Antimicrobial resistance of E. *coli* isolated from commercial layer and backyard chicken farms from two Veterinary Divisions in Kalutara district

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Introduction

Issues relating to use of antimicrobials is increasing globally, with common bacteria being capable of the rapid development of antimicrobial resistance (AMR). Over the past several years the use of antimicrobials in the poultry industry as means of medication or poultry feed has come under heavy criticism. Therefore, use of antimicrobials in poultry production has been greatly restricted in Sri Lanka. However, the usage pattern may be influenced by farm management practices.

Poultry industry is the fastest growing livestock sub-sector in the country. In their review article, Manjula *et al.* 2018, describe the poultry industry in Sri Lanka as being divided into the production of commercial and backyard (indigenous) poultry. The management practices related to commercial layer operations and backyard system are significantly different. Generally, in commercial layer farms, hundreds or sometimes thousands of birds are reared under intensive management system and fed with commercially available feed. On the other hand, backyard poultry production system which is an extensive management, provides shelter but animals are free to move around and depend on scavenging. Locally adapted indigenous chicken or village chicken are reared in backyard farms and their production is lower than that of commercial exotic breeds. According to available statistics backyard poultry production contributes about 5% to the national egg production but there is a high demand for these eggs.

Even though poultry diseases are reported under any type of management, disease occurrence in free range- backyard birds is lower than that seen in intensive – commercial layer production, probably due to natural resistance (Weerasooriya *et al*., 2017). Accordingly, commercial layers appear to be more prone to get exposed to antimicrobials in comparison to backyard chicken. Considering the above facts, the aim of the current study was to compare colonization of antimicrobial resistant *E.coli* in commercial and backyard layer chickens in an identified area.

Methodology

Twenty five backyard farms and all registered commercial layer farms (n=4) located within the selected area, belonging to two adjacent veterinary divisions from Kalutara district (Bandaragama and Millaniya), were included in to the study. Information on management

practices including housing, feeding patterns, and history of antimicrobial usage was gathered by interviewing the farmers with the use of a structured questionnaire.
Cloacal swabs were collected from selected five birds in each selected farm and pooled before testing. Each pooled sample was directly streaked on MacConkey agar plates and five presumptive *E.coli* colonies from each pooled sample were selected and sub cultured on Nutrient agar. Isolates were confirmed as *E.coli* using an array of biochemical tests (Triple Sugar Iron agar (TSI), Citrate, Urease and Indole). All the confirmed isolates were subjected to antimicrobial susceptibility testing (AST) using disk diffusion method as described by the Clinical Laboratory Standards Institute (CLSI, 2013) guidelines.

Results

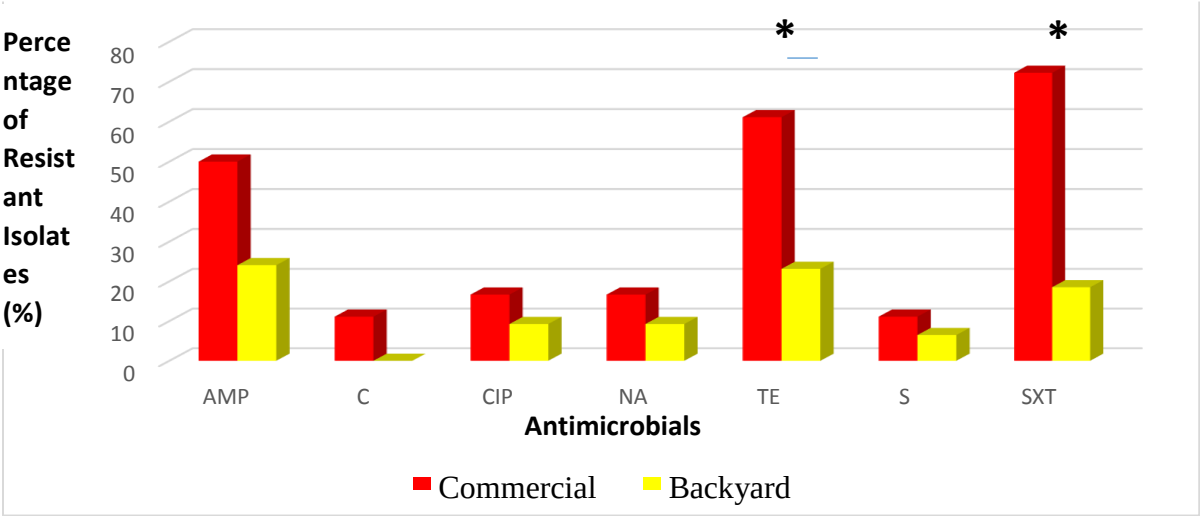
According to the findings of questionnaire survey, mean flock size of a commercial layer farm was 1625 (±478). Deep litter management system and commercial feed were used in these farms and there were several batches within a farm. Further, all farms had history of using antimicrobials. In backyard farms, size of a flock was less than 10 birds, except in two farms. Scavenging was the feeding practice in 13 backyard farms where as in other farms commercial feeds were routinely provided along with scavenging. Twenty four percent (6/25) of farms had used antimicrobials at least once.

Table 1: Resistance profiles of *E.coli* isolated from commercial and backyard farms.

Antimicrobials tested	**Percentage (%)of resistant isolates (Resistant isolates/total no. of isolates)**		
	Commercial Layer (n=18)	**Backyard (n =110)**	
		Commercial feed+ Scavenging	**Scavenging only**
tetracycline	61(11/18)	50 (22/44)	6.1 (4/66)
ampicillin	50 (9/18)	47.7 (21/44)	18.2 (12/66)
sulfamethoxazole - trimethoprime	72 (13/18)	45.5 (20/44)	3 (2/66)
nalidixic acid	27 (5/18)	20.5 (9/44)	3 (2/66)
ciprofloxacin	17 (3/18)	15.9 (7/44)	4.5 (3/66)
streptomycin	83 (15/18)	65.9 (29/44)	62.5 (41/66)
amikacin	0	0	3.03 (2/66)
gentamicin	0	0	0
chloramphenicol	11 (2/18)	0	0
imipenem	0	4.5 (2/44)	0
ceftazidime	0	0	0
ceftriaxone	0	0	0
cefotaxime	0	0	0

Laboratory findings revealed presence of *E.coli* resistant to antimicrobials, colonizing in chickens reared under commercial as well as backyard system. Based on the feeding management practices farms were categorized into three groups and considerable distinction could be seen among the resistance profiles of *E.coli* among the three groups. (Table 1).

Two proportion analysis ($p < 0.05$) revealed, a significantly higher proportion of *E.coli* isolates resistant against tetracycline and sulfamethoxazole-trimethoprim from commercial layers (Figure 1). Furthermore, the same statistical analysis ($p < 0.05$) was used to compare the resistance profiles of *E.coli* isolated from the backyard systems that are practicing two different feeding patterns as mentioned in the table 1. The analysis revealed that the level of resistance seen in *E.coli*, isolated from backyard birds fed with commercial feed, against streptomycin, ampicillin, sulpha-trimethoprime, nalidixic acid and ciprofloxacin, was significantly higher compared to backyard birds that solely depend on scavenging.



(*) show significant difference

Figure 1: Comparison of percentages of antimicrobial resistant *E. coli* isolates from backyard and commercial layers

Discussion

The present study revealed antimicrobial resistance profiles of *E. coli* isolated from poultry reared under commercial and backyard farming systems.

E. coli isolates were mainly resistant against commonly using antibiotics such as ampicillin, sulphamethoxazole-trimethoprim, tetracycline, ciprofloxacin and streptomycin. None of the isolates was resistant against third generation cephalosporins and gentamicin but even two isolates each showing resistance against chloramphenicol and imipenem was a concern. The resistance against tetracycline and sulfamethoxazole – trimethoprim was significantly higher in *E.coli* isolates recovered from commercial layers in compared to that were from backyard poultry. These antimicrobials are commonly used in poultry sector for disease control. Use of some antimicrobial such as sulfamethoxazole – trimethoprim as coccidiostats (Mehdi *et al.*, 2018) has been reported.

It was interesting to note that farms which provide commercial feed having higher number of resistant isolates compared to backyard farms which depend on scavenging (Table 1). Due to the fact that higher numbers of resistant isolates were reported from backyard farms providing commercial feed, a possibility of feed acting as an agent of introducing resistant bacteria or antimicrobials cannot be ignored, even though the antimicrobial usage in poultry has been strictly regulated since 2018. Nevertheless, resistant *E.coli* was isolated from chicken reared in backyard farms that had no history of antimicrobial usage and not providing commercial feed. This could be due to widespread contamination of the environment. Moreover, misuse of antimicrobials in some backyard farms was noted despite the fact that the farmers cannot purchase antimicrobials without a prescription from a veterinarian.

Previous studies conducted in different countries have reported, use of antimicrobials to enhance animal productivity as feed additives leading to AMR. Other than that various factors such as breed of the animals, husbandry practices and hygienic practices have been reported as contributory factors for spreading resistant bacteria in poultry. Even though we have not studied above variables, results of the present study highlight differences between commercial and backyard systems and possible influences of generally unexpected management practices in backyard farming.

Importantly, *E. coli* is considered as a transmitter of resistant genes to other pathogens. Since this study shed light on the colonization of resistant *E.coli* in both layer management systems, commonly practiced in the country, broader studies are proposed to identify risk factors introducing antimicrobial resistant strains/genes to poultry farms.

Conclusions

E.coli, resistant against commonly available antimicrobials are present in association with layer chickens managed under commercial and backyard production systems. Further, there were considerable differences in resistance profiles of *E.coli* isolated from backyard farms practicing scavenging only and providing supplementary commercial feeds.

Reference

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