## Scientific Letters



Poster 9

# Exploring antimicrobial resistance in autochthonous Portuguese hens: a study of *Escherichia coli* on eggshells

<u>Rita Jesus</u> <sup>1,\*</sup>, Virgínia Ribeiro <sup>2</sup>, Rui Dantas <sup>3,4</sup>, Ana R. Freitas <sup>3,5,6</sup>, Nuno V. Brito <sup>3,7</sup>, Sandra Ouinteira <sup>3,8,9,10</sup> and Carla Miranda <sup>3,11</sup>

- <sup>1</sup> University Institute of Health Sciences, Cooperativa de Ensino Superior Politécnico e Universitário, 4585-116 Gandra, Portugal
- <sup>2</sup> AMĪBA-Associação dos Criadores de Bovinos de Raça Barrosã, 4730-260 Vila Verde, Portugal
- <sup>3</sup> UCIBIO—Applied Molecular Biosciences Unit, University Institute of Health Sciences (IH-TOXRUN, IUCS-CESPU), Avenida Central de Gandra 1317, 4585-116 Paredes, Portugal
- <sup>4</sup> ACRC—Associação dos Criadores da Raça Cachena, Parque Empresarial de Paçô, Rua da Roca 107,4970-249 Arcos de Valdevez, Portugal
- <sup>5</sup> UCIBIO—Applied Molecular Biosciences Unit, Faculty of Pharmacy, University of Porto, Rua Jorge de Viterbo Ferreira 228, 4050-313 Porto, Portugal
- <sup>6</sup> Associate Laboratory i4HB, Institute for Health and Bioeconomy, Faculty of Pharmacy, University of Porto, Rua Jorge de Viterbo Ferreira 228, 4050-313 Porto, Portugal
- <sup>7</sup> CISAS—Center for Research and Development in Agrifood Systems and Sustainability, Polytechnic Institute of Viana do Castelo, NUTRIR (Technological Center for AgriFood Sustainability), Monte de Prado, 4960-320 Melgaço, Portugal
- 8 CIBIO—Research Center in Biodiversity and Genetic Resources, InBIO, Research Network in Biodiversity and Evolutionary Biology, Associated Laboratory, University of Porto, Campus de Vairão, Rua Padre Armando Quintas 7,4485-661 Vairão, Portugal
- <sup>9</sup> BIOPOLIS Program in Genomics, Biodiversity and Land Planning, Campus de Vairão, Rua Padre Armando Quintas 7, 4485-661 Vairão, Portugal
- <sup>10</sup> Department of Biology, Faculty of Sciences, University of Porto, Rua do Campo Alegre s/n, 4169-007 Porto, Portugal
- <sup>11</sup> LAQV-REQUIMTE-Associated Laboratory for Green Chemistry, University NOVA of Lisbon, 1099-085 Caparica, Portugal

### **Abstract**

Background: The widespread use of antibiotics in intensive animal farming significantly contributes to antimicrobial resistance (AMR), posing a serious global public health threat [1]. Despite this, little is known about the role of autochthonous Portuguese laying hens as reservoirs of antibiotic-resistant Escherichia coli. Raised in extensive farming systems with reduced antibiotic exposure, these indigenous breeds offer a unique opportunity to study AMR dynamics under absent selective pressure. Investigating their role could yield valuable insights into the natural ecology of resistance and inform strategies for more sustainable livestock management [2]. Objective: To investigate the presence of antibiotic-resistant E. coli and antibiotic resistance genes on the 124 eggshells of autochthonous Portuguese laying hens. Methods: A total of 46 E. coli isolates previously obtained from Preta Lusitânica (n=15), Amarela (n=9), Branca (n=12), and Pedrês Portuguesa (n=10) eggshell swabs were confirmed by PCR. Antimicrobial susceptibility testing for 10 antibiotics was assessed following EUCAST/CLSI guidelines [3]. ESBL phenotype was searched using the Double-Disk Synergy Test. Antibiotic resistance genes (bla<sub>TEM</sub>, bla<sub>CTX</sub>, bla<sub>OXA-1</sub>, bla<sub>SHV</sub>) were searched by PCR in 44 of the 46 E. coli isolates. Results: Escherichia coli eggshell isolates exhibited resistance to gentamicin (96%), tetracycline (41%), ampicillin (24%), amoxicillin/clavulanic acid (4%), amikacin (4%), and trimethoprim/sulfamethoxazole (2%). Resistance to at least one antibiotic was found in 98% of the eggshell isolates, and a multidrug-resistant phenotype was identified in 17% of the isolates (Preta Lusitânica, Branca, Pedrês Portuguesa). Only 2% of the E. coli isolates showed pan-susceptibility (Amarela). ESBL phenotype was not detected. The blaTEM and blaCTX genes were detected in 36% and 11%, respectively, of the 44 E. coli isolates tested, while the blaoxA-1 and the blashy genes were not detected. Conclusion: This study provides insights into AMR in autochthonous hens, highlighting the potential role of environmental factors, such as soil, water, feed, and even human contamination, in AMR transmission to the eggshells. The findings contribute to the understanding of AMR in extensive farming systems and underscore the importance of considering environmental sources in efforts to mitigate AMR, increase consumer confidence, and support the development of sustainable poultry practices.

<sup>\*</sup> Correspondence: ritacarrolo.jesus@gmail.com

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