TITLE: IN VITRO GENOTOXICITY POTENTIAL OF E. MACROPHYLLUS AND E. GRANDIFLORUS LEAVES AQUEOUS EXTRACTS

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Introduction: *E. macrophyllus* and *E. grandiflorus* are native medicinal plants from Brazil known as chapéu-de-couro. As they have been used for centuries in folk medicine, it is necessary to assess toxic potential and ensure their safe use. **Aim:** The aim of this research was to evaluate the *in vitro* genotoxicity potential of leaves aqueous extract of these species in 3T3 cell line by comet assay. **Materials and Methods:** Leaves of both plants were collected, dried in a drying oven and processed in a blade mill. Infusions were prepared following the proportion 1:50 (plant: water), as traditionally used. The concentrations tested in immortalized lineage of mice fibroblasts (3T3) were 5, 10, 20 (popular use) and 50 mg/mL. Total flavonoids content was assessed by chloride aluminium assay. MTT reduction assay

was executed to evaluate concentrations' cytotoxicity potential and cell viability. Alkaline comet assay was used to determine the genotoxicity of both species. LMW assay was performed simultaneously with comet assay as a control. For statistical analyses, Shapiro-Wilk test was performed followed by one-way ANOVA test and Bonferroni *post hoc* test (parametric groups) or Kruskal Walli's test and Dunn´s *post hoc test* (non-parametric groups) ($p<0.05$). SisGen registration nº A938D92. **Results:** *E. macrophyllus* was not genotoxic at concentrations tested, neither at the most concentrated extract ($p<0.001$). Curiously, *E. grandiflorus* aqueous extract showed an

increased genotoxic potential at 5 and 50 mg/mL, while in the intermediate concentrations it decreased. It suggests an hormetic effect. LMW assay did not detect toxicity induced by comet assay execution, guaranteeing the reliability of this test. *E. grandiflorus* has a greater total flavonoid content than *E. macrophyllus* ($p<0.01$). In MTT reduction assay, both plants showed a concentration - dependent cytotoxicity. Its effect was less prominent at *E. grandiflorus*, possibly associated with its relevant flavonoids content. **Conclusions:** Leaves aqueous extract of *E. macrophyllus* was not genotoxic, otherwise *E. grandiflorus* was potentially genotoxic at concentrations tested in 3T3 cell line.

KEY-WORDS: Chapéu-de-couro; 3T3 cell line; Genotoxicity.