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Fowl Typhoid: Present Scenario, Diagnosis, Prevention and Control Measures

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ABSTRACT

Poultry farming is facing complicated diseases including salmonellosis throughout the world. Avian salmonellosis renders heavy economic losses not only by causing high mortality (maybe up to 80%) but also by a reduction in meat and egg production. Salmonella has zoonotic importance. There are more than 2500 species of Salmonella, among these species important for poultry are *Salmonella Gallinarum, S. pullorum, S. Typhimurium, S. enteritidis, etc*. Among these *Salmonella Gallinarum* causes fowl typhoid (FT). FT affects mostly adult chickens resulting in greenish diarrhea and enteritis and high mortality. In the past, only antimicrobial drugs were used to control Salmonella in poultry which was causing also a threat to human beings as residues of the drugs were coming in meat and eggs. Day by day, antimicrobial resistance is developing against Salmonella in poultry, thus new ways and means are being searched for its control. Both live and inactivated vaccines have been in application against *S. Gallinarum*. Previously, Salmonella species were diagnosed by traditional ways of isolation, biochemical reactions, and microscopy. With the passage of time, there are new advancements in its diagnosis. Many techniques have been adopted for the detection of the causative agent. The PCR is useful for the rapid and correct diagnosis of Salmonella serotypes in poultry. Multiplex-PCR, Real-time PCR, PCR-RFLP assay, capillary gel electrophoresis (CGE), PFGE, indirect ELISA, and RAPD DNA are being used for diagnosis. Various strategies have been applied to curtail or control Salmonella infection in poultry. As Salmonella antibiotic resistance is developing, alternative treatment/prevention strategies are being opted for. Organic compounds such as prebiotics, probiotics, and phytobiotics are in use to limit salmonella growth. Organic compounds have several benefits to the chicken, and eggs and meats are free from residues and have public health safety. Omic technologies and bacteriophages as an alternative solution to control Salmonellosis have also been suggested.

Key words: Fowl Typhoid, Etiology, Present Scenario, Diagnosis, Prevention, Control Measures

INTRODUCTION

Commercial poultry production in Pakistan started in the 1960′s and has been playing a key role in decreasing the difference between the demand and supply of animal protein as well as providing an efficient source of income at a small scale (Gul and Alsayeqh, 2022). The poultry industry is an important part of the livestock sector and affords employment chances to over 1.5 million people in Pakistan. This sector has undergone extraordinary growth (7.3% annual growth rate) with investments of more than Rs 1,056 billion (Anonymous, 2022-2023).

Poultry meat is a cheap source of rich proteins (Saleem *et al*., 2022; Akhtar *et al*., 2023), however, so many diseases are hampering it such as bacterial, viral, parasitic, or fungal in origin (Mehmood *et al*., 2020; Soliman *et al*., 2021; Lebdah *et al*., 2022; Abu *et al*., 2022; Ahmed *et al*., 2022; Zaheer *et al*., 2022; Raza *et al*., 2022; Mehnaz *et al*., 2023; Qadir and Irum, 2023; Suardana *et al*., 2023). Poultry farming is facing complicated disease complexes out of which salmonellosis possesses an important place around the globe (Carole *et al*., 2021; Malik *et al*., 2021; Nupur *et al*., 2023).

Avian salmonellosis is one of the severe major septicemic bacterial diseases primarily involving the reticuloendothelial system affecting the poultry industry worldwide and responsible for heavy economic losses not only by causing high mortality but also a reduction in meat and egg production (Basit *et al*., 2022). Fowl typhoid (FT) and pullorum disease (PD) were identified in the 19th century and are amalgamated with high mortality (up to 100%) and declines in poultry production (Mshelbwala *et al*., 2017). Salmonella is a significant human pathogen.

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Poultry products such as eggs and meat are found to be a major cause of human infections (Jibril *et al*., 2020).

There are many salmonella species important for poultry *Salmonella Gallinarum;* S. *enteritidis, S. pullorum, Typhimurium, S. infantis, S. Montevideo*, and *S. Newport, etc* (Asif *et al*., 2017; Tchoupou-Tchoupou *et al*., 2022; Mahmood *et al*., 2022; Beylefeld and Abolnik, 2023; Rabie *et al*., 2023a). *Salmonella pullorum* and *S. Gallinarum* are liable for PD and FT of poultry, respectively (Rabie *et al*., 2023a; 2023b). FT affects mostly adult chickens resulting in either acute enteritis with greenish diarrhea or a chronic disease of the genital tract that causes a reduction in egg production. FT is an important disease of chicks and poults; however, growing and mature chickens and turkeys are also susceptible (Shivaprasad, 2000; Alves Batista *et al*., 2018). Some chicken strains are genetically more resistant or susceptible to Salmonella infection than other infections (Wigley *et al*., 2006). Young chicks during the first 4 days post-hatch are extremely vulnerable to Salmonella infection and become progressively resistant to Salmonella infection with age, however, infection becomes chronic in adult birds (Christensen, 1996).

Salmonella Gallinarum and *S. Pullorum* infect avian species and are considered to pose a zoonotic risk (Shivaprasad, 2000; Sanni *et al*., 2022). Salmonella causes localization in the intestines of chicken with *S.* Enteritidis and *S.* Typhimurium. The importance to control salmonellosis is because these serovars contaminate chicken meat, meat products, and eggs, thus gaining access to the human food chain to cause food-borne zoonotic salmonellosis in humans (Hossain *et al*., 2021; Tariq *et al*., 2022; Tohamy *et al*., 2022; Shakeel *et al*., 2023).

It was 1st occurred in chicken breeders in England and was called infectious enteritis killing after that Curtice studied the disease in Rhode Island Red and named it FT in 1902 (Lee *et al*., 2003; Berhanu and Fulasa, 2020). It is quite rare in the modern poultry industry of advanced countries because of the extensive serological testing and control of breeder birds. However, the disease has occurred in been reported in Asian, African, and South American countries (Lee *et al*., 2003). Increased mortality, off-feed, sudden drop in egg production, drowsiness, yellow or chalky white diarrhea with dehydration and soiled vent, and death are significant clinical signs of Salmonellosis (Berhanu and Fulasa, 2020). Fowl typhoid is endemic in Pakistan with infections leading to high mortality and substantial economic loss (Ayalew et al., 2017; Rehman, 2022). In this article, we reviewed the literature on the status of the prevalence of FT in the world especially in Pakistan and strategies to control, prevent, and diagnose disease in chickens.

Prevalence of Fowl Typhoid

Poultry health and welfare are very important for food supply and safety and significantly influence economic and societal prosperity. In many countries, official data regarding disease prevalence is poor as the diseases could be under-reported, thus, the probability incidence of PD and FT is underestimated (Zanetti *et al*., 2019; de Mesquita Souza Saraiva *et al*., 2022). Though has not been reported from Norway, Finland, Greece, and Estonia. FT has not been reported in Spain since 1991. The last occurrence of FT in Poland was reported in 1997. For many years Denmark has historically been free of FT until outbreaks in the 1990s. Salmonella is estimated to be liable for 1.35 million infections, 26,500 hospitalizations, and 420 deaths in the USA every year (CDC, 2023).

Pullorum disease and fowl typhoid, caused by *Salmonella enterica* subspecies *enterica* serovar *Gallinarum* biovars *Pullorum* (bvSP) and *Gallinarum* (bvSG), respectively, are among the most important diseases listed by World Organization for Animal Health (OIE) and have resulted in considerable economic losses to poultry industry over the world (Shivaprasad, 2000; Zhou *et al*., 2022). bvSP and bvSG have been under complete control or eradication programs for decades in several industrialized countries (Shivaprasad 2000; Wigley 2017; Schat *et al*., 2021). However, they are still common in many regions, particularly in developing countries (Wigley, 2017; Li *et al*., 2022; Zhou *et al*., 2022). *Salmonella Gallinarum*, a restricted pathogen of poultry causing fowl-typhoid in adult birds (Munir *et al*., 2023) and leads to up to 80% mortality (Ilyas *et al*., 2022) and has global distribution (Fig. 1). Zhou *et al*. (2022) analyzed data of *Salmonella Pullorum* variants (bvSP) and *Salmonella Gallinarum* (SG) variants (bvSG) from 1945 to 2021. According to them, the global prevalence of *S. Gallinarum* was 8.54% (95% CI: 8.43–8.65). Pullorum disease was most common in Asia (especially in China, India, and Pakistan) and South America (Argentina), though also present in North America, Africa, and Europe (Fig. 2).

Diagnosis of Fowl Typhoid

The OIE continuously reviews the old and new methods to identify causal organisms and disease surveillance. Many techniques have been adopted for detection. Methods based on the detection of bacterial DNA, such as PCR frequently being used now a day for showing the presence or the absence of bacteria in any sample with accuracy but with no obligation of showing live bacteria. In the same manner for the monitoring of the flock ELISA could be used, indicating whether birds have been exposed to the organism or not. Exploration of new techniques such as microarray is thought to be replacing the existing standard serotyping methods (Barrow *et al*., 2012).

Diagnosis of FT is carried out based on history, clinical signs, and postmortem lesions of the affected birds whereas confirmation of the diagnosis is validated by cultural examination, isolation, Gram's staining, and various biochemical tests, and then etiological agent identification by serotyping of *Salmonella* strains (Celis-Estupiñan *et al*., 2017). In mature birds, the infection is identified by serologic tests, positivity indicates that the flock has Salmonella infection (Patil *et al*., 2023). Suspected samples isolated from the diseased birds have usually been estranged from the results of biochemical reactions. An isolate can be contemplated of *S. Gallinarum* only if it provides a positive reaction in dextrin, maltose, dulcitol, and mucate. The variability shown by phenotypical characterization indicated that biotyping was a good tool for backtracing the disease. Through plasmid profiling, 85-kb plasmid in *S. Gallinarum* must be found to declare systemic disease (Pal *et al*., 2017).

The dulcitol fermentation test is performed to confirm the differentiation of *S. pullorum* and *S. Gallinarum*. The

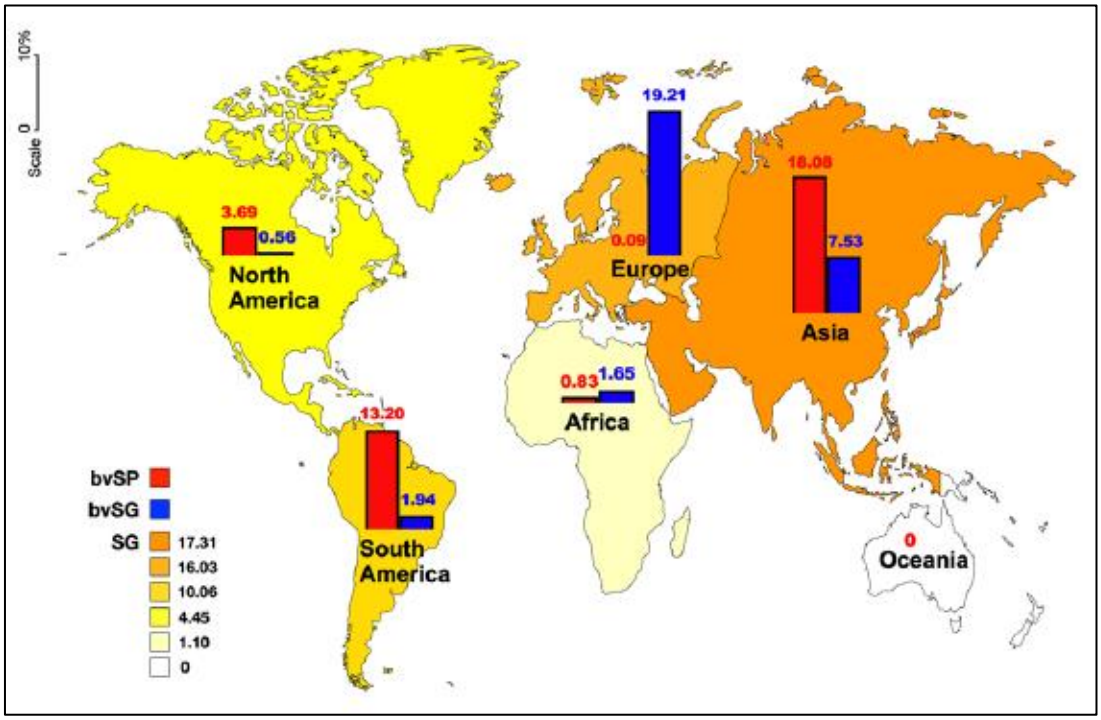

Fig. 1: Global distribution of Salmonellosis including bvSP, bvSG and SG (Zhou *et al.*, 2022).

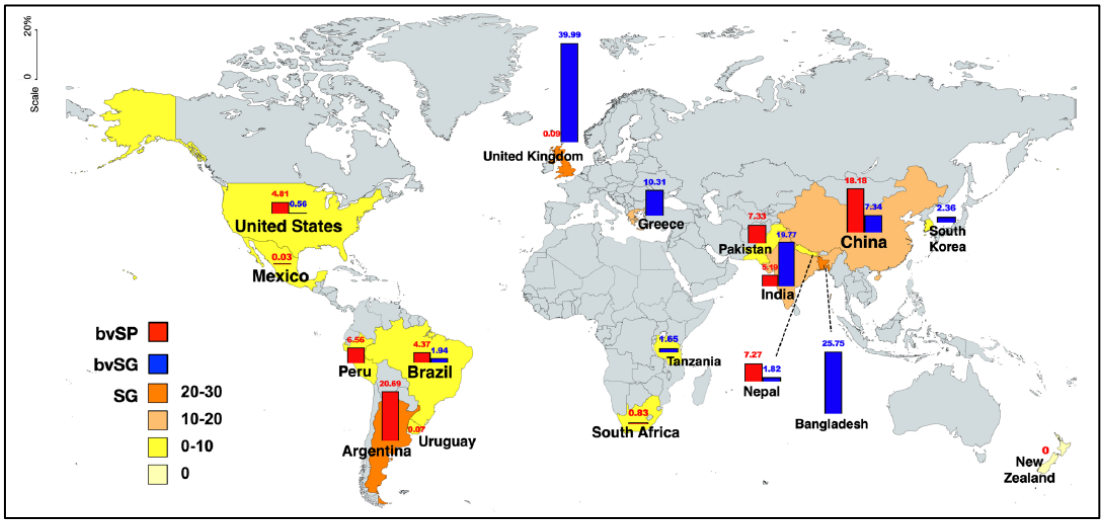

Fig. 2: Distribution of bvSP, bvSG and SG country-wise (Zhou *et al*., 2022).

whole blood plate agglutination test used under field conditions is rapid. Drawbacks include its unsteadfastness in ducks and turkeys as many uninfected birds may react false positive, so not in frequent use nowadays as new techniques are gaining popularity among progressive farmers because of more reliable results, though not cost-effective. Serum agglutination test is used in the laboratories. As microagglutination test is used for poultry especially, as it can provide false-positive findings with turkey sera, however, positive reactors can be confirmed by isolation/culture at necropsy (Mdegela *et al*., 2000).

Maiti *et al*. (2022) have formulated a single novel trivalent invasive non-typhoidal *Salmonella* (iNTS) serovars outer membrane vesicle (OMVs) based immunogen which can confer long-term broad-spectrum protection against the most prevalent iNTS serovars. They isolated OMVs from *Salmonella* Typhimurium, *Salmonella* Enteritidis, and *Salmonella Gallinarum* and used ELISA to measure immunogen.

The PCR is a useful tool for the rapid and definitive detection of avian Salmonella serotypes. Real-time PCR system for reliable and rapid genus and serovar-specific detection of *Salmonella* for monitoring purposes in the poultry (Szmolka *et al*., 2006) but PCR-RFLP assay has mainly been used to make a differential diagnosis of *S. pullorum* and *S. Gallinarum* (Hosen *et al*., 2019; Wen *et al*., 2021). The use of capillary gel electrophoresis (CGE) based on a*rfb*S allele-specific PCR for the analysis and simultaneous detection of *S. Gallinarum* is also giving good results (Jeon *et al*., 2007). Multiplex-PCR has successfully been used for the diagnosis of *Salmonella* spp (Samad *et al*., 2019).

The use of DNA-related techniques such as plasmid analysis (Pal *et al*. 2017), ribotyping (Usera *et al*.,1994),

PFGE (Zhang *et al*., 2021), an indirect ELISA using soluble whole-cell antigen (Berchieri *et al*.,1995) and molecular typing by RAPD DNA (Ji-Dong *et al*., 2006) have been demonstrated as a useful tool in differentiating Salmonella species.

Prevention and Control of Fowl Typhoid

Various strategies have been applied to curtail or control Salmonella infection in poultry. As Salmonella antibiotic resistance is developing day by day, thus the addition of prebiotics (Al-Khalaifah, 2018), probiotics (Khan and Chousalkar, 2020; Hameed *et al*., 2022; Rashid *et al*., 2023), and phytobiotics (Abdel-Wareth and Lohakare, 2014) via feed have been used to enhance food, growth promoters and to curtail Salmonella growth. El-Saadony *et al*. (2022) are also of the view to control salmonellosis in poultry by organic agents (Fig. 3).

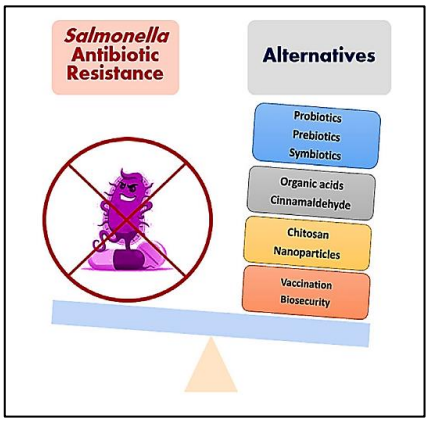

Fig. 3: As Salmonella spp. is developing antibiotic resistance, thus the use of organic compounds is increasing to curtail the growth of salmonella species (El-Saadony *et al*., 2022).

Organic compounds have several benefits to the chicken as well as egg and meats are free from residues (Fig. 4) and ultimately these organic compounds play a significant role in public health safety. Even thoughts are developing to use nanoparticles for vaccine delivery (Fig. 5) to fight against this bacterial species (Acevedo-Villanueva *et al*., 2021; Azam *et al*., 2022).

Fig. 4: Organic compounds fed to the chickens for safeguarding from salmonella infection have so many benefits to the poultry and also to public health.

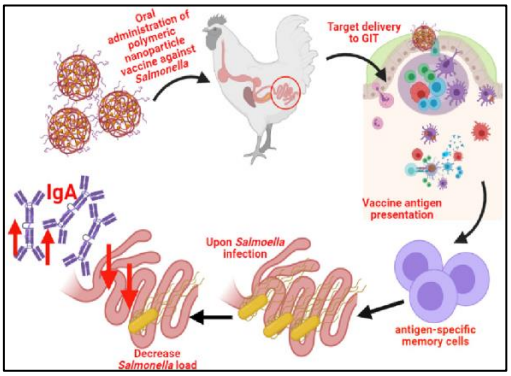

Fig. 5: Immune response of chicken upon the oral administration of biodegradable polymeric nanoparticles vaccine against Salmonella (Acevedo-Villanueva *et al*., 2021).

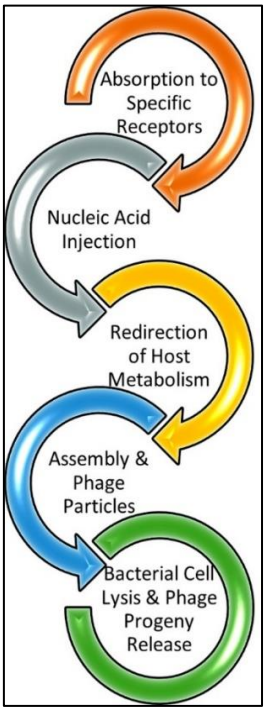

Fig. 6: Bacteriophages have various steps for the lysis of bacterial cells and the release of phage progeny.

Other than these, in ovo applications (Givisiez *et al*., 2020), bacteriophages (Wernicki *et al*., 2017; Jamil *et al*., 2023; Fig. 6), and vaccines (Hofacre *et al*. 2021) have been used. Omic technologies (Ruvalcaba-Gómez *et al*., 2022) are the latest tools in poultry to generate information that can result in tailoring medicinal policies and identifying patterns of resistance to antibiotics for lowering *Salmonella* presence and also costs of production (Vaid *et al*., 2021).

Vaccination use in poultry against Salmonella has been proven to be very beneficial in terms of reducing infection. Killed vaccines have proved to be very efficient but with the drawbacks that they may be destroyed and eliminated rapidly from the body, if not, may cause a poor immunogenic response in unprimed hosts and be unable to induce the production of cytotoxic T cells. There are many questions regarding the identification of the major protective immunogens and the nature of the immune response in the chicken yet to be answered so production of more efficient killed or subunit vaccine in future is

deemed up till now. Nevertheless, live vaccines are adventitious i.e., more effective, and have additional protective effects when administered orally by increasing lymphocyte proliferation (Hajam *et al*., 2018). Recently, Akter *et al*. (2022) used SG-9R Strain owl typhoid live lyophilized vaccine (Komipharma, South Korea) and reported 100% clinical protection and reduced the *Salmonella* shedding in the feces and eggs of the challenged birds.

According to Rehman (2022), currently, *Salmonella Gallinarum* infections in Pakistan poultry are controlled with antibiotics. The continued emergence of antibiotic resistance, however, has led to global initiatives to reduce the use of antibiotics in both human and veterinary medicine. Concurrently, the Pakistani government recently introduced new national policies that limit the use of antibiotics for performance in livestock and poultry production. As such, controlling bacterial infections in poultry without increasing the likelihood of antibiotic use could ensure the sustainability of Pakistan's poultry production without posing risks to public health. In this regard, Rehman (2022) has given an alternative solution to control Salmonellosis via bacteriophages. Rehman (2022) reported that phage concentrations were reduced to undetectable levels when exposed to SGF for more than 5 minutes. However, exposure to SIF did not result in appreciable reductions in phage concentrations. To mitigate the potential effects of gastric environments, phages were encapsulated using a sodium alginate-based method.

In developing countries, however, it is still a major threat; it could be controlled by adopting good management, hygiene, biosecurity, supervising, and properly eliminating infected flocks. The case is quite different in the developed countries where FT has been eradicated from commercial poultry in the USA, Wales, and the UK, because of the adaptation of test-and-slaughter methods of disease control under these surveillance programs, namely: the National Poultry Improvement Plan and the Poultry Health Scheme, respectively (Rabsch *et al*., 2000).

Another problem with poultry after an outbreak is that a large proportion of chicken that survives an outbreak becomes a carrier and transmits the disease to the next generation then the most efficient way of control is a combination of stringent management procedures and eradication (Lee *et al*., 2020).

Control strategies may face complications in the countries where the FT vaccine is being used continuously not to prevent infection but merely to reduce the clinical disease and allow the infected flock to continue its production. In this scenario, in some countries where FT has been eradicated the use of commercially available 9R vaccines has been banned. In ideal conditions, the aim should be the eradication of the organism which is obligatory to guarantee continued freedom from infection by culling the flock rather than acceptance of ongoing infection, but this is not often economical (Lee *et al*., 2020).

Conclusion

Avian salmonellosis renders heavy economic losses, may be high mortality, and also a reduction in meat and egg production. Salmonella has zoonotic importance. There are more than 2500 species of Salmonella, among these species important for poultry are *Salmonella Gallinarum, S. pullorum, S. Typhimurium, S. enteritidis, etc.* Among these *Salmonella Gallinarum* causes fowl typhoid (FT). FT affects mostly adult chickens, resulting in greenish diarrhea, enteritis, and high mortality. Day by day, antimicrobial resistance is developing against Salmonella in poultry, thus new ways and means are being searched for its control. Both live and inactivated vaccines have been in application against *S. Gallinarum*. The rough 9R strain is most widely used for the preparation of the vaccine.

Though traditional methods such as ways of isolation, biochemical reactions, and microscopy are in practice, however, many molecular techniques have been adopted, such as multiplex-PCR, Real-time PCR, PCR-RFLP assay, capillary gel electrophoresis (CGE), PFGE, indirect ELISA and RAPD DNA. As Salmonella antibiotic resistance, alternative treatment/prevention strategies are being opted for. Organic compounds such as prebiotics, probiotics, and phytobiotics are in use to limit salmonella growth. These organic compounds not only benefit the chicken and eggs and meats are free from residues and have public health safety. It is speculated that in the future, non-antibiotic therapies will gain importance for the control of Salmonella in poultry. Such organic compounds could be prebiotics, probiotics, and phytobiotics. Bacteriophages and omic technologies will gain importance as these are the latest tools in poultry to generate information that can result in tailoring medicinal policies and identifying patterns of resistance to antibiotics for lowering *Salmonella* presence and also costs of production.

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