RESUMO SIMPLES - ECTOPARASITOS: SISTEMÁTICA, EPIDEMIOLOGIA E CONTROLE

PROTEIN CHARACTERIZATION AND TOTAL HEMOCYTE QUANTIFICATION IN THE HEMOLYMPH OF COCHLIOMYIA HOMINIVORAX LARVAE: IMPLICATIONS FOR INNOVATIVE CONTROL STRATEGIES

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Primary myiasis caused by larvae of Cochliomyia hominivorax represents a sanitary, economic, and animal-welfare challenge across livestock-producing regions of the Americas, leading to significant productivity losses associated with tissue destruction, reduced zootechnical efficiency, the need for repeated treatments, and a higher risk of secondary infections. These impacts highlight the importance of deepening our understanding of the biology of this species, particularly regarding cellular responses observed through the study of hemolymph and hemocytes, in order to clarify the immunological and physiological mechanisms involved in potential resistance processes and to support the development of effective control measures. Despite this relevance,

baseline information on the composition of C. hominivorax hemolymph,especially parameters such as hemocyte dynamics and total protein concentration, which remains scarce, limiting experimental analyses and hindering the advancement of innovative control strategies. To address this gap, the present study aimed to quantify circulating hemocytes in the hemolymph of third-instar larvae of C. hominivorax under different preservation conditions, as well as to determine total hemolymph protein concentrations using refractometry, applying a standardized and reproducible protocol. Larvae originated from the colony (CEUA: 8634020223) maintained at the Center for Clinical Research and Technological Innovation in Veterinary Medicine – Laerte Grisi, formerly the Laboratory of Experimental Chemotherapy in Veterinary Parasitology, DPA-IV-UFRRJ. For total hemocyte counts, 10 µL of hemolymph were collected from 60 L3 larvae by posterior incision after antisepsis and immobilization by cooling, and distributed into three experimental groups: G1, fresh undiluted hemolymph; G2, hemolymph diluted 1:1 in PBS containing anticoagulant (98 mM NaOH, 145 mM NaCl, 1.7 mM EDTA, and 41 mM citric acid); and G3, hemolymph diluted 1:1 in a modified anticoagulant solution (0.392 g NaOH, 0.853 g NaCl, 0.632 g EDTA, and 1.206 g citric acid in 100 mL of Milli-Q water). Cell aggregation was most pronounced in G1, which showed an average of 2.4×10^5 cells/mL; greater cellular dispersion was observed in G2, resulting in an average of 4.8×10^5 cells/mL; and the best performance occurred in G3, which demonstrated superior structural preservation and greater reproducibility among replicates, reaching 6.3×10^5 cells/mL. For protein analysis, hemolymph from 50 larvae was collected and diluted 1:1 in Milli-Q water prior to refractometric measurement, yielding corrected protein values ranging from 6.0 to 21.2 mg/mL, with a mean of 14.8 mg/mL, a median of 14.8 mg/mL, and a standard deviation of 2.96 mg/mL. The marked difference between the lowest and highest concentrations suggests that intrinsic factors related to larval development may directly influence hemolymph composition. This study establishes an initial foundation of information on hemolymph composition and cellular dynamics in C. hominivorax larvae, providing essential data for future studies, including those assessing hemolymph modulation in response to insecticides, and contributing to the development of more effective and selective control strategies aligned with livestock production and animal health needs.

Palavras-chave: applied biotechnology; hemolymph; fly; myiasis; protein.