Evento	Salão UFRGS 2014: SIC - XXVI SALÃO DE INICIAÇÃO CIENTÍFICA DA UFRGS
Ano	2014
Local	Porto Alegre
Título	Antioxidant Activity and Antiproliferative Action of Ethanolic Extract of Echinodorus grandiflorus in the Yeast Saccharomyces cerevisiae and Human Cancer Cell Lines
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INTRODUCTION: *Echinodorus grandiflorus* is popularly known as "chapéu-de-couro". This plant is an aquatic or semi-aquatic herb with a milky sap. It has been used in the folk medicine as anti-inflammatory and diuretic. To give a scientific basis for traditional usage of this medicinal plant, the leaf extracts were evaluated for their antioxidant and antiproliferative activities.

METHODS: In this work, we report the in vivo antioxidative properties of the crude ethanolic extract of *E. grandiflorus* studied by using *Saccharomyces cerevisiae* strains proficient and deficient in antioxidant defences. In addition, the ability of deactivating free radicals was investigated with in vitro biochemical method 1, 1- Diphenyl-2-picrylhydrazyl (DPPH) scavenging assay. Furthermore, The *E. grandiflorus* cytotoxicity action was investigated on MRC5, MCF-7, HepG2, T-24, PC-3, 22Rv-1, HCT-116, HT-29, and CACO-2 cells by XTT assay, after 24, 48, 72, 96 and 120 h of treatment. The mechanism of cell death as well as DNA damage induction was investigated by flow cytometry and comet assay, respectively. Moreover, to further understand the biological mechanism of the cytotoxic effect of *E. grandiflorus*, we also investigated its mutagenic effect on XV185-14c haploid yeast.

RESULTS: *E. grandiflorus* at doses 10 – 100 μg/mL was able to protect *sod1*Δ, *sod2*Δ and *sod1*Δ*sod2*Δ mutants against H_2O_2 cytotoxicity and this activity was more effective in the *sod1*Δ*sod2*Δ double mutant ($P < 0.001$). However, *E. grandiflorus* did not protect wild type yeast strain. In addition, the ethanolic extract of *E. grandiflorus* showed significant radical scavenging activity by DPPH reduction. The ethanolic extract was not mutagenic in strain XV185-14c. *E. grandiflorus* induced a time- and dose-dependent growth inhibition in all human cancer cell lines. However, in the T-24 bladder cancer cells the toxicity was more pronounced with an IC50 at 24 h of the 12.6 μg/mL with a higher potency, five fold more potent in this cell than normal cells (MRC5). Comparatively, Mitoxantrone (MXT), an anticancer drug used in this study as a positive control, demonstrated IC50 values in the tumor cell lines ranging from 0.61-3.5 μg/mL. We therefore evaluated the cell death mechanism induced by this extract (IC50) in T-24 cells at 24 h. We found that the treatment with IC50 *E. grandiflorus* for 24 h induced apoptosis in 19% of T-24 cells. The percentages of 7-AAD positive cells were less than 3% for IC50 treatment, and the dominant cell death type was apoptosis.

CONCLUSION: Apoptosis is one of the body's most potent defences against cancer. The apoptotic potency of *E. grandiflorus* suggests that it may be an effective compound in therapy for the treatment of bladder cancer. This work provides a scientific support for the high antioxidant and antiproliferative activity of this plant and thus it may have potential applications in the treatment of the diseases caused by ROS. Further studies are needed to confirm in vivo anti-tumorgenicity and subsequent chemical characterization of its active molecule(s).

Financial Support: CAPES, CNPq and FAPERGS - Brazil.