Assessment of essential oil-loaded nanofibrous mats against the *Escherichia* virus MS2, a mimic of SARS-CoV-2, for potential applications as inner layers in individual protection masks

Samira F. da Silva, Joana Domingues, Joana C. Antunes and Helena P. Felgueiras*

Centro de Ciência e Tecnologia Têxtil (2C2T), Universidade do Minho, Portugal

*helena.felgueiras@2c2t.uminho.pt

Introduction

In December 2019, a novel strain of coronavirus, SARS-CoV-2, was identified. Infected patients revealed symptoms of fever, cough (dry), sore throat, and fatigue, which began manifesting after 5 days of incubation. Hoping to prevent transmission, many countries adopted a mandatory mask use in closed public spaces. However, most mask options display a passive action against COVID-19. To overcome such restrictions, this work proposes the incorporation of anti-viral essential oils (EOs) loaded onto a nanofibrous layer that can be adapted to both community and commercial masks.

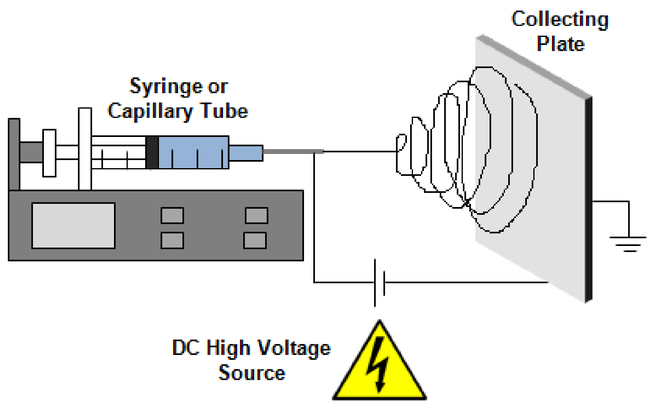
EOs Drawbacks

- cytotoxic at increased concentrations, which prevents systemic delivery;
- present low resistance to degradation by external factors (e.g., temperature, light, moisture);
- highly volatile in their free, unloaded form.

Materials and Methods

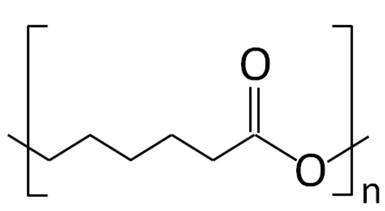
Electrospinning

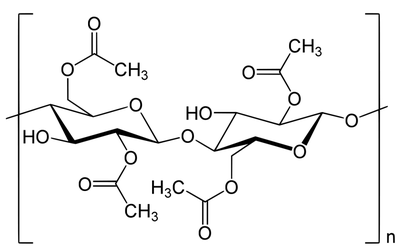
Spinning technique that allows the production of continuous, homogeneous nanofibers.



Polymeric solution preparation

Polycaprolactone (PCL) + Cellulose Acetate (CA)





PCL 14%wt in chloroform/dimethyl formamide (9/1 v/v)

PCL/CA 14%/10%wt (3/1 ratio) in acetone/dimethyl formamide (2/1 v/v)

Processing conditions

PCL = 12.0 kV, speed of 0.7 mL/h, and 17 cm of distance to collector.
PCL/CA = 24.7 kV, speed of 3.2 mL/h, and 21 cm of distance to collector

EOs Minimum Inhibitory Concentrations (MICs)

20 EOs were tested – selected based on their antimicrobial action:
Amyris; Cajeput; Cinnamon leaf; Citronella; Clove; Eucalyptus; Frankincense; Geranium; Himalayan cedar; Lavandin; Lemongrass; Niaouli; Orchid; Palmarosa; Patchouli; Rosemary; Sage; Star anise; Tea tree oil; Wintergreen.

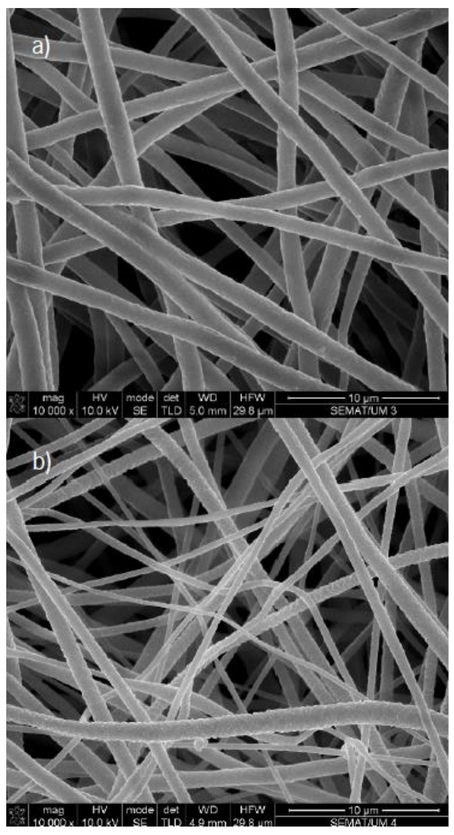
Results and Discussion

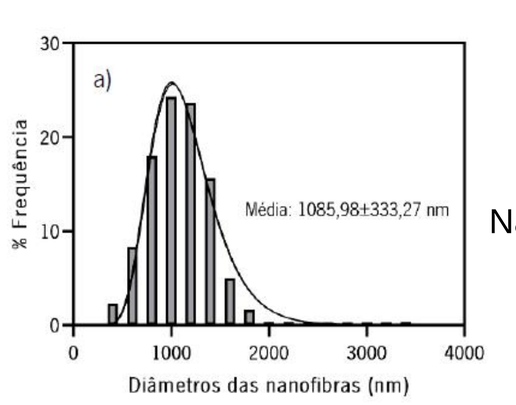
Minimum inhibitory Concentrations (MIC)

List of most effective oils tested against MS2 virus at $1x10^7$ PFUs/mL:

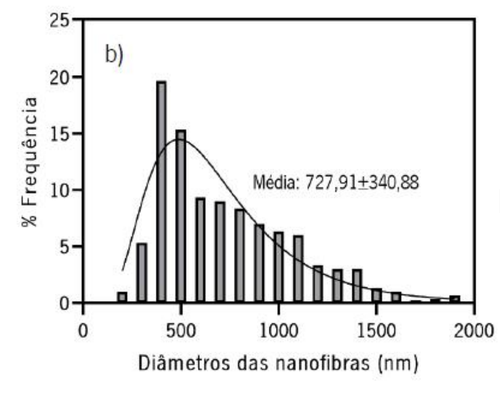
EOs	*MS2 virus* (mg/mL)
Lemongrass	356.0
Niaouli	365.2
Eucalyptus	586.0

Nanofiber Morphology and Diameters





PCL Nanofibers

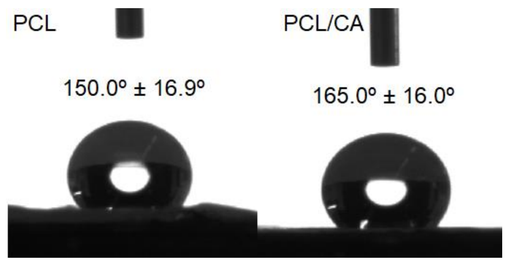


PCL/CA Nanofibers

Degree of Swelling (DS) and Hydrophobicity

DS determined in simulated body fluid:
- DS of PCL = 74.84%
- DS of PCL/CA = 55.30%

Hydrophobicity in distilled water →



EOs Incorporation and Antimicrobial Testing (ongoing)

Mats were loaded with the EOs by immersing the mats in a MIC concentrated EO/ethanol solution for 72 h (saturation). Only 10% of the oil was bonded to the fibers.
Antimicrobial testing via halo determination, verified their diffusion abilities. More importantly, time-kill kinetics testing of the loaded mats attested to the EOs capability to fight the virus MS2 even when bonded to the nanofibers – data based on one repetition (require confirmation).

Conclusions: Data demonstrated the potential of these EOs-loaded PCL/CA nanofiber mats to work as COVID-19 active barriers for individual protection masks.

Acknowledgments
This work is financed by FEDER funds through COMPETE and by national funds through FCT via the projects POCI-01-0145-FEDER-028074 and UID/CTM/00264/2021.