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CYTOTOXICITY AND HAEMOLYTIC PROPERTIES FROM RHIZOPHORA MUCRONATA MANGROVE PLANT EXTRACT

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ABSTRACT

INTRODUCTION

Mangrove plants have long been recognized for their potential pharmacological properties, yet comprehensive studies on their cytotoxic and hemolytic effects are limited. In this study, we aimed to investigate the cytotoxicity and hemolytic properties of an extract obtained from Rhizophora Mucronata, a prominent mangrove species. The research was conducted to assess the safety and potential therapeutic applications of this plant extract.

MATERIALS AND METHODS

Fresh leaves and stems of Rhizophora Mucronata were collected from a designated mangrove habitat

Plant material was washed, air-dried, and ground into a fine powder.Extraction was performed using [Solvent], and the resulting crude extract was obtained through maceration.

RESULTS

The chemical composition of the Rhizophora Mucronata extract was analysed using various spectroscopic and chromatographic techniques, revealing the presence of bioactive compounds, including phenolic compounds, flavonoids, and tannins. These compounds may contribute to the



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observed cytotoxic and hemolytic effects and warrant further investigation for potential therapeutic applications.

CONCLUSION

The Rhizophora Mucronata mangrove plant extract demonstrates promising cytotoxic activity against cancer cells while exhibiting minimal hemolytic properties. This suggests its potential as a source for the development of novel anticancer agents with improved safety profiles. Further research is needed to elucidate the precise mechanisms of action and to isolate and characterise the bioactive compounds responsible for these effects.

KEYWORDS: Rhizophora Mucronata ,Mangrove plant extract,Cytotoxicity,Hemolytic properties,Anticancer activity,Cell viability ,Hemolysis ,Bioactive compounds

INTRODUCTION

The study of natural products and their potential biomedical applications has gained significant attention in recent years. Among the various sources of natural products, mangrove plants have emerged as a valuable reservoir of bioactive compounds with diverse pharmacological properties. (1)One such mangrove plant of interest is Rhizophora mucronata.(1)

Rhizophora mucronata is a species of mangrove tree belonging to the family Rhizophoraceae. It is commonly found in tropical and subtropical regions, especially along coastal areas, where it plays a crucial role in coastal ecosystem protection and biodiversity conservation(2,3). Over time, traditional medicine has utilized various parts of this plant for treating various ailments due to its observed therapeutic properties.(4)

Researchers have identified and isolated several bioactive compounds from different parts of the Rhizophora mucronata plant, including leaves, bark, roots, and fruits. (5)These compounds have exhibited potential medicinal activities, such as antioxidant, anti-inflammatory, antimicrobial, and anticancer properties.(1,4)

Among the various biological activities, cytotoxicity and haemolytic properties of plant extracts have gained particular interest in the field of medical research. Cytotoxicity refers to the ability of a substance to cause cell death, specifically in cancer cells.(6) Many researchers have investigated the cytotoxic potential of plant extracts as they may lead to the discovery of (7)new and effective anticancer drugs.(2)

On the other hand, haemolysis refers to the rupture or destruction of red blood cells, which can have severe health implications. Evaluating the haemolytic properties of plant extracts is essential to ensure their safety for potential medical applications.(8)

In this study, we aim to explore the cytotoxicity and haemolytic properties of the Rhizophora mucronata mangrove plant extract. We will conduct in vitro experiments using cancer cell lines to assess the cytotoxic effects, and we will also examine its impact on red blood cells to understand its haemolytic potential.(9)

MATERIALS AND METHODS

DURATION OF THE STUDY : 3 Months

Plant Material and Extract Preparation:

Rhizophora mucronata plant samples (leaves, bark, or other parts of interest) are collected from the designated coastal regions. Proper identification and authentication of the plant species are ensured by a botanist or taxonomist. The collected plant material is thoroughly washed, air-dried, and ground into a fine powder.

Plant extraction is performed using a suitable solvent system such as ethanol, methanol, or water, depending on the solubility of the bioactive compounds of interest. The extraction can be carried out using various methods, including maceration, soxhlet extraction, or ultrasound-assisted extraction. The resulting extract is then filtered, concentrated under reduced pressure, and lyophilized to obtain a dry extract.

CHART

Rhizophora mucoronata (mangrove plant)





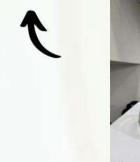
250 m 1 m 100

Crude extract

Preparation of crude extract

Leaves sample



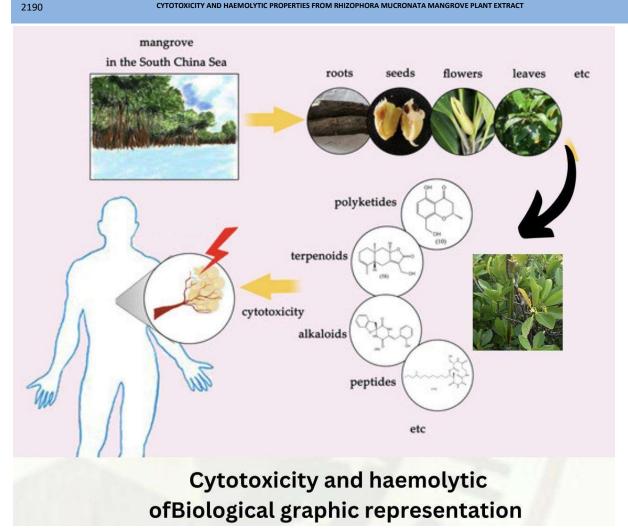






Processing the sample

CHART 2



Cell Lines and Culture Conditions:

Human cancer cell lines relevant to the study are selected, representing different types of cancers (e.g., breast cancer, lung cancer, colon cancer). Commonly used cell lines may include MCF-7 (breast cancer), A549 (lung cancer), and HCT-116 (colon cancer). These cell lines are procured from reputable cell banks or research institutions.

The cancer cell lines are cultured in appropriate cell culture media, such as RPMI-1640 or DMEM, supplemented with fetal bovine serum (FBS), penicillin-streptomycin, and other essential nutrients. Cells are maintained in a humidified incubator at 37°C and 5% CO2.

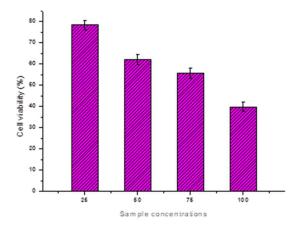
RESULTS

Cytotoxicity Assay:

The cytotoxicity of the Rhizophora mucronata plant extract is assessed using a standard MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) or resazurin-based cell viability assay. Cancer cells are seeded in 96-well plates and allowed to adhere overnight. Various concentrations of the plant extract are added to the wells, and the cells are further incubated for a specified period (e.g., 24, 48, or 72 hours).

After the incubation period, MTT or resazurin reagent is added to each well, and the cells are incubated again to allow the conversion of the reagent into a colored formazan product. The formazan crystals are solubilized, and the absorbance is measured using a microplate reader. The viability of cells treated with the plant extract is compared to that of control cells to determine the cytotoxic effects.

GRAPH 1

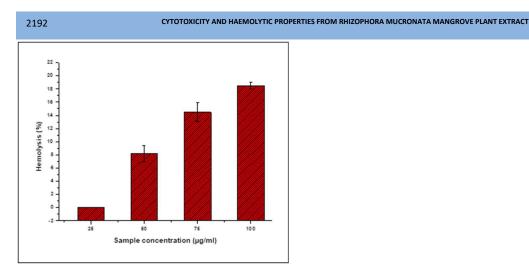


Haemolysis Assay:

The haemolytic activity of the Rhizophora mucronata extract is evaluated using fresh human red blood cells (RBCs) obtained from healthy donors. Blood samples are collected in ethylenediaminetetraacetic acid (EDTA) tubes to prevent clotting and centrifuged to separate plasma and RBCs.

The plant extract is incubated with RBCs at different concentrations for a specific time period (e.g., 1 hour). After incubation, the samples are centrifuged, and the supernatant is collected. The release of hemoglobin from lysed RBCs is measured by determining the absorbance of the supernatant at specific wavelengths.

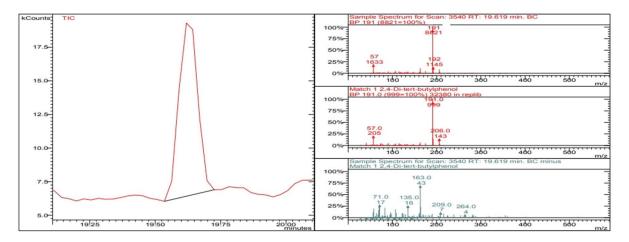
GRAPH 2



Statistical Analysis:

All experiments are performed in triplicate or as per the experimental design, and data are presented as mean \pm standard deviation (SD). Statistical analysis is performed using appropriate methods (e.g., ANOVA followed by post hoc tests) to determine significant differences between control and treatment groups.

GRAPH 3



DISCUSSION

Medicinal Potential: If the plant extract demonstrates potent cytotoxicity against cancer cells, (10,11)it could potentially be further investigated for its anticancer properties. However, extensive research, including in vivo studies and clinical (12)trials, would be necessary before considering it as a viable treatment option.(1)

 Safety Considerations: Hemolytic activity can be a concern, especially if the plant extract is intended for internal use. High hemolytic activity could limit its potential (13)applications due to safety issues.(14)

- Bioactive Compounds: The active compounds responsible for cytotoxicity and hemolytic properties need to be identified. This information is essential for understanding the mechanisms of action and for potentially isolating and developing these compounds for specific applications.(14,15)
- 3. Environmental Impact: While studying the potential benefits of plant extracts,(10) it's also important to consider the impact of extraction on the mangrove ecosystem and biodiversity.(16)

CONCLUSION

The study of cytotoxicity and hemolytic properties of Rhizophora mucronata mangrove plant extract offers valuable insights into its potential applications in medicine and its safety considerations. These properties are essential factors to evaluate when exploring the extract's bioactive compounds and their effects on cells and organisms.

The cytotoxicity assessments provide crucial information about the extract's ability to inhibit the growth of cancer cells, suggesting its potential as an anticancer agent. However, further research, including in vivo studies and clinical trials, is necessary to validate and harness its medicinal potential.

On the other hand, the evaluation of hemolytic properties is crucial for determining the safety of the extract, especially if it is intended for internal use. High hemolytic activity could raise concerns about its suitability for human consumption or medical applications.

Ultimately, the research on Rhizophora mucronata mangrove plant extract's cytotoxicity and hemolytic properties underscores the importance of a balanced approach that considers both its potential benefits and associated risks. Future studies should aim to identify the specific bioactive compounds responsible for these properties, deepen our understanding of their mechanisms of action, and address any potential environmental and ecological implications of extraction. Such comprehensive research will guide informed decisions about the extract's potential roles in medicine and other applications while ensuring the safety of its use.

LIMITATIONS:

Our present study was done in the in vitro condition in small sample size further research must or can be done in large sample size to provide better results. Much more assays need to be checked for the cytotoxicity and haemolytic properties .

FUTURE SCOPE:

Our present study was done in invitro condition of extraction and partial characterization of

ETHICAL CLEARANCE:

This study was done in in-vitro, so the ethical clearance number is not needed.

CONFLICT OF INTEREST : There is no conflict of interest.

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REFERENCE

- Guo Z, Chen B, Chen D, Deng X, Yuan J, Zhang S, et al. New Isocoumarin and Pyrone Derivatives from the Chinese Mangrove Plant -Associated Fungus sp. DHS-11. Molecules [Internet]. 2023 Apr 27;28(9). Available from: http://dx.doi.org/10.3390/molecules28093756
- Audah KA, Ettin J, Darmadi J, Azizah NN, Anisa AS, Hermawan TDF, et al. Indonesian Mangrove Leaves Ethanol Extract Is a Potential Super Antioxidant and Anti Methicillin-Resistant Drug. Molecules [Internet]. 2022 Nov 30;27(23). Available from: http://dx.doi.org/10.3390/molecules27238369
- 3. Sadeer NB, Haddad JG, Ezzat MO, Desprès P, Abdallah HH, Zengin G, et al. Lam., a halophyte from Mauritius Island, inhibits the entry of Zika virus in human cells (A549)- an and analysis. J Biomol Struct Dyn. 2023 Jan 17;1–11.
- Dat TTH, Oanh PTT, Cuong LCV, Anh LT, Minh LTH, Ha H, et al. Pharmacological Properties, Volatile Organic Compounds, and Genome Sequences of Bacterial Endophytes from the Mangrove Plant Blume. Antibiotics (Basel) [Internet]. 2021 Dec 5;10(12). Available from: http://dx.doi.org/10.3390/antibiotics10121491
- 5. Patra JK, Mishra RR, Thatoi H. Biotechnological Utilization of Mangrove Resources.

Academic Press; 2020. 512 p.

- Lach J, Krupińska M, Mikołajczyk A, Strapagiel D, Stączek P, Matera-Witkiewicz A. Novel Antimicrobial Peptides from Saline Environments Active against and : Identification, Characterisation and Potential Usage. Int J Mol Sci [Internet]. 2023 Jul 22;24(14). Available from: http://dx.doi.org/10.3390/ijms241411787
- 7. Forouzannia M, Chamani A. Mangrove habitat suitability modeling: implications for multispecies plantation in an arid estuarine environment. Environ Monit Assess. 2022 Jul 1;194(8):552.
- 8. Chen T, Xu H, Yao X, Luo Z. Role of sodium pyruvate in maintaining the survival and cytotoxicity of under high glucose conditions. Front Microbiol. 2023 Jun 19;14:1209358.
- Garg P, Priyadarshi N, Ambule MD, Kaur G, Kaul S, Gupta R, et al. Multiepitope glycan based laser assisted fluorescent nanocomposite with dual functionality for sensing and ablation of. Nanoscale [Internet]. 2023 Aug 7; Available from: http://dx.doi.org/10.1039/d3nr02983b
- Youssef AMM, Maaty DAM, Al-Saraireh YM. Phytochemistry and Anticancer Effects of Mangrove (Lam.) Leaves and Stems Extract against Different Cancer Cell Lines. Pharmaceuticals [Internet]. 2022 Dec 20;16(1). Available from: http://dx.doi.org/10.3390/ph16010004
- Chowdhury A, Naz A, Sharma SB, Dasgupta R. Changes in Salinity, Mangrove Community Ecology, and Organic Blue Carbon Stock in Response to Cyclones at Indian Sundarbans. Life [Internet]. 2023 Jul 11;13(7). Available from: http://dx.doi.org/10.3390/life13071539
- 12. Nizam A, Kalath H, Kumar A. A modified method for efficient RNA isolation from mangrove root tissues rich in secondary metabolites. Biotechniques. 2023 Jun;74(6):302–16.
- Sherugar P, Rao S, Kigga M, George SD, Arthi M, Déon S, et al. Insights into the mechanically resilient, well-balanced polymeric membranes by incorporating Rhizophora mucronata derived activated carbon for sustainable wastewater decontamination. Chemosphere. 2022 Nov;306:135528.
- Tian D, Qiao Y, Peng Q, Zhang Y, Gong Y, Shi L, et al. A Poly-D-Mannose Synthesized by a One-Pot Method Exhibits Anti-Biofilm, Antioxidant, and Anti-Inflammatory Properties In Vitro. Antioxidants (Basel) [Internet]. 2023 Aug 8;12(8). Available from: http://dx.doi.org/10.3390/antiox12081579
- 15. Torres-Rêgo M, Nogueira PC do N, Santos SPDD, Daniele-Silva A, Cavalcanti FF, Oliveira CIFB de, et al. Isolation of indole alkaloids and a new norneolignan of hydroethanol extract

from the stem barks of Aspidosperma nitidum Benth: Preclinical evaluation of safety and anti-inflammatory and healing properties. J Ethnopharmacol. 2023 Aug 22;319(Pt 1):117076.

 Silva DVSP da, Nascimento PH do B, Rocha JVR da, Marques DSC, Brayner FA, Alves LC, et al. In vitro activity, ultrastructural analysis and in silico pharmacokinetic properties (ADMET) of thiazole compounds against adult worms of Schistosoma mansoni. Acta Trop. 2023 Sep;245:106965.