehensively manage and mitigate the environmental impact of ARGs[15].

6. Conclusions and prospects

6.1. Summary of the study

The generation and spread of antibiotic resistance genes (ARGs) in livestock and poultry farming environments is a complex issue involving multiple links and factors. In this paper, we review the causes, transmission routes and environmental impacts of ARGs. Studies have shown that the high use of antibiotics in livestock and poultry farming is one of the main causes of ARGs. The misuse of antibiotics leads to the enrichment and spread of drug-resistant strains and their resistance genes, and spreads among different microorganisms through horizontal gene transfer (HGT). Livestock and poultry manure are important reservoirs and vectors for ARGs, polluting soil and water bodies through direct discharge into the environment or as fertilizer applied to farmland. In addition, airborne transmission is also one of the important ways for ARGs to spread. ARGs in the environment not only have a negative impact on ecosystems, but also pose a potential threat to human health through the food chain. Existing detection methods and control technologies can monitor and mitigate the spread of ARGs to some extent, but they still need to be further refined and developed to meet the changing challenges.

6.2. Future research directions and suggestions

Future research should focus on the following aspects: firstly, it is necessary to study the specific propagation mechanism of ARGs and their dynamic changes in different environmental media in order to predict their environmental behavior more accurately; Secondly, the research and development of new and efficient detection technologies should be strengthened, especially fast, sensitive and low-cost methods, to improve the monitoring efficiency. Thirdly, it is necessary to explore more scientific and reasonable control strategies, such as developing green and environmentally friendly alternatives and optimizing breeding models, so as to fundamentally reduce the generation and spread of ARGS. In addition, it is necessary to strengthen international cooperation to jointly address this global challenge, and promote greater progress in the prevention and control of ARGS pollution around the world by sharing data resources and technological achievements. Last but not least, it is important to raise public awareness of the importance of the rational use of antibiotics and to actively participate in prevention and control efforts to protect the safety and health of the environment on which we all depend. Only by working together can we effectively curb the harm caused by ARGS in the livestock and poultry breeding environment and ensure the long-term health of human beings and the earth.

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