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Incubation of nanopollutants in a circulation-mimicking device : impact on protein corona and endothelial cells

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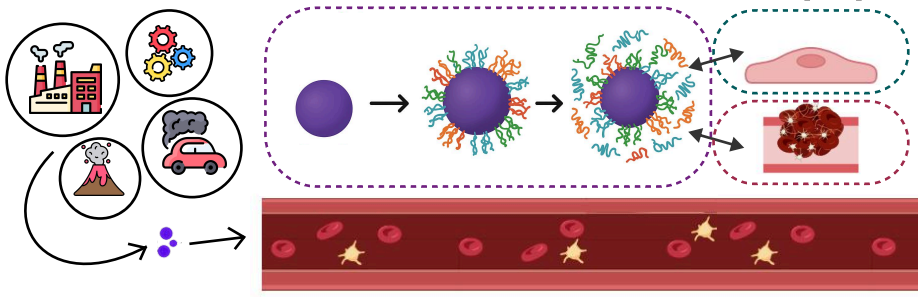
Background

Airborne **nanopollutants (NPs)** are linked to **cardiovascular** disease and can enter the bloodstream due to their small size [1–2].



Mechanism

In biological fluids, NPs form a **protein corona** that alters their physicochemical properties and **cellular interactions.**

Challenge

Understanding NP effects is limited by:

- Dynamic corona evolution
- Complex biofluid interactions
- Lack of physiologically relevant models

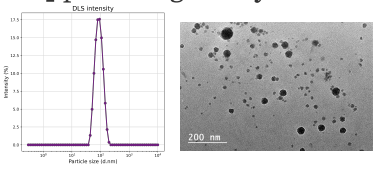
Approach

A first **microphysiological** system to form NP protein corona in **circulation-mimicking,** shear-controlled flow.

Two other devices for evaluation of NP impact on **endothelial response** and **primary haemostasis.**

Nanoparticles (NPs)

SiO_2 polishing slurry



Physicochemical characterization of SiO_2 polishing slurry nanoparticles in PBS by DLS size distribution and TEM imaging.

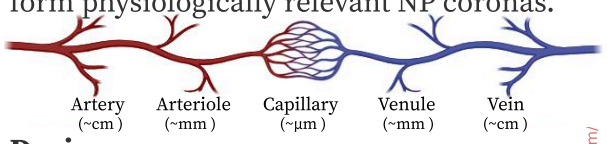
Hydrodynamic diameter: ~90 nm

High polydispersity

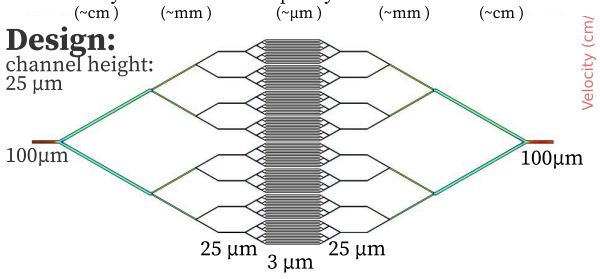
Concentration ~ 2 mg/mL

Physiologically relevant circulation of NPs

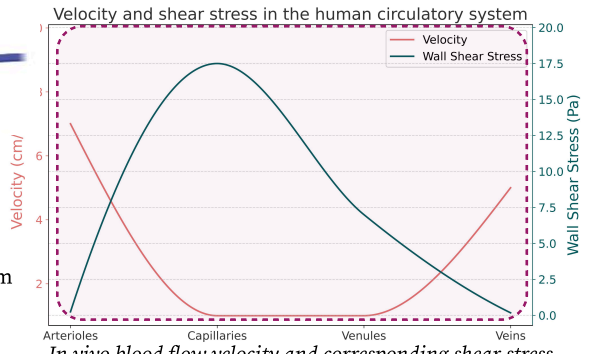
Purpose:
Simulate the human circulatory system to form physiologically relevant NP coronas.



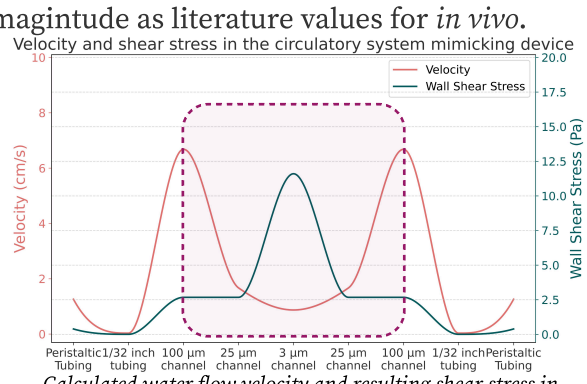
Design:
channel height: 25 µm



Validation:
Simulations of device in the same order of magintude as literature values for *in vivo*.



In vivo blood flow velocity and corresponding shear stress across different regions of the circulatory system. [3]

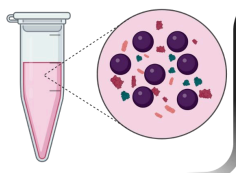


Calculated water flow velocity and resulting shear stress in the circulation-mimicking device.

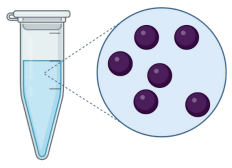
Experimetal Workflow

1. SiO_2 nanoparticles (NPs)

In PBS + 1% Fetal Bovine Serum (FBS)

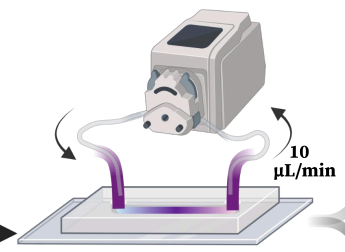


In Phosphate Buffered Saline (PBS)

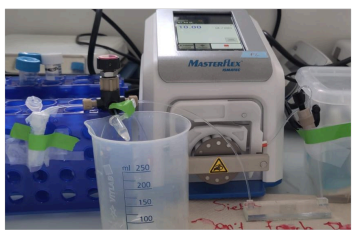


2. Incubation

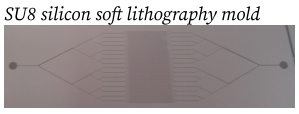
Static incubation



Dynamic incubation in a circulation mimicking microfluidic device

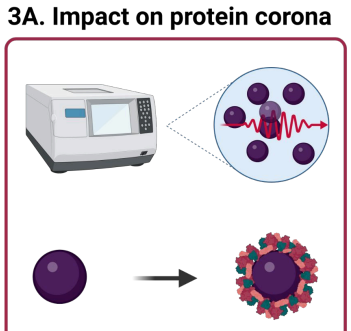


SU8 silicon soft lithography mold



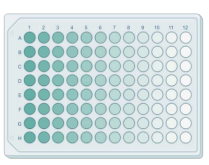
Microfluidic chip: Moulded PDMS on glass slide

3A. Impact on protein corona



Increase in hydrodynamic diameter of the particles upon protein corona formation

3B. Impact on HUVEC

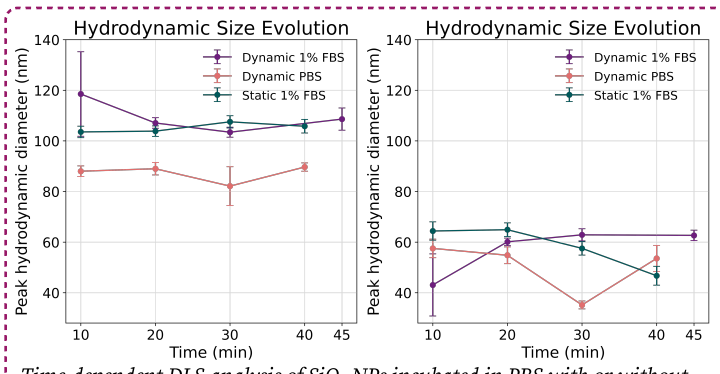


MTT — % Viability

LDH — % Cytotoxicity

HUVEC passage 10 in EGM2 + ECGS
10k cells per well
24h incubation before exposure to NP

Results

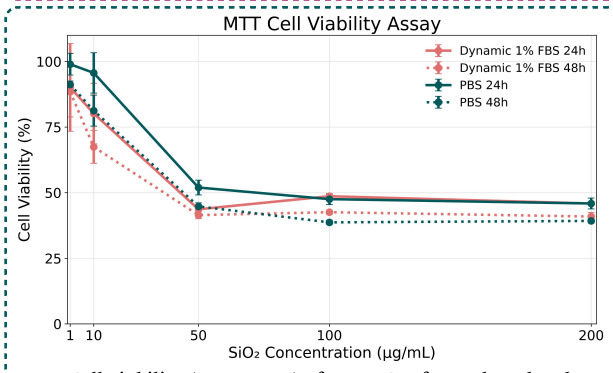


Time-dependent DLS analysis of SiO_2 NPs incubated in PBS with or without 1% FBS showing peak hydrodynamic diameter as determined from both scattering intensity (left) and particle number (right).

1% FBS → increases hydrodynamic diameter by ~10-20 nm

Dynamic vs static → no significant effect

⚠ Particle n.o. correction → inconclusive outcome



Cell viability (MTT assay) of HUVECs after 24 h and 48 h exposure to SiO_2 NPs at increasing concentrations.

Increased NP concentration → decreased HUVEC viability

PBS vs 1% FBS → no significant effect

⚠ LDH assay → no signal above background → excluded from analysis

Key Findings

Protein corona formation increased hydrodynamic diameter

Microfluidic limitation: Minimum incubation time insufficient to resolve size increase

Cytotoxicity: SiO_2 NPs cause decreased HUVEC viability (concentration-dependent)

Circulation effect: No significant change in size or viability after dynamic incubation

Next Steps

Microfluidic blood circulation mimick

On-chip mixing → Dynamic NP–protein interactions

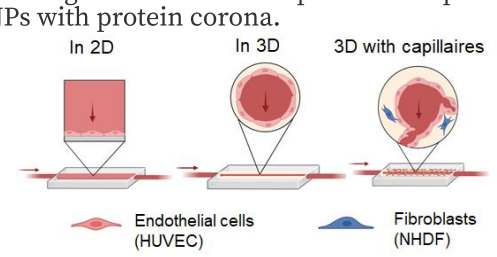
Device redesign → Increase circulation time in microchannel

Blood rheology → Include RBC-driven flow effects

Particle variation → Compare SiO_2 NP sizes

Endothelium-on-Chip

Investigate endothelial responses to exposure of NPs with protein corona.



References

[1] European Environment Agency, Harm to human health from air pollution in Europe: burden of disease 2023, Briefing No. 23/2023 (2023).
[2] H.-S. Kwon, M. H. Ryu, and C. Carlsten, Exp. Mol. Med. 52, 318 (2020).
[3] J. G. Betts et al., Anatomy and Physiology, OpenStax (2022).

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