TITLE: MINIMUN INHIBITORY CONCENTRATION OF OREGANO ESSENCIAL OIL AGAINST *Clostridium perfringens* CAUSER OF NECROTIC ENTERITIS

AUTHORS: MENCK, M.F. *1; JUSTINO, L.¹; SANTOS, B.Q.¹; SOUZA, M.¹; BERBERT, J.V.M.¹; HUIJBOOM, J.A.A. ¹; MATTÉ, F.²; KOBAYASHI, R.K.T.³; NAKAZATO, G. ³; BAPTISTA, A.A.S.¹

INSTITUTION: ¹Avian Medicine Laboratory, Department of Preventive Veterinary Medicine, State University of Londrina - UEL (Rodovia Celso Garcia Cid – Pr 445 Km 380, Campus Universitário, Londrina – PR). ²Vetanco Company. ³ Laboratory of Basic and Applied Bacteriology, Department of Microbiology, State University of Londrina- UEL (Rodovia Celso Garcia Cid – Pr 445 Km 380, Campus Universitário, Londrina – PR).

Clostridium perfringens (CP) is an important pathogenic producer of toxins and responsible for necrotic enteritis (EN) in birds, a disease that reduces performance and increases mortality, besides that it is a zoonotic risk factor. The use of oregano oil (OEO) has become common practice in the control of microorganisms. The principle acting substances, carvacrol and thymol, have an active/synergic effect on bacteria and cause bacterial growth reduction. The objective of this present study is to evaluate the ability of essential oregano oil to inhibit growth of *Clostridium perfringens* strains isolated from by nectrotic enteritis affected intestine. The minimal inhibitory concentration (MIC) was determined in accordance with the CLSI. The essential oregano oil was at first diluted in DSMO (50%) and there after diluted to a 1%; 0.8%; 0.7%; 0.6%; 0.5%; 0.4%; 0.3%; 0,2%; 0,1%; 0,08% and 0,06% solution. The bacterial inoculum was obtained by CP-strains that were grown on a Mueller Hinton 5% Sheep Blood agar plate and subsequently diluted to 0,5 on the scale of Mac Farland (1x10⁸ UFC/mL), causing the final bacterial dilution to be 1x10⁶. The different dilutions of the essential oil were anearobically incubated with the bacterial inoculum at 37°C for 20 hours in microtubes, containing 50 µl of bacterial suspension and 50 µl of OEO solution in one of the different concentrations. The MIC of the OEO on the CP-strains was determined by assessing the turbidity of the various samples. The Minimal Inhibitory Concentrations proved to be 0,1%; 0,1%; 0,4%; 0,4% for the strains CP4, CP5, CP10 and CP148 (volume/volume), respectively being 0,954 mg/ml for 0,1% and 3,816mg/ml for 0,4% (mass/volume). Earlier performed studies found a MIC for a CP-strain to be 0,750mg/ml, which is corresponding with the outcome of this study for the CP-strains 4 and 5. The use of essential oregano oil proved to be effective in order to inhibit growth of *Clostridium perfringens* strains.

Keywords: Antimicrobial, CIM, Clostridium perfringens, Oregano essencial oil.