

ARATI-LESS TUMOR: In Vivo Tumoricidal Activity of Aratiles leaves ethanolic extract with Chitosan derived from Dalagang Bukid against UVB-induced Tumor in Fruit Flies

Courtney Tingson, Ma. Gretchen M. Medianista

TNCHS STEM DEPARTMENT

INTRODUCTION & AIM

Cancer remains a critical global health challenge, with the incidence of UV-induced skin tumors steadily rising, particularly in tropical regions such as the Philippines. Although conventional chemotherapeutic agents are effective in suppressing tumor growth, their high cost, systemic toxicity, and severe side effects limit long-term use and accessibility. These limitations underscore the urgent demand for innovative, safer, and sustainable tumor management strategies. Natural products derived from medicinal plants have emerged as promising alternatives due to their bioactive compounds and lower toxicity profiles. Aratiles (*Muntingia calabura L.*), a locally abundant plant, is rich in phytochemicals such as alkaloids, flavonoids, and triterpenoids known for their tumoricidal and apoptosis-inducing properties. However, the therapeutic potential of these compounds can be limited by instability and inefficient delivery. To address this, chitosan derived from discarded fish scales of Dalagang Bukid (*Caesio cuning*) was utilized as a biodegradable and biocompatible stabilizing agent. This approach not only enhances bioavailability but also promotes environmental sustainability through waste valorization. Using *Drosophila melanogaster Meigen* as an in vivo tumor model, this study evaluates the tumoricidal efficacy of Aratiles leaf ethanolic extract with chitosan against UVB-induced tumors. Overall, the research aims to contribute a competitive, low-cost, and eco-friendly alternative for tumor suppression while advancing sustainable biomedical innovation.

METHOD



Fresh *Muntingia calabura L.* leaves and *Caesio cuning* fish scales were collected, authenticated, and processed following standard laboratory procedures. All reagents, materials, and equipment were sterilized prior to experimentation to ensure accuracy and prevent contamination. Chitosan was extracted from *Caesio cuning* fish scales through sequential deproteinization, demineralization, and deacetylation using alkaline and acidic treatments. The resulting chitosan was washed to neutral pH, air-dried, and stored under sterile conditions until use. Air-dried *Muntingia calabura L.* leaves were pulverized and subjected to maceration in 95% ethanol to extract bioactive compounds. The filtrate was concentrated using rotary evaporation to obtain the crude ethanolic extract.

Drosophila melanogaster Meigen were maintained on a sweet potato-based medium under controlled temperature and photoperiod conditions. Third instar larvae were selected to ensure uniformity in tumor induction and experimental treatment.



Third instar larvae were exposed to ultraviolet B (280–315 nm) radiation for a fixed duration over consecutive days to induce tumor formation. Exposure parameters were kept constant across all experimental groups.



Tumor development was assessed via microscopic examination based on visible phenotypic abnormalities. Tumors were classified according to size, number, and anatomical location for quantitative analysis.



Tumor development was assessed via microscopic examination based on visible phenotypic abnormalities. Tumors were classified according to size, number, and anatomical location for quantitative analysis. Tumor-induced larvae were treated with varying concentrations of *Muntingia calabura* ethanolic leaf extract stabilized with chitosan incorporated into the culture medium. Tumor regression and survival rate were monitored throughout the treatment period.



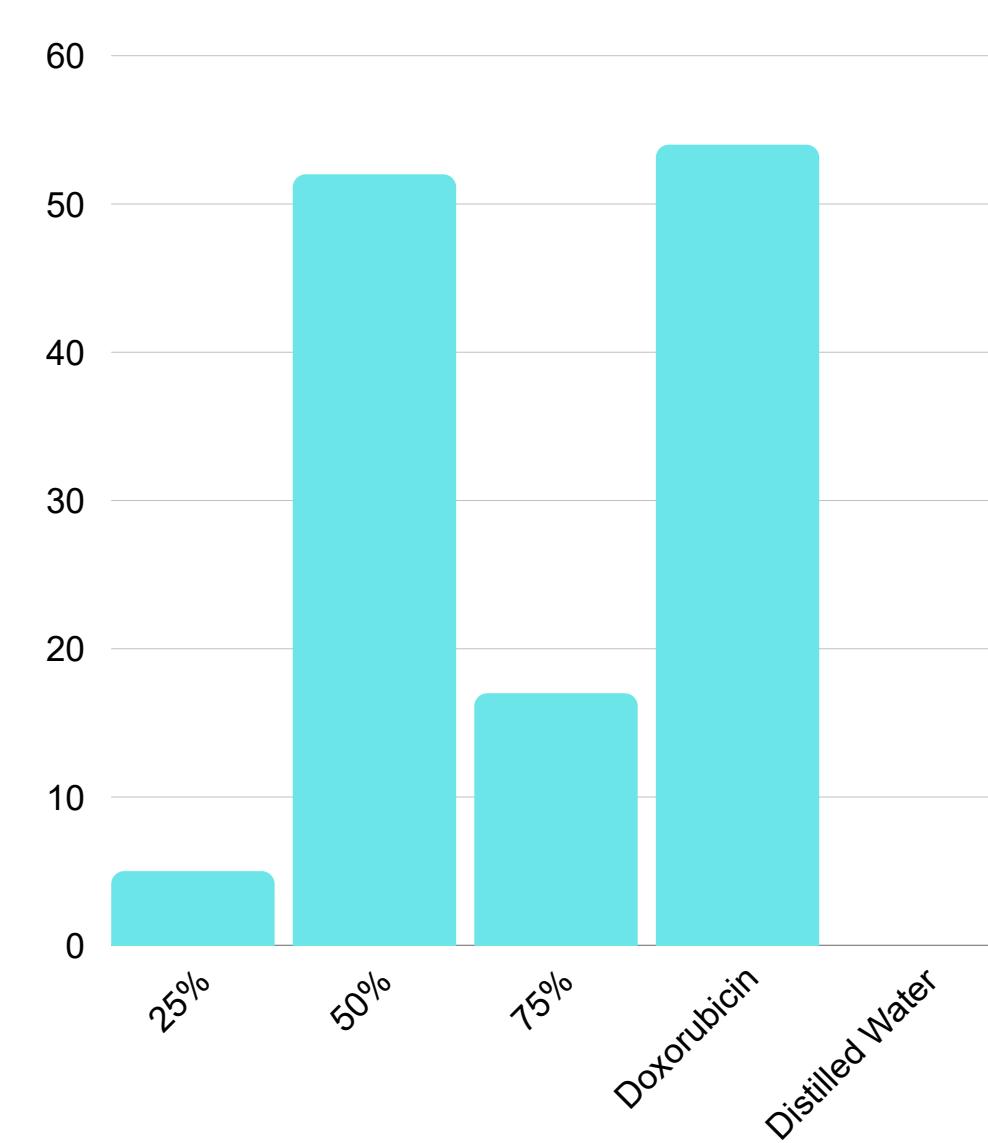
RESULTS & DISCUSSION

Table I. Phytochemical Screening Results

Phytochemical	Test type	Presence
Flavonoids	Shinoda Test	+
Alkaloids	Wagner's Test	+
Triterpenes	Liebermann-Burchard Test	+

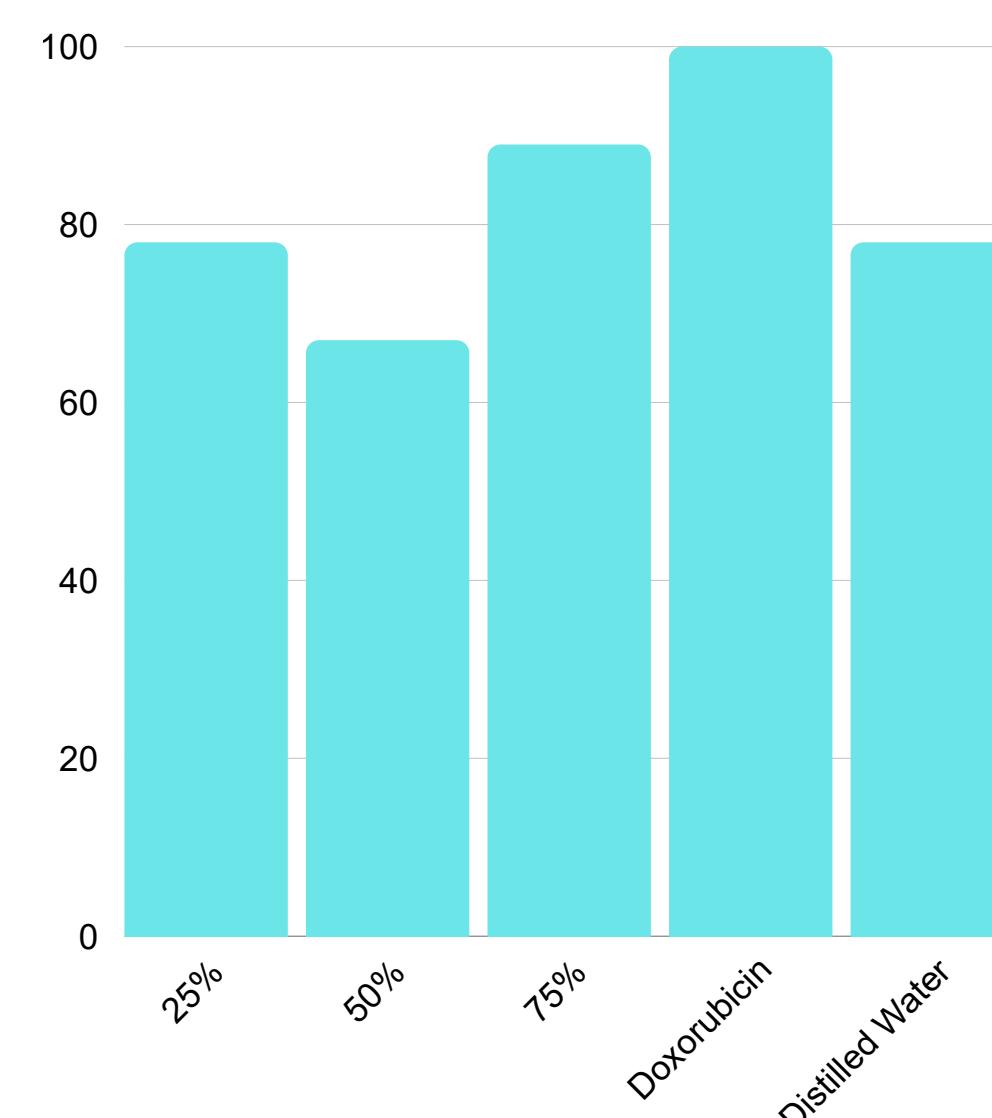
Screening results of Aratiles ethanolic leaf extract confirms the presence of flavonoids, alkaloids, and triterpenes which are compounds linked to their biological activities, particularly their antioxidant, anti-inflammatory, and anticancer properties. The positive results obtained from the Shinoda, Wagner's, and Liebermann-Burchard tests confirm that the extraction method was effective in isolating bioactive compounds from the plant material. The presence of these phytochemicals supports the potential tumoricidal activity observed in the study. Moreover, these compounds are known to induce apoptosis and inhibit abnormal cell proliferation, which may explain the tumor regression seen in treated fruit flies.

Table II. Tumor Regression Assay Results



The tumor regression assay demonstrated varying degrees of tumor reduction across the different concentrations of Aratiles ethanolic extract with chitosan. Among the treatments, the 50% extract concentration showed the highest tumor regression, indicating an optimal balance between efficacy and biological tolerance. The 25% concentration resulted in minimal tumor reduction, suggesting that the amount of bioactive compounds was insufficient to produce a strong tumoricidal effect. In contrast, the 75% concentration showed inconsistent regression, which may be attributed to possible toxicity or physiological stress on the organism. These findings suggest that higher concentrations do not necessarily lead to better outcomes and highlight the importance of dose optimization.

Table III. Mortality Percent Assay Results



The mortality assay results revealed differences in survival rates among the treatment groups. Doxorubicin exhibited the highest mortality rate, indicating its strong cytotoxic effect on both tumor and non-tumor cells. In comparison, the Aratiles extract-chitosan treatments showed lower mortality rates, suggesting a less toxic effect on the fruit flies. The 50% extract concentration demonstrated moderate mortality while maintaining high tumor regression, indicating a favorable therapeutic profile. This result implies that the plant-based formulation may reduce harmful side effects while still effectively suppressing tumor growth. Overall, the mortality data highlight the potential advantage of the extract as a safer alternative to conventional chemotherapy.

CONCLUSION

Aratiles (*Muntingia calabura L.*) leaves ethanolic extract combined with chitosan derived from Dalagang Bukid demonstrated significant tumoricidal activity against UVB-induced tumors in *Drosophila melanogaster*. The 50% extract concentration showed tumor regression comparable to doxorubicin while exhibiting lower mortality, indicating reduced toxicity. These findings help to discover possibilities and to establish a scientific basis suggesting that aratiles leaves with chitosan may have potential relevance for future studies on malignancy or further—cancer treatment.

FUTURE WORK / REFERENCES

Zakaria, Z. A., et al. (2011). In vitro antiproliferative and antioxidant activities of the extracts of *Muntingia calabura* leaves. *Journal of Medicinal Plants Research*, 5(11), 2458–2464.

Amin, S. A., et al. (2017). Antiproliferative activity and apoptosis induction of *Muntingia calabura* L. leaf extract on human cancer cell lines. *Acta Histochemica*, 119(8), 735–744.

Adhikari, R., et al. (2018). Anticancer activity of chitosan and chitosan derivatives: A review of the mechanisms. *International Journal of Biological Macromolecules*, 113, 1076–1086.

Dash, M., et al. (2011). Chitosan—A versatile semi-synthetic polymer in biomedical applications. *Progress in Polymer Science*, 36(8), 981–1014.

Vidal, M., & Cagan, R. L. (2006). *Drosophila* models for cancer research. *Current Opinion in Genetics & Development*, 16(1), 10–16. (Fruit flies are used to study tumor suppressor and oncogene function in cancer biology.)

Vidal, M., & Cagan, R. L. (2006). *Drosophila* models for cancer research. *Current Opinion in Genetics & Development*, 16(1), 10–16. (Fruit flies are used to study tumor suppressor and oncogene function in cancer biology.)

Karger Publishers.

Uhlirova, M., & Bohmann, D. (2006). JNK- and Fos-regulated *Mmp1* expression cooperates with Ras to induce invasive tumors in *Drosophila*. *EMBO Journal*, 25(22), 5294–5304.

Lippincott Journals.

Readout, R., & Smith, O. (2019). *Drosophila melanogaster*: A Model Organism to Study Cancer. *Frontiers in Genetics*, 10, 51. (Comprehensive review of the utility of fruit flies in cancer research, including conserved pathways and drug discovery potential.)

Frontiers.

Waszak, S. M., & Korbel, J. O. (2014). Applications and Advantages of *Drosophila melanogaster* in Cancer Research. *Oncology Reports*. (Review discussing tumorigenesis, invasion, and metastasis in cancer research using *Drosophila melanogaster*.)