THE SCIENCE WORLD
A Monthly e Magazine
ISSN:2583-2212
Short Communication
January, 2026 Vol.6(1), 106-110

Bacteriological Isolation and Identification of Potential Pathogens Associated with Swollen Head Syndrome in Broiler Chickens

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DOI:10.5281/ScienceWorld.18358917

Abstract

This study was carried out to isolate and identify potential bacterial pathogens from broiler birds affected with swollen head syndrome in and around Anand, Gujarat. Over a six-month period, 40 swab samples were collected from 20 commercial broiler farms showing clinical signs of Swollen head syndrome. *Escherichia coli* was detected in all twenty flocks examined. In total, 83 bacterial isolates were recovered, comprising *Escherichia coli* (34.94%), *Staphylococcus aureus* (32.53%), *Proteus* spp. (6.02%), *Corynebacterium* spp. (7.23%) and other Gram-negative rods (19.28%). Among these, *E. coli* emerged as the most prevalent pathogen associated with swollen head syndrome in broilers during the study period.

Keywords: Broilers, Bacterial isolation and identification, Swollen head syndrome

INTRODUCTION

The Indian poultry industry has shown remarkable growth and now represents a major segment of the agro–animal industry. Swollen head syndrome (SHS) is considered a complex, multifactorial disease in poultry, with viral infections initiating the condition and secondary bacterial involvement exacerbating lesions. SHS is regarded as a localized form of

colibacillosis and is characterized by inflammation of the periorbital and surrounding subcutaneous tissues of the head (Barnes & Gross, 1997). *Escherichia coli* has been frequently isolated from multiple organs, including the brain and middle ear, during SHS outbreaks in broilers and breeders with pneumovirus exposure (Pattison *et al.,* 1989).

Avian pneumovirus, classified under *Pneumoviridae*, genus *Metapneumovirus*, plays a significant role in respiratory diseases of poultry (Cook and Cavanagh, 2002). Avian metapneumovirus is the causative agent of turkey rhinotracheitis and is also associated with swollen head syndrome in chickens. Additionally, *Mycoplasma gallisepticum* and *Mycoplasma synoviae* act as important co-infecting agents, intensifying respiratory disease severity and contributing to chronic respiratory disease and air sac pathology (Lockaby *et al.,* 1999).

MATERIAL AND METHODS

The study was conducted on broiler flocks affected with swollen head syndrome in and around Anand district of Gujarat, India. A total of twenty broiler flocks were investigated during disease outbreaks. From these flocks, forty samples comprising swabs from subcutaneous edema and sinus exudates were collected. All samples were collected aseptically using sterile cotton swabs to avoid contamination. The samples were inoculated onto MacConkey agar and blood agar and incubated aerobically at 37°C for 24 hours. After incubation, the plates were examined for bacterial growth, and representative colonies were further subcultured onto selective media.

Bacterial identification was performed based on colony morphology, Gram staining, cultural characteristics, and biochemical reactions in accordance with Bergey's Manual of Determinative Bacteriology. Microscopic examination revealed gram-negative, pink-stained bacilli indicative of *Escherichia coli*. The isolates were further confirmed by IMViC tests comprising Indole, Methyl Red, Voges–Proskauer, and Citrate utilization tests. The results were subjected to descriptive statistical analysis.

RESULTS AND DISCUSSION

The occurrence of the bacterial isolates is presented in Table 1. A total of 83 bacterial isolates representing different genera were identified: *Escherichia coli* 29 (34.94%), *Staphylococcus aureus* 27 (32.53%), Gram negative rods 16 (19.28%), *Corynebacterium* spp. 6 (7.23%) and *Proteus* spp. 5 (6.02%). Results suggest that *Escherichia coli* are the predominant pathogen responsible for causing swollen head syndrome in broilers.

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Published 16/01/2026

Table 1. Bacterial isolates from SHS-affected broiler samples

Bacteria	Isolates (%)
Escherichia coli	29 (34.94)
Staphylococcus aureus	27 (32.53)
Gram-negative rods	16 (19.28)
Corynebacterium spp.	6 (7.23)
Proteus spp.	5 (6.02)
Total	**83 (100.00)**

Table 2. Cultural features of bacterial isolates from SHS-affected broilers

Isolate	Blood agar	*MacConkey agar*	Eosin Methylene Blue (EMB) Agar	Mannitol salt agar
E. coli	Greyish-white, NH	Pink LF	Greenish metallic sheen	–
S. aureus	Yellowish-grey, mucoid	–	–	Yellow (mannitol +) / pink (mannitol –)
Proteus spp.	Swarming	–	–	–
Corynebacterium spp.	Small white	–	–	–
Gram-negative rods	White opaque	Pale NLF	–	–

NH = non-haemolytic; LF = lactose fermenting; NLF = non-lactose fermenting

Isolation results showed that *Escherichia coli* was present in all twenty flocks studied, while *Staphylococcus aureus* was identified in sixteen flocks. Gram-negative rods, *Proteus* spp., and *Corynebacterium* spp. were recovered from eleven, four, and four flocks, respectively. Nineteen flocks were affected by mixed bacterial infections, and only one flock showed a pure *E. coli* infection. Sample-wise distribution indicated that mixed infections were present in 33 of the 40 samples. Single bacterial isolates were detected in only seven samples, with *E. coli*, *Staphylococcus aureus*, Gram-negative rods, and *Proteus* spp. isolated individually in 2, 2, 2, and 1 samples, respectively. Mixed infections involving *E. coli* were predominant.

The involvement of *Escherichia coli* as a secondary bacterial pathogen in the development of swollen head syndrome has been documented by several researchers. Georgiades *et al.* (2001) reported *E. coli* as the most frequently isolated organism from SHS cases, followed by *Staphylococcus aureus*. Similarly, Radwan *et al.* (2018) and Abdelmoez et al. (2019) described mixed bacterial infections in SHS-affected flocks, including *Escherichia coli*, *Streptococcus dysgalactiae*, *Pseudomonas aeruginosa*, and *Proteus mirabilis*. In

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contrast, the present study identified *E. coli* as the predominant isolate followed by *Staphylococcus aureus*, while *Streptococcus* spp. were not detected.

Five different bacterial isolates were recovered during the present study. Their identification was based on colony morphology observed on different culture media, as summarized in Table 2. Greyish-white colonies on blood agar were identified as *Escherichia coli*, while yellowish-white non-haemolytic colonies indicated *Staphylococcus aureus*. Swarming growth, small white colonies, and white opaque colonies were characteristic of *Proteus* spp., *Corynebacterium* spp., and Gram-negative rods, respectively.

E. coli produced metallic sheen colonies on EMB agar and lactose-fermenting pink colonies on MacConkey agar. On Mannitol Salt Agar, *Staphylococcus aureus* showed yellow colonies due to mannitol fermentation or pink colonies in non-fermenting strains. The IMViC reactions for *E. coli* and *Proteus* spp. were (+,+,−,−) and (−,+,−,+), respectively, which agree with previous observations.

CONCLUSION

Escherichia coli was identified as the predominant bacterial pathogen in broiler flocks affected with swollen head syndrome. Other bacteria, including *Staphylococcus aureus*, *Proteus* spp., *Corynebacterium* spp., and Gram-negative rods, were also isolated, indicating the polymicrobial nature of the disease. These findings emphasize the importance of targeted bacterial management to control SHS in broiler farms.

ACKNOWLEDGEMENT

Authors Thank the Dean of Veterinary College and authorities of Anand Agricultural University, Anand for the fund and facilities provided for this research work.

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Pattison *et al.* (1989) carried out serological examination of *avian metapneumovirus (AMPv)* in boiler birds. Sera were collected from Ninety-nine broiler flocks were tested using commercial ELISA kit of which only 20 were serologically positive for *avian metapneumovirus (AMPv)* antibodies.

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Published 16/01/2026

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