

Avian pathogenic *Escherichia coli* biofilm formation ability at different temperatures (37°C and 42°C)

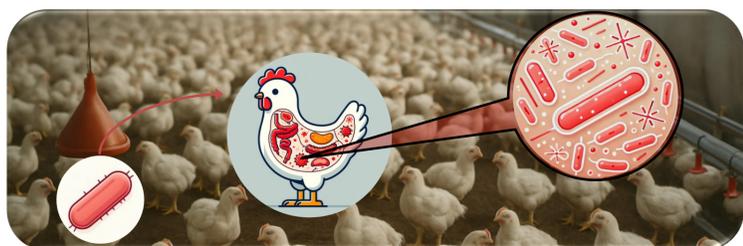
Enea Ovedani¹, Alessandra Piccirillo¹, Roberta Tolosi¹, Luca Bano², Luca Zandonà², Andrea Laconi¹

¹Department of Comparative Biomedicine and Food Science, University of Padua, Viale dell'Università 16, 35020 Legnaro, Italy

²Veterinary Diagnostic Laboratory, Istituto Zooprofilattico Sperimentale delle Venezie, 31020 Villorba (TV), Italy

INTRODUCTION & AIM

- Avian pathogenic *Escherichia coli* (APEC) causes **colibacillosis** in poultry and can form **biofilms** that aid in **antimicrobial resistance** spread
- Understanding APEC's biofilm formation and persistence in poultry environments is essential for creating effective control strategies
- This study aimed to assess the biofilm formation ability of APEC and the influence of temperature variations on this property

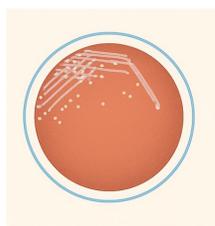


METHOD

- 34 APEC** strains were isolated from **diseased chickens** (n = 27) and **turkeys** (n = 7) from different farms located in Northern Italy (fig.1)

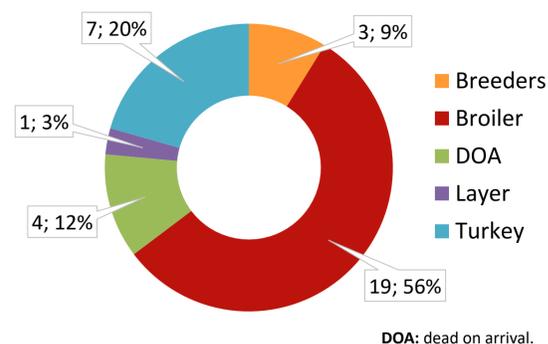
Biofilm formation assay¹

Day 1

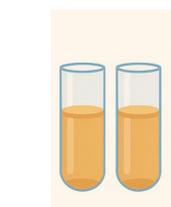


Bacteria resuscitation on Blood Agar (24 h at 37°C)

Figure 1: Graphical representation of the origin of the samples.

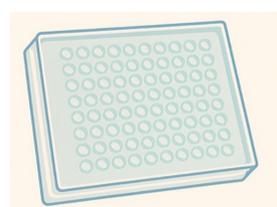


Day 2



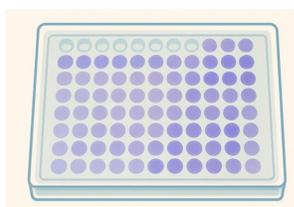
BHI inoculation (24 h at 37°C or 42°C)

Day 3



Microtiter plate inoculation (1 McFarland) and incubation (24 h at 37°C or 42°C)

Day 4



Biofilm fixation, staining and Optical Density (OD) measurements

- The mean OD was calculated for each strain (mean of six independent replicates)

- Strains classified into **4 different categories** (optical density of the samples (OD_S) vs optical density of the negative control (OD_{NC}):

- Non-producer** (OD_S ≤ OD_{NC})
- Weak producer** (OD_{NC} < OD_S ≤ 2OD_{NC})
- Moderate producer** (2OD_{NC} < OD_S ≤ 4OD_{NC})
- Strong producer** (4OD_{NC} ≤ OD_S)

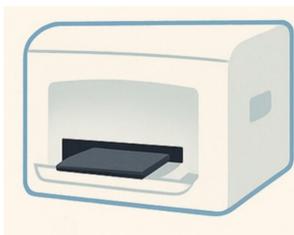


Figure 2: Graphical representation of the workflow of the biofilm assay.

RESULTS & DISCUSSION

- Results at **37°C** (fig.3): 70.6 % of the samples (24/34) were weak producers, 23.5 % (8/34) moderate producers, and the remaining 5.9 % strong biofilm producers
- Results at **42°C** (fig.4): 82.3% of the samples (28/34) were weak producers, 14.7% (5/34) moderate producers, and 3% (1/34) strong biofilm producers
- All strains isolated from carcasses were weak biofilm producers
- Increased temperature** results in **decreased** or **increased biofilm production** in 8 (23.5%) and 2 (5.9%) strains, respectively

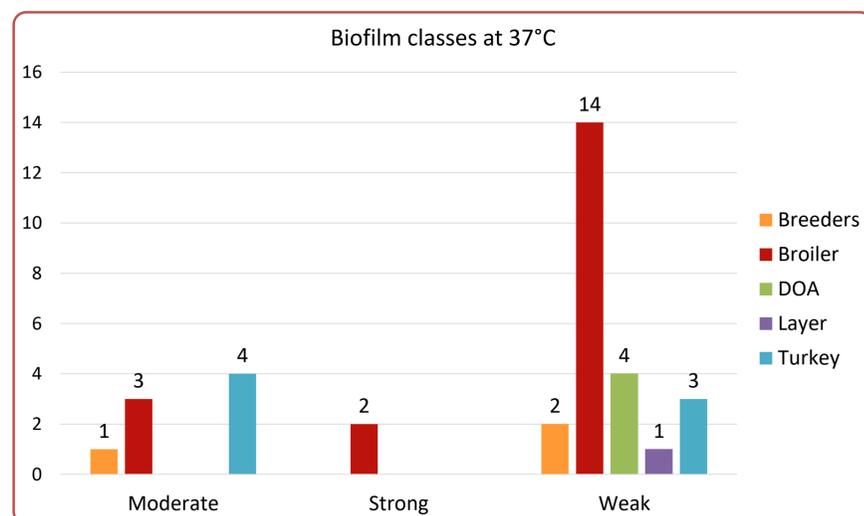


Figure 3: Stacked bar graph reporting the number of strains in each biofilm category at 37°C divided according to the strain origin.

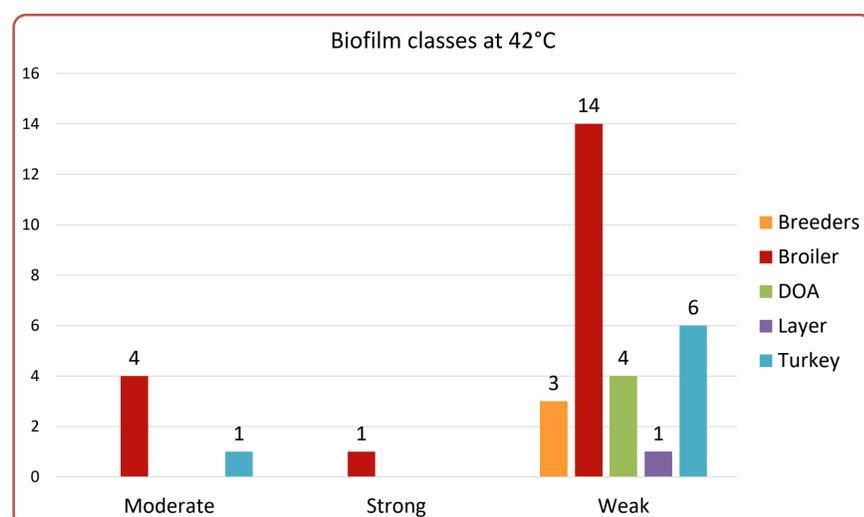


Figure 4: Stacked bar graph reporting the number of strains in each biofilm category at 42°C divided according to the strain origin.

CONCLUSION

- All strains** demonstrated the ability to **form biofilms**
- This represents a concern for poultry health, since biofilm can **enhance APEC persistence** in the farm environment
- Increased temperature** seems to **decrease** APEC biofilm formation ability

FUTURE WORK / REFERENCES

- Research should focus on elucidating the **genetic background** underlying the APEC biofilm-forming ability and clarify the **effect of the temperature** on this phenotypic feature

¹Laconi, A., Tolosi, R., Apostolakis, I., Piccirillo, A. (2023). Biofilm formation ability of ESBL/pAmpC-producing *Escherichia coli* isolated from the broiler production pyramid. *Antibiotics*, 12(1). <https://doi.org/10.3390/antibiotics12010155>