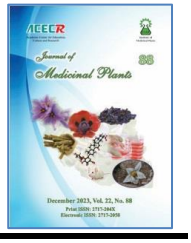
J. Med. Plants 2023; 22(88): 1-9

Institute of Medicinal Plants

Journal of Medicinal Plants

Journal homepage: www.jmp.ir



Research Article

Effects of ethanolic extract and cernumidine compound obtained from *Solanum cernuum's* Vell. leaves on benign prostatic hyperplasia

Mariza Abreu Miranda[1,*], Claudia Sares[2], Augusto Faria Andrade[3], Kleiton Silva Borges[3], James Dewey McChesney[4], Jairo Kenupp Bastos[1], Rodolfo Borges dos Reis[2]

[1] *School of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo, São Paulo, Brazil*
[2] *Department of Surgery and Anatomy, Ribeirão Preto Medical School, University of São Paulo, Ribeirão Preto, São Paulo, Brazil*
[3] *Department of Pediatrics, Ribeirão Preto Medical School, University of São Paulo, Ribeirão Preto, São Paulo, Brazil*
[4] *Ironstone Separations, Inc., Oxford, USA*

ARTICLE INFO

Keywords:
Cytotoxicity
Prostate muscle cells
Apoptosis
Alkaloid
Solanaceae

ABSTRACT

Background: *Solanum cernuum* Vell. (Solanaceae) is a native plant from Brazil traditionally used for treating ulcers, liver damage, skin infections, gonorrhea, and benign prostatic hyperplasia (BPH). **Objective:** To evaluate the impact of hydroalcoholic extract and cernumidine obtained from *S. cernuum* Vell. leaves against prostate primary smooth muscle cell culture and assess their potential effect in inducing apoptosis (annexin V assay by flow cytometry). **Methods:** The hydroethanolic extract of *S. cernuum* was obtained by macerating powdered dried leaves followed by fractionation, furnishing 4.8 % w/w of cernumidine alkaloid as established by high performance liquid chromatographic analysis. Primary BPH smooth muscle cell culture was used in cell proliferation and apoptosis assays, whereby prostate smooth muscle cells were subjected to 0.125-5.0 mg/mL of hydroethanolic extract and 0.025-1.0 mg/mL of cernumidine for 48, 72, and 96 h. **Results:** Hydroethanolic extract (2 mg/mL) and cernumidine (1 mg/mL [3.3×10^{-6} M]) inhibited the cell growth by 60 % and 62 % at 96 h, respectively, and eventually led to cell death by apoptosis. On the other hand, only cernumidine (1 mg/mL) induced significant death by necrosis compared to control. **Conclusion:** The obtained findings corroborate the use of *S. cernuum* in native medicine and indicate that cernumidine is a promising candidate for further studies focusing on BPH treatment.

1. Introduction

Benign prostatic hyperplasia (BPH) occurs frequently in middle-aged and older men. It is the most common benign neoplasm affecting men and is associated with increased proliferation of prostatic epithelial and stromal cells by increasing their size and number, causing prostate enlargement. It is estimated that 14

Abbreviations: HPLC, High Performance Liquid Chromatography; BPH, Benign Prostatic Hyperplasia; PBS, Phosphate Buffer Saline; HPV, Human Papilloma Virus

*Corresponding author: marizamiranda@ufl.edu, marizamiranda2003@yahoo.com.br

doi:
Received 6 December 2023; Received in revised form 8 April 2024; Accepted 13 April 2024

million men in the United States have symptoms of BPH [1, 2]. Worldwide, this number reaches approximately 30 million [3], and is rapidly rising, particularly in low- and middle-income countries [4].

Due to the increase in life expectancy, global BPH prevalence has also increased. The available treatments comprise the use of α1-adrenoceptor antagonists and phosphodiesterase-5 and 5α-reductase inhibitors. If not treated, BHP leads to severe complications, such as infections, kidney failure, and impairment of life quality [5].

Solanum cernuum is an herbal medicine from southern and southeastern Brazil [6]. It is popularly known as "panacea," as it is believed to have many beneficial actions against a variety of medical conditions. While it remains insufficiently studied, flavonoids, guanidinic alkaloids, saponins, and triterpenoids have been isolated from *S. cernuum* [7–12].

Many folk uses have been reported for infusions of the aerial parts of *S. cernuum*, such as anti-inflammatory, gastroprotective, antitumor, diuretic, anti-hemorrhagic, and anti-arrhythmic [6]. It is also used to treat liver damage, skin infections, gonorrhea, and in the prevention and treatment of BPH. However, no report in the literature corroborates its use for BPH.

The literature reports that alkaloids with *in vivo* activity reduced the inflammation associated with BPH, such as demethyleneberberine, palmatine, and berberine isolated from *Cortex Phellodendri* [13], berberine from *Coptis chinensis Franch* [14], isocorynoxeine, an indole alkaloid from *Uncaria rhynchophylla* [15], and yohimbine from the *Pausinystalia yohimbe* bark [16].

Guided by this evidence, the purpose of this study was to assess the efficacy of *S. cernuum* hydroethanolic extract and its isolated alkaloid, cernumidine, in reducing BPH symptoms. Accordingly, we evaluated their inhibitory effect on a BPH primary smooth muscle cell culture and their apoptosis-inducing effect *in vitro*.

2. Materials and methods

2.1. Plant Material

The leaves of *S. cernuum* were collected in Teresópolis, State of Rio de Janeiro, Brazil, in January 2021. A voucher specimen is kept under registration number SPFR: 15118 with the Herbarium of the Biology Department of the University of São Paulo, the Ribeirão Preto, São Paulo State Brazil campus.

The plant name was confirmed on www.theplantlist.org on the 1st of February 2021. The SISBIO ICMBio authorized collecting the plant material under 18458–1. An ethical clearance (approval number 010221/2012-3) from the Brazilian Ministry of Environment, under the auspices of the Council for Genetic Heritage Management (CGEN/MMA) was obtained to access the traditional knowledge associated with genetic resources for research purposes from the traditional communities.

2.2. Preparation of the Extract, Isolation of cernumidine and HPLC analysis

Extract preparation and cernumidine isolation were previously described in detail [17]. Fresh leaves were briefly dried in an oven at 40 ◦C with circulating air for 24 h and powdered in a knife mill. Then, 2 kg of dried and powdered leaves were macerated in 70 % aqueous ethanol at room temperature for 48 h, followed by percolation, yielding 325.74 g of crude hydroethanolic extract after undergoing concentration and lyophilization processes.

Cernumidine was isolated by digesting the crude extract with *n*-butanol, followed by chromatography methods, including open chromatographic columns and preparative HPLC. The detailed chromatographic process,

including the HPLC-UV the *S. cernuum* hydroethanolic extract chromatographic profile and quantitative analysis of cernumidine (4.8 % W/W) was previously reported using a Synergi Polar-RP (4 µm, 15 cm (h), 0.46 cm (i.d.) Phenomenex column. The mobile phase comprised 0.1 % formic acid in water and acetonitrile [12, 17].

2.3. Assays with the primary culture of smooth muscle cells of BPH

2.3.1. Primary culture

Primary BPH smooth muscle culture cells were purchased from the LONZA® company: CC-2587 PrSMC (Prostate Smooth Muscle Cells). The primary culture was maintained in SmGmTM medium - 2Bullet KitTM (CC-3182) (LONZA®) and incubated in a humid atmosphere containing 5 % CO_2 at 37 °C [18].

2.3.2. Cell Proliferation Assay

Cells from the primary culture were seeded in 96-well plates (800 cells/well). After 24 h of incubation, the culture medium was removed, and the cells treated with hydroethanolic extract (5; 4; 3; 2; 1; 0.5; 0.25 and 0.125 mg/mL) solubilized in 0.5 % of DMSO or with cernumidine alkaloid (1; 0.9; 0.8; 0.7; 0.6; 0.5; 0.4; 0.3; 0.2; 0.1; 0.05 and 0.025 mg/ml) solubilized in 0.14 % dimethyl sulfoxide. The cells were treated for 48, 72, or 96 h. After the respective exposure times, the culture medium was removed, and 195 µL of medium containing 5 µL of resazurin was added to each well. Then, the plates were incubated for 4 h, and the absorbance was read using iMark Microplate Absorbance Reader (BioRad Laboratories®). The reading was performed at wavelengths of 570 and 595 nm. The experiments were done in triplicate, considering all evaluated times. Dimethyl sulfoxide at 0.5 % and 0.14 % were used as negative controls [19].

2.4. Apoptosis assay

The assay to detect cell death was performed by labeling apoptotic cells with Annexin V Cy-5 (BD Biosciences Pharmigen®) and necrotic cells with propidium iodide (PI). Primary smooth muscle culture cells were seeded into 6-well plates containing 4 × 104 cells/well. The concentrations of hydroethanolic extract were 2, 0.5, and 0.125 mg/ml. Cernumidine was tested at 1 mg/mL (3.3 × 10-6 M), 0.5 mg/ml (1.6 × 10 -6 M), and 0.05 mg/mL (1.6 × 10 -7 M). The evaluated exposure time was 96 h. Next, the cells were trypsinized and centrifuged at 1000 rpm for 5 min at 4 °C, washed with ice-cold PBS, and resuspended in 300 µL of diluted binding buffer (1x) (BD Biosciences Pharmingen®). 5 µL of annexin V Cy5 was added to each sample tested. Tubes were incubated at room temperature and protected from light for 15 min. After incubation, 50 µl of a PI solution (50 µg/ml) was added, and the cells were analyzed immediately by BD FACSCaliburTM flow cytometry (BD Biosciences®). Cells in culture medium and culture medium with 0.5 % or 0.14 % dimethyl sulfoxide were used as a negative control. The test was performed in triplicate. Statistical analysis for primary cell culture assays was performed using GraphPad Prism® software. ANOVA followed by Tukey's test was used [20].

2.5. Statistical analysis

The data for cell proliferation and apoptosis assays were expressed as mean+-standard error using one-way analysis of variance (ANOVA), followed by Tukey test and *P < 0.05, **P < 0.01, ***P < 0.001 considered as significant.

3. Results

Figs. 1a and 1b show the effect of hydroethanolic extract and cernumidine, respectively, after 96 h of exposure on cell proliferation in BPH primary smooth muscle culture. It is evident that the extract in 0.5−5 mg/mL concentrations significantly reduced cell proliferation ($P < 0.05$) compared to the control group, with a maximum inhibition effect of 60 %. Likewise, the alkaloid applied in 0.4−1 mg/mL concentrations reduced proliferation ($P < 0.05$), inhibiting cell growth by up to 62 %. As shown in Fig. 1b, the concentration−response effect was also observed.

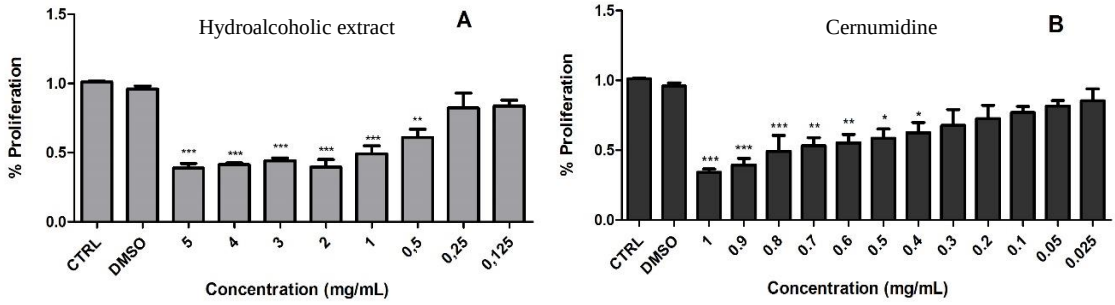


Fig. 1. Inhibitory effect of hydroalcoholic extract (a) and cernumidine (b) on benign prostatic hyperplasia muscle cell proliferation. Statistical analysis ANOVA followed by Tukey test. Significant difference compared to the control group (CTRL) *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

There was no statistically significant difference in cell proliferation in the assays performed at 48 and 76 h for either the extract or cernumidine (data not shown).

3.1. Effect of hydroalcoholic extract and cernumidine on apoptosis induction

To verify the cytotoxic effect of decreasing cell proliferation, the percentage of apoptotic cells was also evaluated after 96 hours of the following treatments: hydroalcoholic extract (0.125, 0.5, and 2 mg/mL) and cernumidine (0.05 mg/mL [1.6×10^{-7} M], 0.5 mg/mL [1.6×10^{-6} M], and 1 mg/mL [3.3×10^{-6} M]). These concentrations were selected based on the preliminary cell proliferation tests to verify if the extract and the alkaloid would present a concentration−response effect.

For the hydroalcoholic extract, a significant induction of apoptosis was observed at 0.5 and 2 mg/mL, whereby a maximum effect of 53 % was obtained at the highest concentration ($P < 0.05$) (Fig. 2a).

In cells treated with cernumidine, a high induction of cell death by apoptosis was observed at all concentrations compared to the control (52.93 %, 44.06 %, and 32.05 % for 1, 0.5, and 0.05 mg/mL, respectively). However, only the highest concentration (1 mg/mL) resulted in a significant difference (16.94 %) in cell death by necrosis compared to the control (Fig. 2b).

Cell death by apoptosis was also observed to exhibit a concentration−response effect correlation (Fig. 2a and 2b) and yielded good flow cytometry separation (Fig. 3).

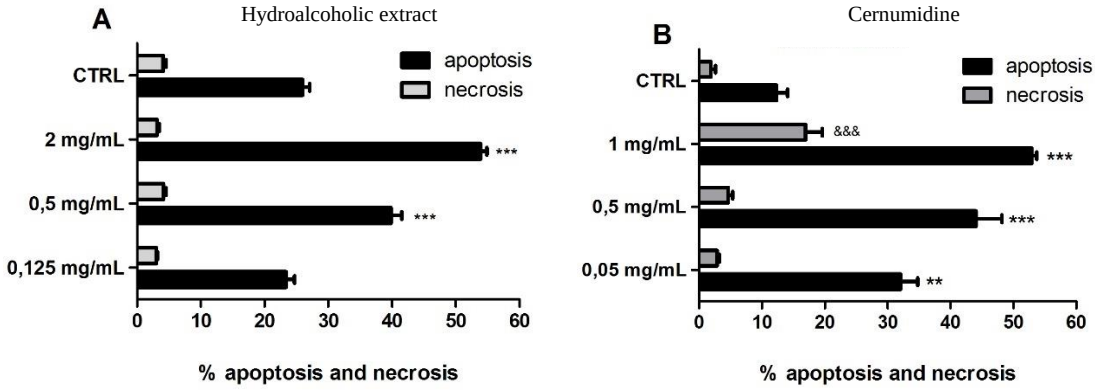


Fig. 2. Effect of hydroalcoholic extract and cernumidine at different concentrations on apoptosis induction. Statistical analysis ANOVA followed by Tukey test. Significant difference compared to the control group (CTRL) **P < 0.01 and ***P < 0.001.

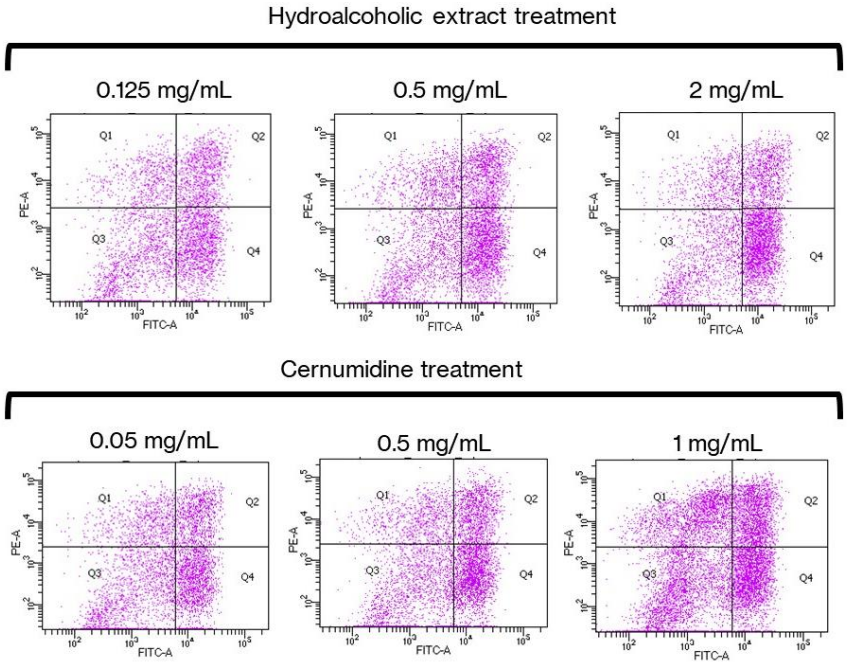


Fig. 3. Flow cytometry plots for hydroalcoholic extract and cernumidine treatment. Q1: damaged cells or naked nuclei cells; Q2: necrosed cells; Q3: live cells; Q4: apoptotic cells.

4. Discussion

It is known that toxic or deleterious effects on the cell can trigger cell death by necrosis or apoptosis, whereby these processes are differentiated by cellular morphology and biochemical pathways, with necrosis referring to a violent form of cell death [21]. During necrosis, cells transform, resulting in increased cell volume, chromatin aggregation, cytoplasmic disorganization, loss of plasma membrane integrity, and consequent cell disruption (cytolysis) [22, 23]. On the other hand, apoptosis is a regulated physiological process of programmed cell death that plays an essential role in the homeostasis of different tissues in response to numerous stimuli [21]. It is

characterized by alterations that include chromatin condensation and DNA fragmentation, forming extensions in the plasma membrane, and forming apoptotic bodies that are rapidly phagocytosed by macrophages and removed without causing an inflammatory response [24, 25].

As a part of their investigation, Yasuhara *et al*. assessed the potential apoptosis-inducing effect of guanidine alkaloid nitensidine A in SiHa strains (cervical carcinoma strain immortalized by HPV-16) and C33A (strain not immortalized by HPV) [26]. At 24 and 48 h following treatment with 0.25, 0.74, 2.22, 6.66, and 20.0 µg/mL of the compound, the authors detected high induction of cell death by apoptosis. In our study, we found that treatment with *S. cernuum* extract and isolated cernumidine resulted in a significant increase in cell death through apoptosis.

Lopes et al. [7] reported that cernumidine was inactive in a prostate tumor cell line (PC-3). However, in extent literature, natural products are typically tested against several cancer cell lines, as they are easy to grow and require less expensive culture-media conditions. Nevertheless, primary cultures, especially for assessing BPH, provide advantages by better representing the aspects associated with the pathology of the disease [27-33].

In their literature review, Allkanjari & Vitalone described the use of herbal medicine in treating BPH. They demonstrated that several parts of different species, such as *Serenoa repens* (Aricaceae), *Pygeum africanum* (Rosaceae), *Curcubita pepo* (Cucurbitaceae), and *Urtica dioica* (Urticaceae), have been used in patients with mild to moderate BPH symptoms [34]. Based on preclinical studies involving these species, several mechanisms of action have been proposed, including 5α-reductase inhibition, α-adrenergic antagonism, and estrogen receptor inhibition. Findings yielded by randomized clinical trials involving these plant products also indicate significant efficacy in improving urinary symptoms and diminishing BPH adverse effects.

Our findings further indicate that *S. cernuum* hydroalcoholic extract and its isolated cernumidine compound induce cell apoptosis at lower concentrations and cell necrosis at higher concentrations. However, these findings should be confirmed by additional *in vitro* and *in vivo* experiments. The cell death process by necrosis is not controlled and may affect large tissue areas, and it would not be a desired treatment outcome. Therefore, lower concentrations of cernumidine might be a better strategy, as this would ensure that only apoptosis is induced.

Regarding the safe oral use of *S. cernuum* extracts, Almança et al. [35] performed orally acute and sub-chronic toxicological evaluation in mice with increasing doses of hydroethanolic extract obtained from *S. cernuum* leaves (2, 4, 8, 12, 16, 20, and 25 g per kg of animal body mass). These authors estimated the LD_{50} at 14.50 g/kg and concluded that oral consumption of this plant is highly safe, which supports clinical applicability.

It is important to highlight that this plant has been used in folk medicine as a tea for a long time. Therefore, its crude extract standardization should be performed via further studies involving both *in vivo* preclinical assays and clinical assays, allowing its eventual adoption as phytotherapeutic medicine. Likewise, the guanidine alkaloid cernumidine could be a suitable starting compound for producing some active semisynthetic derivatives in the future.

5. Conclusion

The results obtained in this study support the hypothesized effect of *Solanum cernuum* on the prostate primary smooth muscle cells, with apoptosis as the suggested mechanism of action,

corroborating the folk use of this plant for ameliorating benign prostate hyperplasia (BHP). The hydroethanolic extract of *S. cernuum* and its isolated alkaloid cernumidine reduced cell proliferation of BPH primary smooth muscle cell culture. Cernumidine's potential for reducing BPH symptoms thus deserves further investigation, including experimental studies aiming to confirm its mechanism of action in different assays, including *in vivo* models.

Author contributions

MM: Methodology, Validation, Investigation, Writing Original draft preparation, Reviewing and Editing. CS, KB, AA: Methodology, Investigation. JB: Methodology, Resource, Investigation. JM: Methodology, Investigation, Formal analysis. RR: Conceptualization, Supervision, Writing- Reviewing and Editing.

Conflicts of interest

We declare no conflict of interest.

Acknowledgment

Mariza Miranda thanks the São Paulo Research Foundation (FAPESP) for the fellowship and bench grant, process. Fellowship # 2020/03078-9.

References

1. Lim K.B. Epidemiology of clinical benign prostatic hyperplasia. *Asian J. Urol.* 2017; 4(3): 148-151. doi: 10.1016/j.ajur.2017.06.004.

2. Egan K.B. The Epidemiology of Benign Prostatic Hyperplasia Associated with Lower Urinary Tract Symptoms: Prevalence and Incident Rates. *Urol. Clin. North Am*. 2016; 43(3): 289-297. doi: 10.1016/j.ucl.2016.04.001.

3. Bearelly P, Avellino G.J. The role of benign prostatic hyperplasia treatments in ejaculatory dysfunction. *Fertil. Steril*. 2021; 116(3): 611-617. doi: 10.1016/j.fertnstert.2021.07.1199.

4. GBD 2019 Benign Prostatic Hyperplasia Collaborators. The global, regional, and national burden of benign prostatic hyperplasia in 204 countries and territories from 2000 to 2019: a systematic analysis for the Global Burden of Disease Study 2019. *Lancet Healthy Longev*. 2022; 3: e754-76. doi: 10.1016/S2666-7568(22)00213-6.

5. Tamalunas A, Keller P, Schott M, Atzler M, Ebner B, Hennenberg M, Stief C G and Magistro G. Benign prostate Hyperplasia current medical therapy, new developments, and side effects. *Ther. Umsch.* 2023; 80(3): 113-122. doi: 10.1024/0040-5930/a001423.

6. Brandão MGL, Pignal M, Romaniuc S, Grael CFF and Fagg CW. Useful Brazilian plants listed in the field books of the French naturalist Auguste de Saint-Hilaire (1779-1853). *J. Ethnopharmacol.* 2012; 143(2): 488-500. doi: 10.1016/j.jep.2012.06.052.

7. Lopes LC, Roman B, Medeiros MA, Mukhopadhyay A, Utrilla P, Gálvez J, Mauriño SG, Moltiva V, Lourenço A and Feliciano AS. Cernumidine and isocernumidine, new type of cyclic guanidine alkaloids from *Solanum cernuum*. *Tetrahedron Lett*. 2011; 52(48): 6392-6395. doi: 10.1016/j.tetlet.2011.09.060.

8. Lopes LC, De Carvalho JE, Kakimore M, Vendramini-Costa DB, Medeiros MA, Spindola HM, Ávila-Román J, Lourenço AM and Motilva V. Pharmacological characterization of *Solanum cernuum* Vell.: 31-Norcycloartanones with analgesic and anti-inflammatory properties. *Inflammopharmacol.* 2014; 22: 179-185. doi: 10.1007/s10787-013-0182-8.

9. Grando R, Antônio M A, Araújo C E P, Soares C, Medeiros M A, Carvalho J E de Lourenço A M, and Lopes L C. Antineoplastic 31-Norcycloartanones from *Solanum cernuum* Vell. *Z. Naturforsch C.* 2008; 63: 507-514. doi:

10.1515/znc-2008-7-807.

10. Ramos AC and de Oliveira RR. A new alkaloid and flavonoids isolated from *Solanum cernuum* leaves by high-performance countercurrent chromatography. *Nat. Prod. Res.* 2017; 31(20): 2405-2412. doi: 10.1080/14786419.2017.1311889.

11. Ramos A C and De Oliveira R R. One-step separation of terpenoids from leaves extracts of *Solanum cernuum* by high-performance countercurrent chromatography. *J. Liq. Chromatogr. Relat. Technol.* 2016; 39(1): 8-12. doi: 10.1080/10826076.2015.1115767.

12. Miranda MA, Mondal A, Sachdeva M, Cabral H, Neto YAAH, Khan I, Groppo M, McChesney JD and Bastos JK. Chemosensitizing Effect of Cernumidine extracted from *Solanum cernuum* on bladder cancer cells *in vitro*. *Chem. Biodivers.* 2019; 16(10): 16-24. doi: 10.1002/cbdv.201900334.

13. Wang S, Lee DY, Shang Y, Liao J, Cao X, Xie L, Zhang T, Liu J and Dai R. The bioactive alkaloids identified from Cortex Phellodendri ameliorate benign prostatic hyperplasia viaLOX-5/COX-2 pathways. *Phytomedicine* 2021; 93: 15-25. 153813 doi: 10.1016/ j.phymed.2021. 153813.

14. Miao L, Yun X, Yang X, Jia S, Jiao C, Shao R, Hao J, Chang Y, Fan G, Zang J, Geng Q, Wichai N and Gao X. An inhibitory effect of Berberine from herbal Coptis chinensis Franch on rat detrusor contraction in benign prostatic hyperplasia associated with lower urinary tract symptoms. *J. Ethnopharmacol*. 2021; 268: 113-126. doi: 10.1016/j.jep.2020.113666.

15. Li T, Xu K, He J, Jahan N, Song J and Wang S. Effects of isocorynoxeine, from Uncaria, on lower urinary tract dysfunction caused by benign prostatic hyperplasia via antagonism of α_{1A}-adrenoceptors. *Toxicol. Appl. Pharmacol*. 2019; 376: 95-106. doi: 10.1016/j.taap.2019.05.018.

16. Zhao Y, Zhang Y, Li Y, Yang M, Yuan J, Cao Y, Xu L, Ma X, Lin S, An J and Wang S Yohimbine hydrochloride inhibits benign prostatic hyperplasia by downregulating steroid 5α-reductase type 2. *Eur. J. Pharmacol.* 2021; 908: 175-183. doi: 10.1016/ j.ejphar.2021. 174334.

17. Miranda MA, Lemos M, Cowart KA, Rodenburg D, Mcchesney JD, Radwan MM, Furtado NAJC and Bastos JK. Gastroprotective activity of the hydroethanolic extract and isolated compounds from the leaves of *Solanum cernuum* Vell. *J. Ethnopharmacol.* 2015; 172: 421-429. doi: 10.1016/j.jep.2015.06.047.

18. Hamakawa T, Sasaki S, Shibata Y, Imura M, Kubota Y, Kojima Y and Kohri K. Interleukin-18 may lead to benign prostatic Hyperplasia via Thrombospondin-I production in prostatic smooth muscle cells. *The Prostate*. 2014; 74(6): 590-601. doi: 10.1002/pros.22773.

19. Kamiloglu S, Sari G, Ozdal T and Capanoglu E. Guidelines for cell viability assays. *Food Front*. 2020; 1(3): 332-349. doi: 10.1002/fft2.44.

20. Lakshmanan I and Batra SK. Protocol for Apoptosis assay by Flow Cytometry using Annexin V staining method. *Bio Protoc.* 2013; 3(6): 1-3. doi: 10.21769/BioProtoc.374.

21. Bras M, Queenan B and Susin SA. Programmed cell death via mitochondria: Different modes of dying. *Biochem.* 2005; 70: 231-239. doi: 10.1007/s10541-005-0105-4.

22. Boujrad H, Gubkina O, Robert N, Krantic S and Susin AS. AIF-mediated programmed necrosis: A highly regulated way to die. *Cell Cycle* 2007; 6(21): 2612-2619. doi: 10.4161/cc.6.21.4842.

23. Hail N, Carter B Z, Konopleva M and Andreeff M. Apoptosis effector mechanisms: A requiem performed in different keys. *Apoptosis* 2006; 11: 889-904. doi: 10.1007/s10495-006-6712-8.

24. Liao X, Tang S, Thrasher J B, Griebling T L and Li B. Small-interfering RNA-induced androgen receptor silencing leads to apoptotic cell death in prostate cancer. *Mol. Cancer Ther*.

2005; 4(4): 505-515. doi: 10.1158/1535-7163.MCT-04-0313.

25. Yasuhara S, Zhu Y, Matsui T, Tipirneni N, Yasuhara Y, Kaneki M, Rosenzweig A and Martyn J A J. Comparison of comet assay, electron microscopy, and flow cytometry for detection of apoptosis. *J. Histochem. Cytochem.* 2003; 51(7): 873-885. doi: 10.1177/002215540305100703.

26. Souza F O, Sorbo J M, Regasini L O, Bolzani V S, Rosa J C, Czernys E S, Valente V, Moreira T F, Navegante G, Fernandes B C and Soares C P. Nitensidine B affects proteins of the glycolytic pathway and induces apoptosis in cervical carcinoma cells immortalized by HPV16. *Phytomedicine* 2018; 48: 179-186. doi: 10.1016/j.phymed.2018.05.016.

27. Russell P J, Russell P, Rudduck C, Tse B W C, Williams E D and Raghavan D . Establishing prostate cancer patient derived xenografts: Lessons learned from older studies. *Prostate* 2015; 75(6): 628-636. doi: 10.1002/pros.22946.

28. Ellis L M and Fidler I J. Finding the tumor copycat: Therapy fails, patients don't. *Nat. Med.* 2010; 16: 974-975. doi: 10.1038/nm0910-974.

29. Gillet J P, Calcagno A M, Varma S, Marino M, Green L J, Vora M I, Patel C, Orina J N, Eliseeva T A, Vineet S, Padmanabhan R, Davidson B, Ganapathi R, Sood A K, Rueda B R, Ambudkar S V and Gottesman M M. Redefining the relevance of established cancer cell lines to the study of mechanisms of clinical anti-cancer drug resistance. *Proc Natl Acad Sci. USA* 2011; 108(46): 18708-18713. doi: 10.1073/pnas.1111840108.

30. Whittle J R, Lewis M T, Lindeman G J and Visvader J E. Patient-derived xenograft models of breast cancer and their predictive power. *Breast Cancer Res.* 2015; 17: doi: 10.1186/s13058-015-0523-1.

31. Sajjad H, Imtiaz S, Noor T, Siddiqui YH, Sajjad A, Zia M. Cancer models in preclinical research: A chronicle review of advancement in effective cancer research. *Animal Model. Exp. Med.* 2021; 4(2): 87-103. doi: 10.1002/ame2.12165.

32. Ibarra-Sánchez M J, Martínez-Aguilar J F and Esparza-López J. Deriving primary cancer cell cultures for personalized therapy. *Rev. Invest Clin*. 2019; 71(6): 369-380. doi: 10.24875/ric.19002832.

33. Killekar K, Puranik S I, Akbar A A, Ghagane S C, Nerli R B, Hiremath M B. Overview of primary cell culture models in preclinical research of prostate and bladder cancer [Internet]. Cell Culture - Advanced Technology and Applications in Medical and Life Sciences. IntechOpen; 2022. doi: 10.5772/intechopen.99493.

34. Allkanjari O and Vitalone A. What do we know about phytotherapy of benign prostatic hyperplasia? *Life Sci.* 2015; 126: 42-56. doi: 10.1016/j.lfs.2015.01.023.

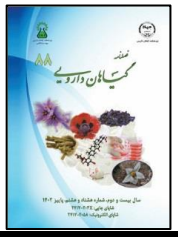
35. Almança C C J, Saldanha S V, Sousa D R, Trivilina L O, Nunes L C, Porfírio L C and Marinho B G. Toxicological evaluation of acute and sub-chronic ingestion of hydroalcoholic extract of *Solanum cernuum* Vell. in mice. *J. Ethnopharmacol*. 2011; 138(2): 508-512. doi: 10.1016/j.jep.2011.09.045.

How to cite this article: Miranda MA, Sares C, Andrade AF, Borges KS, McChesney JD, Bastos JK, dos Reis RB. Effects of ethanolic extract and cernumidine compound obtained from *Solanum cernuum's* Vell. leaves on benign prostatic hyperplasia. ***Journal of Medicinal Plants*** 2023; 22(88): 1-9.
doi:

فصلنامه گیاهان دارویی

Journal homepage: www.jmp.ir

پژوهشکده گیاهان دارویی
جهاد دانشگاهی



مقاله تحقیقاتی

اثرات عصاره اتانولی و ترکیب سرنومیدین بدست آمده از *Solanum cernuum*'s Vell. بر روی هیپرپلازی خوش‌خیم پروستات

ماریزا آبرو میراندا[۱،*]، کلودیا سارس[۲]، آگوستو فاریا آندراد[۳]، کلیتون سیلوا بورجس[۳]، جیمز دیویی مک‌چسنی[۴]، ژایرو کناپ باستوس[۱]، رودولفو بورجس دوس ریس[۲]

[۱] *دانشکده علوم دارویی ریبیرائو پرتو، دانشگاه سائوپائولو، سائوپائولو، برزیل*
[۲] *گروه جراحی و آناتومی، دانشکده پزشکی ریبیرائو پرتو، دانشگاه سائوپائولو، سائوپائولو، برزیل*
[۳] *گروه اطفال، دانشکده پزشکی ریبیرائو پرتو، دانشگاه سائوپائولو، سائو پائولو، برزیل*
[۴] *موسسه تجاری جداسازی ترکیبات طبیعی، آکسفورد، ایالات متحده آمریکا*

اطلاعات مقاله

گل‌واژگان:
سمیت سلولی
سلول‌های عضلانی
پروستات
آپوپتوز
آلکالوئید
Solanaceae

چکیده

مقدمه: گیاه *Solanum cernuum* Vell. (Solanaceae)، بومی برزیل بوده و به طور سنتی برای درمان زخم، آسیب کبدی، عفونت‌های پوستی، سوزاک و هیپرپلازی خوش خیم پروستات (BPH) استفاده می‌شود. **هدف:** بررسی تأثیر عصاره هیدروالکلی و ترکیب سرنومیدین به‌دست‌آمده از برگ گیاه *S. cernuum* Vell. در برابر کشت سلولی عضله صاف اولیه پروستات و ارزیابی اثر بالقوه آنها به روش فلوسیتومتری. **روش بررسی:** عصاره هیدرواتانولی *S. cernuum* از برگ‌های خشک و پودر شده گیاه و به روش خیساندن حاصل گردید. این عصاره حاوی ۴/۸ درصد وزنی-وزنی آلکالوئید سرنومیدین که به روش کروماتوگرافی مایع با کارایی بالا مشخص گردید، بود. کشت سلولی عضله صاف اولیه سرطان خوشخیم پروستات در سنجش تکثیر سلولی و آپوپتوز مورد استفاده قرار گرفت. سلول‌های عضله صاف سرطان پروستات خوشخیم به مدت ۴۸، ۷۲ و ۹۶ ساعت در معرض ۰/۵ - ۰/۱۲۵ میلی‌گرم بر میلی‌لیتر عصاره هیدرواتانولی و ۰/۱ - ۰/۰۲۵ میلی‌گرم بر میلی‌لیتر سرنومیدین قرار گرفتند. **نتایج:** عصاره هیدرواتانولی (۲ میلی‌گرم در میلی‌لیتر) و سرنومیدین (۱ میلی‌گرم در میلی‌لیتر [$۳/۳ \times ۱۰^{-۶}$M]) به ترتیب ۶۰ و ۶۲ درصد رشد سلولی را در ۹۶ ساعت مهار کردند و درنهایت منجر به مرگ سلولی با آپوپتوز شدند. از سوی دیگر، تنها سرنومیدین (۱ میلی‌گرم در میلی‌لیتر) باعث مرگ قابل توجهی در اثر نکروز نسبت به شاهد شد. **نتیجه‌گیری:** یافته‌های به‌دست‌آمده از *S. cernuum* در طب بومی را تأیید می‌کند و نشان می‌دهد که سرنومیدین یک ترکیب مناسب برای مطالعات بیشتر با تمرکز بر درمان سرطان خوشخیم پروستات می‌باشد.

مخففها: HPLC، کروماتوگرافی با کارآیی بالا؛ BPH، سرطان خوش‌خیم پروستات؛ PBS، اندازه غیرطبیعی گلبول‌های قرمز؛ HPV، ویروس پاپیلومای انسانی

* نویسنده مسؤول: marizamiranda@ufl.edu ،marizamiranda2003@yahoo.com.br

تاریخ دریافت: ۱۵ آذر ۱۴۰۲؛ **تاریخ دریافت اصلاحات:** ۲۰ فروردین ۱۴۰۳؛ **تاریخ پذیرش:** ۲۵ فروردین ۱۴۰۳

doi: