

Galleria mellonella as a model to evaluate the effect of tetracycline against *Campylobacter coli* strains

Ashly Chagua ¹, César Lazaro ¹, Pedro Angulo ¹, Diego Diaz ¹

¹ Laboratorio de Farmacología y Toxicología Veterinaria, Facultad de Medicina Veterinaria, Universidad Nacional Mayor de San Marcos, Lima 03-5137, Perú.

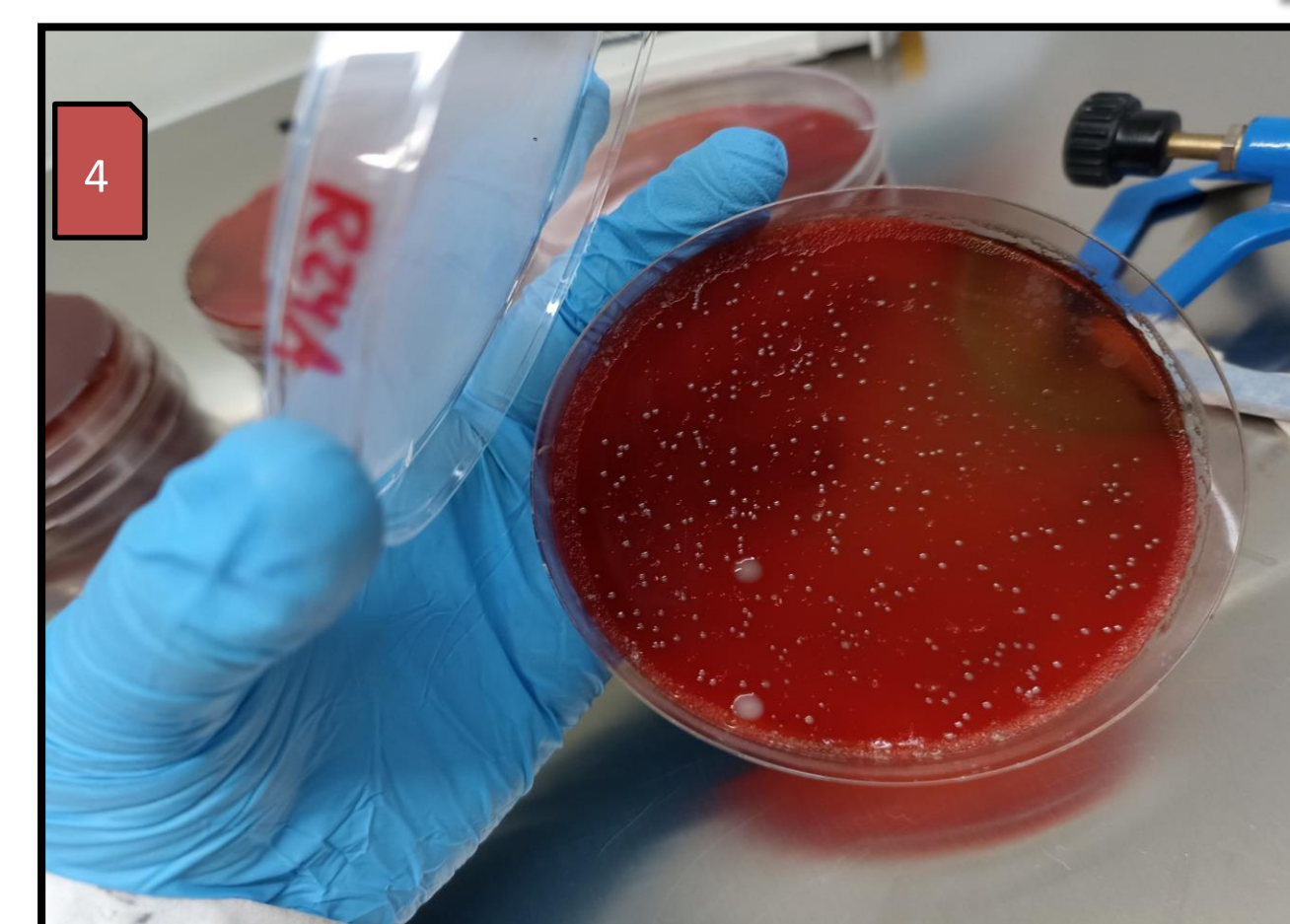
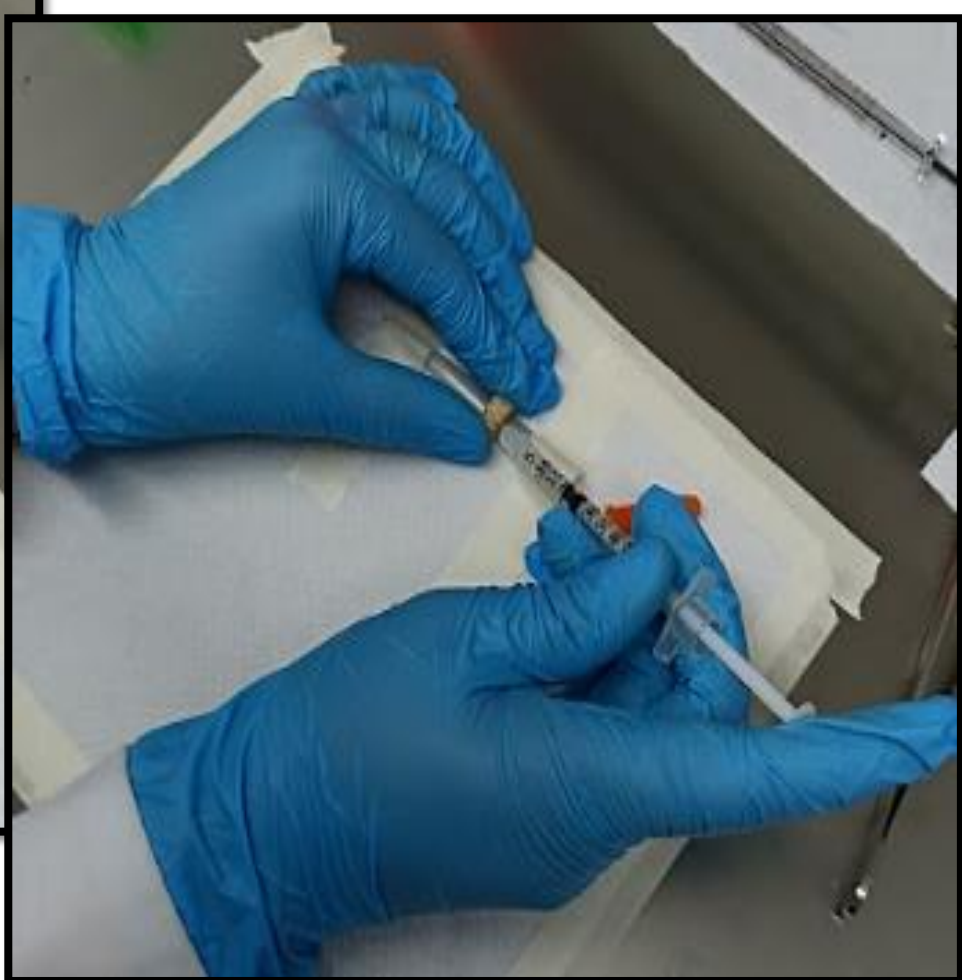
INTRODUCTION & AIM

Campylobacter spp. is one of the leading causes of foodborne bacterial gastrointestinal illnesses in humans, primarily through chicken meat. The prevalence of antibiotic-resistant *Campylobacter* strains in chickens is increasing worldwide. This makes it necessary to evaluate antimicrobial efficacy, particularly using in vivo methods, such as models such as *Galleria mellonella* larvae (GML).

METHOD



1 GML larvae infected with *C. coli* strains and subsequently inoculated with ATB tetracycline (8, 12, 15, 20 y 40 mg/mL)



3 Hemolymph extraction and deposited in Eppendorf tubes (1.5 ml) kept at 4°C to avoid melanization. Processing within 10 minutes after extraction.

4 Serial dilutions of 9 in 1 were made (900µl of PBS and 100µl of hemolymph). 100 µl were seeded on Mueller Hilton Agar blood at 37°C for 18-24 hours and the number of CFU was recorded.

5 Serial dilutions were made by placing 90 µl of insect physiological saline (IPS) and 10 µl of hemolymph. The dilution was homogenizing and 10 µl of the solution were placing in a hemocytometer. Quantification was performed using a compound microscope.

RESULTS & DISCUSSION

The results showed that the group treated with 8 and 12 mg/mL of tetracycline had its mortality reduced to less than 10% in the first 3 days. In contrast, higher doses (15, 20 and 40 mg/ml) showed no significant differences compared to the infected group without antibiotic treatment. The bacterial load decreased by more than 3 log 10 in the first 0.5 h in the treated groups. No differences in the number of hemocytes were observed between treated and untreated larvae. Treatment with tetracycline at 8 mg/ml was the most effective in reducing larval mortality and controlling *C. coli* infection. These results suggest the usefulness of the *Galleria mellonella* larval model to evaluate antibiotic doses against *C. coli*

The present study evaluated the viability of *Galleria mellonella* larvae as an alternative in vivo model to study the pathogenicity of *Campylobacter coli* strains and the efficacy of the antibiotic tetracycline hydrochloride. This model has gained popularity in recent years as a useful tool in medical and veterinary microbiology due to its low cost, easy handling, absence of ethical restrictions, and an innate immune response surprisingly comparable to that of mammals in terms of cellular and humoral functionality (Tsai et al., 2016; Desbois & Coote, 2011).

CONCLUSION

Galleria mellonella has established itself as a valid and reproducible experimental model for antibiotic efficacy studies against *Campylobacter coli*. A tetracycline concentration of 8 mg/ml proved effective and nontoxic, providing an experimental framework for future evaluations in superior models. This approach may contribute to optimizing antimicrobial use in veterinary medicine and to combating antimicrobial resistance.

FUTURE WORK / REFERENCES

Tsai, C. J. Y., Loh, J. M. S., & Proft, T. (2016). *Galleria mellonella* infection models for the study of bacterial diseases and for antimicrobial drug testing. In *Virulence* (Vol. 7, Issue 3, pp. 214–229). Taylor and Francis Inc. <https://doi.org/10.1080/21505594.2015.1135289>

This work was funded by the Universidad Nacional Mayor de San Marcos with the projects PCONFIGI A23080991, PINTERDIS A2108001i and by CONCYTEC-PROCIENCIA within the framework of the competition "Undergraduate and Postgraduate Theses in Science, Technology and Technological Innovation - Competition E073-2024-01; PE501089669-2024"