POSITIVE

Contact information:
www.fp7positive.eu

SEVENTH FRAMEWORK PROGRAMME

A highly integrated and sensitive POrous SIlicon based lab on a chip for multiple quantitaTIVE monitoring of Food allergies at point of care.

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Keywords:

Lab on a chip
Rapid cost-effective multiplexed biochip
integrated sample preparation
Microfluidic
Porous silicon
portable label-free multiallergy diagnostic
biomolecular recognition optical sensor

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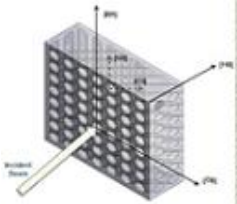

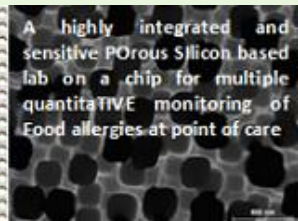

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Nanoscience Laboratory

Consiglio Nazionale delle Ricerche

PHYLOGENE

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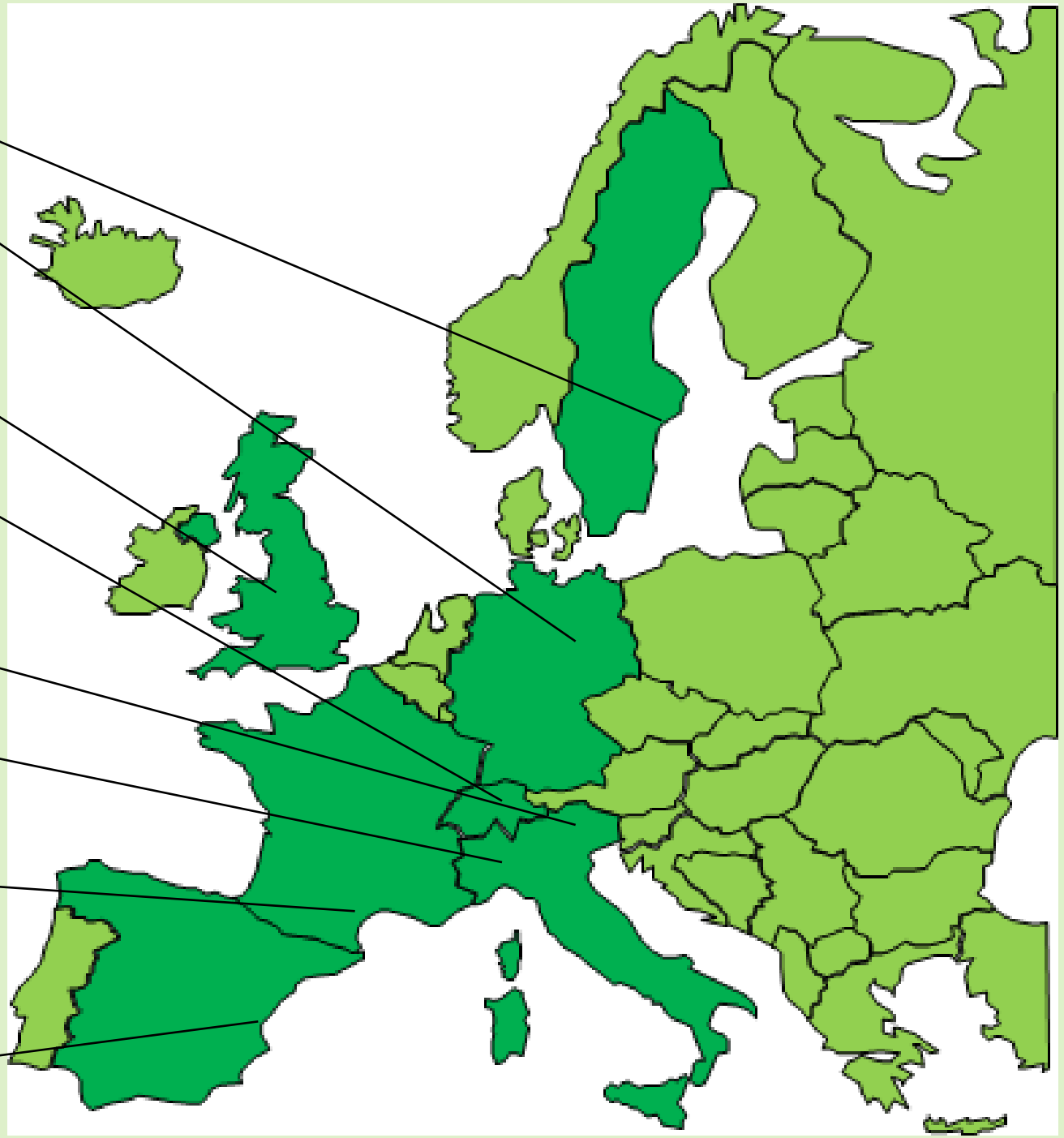

About food allergies

Food allergies – sensitization to food products

Life threatening:
Can provoke clinical reactions (e.g. anaphylaxis), with respiratory and/or cardiovascular problems that might result in death.

- 8% of children
- Over **15 million people in Europe.**
- Increasing prevalence.

Current diagnostic technology:

- Skin prick test - limited.
- Alternatives tests are blood/lab based –
  FEIA, RAST and ELISA
- PoC devices - only a few allergens per test,
  at most a semi-quantitative determination.

<u>POSITIVE technology:</u>
100µL whole blood sample
Sensitization determination to 0.1kU/L (0.24ng/ml)
for 10 food allergies in 15'

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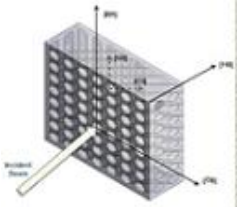
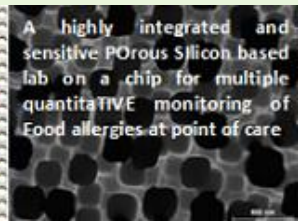




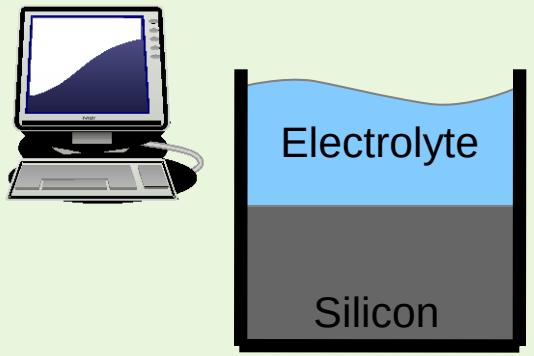
(1) Nanoporous membranes

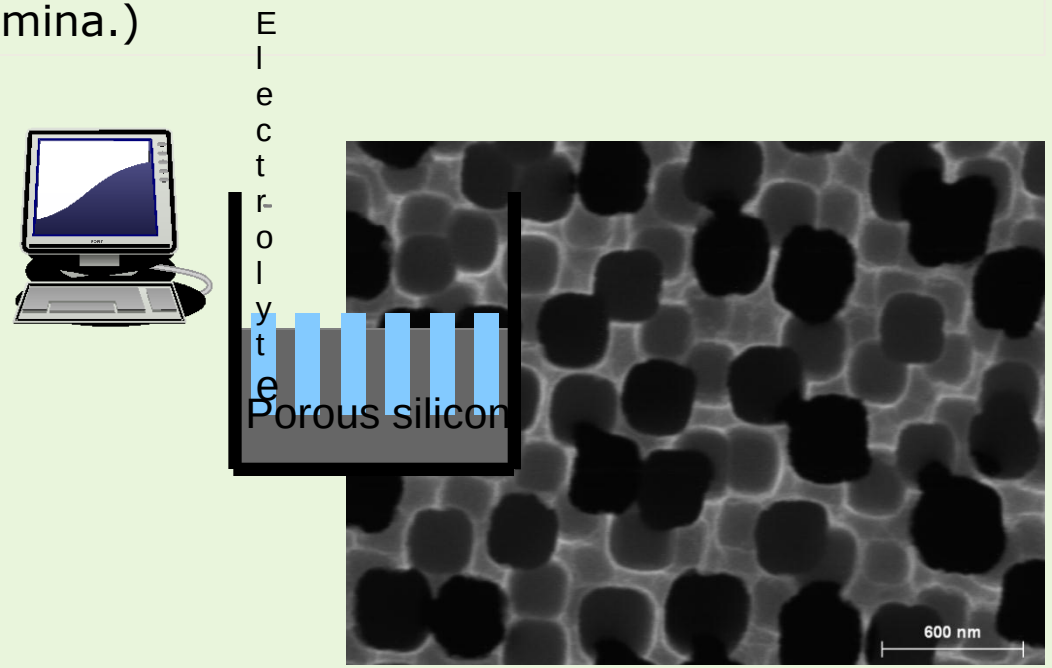
Increasing importance of <u>nanoporous membranes</u> in sensing applications:

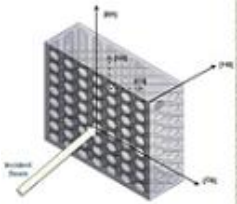
- ✓ Typical length scales maximise formation of bioassay.
- ✓ Enhance response of sensor over planar sensors.
- ✓ Selective size filtering.

Sensing performance directly correlated to quality/reproducibility of structures.

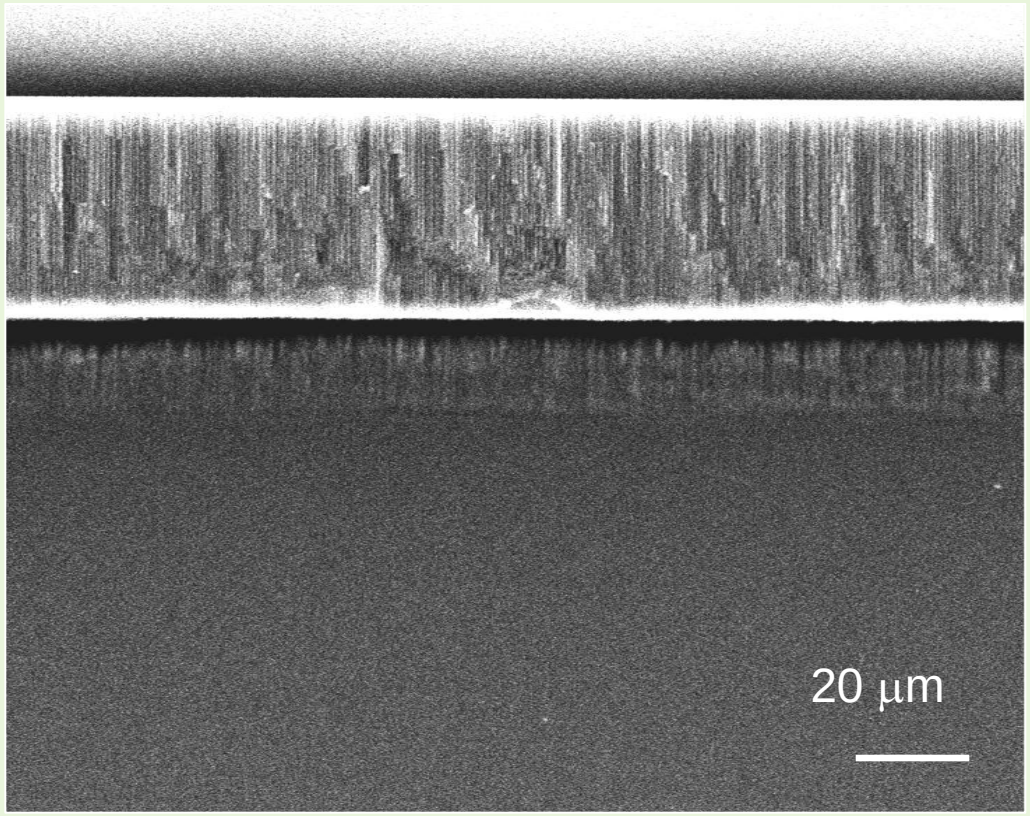
Examples: (<u>Porous silicon</u>….porous alumina.)





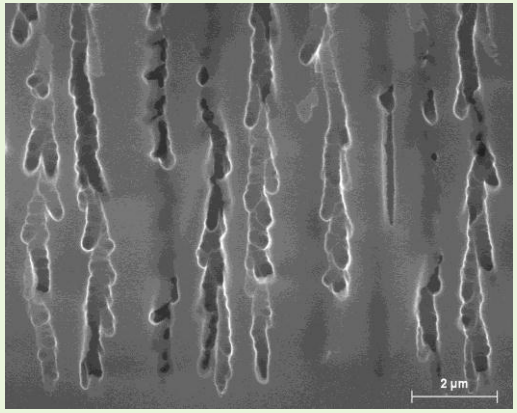


(1) Nanoporous membranes



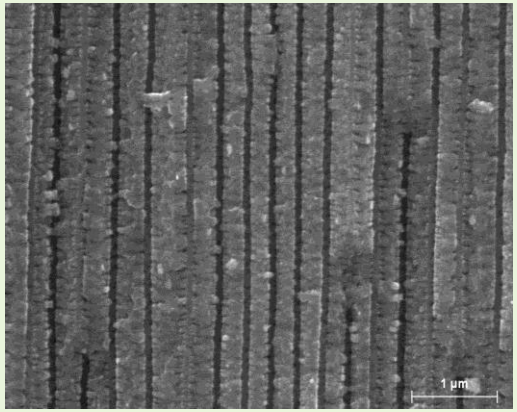
(1) Nanoporous membranes

Resist. > 1 Ωcm



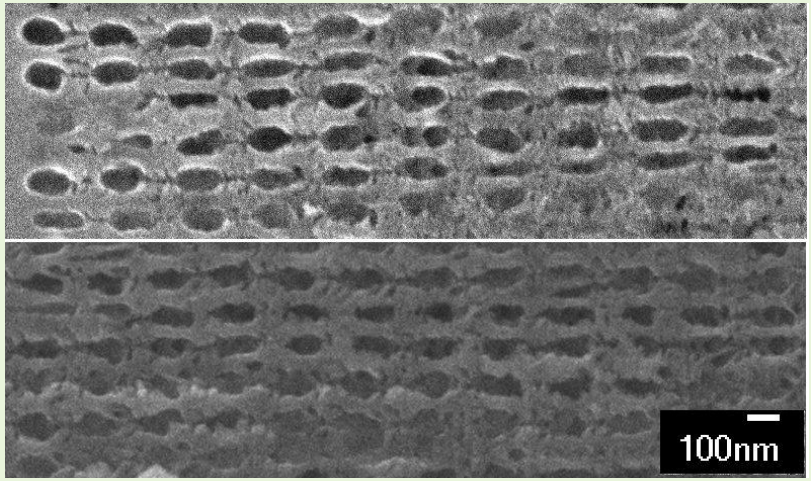
Low porosity, branched pores

Resist. = 0.1 Ωcm



Optimal pore size, low porosity

Resist. = 0.01 Ωcm

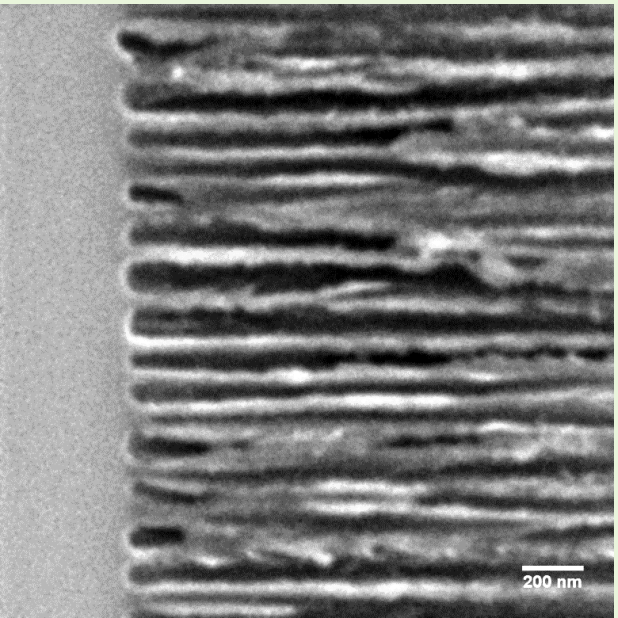


Good pore size & porositiy

Optimal etching conditions → high control over etching process

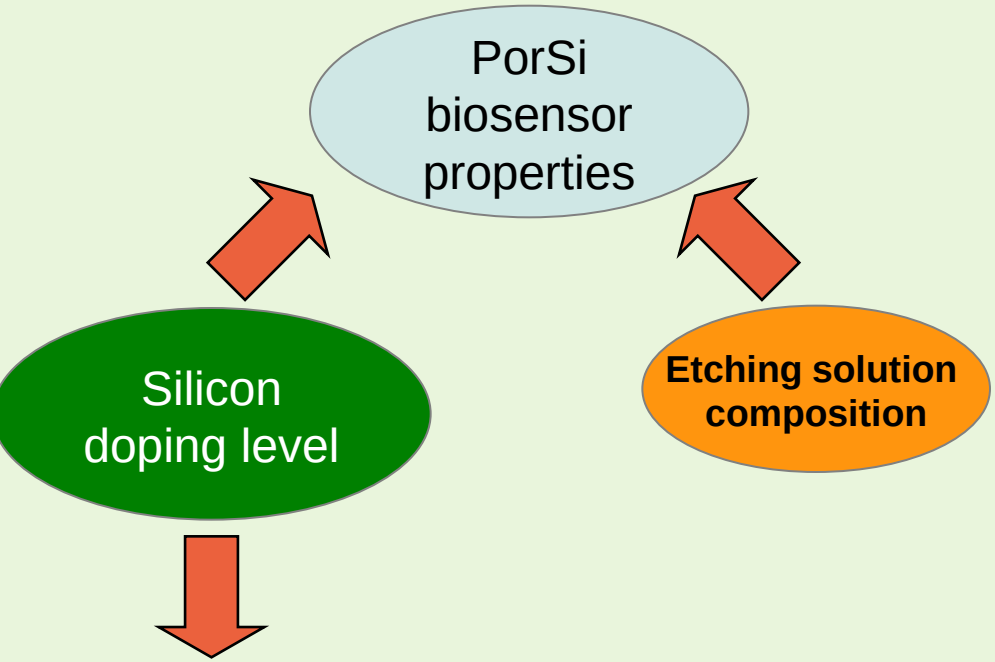
(1) Nanoporous membranes

Solution	Etching rate (um/s)	Pore size (nm)	Refractive index	Membrane thickness
a%HF + b% Ethanol	0.06	50 - 60	2.8135	20
c% HF + d% Ethanol + e% H_2O_2	0.05	30 – 40	2.7862	10
f% HF + g% Ethanol	0.04	70 - 80	1.9279	10



Extremely thin free standing membranes can be realized, down to 3 μm thick and area of around 1 cm^2.

(1) Nanoporous membranes



Greatly dependent on:

1.Etchant solution composition – varied at will.

2.Starting material – limited

[100] n-type commonly 0.01 Ωcm or 0.001 Ωcm, but not 0.1 Ωcm

[111] n-type commonly 0.001, 0.01, 0.1 Ωcm
→ but [111] has no optical anisotropy

Need a larger variety of substrate types on the market to develop OPTIMIZED biosensors

Furthermore nanoporous membranes are of great interest for many analytical uses e.g. DNA translocation!

(1) Nanoporous membranes

PorSi Advantages:

- High refractive index
- Flexible pore tunability
- Well known chemistry
- Good chemical resistance
- No intrinsic photoluminescence

PorSi Disadvantages:

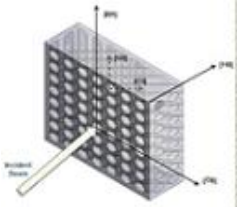
- Rough internal pore surfaces (difficult fluidics)
- Pore size dispersion

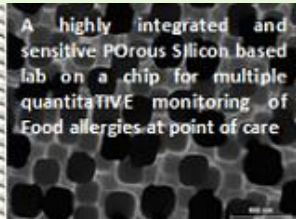
PorAl Advantages:

- Smoother pore surface
- Monodisperse pores size
- Higher optical transparency in the Vis range

PorAl Disadvantages:

- Low refractive index
- Intrinsic photoluminenscence (no labelled assay, no Raman analysis)
- Lower stability in biological environment





(2) Polymer to silicon packaging

To convey advantages of membrane to LoC world for:

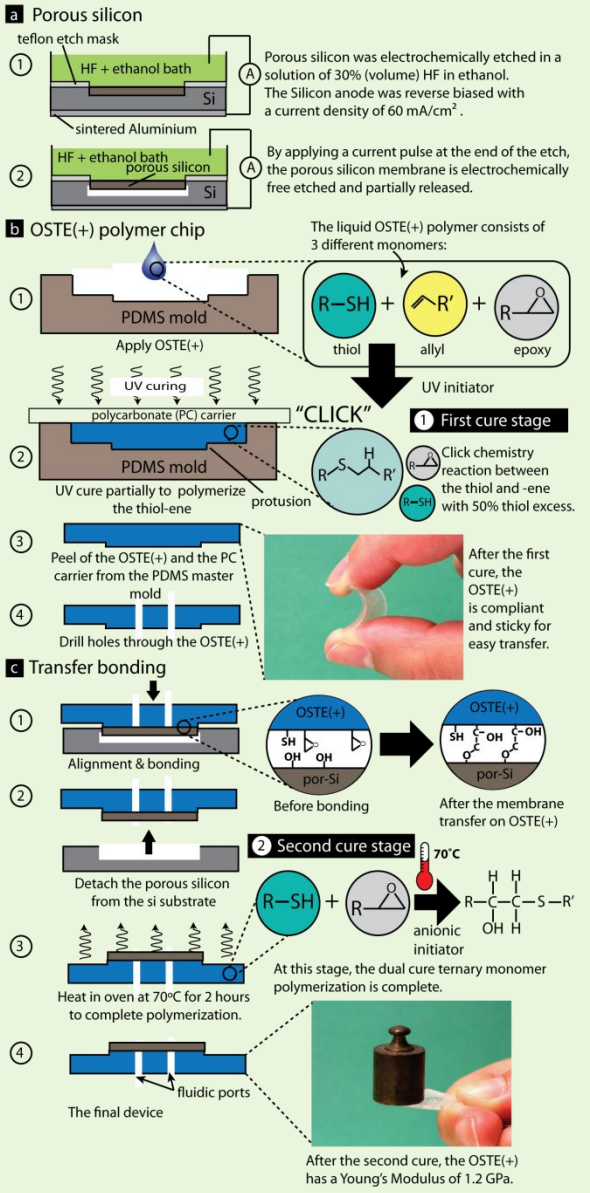
- sensing,
- filtering,
- bioassay scafffoling etc

need **<u>suitable/cost-effective chip technology</u>**
e.g. a novel polymer to Si packaging technologies (RHS).

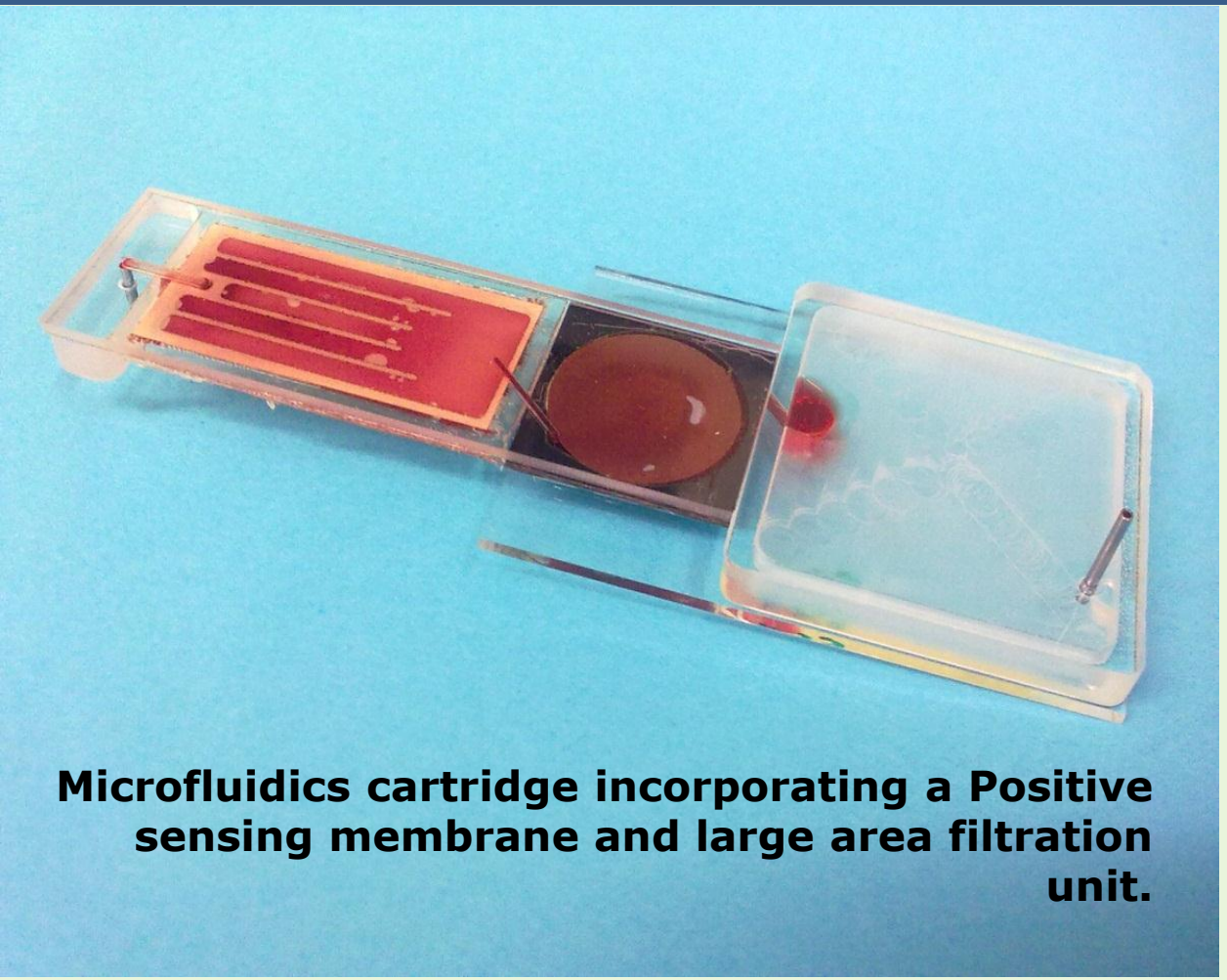
- Biochips - 15×15×1 mm^3 microfluidic OSTE(+) chips, with 6 mm diameter, 30 µm thick porSi membranes (50% porosity, 70 nm pore size) bonded. [1].

- Spin-out Mercenelabs (www.mercenelabs.com) is currently commercializing the novel polymer used.

[1] Saharil, F. Gylfason, K.B. ; Liu, Y. ; Haraldsson, T. ; Bettotti, P.; Kumar, N. ; van der Wijngaart, W. "Dry transfer bonding of porous silicon membranes to OSTE(+) polymer microfluidic devices", MEMS 2012, Paris



(3) Blood filtering technology



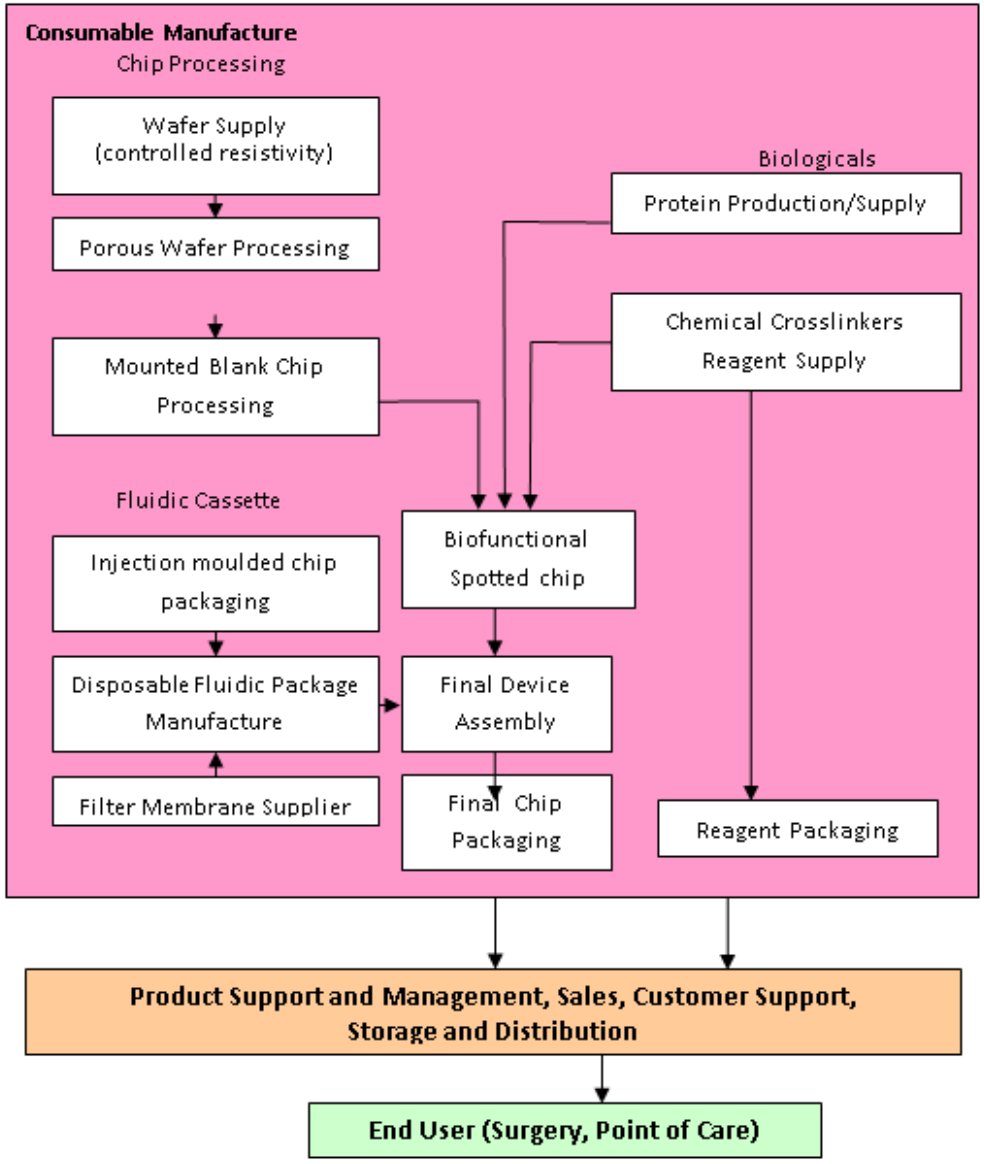
Microfluidics cartridge incorporating a Positive sensing membrane and large area filtration unit.

In Positive filter paths quickly blocked by solids. Instead:

- Guided flow along large area commericial membrane filters.

  -> *several 100 ul of whole blood* filtered and plasma generated for subsequent analysis .
  -> *Low cost implementation* for Positive and ?

Analysis of the supply chain



<u>Two discrete items in product supply chain.</u>

- Reader supply/distribution chain is of that of a conventional system and is well established in the market.
- Diagram shows the supply chain for the consumable device/associated reagents.

Analysis of the supply chain

Companies serving the different steps and identification of gaps:

➢ Chip Processing

Highly volume dependent.
Different suppliers, slightly different ranges of processes, how generic are the process steps?

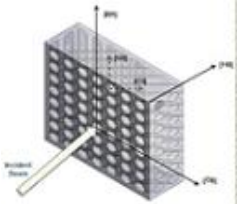
- *Small scale batch silicon semiconductor processing and device packaging. - Tyndall, Cork, Ireland.*
- *Medium scale silicon semiconductor processing and device packaging. - INEX, Newcastle, UK*
- *Porous Chip Manufacture.- SmartMembranes GmbH, Halle, Germany.*

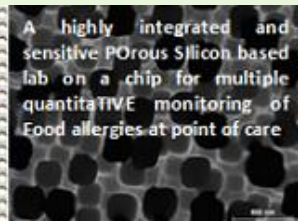
➢ Fluidic Cassette

- *Filter Membrane Suppliers - Millipore Corporation, Billerica, MA, USA. Whatmann- GE Healthcare.*
- *Microfluidic Laminated components –Epigem, UK*

➢ Biologicals

- *Protein Array Spotting - Biotools B&M Labs, S.A.*





Analysis of the supply chain

Supply chain problems (e.g. lack of standard, common language, interest, etc).

1. Semicon manufacturing by 'recipes', don't understand/have control outside:
   - No economic supply of wafers for membrane production with reproducible properties.

2. Biological reagents from different suppliers and even from batch to batch at same supplier will vary in quality and concentration
   - individual batches will need to be verified on a 'standard Positive platform', or the assay has to be designed to accommodate this variation.

3. Quality control in spotting of all 'wells' in each biochip (miss one and chip needs be rejected).

4. Spotted biochips may fail in tests at limit of specification.

5. Lack of standards, for quantifying biosensor performance and characterisation.

6. Lack of standards, only guidelines for activity differences between IgG and IgE.

7. General process development for reproducible and in specification, consumables.

8. General miscomprehension due to different meanings of terminology from biology to medicine, physics and chemistry.

POSITIVE

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Thank you for your attention!

And thank you to

All of the Positive team.

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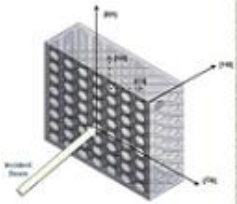

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