Evolution of Antimicrobial Resistance and Virulence in Avian Pathogenic *Escherichia coli*: A Dual Threat to Animal and Human Health

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INTRODUCTION & AIM

Colibacillosis is a widespread bacterial disease in poultry, primarily caused by Avian Pathogenic *Escherichia coli* (APEC). The pathogenicity of these strains is linked to various virulence factors. The intensive and often unregulated use of antibiotics in poultry farming has significantly contributed to the emergence of antibiotic-resistant APEC strains. The rise of this multidrug-resistant (MDR) isolates is alarming, as it threatens animal and public health.

Objectives:

➢ Isolation and identification of APEC strains.

➢ Evaluation of antimicrobial resistance profiles;

➢ Detection and confirmation of colistin resistance;

➢ Characterization of virulence phenotypes;

➢ Investigation of the transferability of resistance genes;

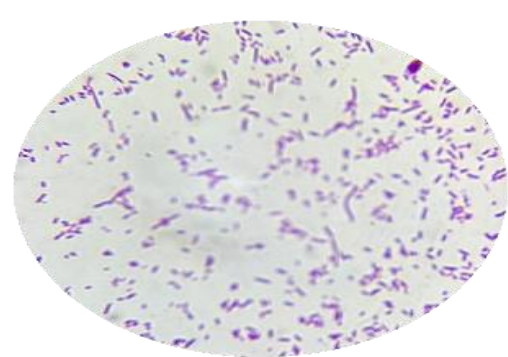➢ Assessement of correlations between virulence and resistance.

METHODS

1- Isolation and identification of *Escherichia coli*
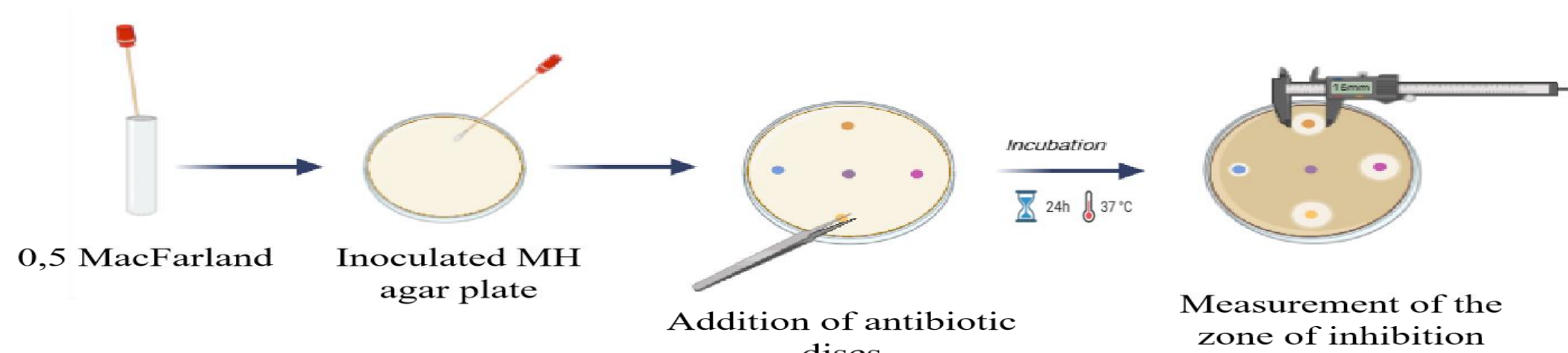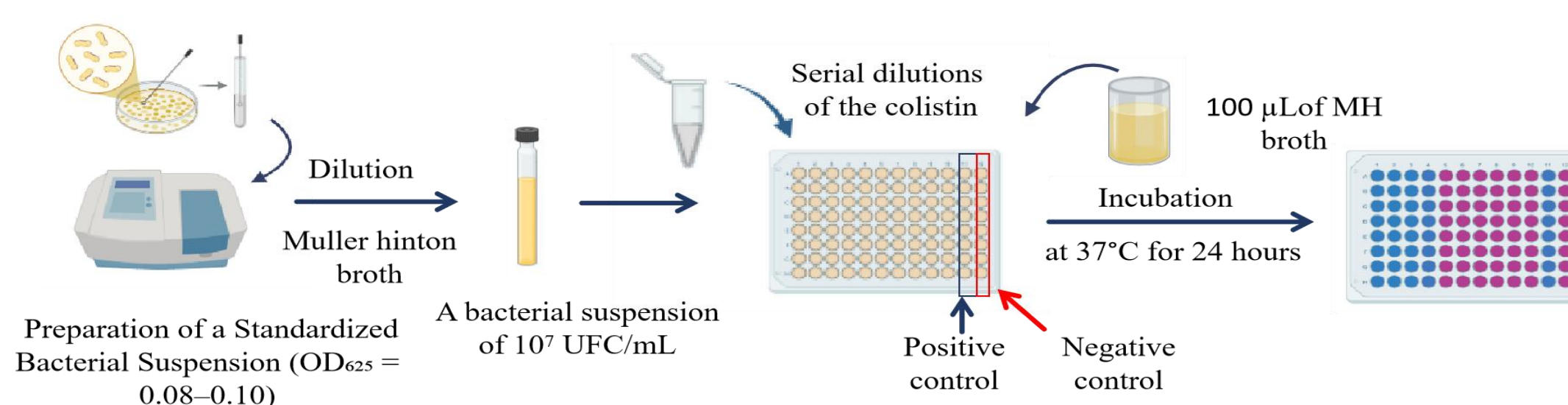
Identification
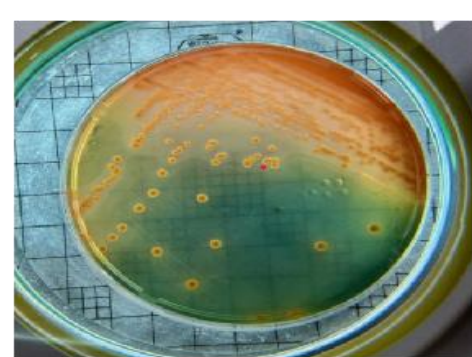
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2- Antibiotic susceptibility testing (CLSI 2023) and Colispot test

0,5 MacFarland → Inoculated MH agar plate → Addition of antibiotic discs → Incubation 24h 37°C → Measurement of the zone of inhibition

3- Determination of antibiotic MICs (CLSI 2023)

Dilution → Muller hinton broth → A bacterial suspension of 10^7 UFC/mL → Serial dilutions of the colistin → 100 µL of MH broth → Incubation at 37°C for 24 hours → Positive control / Negative control

Preparation of a Standardized Bacterial Suspension (OD_{625} = 0.08–0.10)

4- Conjugation assays: Conducted to assess the transferability of antibiotic resistance genes **(BAKOUR et al., 1983).**

5- Virulence tests

➢ Congo red binding was assessed on TSA supplemented with 0.02% Congo red dye **(AL-SAIEDI et AL-MAYAH, 2014).**

➢ Hemolysin production was evaluated on Columbia agar supplemented with human blood **(AL-SAIEDI et AL-MAYAH, 2014).**

➢ Serum resistance was determined using the spot test technique on Mueller-Hinton agar. **(Fierer *et al*., 1972).**

6- Statistical analysis : The chi-square test was used to determine the presence of a significant correlation between virulence factors and antibiotic resistance.

RESULTS & DISCUSSION

➢ Among the 104 avian isolates tested, only 50 were identified as *Escherichia coli* based on species-specific identification criteria.
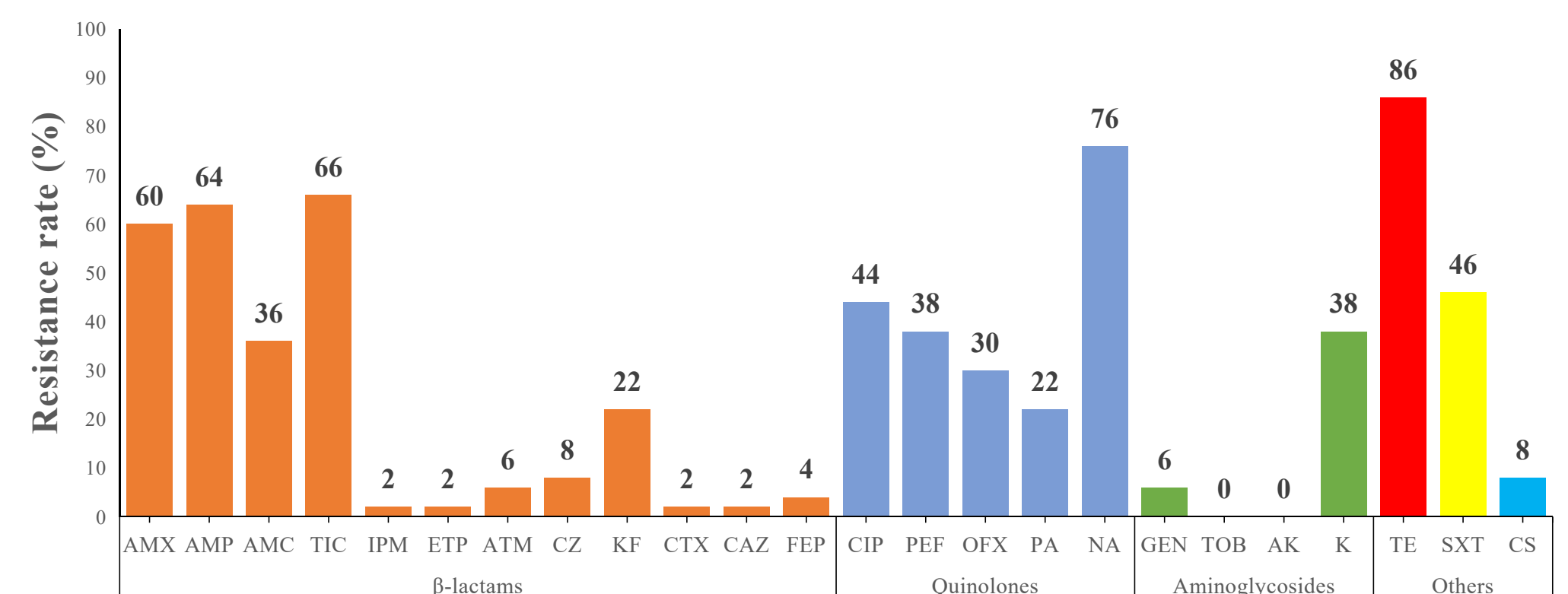

Figure 1. Graphical Representation of Antibiotic Resistance Rates in *Escherichia coli* strains.

➢ Among all resistant strains, 98% exhibited multidrug resistance, ranging from resistance to 3 to 18 antibiotics, with respective rates of 16.67% and 4.17%.

➢ Colistin resistance was detected in 8% of APEC isolates by the Colispot test and confirmed by MIC determination, with values exceeding 2 µg/mL.
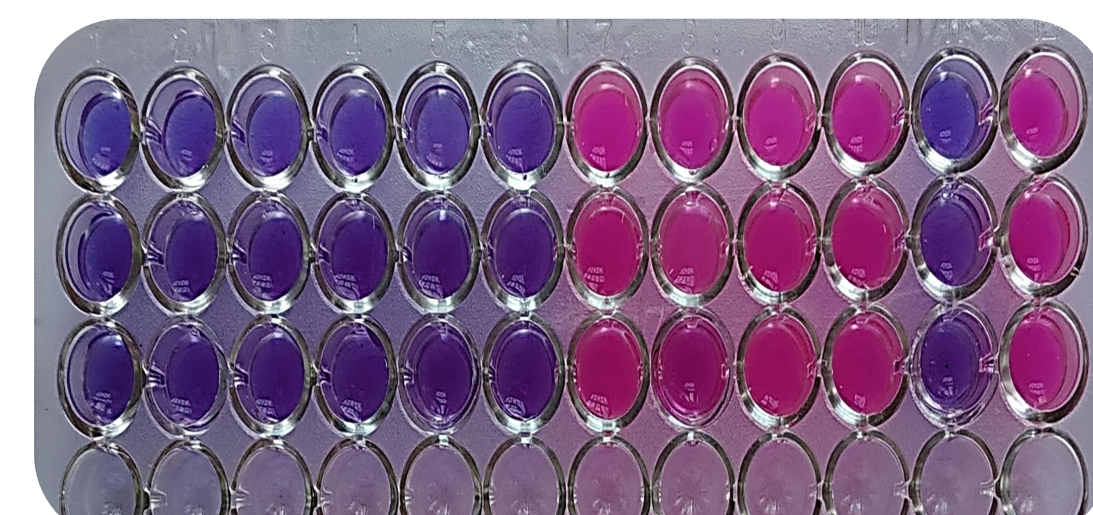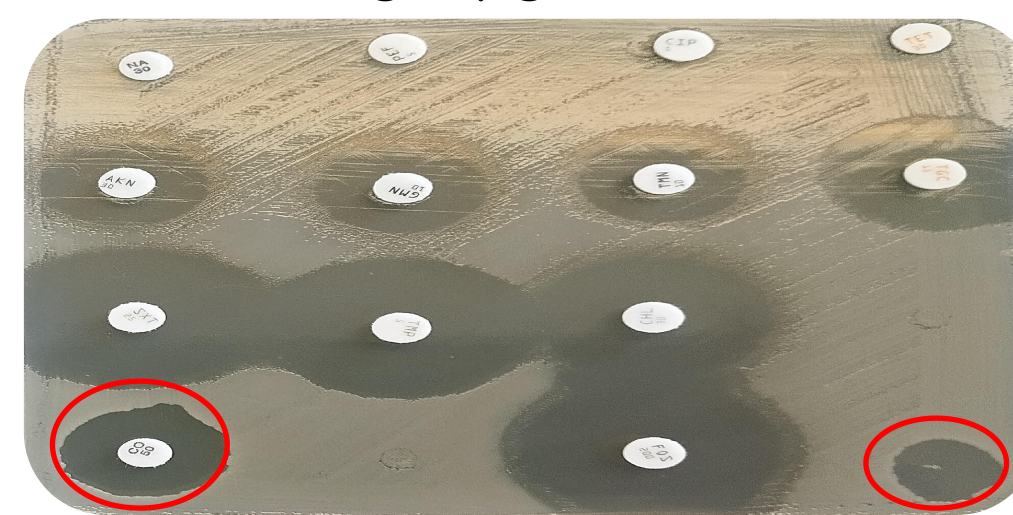
➢ Conjugation assays revealed that amoxicillin resistance was plasmid-mediated in all tested isolates, while nalidixic acid resistance was transferable in two strains and chromosomally encoded in two others.
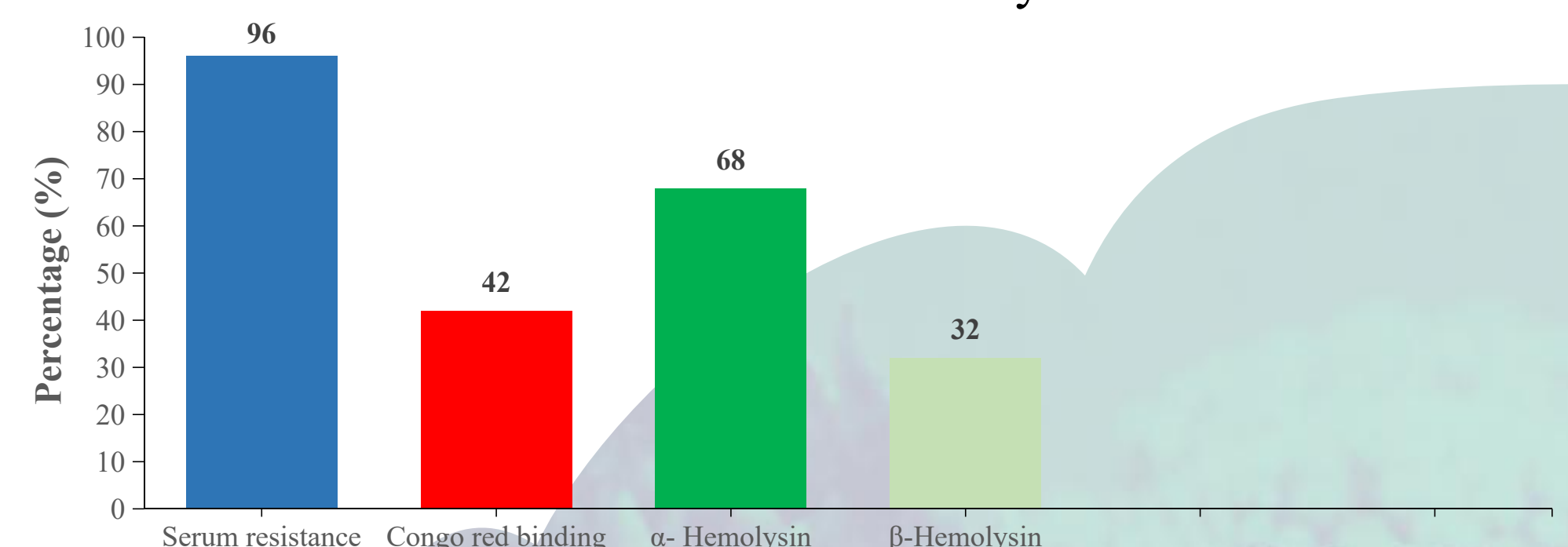

Figure 2. Distribution of virulence factors among *Escherichia coli* isolates.

➢ The Chi-squared test yielded a p-value $< 2.2 \times 10^{-16}$, confirming a highly significant correlation between antibiotic resistance and the virulence of the studied strains

CONCLUSION

This study underscores the urgent threat posed by the emergence of potentially pathogenic, highly virulent, and multidrug-resistant avian strains. These findings constitute a significant warning regarding the escalating risks to public health and emphasize the need for stricter regulation and more judicious use of antibiotics in poultry farming.

FUTURE WORK / REFERENCES

➢ BAKOUR R., LAROCHE Y. and CORNELIS G. (1983).Studies of the incompatibility and replication of the 70kb virulence plasmides of Yersinia.Plasmide. 10: 279-289.

➢ AL-SAIEDI R. L. R. and AL-MAYAH A. A. S. (2014). Pathogenicity Testing of Several APEC Isolates Obtained from Naturally Infected Broiler Birds Reared in Basrah. International Journal of Poultry Science 13 (7): 374-378.

➢ FIERER J., FINLAY F. and BRAUNE A. L. (1972).A plaque assay on agar for detection of Gram negative bacilli sensitive to complement. I. Immunology 109, 1156.

➢ Clinical and Laboratory Standards Institute 2023.