Advancement in treatment, prevention and control strategies of avian pathogenic Escherichia coli : A literature review

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Abstract

Avian pathogenic *Escherichia coli* (APEC) poses significant health risks in poultry, leading to infections that impact productivity. APEC strains exhibit antibiotic resistance, complicating treatment options. Current vaccination strategies are limited in their protective scope, necessitating the exploration of innovative alternatives such as probiotics, bacteriophages, immune stimulants, and antimicrobial peptides (AMPs). AMPs show promise due to their rapid action against resistant bacteria and minimal resistance development. Additionally, small molecules have demonstrated effectiveness against various APEC serotypes, supporting the development of new antimicrobial therapies. Overall, a multifaceted approach addressing APEC's virulence factors and incorporating novel therapies is crucial for controlling colibacillosis in poultry and mitigating risks to human health.

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ABSTRACT

Key Clinical Message

Avian pathogenic *Escherichia coli* (APEC) poses significant health risks in poultry, leading to infections that impact productivity. APEC strains exhibit antibiotic resistance, complicating treatment options. Current vaccination strategies are limited in their protective scope, necessitating the exploration of innovative alternatives such as probiotics, bacteriophages, immune stimulants, and antimicrobial peptides (AMPs). AMPs show promise due to their rapid action against resistant bacteria and minimal resistance development. Additionally, small molecules have demonstrated effectiveness against various APEC serotypes, supporting the development of new antimicrobial therapies. Overall, a multifaceted approach addressing APEC's virulence factors and incorporating novel therapies is crucial for controlling colibacillosis in poultry and mitigating risks to human health.

2. DISCUSSION

2.1 Control and biosecurity measures

E. coli can spread between flocks mainly through dirty hatching eggs, so preventing contamination is crucial. Key steps include collecting eggs often, discarding damaged or soiled ones, and disinfecting eggs within two hours of laying. Cleaning the egg surface and using electrostatic sprayers can help reduce bacteria. UV light can kill *E. coli* without harming the eggs. Handling eggs carefully is important, as broken eggs can contaminate others. Good airflow in incubators and avoiding cross-contamination are also important. Chicks that may carry *E. coli* should be kept warm and fed by hand.[4,5] To control APEC infections in poultry, several strategies are essential. Antibiotics and vaccines are commonly used, but newer options like probiotics and bacteriophages are being explored. Other innovative treatments such as immune stimulants, virulence inhibitors, and antimicrobial peptides target the bacteria directly or strengthen the chicken's natural defenses. Minimizing stress in poultry is crucial—keeping ammonia and dust levels low, providing good airflow, and maintaining comfortable temperature, humidity, and space helps birds stay healthier. Vaccinating against certain diseases and ensuring balanced nutrition also boosts immunity, further protecting chickens from infection.[1,6] To prevent vertical APEC transmission in poultry breeding, breeders focus on developing resistant breeds, ensuring clean hatching eggs, and avoiding the use of eggs laid on the floor. Horizontal transmission is managed through controlled production cycles, removing weak chicks early, and maintaining rigorous sanitation. Strong biosecurity is vital—this includes chlorinating feed and water, disinfecting poultry houses and equipment, and restricting entry points for potential carriers like houseflies, wild birds, and rodents to keep APEC out of facilities.[1,6,7]

2.2 Antibiotics

APEC strains are resistant to most antibiotics, with only a few carbapenems still effective—although resistance to imipenem has started appearing. These strains often withstand drugs like ampicillin, tetracycline, trimethoprim, sulfamethoxazole, and streptomycin. The high resistance to critical antibiotics, especially β-lactams and colistin, raises serious concerns about spreading these resistant bacteria and genes to humans through the food chain.[8] The US and EU restrict non-therapeutic antibiotic use for growth and limit critical antibiotics in animal farming to help reduce antibiotic resistance risks.[9] A study showed chickens treated with enrofloxacin showed better feed efficiency, lower death rates, healthier organs, and less bacterial presence than those given oxytetracycline. Oral enrofloxacin was especially effective, providing strong protection within 2 hours and lasting all day.[10] Colistin, a last-resort antibiotic for Gram-negative infections, becomes more effective and less prone to resistance in *E. coli* when combined with small molecule adjuvants that target the pmrAB system.[11]

2.3 "No Antibiotics Ever" Broiler Production Strategy

Pathogenic *E. coli* is present in both no antibiotics ever (NAE) broilers and their environment, which can increase the likelihood of colibacillosis outbreaks, particularly when birds are under stress. NAE programs can lower feed efficiency, slow growth, and negatively impact gut health, ultimately reducing overall poultry production. Research shows that raising broilers without antibiotics can lead to more pathogens, higher stress levels, and poorer growth. Additionally, stress makes birds more vulnerable to infections like colibacillosis, highlighting the challenges of NAE approaches.[12,13]For decades, antimicrobial growth promoters were the primary approach to managing APEC, but the move to NAE production has resulted in more cases of colibacillosis. The prevalence of APEC-like virulent strains tends to be high overall, but it varies with the seasons, reaching peak in spring and dropping during the hotter months. Key environmental factors such as temperature, humidity, and housing conditions significantly influence APEC levels.[12,13]

2.5 Probiotics and Prebiotics

Beneficial live microbes, known as probiotics, play a key role in preventing infections, while prebiotics serve as non-digestible ingredients that encourage the growth of healthy gut bacteria. Research indicates that including Lactobacillus plantarum B1 in the diets of broilers enhances levels of ileal mucosal secretory IgA and decreases pro-inflammatory cytokines. This not only improves growth performance but also reduces cecal *E. coli* counts. Specifically, L. plantarum B1 has been effective in lowering *E. coli* levels and bolstering immune responses.[20,21] Other strains, such as Lactobacillus plantarum 15-1 with fructooligosaccharides and

Enterococcus faecalis-1, have also shown benefits, resulting in reduced mortality and improved immunity against APEC infections. A combination of multiple probiotic strains was able to eliminate deaths from APEC O78 and decrease bacterial levels in the liver and spleen. Furthermore, a commercial probiotic tested alongside a recombinant Salmonella vaccine demonstrated potential in enhancing both growth and immunity in poultry.[2]

2.6 Bacteriophages

The effectiveness of phage mixtures against APEC infections in chickens was tested using various methods of administration. Phages SPR02 and DAF6 significantly lowered mortality rates when given before or 24-48 hours after the APEC challenge.[22] An oral or spray phage cocktail (phi F78E, phi F258E, phi F61E) reduced mortality in both experimental and natural infections. Another intramuscular cocktail (TM1, TM2, TM3, TM4) also decreased mortality, lowered APEC levels in the lungs, and improved body weight. Additionally, phage-loaded chitosan nanoparticles (C-ΦKAZ14 NPs) given orally reduced mortality and intestinal APEC levels while improving health and weight. Overall, these studies show that bacteriophages could be a promising way to control APEC infections in chickens, especially with the C-ΦKAZ14 NP treatment.[2,22]

2.8 Virulence inhibitors

Virulence inhibitors function by disabling or weakening pathogens instead of just stopping their growth like antibiotics. They target virulence mechanisms, such as the quorum sensing (QS) system. These inhibitors can overcome issues associated with traditional antibiotics, like antibiotic resistance and harm to helpful bacteria, while making pathogens more vulnerable to the host's immune system. This makes them a promising alternative to standard antibiotics.[2,23]

2.9 Growth Inhibitors

Growth inhibitors targeting bacterial membranes are promising new antibacterial agents with a lower risk of resistance. Baicalin, from*Scutellaria baicalensis* , reduced mortality and lung damage in chickens with APEC lung injury. Rutin lowered AI-2 secretion, reduced biofilm formation, and protected chicken lung cells. Additionally, andrographolide from *Andrographis paniculata* lowered inflammation in chicken lung cells.[11,24] Small molecules (SMs) are small compounds (200–500 Da) that can stop bacterial growth and enzymes. They work against different APEC types, including antibiotic-resistant ones, without harming eukaryotic cells. These findings support new treatments for APEC infections in poultry, which could also lower human ExPEC infections and reduce antibiotic resistance. Screening a small molecule library found several APEC growth inhibitors that effectively killed bacteria at low doses, improved survival in wax moth larvae, and reduced APEC levels.[2,11,24]

2.10 Antimicrobial peptides

AMPs, also known as host defense peptides, are short, positively charged proteins found in many organisms, including humans. They have the ability to directly eliminate harmful microbes or enhance the body's immune response. With the growing issue of antibiotic resistance, AMPs are being recognized as potential therapeutic solutions. They provide quick and targeted action against bacteria that are resistant to traditional antibiotics and have a low risk of developing resistance, positioning them as excellent options for antibacterial treatments.[25] Bacteria often produce AMPs to fight off rival bacteria within their environment.[26]Gram-positive and Gram-negative bacteria have membranes with negatively charged lipids that attract the positively charged AMPs. In Gram-negative bacteria, AMPs first need to breach the outer membrane, which is strengthened by cations like calcium and magnesium attached to lipopolysaccharides. AMPs likely use a method where they displace these cations, creating temporary openings in the outer membrane for entry.[25,27] Antimicrobial peptides (AMPs) fight bacteria mainly by interacting with their membranes in different ways. Some peptides insert into the membrane to create pores, while others coat the membrane surface and disrupt it like a detergent when they reach a high concentration. Additionally, some peptides cause the membrane to bend around a central opening. AMPs can also block the creation of cell walls and inhibit the production of proteins or genetic material. Among the most studied AMPs are insect-derived peptides, particularly cecropins, which effectively target bacteria like *E. coli* by damaging their membranes.[28] AMPs causes ion leakage and depolarization that lead to membrane dysfunction and rapid cell lysis.[29] Although bacteria are less likely to resist AMPs due to their multiple targets, recent studies show that resistance can develop under selective pressure, and AMPs also face challenges due to low stability and high production costs.[25,30] In ovo treatment with D-CATH-2 reduced APEC mortality and bacterial load in chickens. Combining surfactin with amoxicillin enhanced its effectiveness, lowering mortality and APEC levels while boosting cytokine genes. Peptides like A3 and cecropin A-D-Asn also decreased*E. coli* in the chicken gut, suggesting antimicrobial peptides as alternatives or supplements to antibiotics in poultry.[2,31]

3. CONCLUSION AND FUTURE PROSPECTIVES

APEC's diverse virulence factors interact to cause systemic infections in poultry, necessitating a comprehensive strategy that targets iron acquisition, quorum sensing, bacterial metabolism, and secretion systems for effective therapeutic development. APEC strains, especially ST95, ST131, and serogroups O1, O2, and O18, pose a risk of extra-intestinal infections in humans. With rising antibiotic resistance and the potential for transmission of resistant bacteria and genes to humans, developing antibacterials for animals that avoid cross-resistance with existing antibiotics is essential. An effective APEC vaccine offering cross-protection against multiple serotypes is also needed, and insights into APEC's virulence should guide the identification of new vaccine candidates. Additionally, alternative therapies, such as small molecule inhibitors and antimicrobial peptides targeting novel pathways, should be explored to control colibacillosis in poultry.

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Priyanka Devkota : Conceptualization; data curation; supervision; validation; visualization; writing – original draft; writing – review and editing.

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Bhuminanda Devkota: Supervision; validation; visualization; writing- review and editing

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