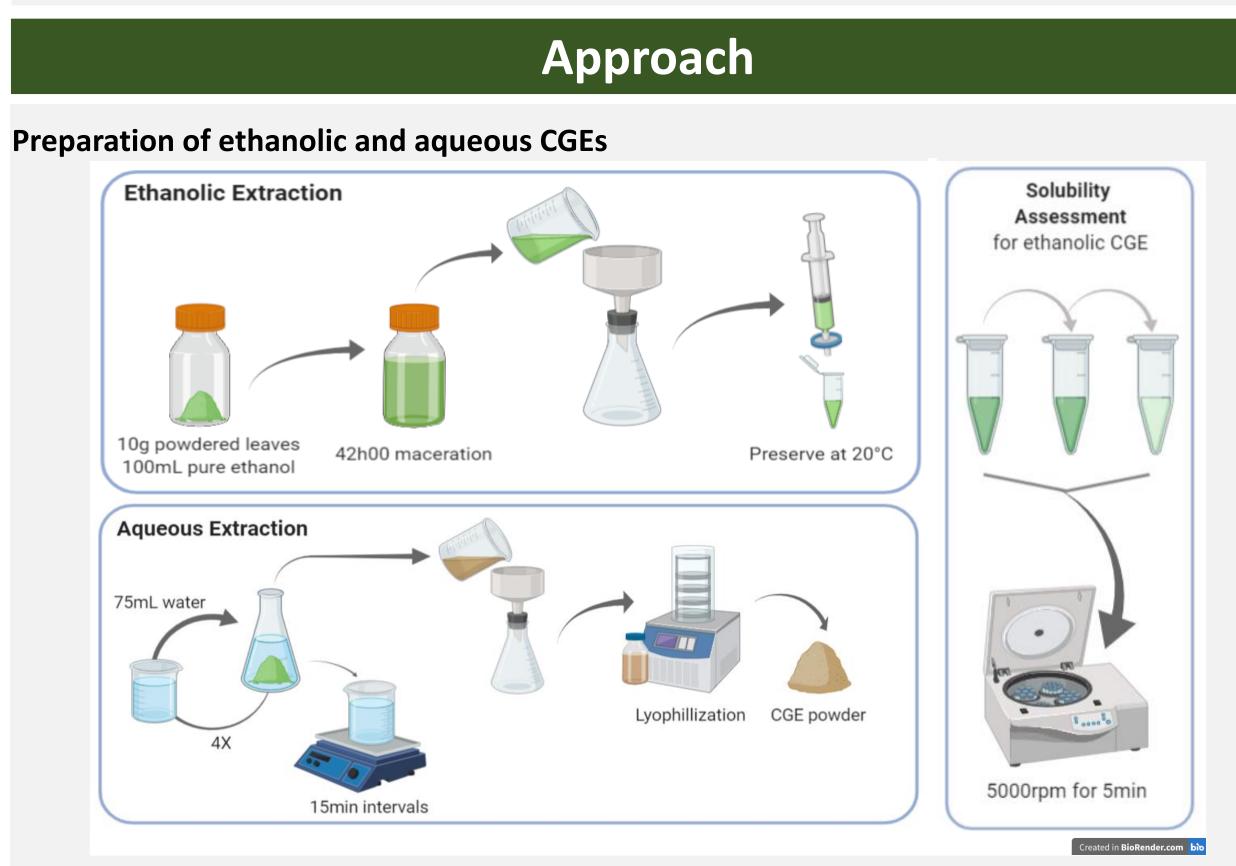


# Antiproliferative properties of Ethanolic and Aqueous Graviola leaf extracts on tongue Squamous Cell Carcinoma cell line-25

### Introduction

Head and Neck cancer (HNC) is a group of cancers which arise in the mouth, nose, throat, larynx, sinuses, or salivary glands. Squamous Cell Carcinoma HNC (HNCSCC), accounts for over 90% of all HNC and represents the seventh most common type of cancer worldwide. Due to their tendency to inhibit cancer-specific pathways such as increased glycolysis, ATP production, and uncontrolled cell proliferation, plantextracted molecules are considered excellent potential therapeutic alternatives to classical chemotherapeutic treatments. Annona muricata, also known as Graviola or soursop, is an evergreen tree native to the tropical regions of the Americas and the Caribbean which antiproliferative properties have been reported in a variety of cancer cell types and models. Here we report the antiproliferative/anticancer properties of ethanolic and aqueous crude Graviola leaf extracts (CGEs) on an in vitro model for HNCSCC. Our results show that the ethanolic CGE significantly inhibits the growth of the Squamous Cell Carcinoma cell line-25 (SCC-25), a property that could be exploited for therapeutic benefits against HNC.

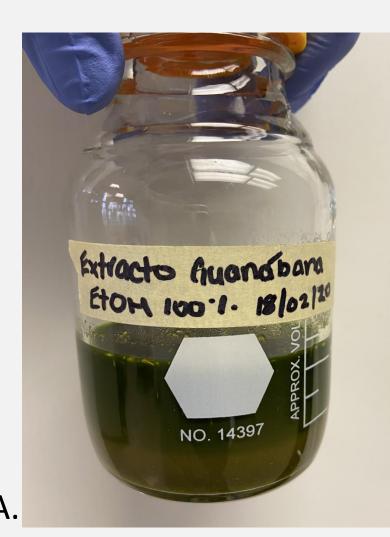


#### **Cell viability Assays**

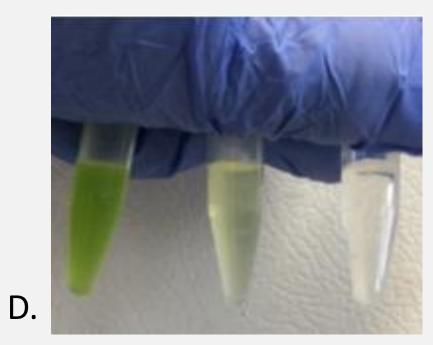
SCC-25 cells were treated with increasing concentrations of CGEs for 24h. The halfviability CGEs doses (GI<sub>50</sub>) were graphically determined and compared to Camptothecin, a positive antiproliferative control. SCC-25 growth inhibition was also assessed microscopically after a 24h treatment with 100 ug/mL of the CGEs.

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#### Results

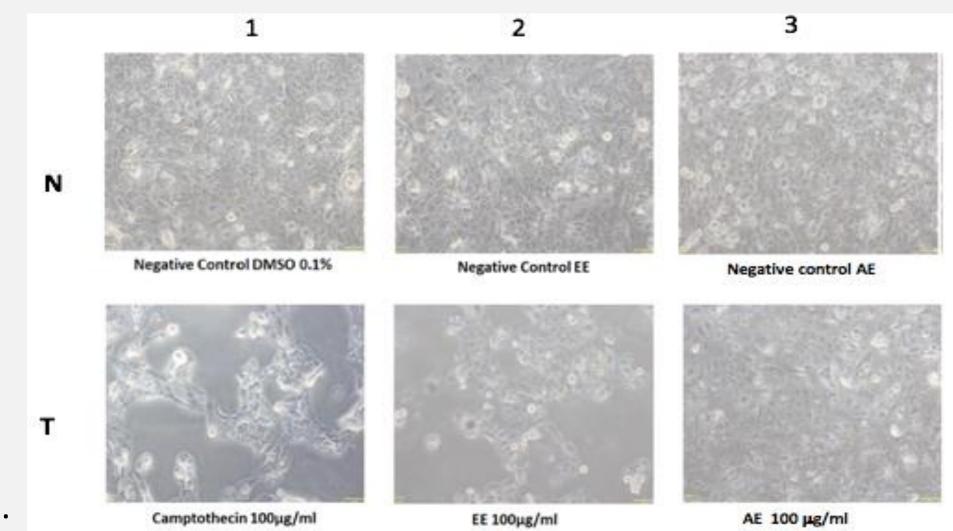


**A** Graviola maceration extraction using pure ethanol as a solvent. **B** Graviola pulverized aqueous extract after lyophilization process.

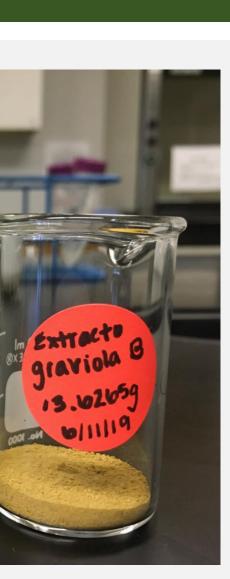




**C** Solubility tests of Graviola ethanolic extracts. **D** Dilutions after centrifugation. No pellet was observed for all three (0.1, 1 and 10%, v:v) dilutions, showing CGEs solubility in water.



**E** Microscopy images of SCC-25 cells before (**line N**) and after (**line T**) treatment with Camptothecin (column 1), ethanolic CGE (column 2) or aqueous (column 3).



Camptothecin

Gl₅₀ (µg/mL)

30.0

**F** Half growth inhibition doses for positive control Camptothecin, ethanolic, and aqueous CGE.

# Conclusion

- Compounds extracted from pure ethanolic CGE were soluble in water at, and 0.1%, 1%, 10% (v:v) dilutions.
- Ethanolic CGE showed a 4 times higher antiproliferative activity than the aqueous CGE on SCC-25 cells.
- We hypothesize that the Graviola leaf contains organic phytocompounds with antiproliferative activities that are present only in the ethanolic extract.

# Future steps

- To characterize and compare the chemical phytoprofiles of the CGEs.
- To test the antiproliferative activity of CGEs on other SCC cell line
- To compare the antiproliferative activity of CGEs on SCCs. vs. non-transformed human gingival fibroblast cells (HGF-1).

### Acknowledgments

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#### PRINBRE HEMTOX LABORATOR

#### Reference

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Ethanol CGE

Aqueous CGE

61.7

274.6

