

## **Título: *IN VITRO* CHOLINERGIC ENZYMATIC EVALUATION OF FERULIC ACID PARTICLES**

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**INTRODUCTION:** The cholinergic system plays a crucial role in neuroimmune communication, and its dysregulation is associated with autoimmune and neurodegenerative diseases. Acetylcholinesterase (AChE), the enzyme responsible for degrading acetylcholine, is an important therapeutic target, especially in the treatment of Alzheimer's disease. In this context, ferulic acid (FA), a phenolic compound found in medicinal herbs and the daily diet, has gained attention for its antioxidant effects and its potential to inhibit AChE, suggesting a possible neuroprotective action. **OBJECTIVE:** This study aims to evaluate *in vitro* effects of FA particles on AChE activity. **MATERIALS AND METHODS:** The nanocapsule suspension (n=3) was prepared by interfacial deposition of a preformed polymer as Rampelotto et al. (2022). After the addition of FA and solvent evaporation, FA-loaded nanocapsules (NCFA) were obtained at a concentration of 0.5 mg/mL, and blank nanocapsules (NCBR) or nanoemulsion (NEBR) were also prepared for comparison. Nanoemulsions (NEFA) were prepared by spontaneous emulsification and the FA concentration incorporated was 1.5 mg/mL. *In vitro* AChE activity was assessed using a colorimetric assay based on the DTNB reaction, with absorbance measured at 412 nm. The enzyme isolated from *Electrophorus electricus* was diluted in Tris-HCl buffer to a final 10x concentration. The experiments included blank control (without FA) and experimental groups with different concentrations of NCFA (1.875; 3.75 or 7.5 ug/mL) or NEFA (5.625; 11.25 or 22.5 ug/mL). The microplate reader was set to 30°C and each sample was treated with potassium phosphate buffer (TFK), Milli-Q water, and the enzyme, followed by a 5-minute incubation. DTNB and the substrate were then added, and absorbance readings were taken six times at one-minute intervals. **RESULTS:** No statistically significant differences were observed among the tested concentrations of blank (NCBR or NEBR), NEFA and NCFA. **CONCLUSION:** Ferulic acid (FA) particles, at the tested concentrations, did not show inhibitory potential against *in vitro* AChE activity.