



# **FIB** MICRO/NANO FABRICATION OF SILICON MASTER FOR BIOMEDICAL APPLICATIONS

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## CrossBeam® 1540XB by Zeiss

### ❑ Scanning Electron Microscopy (FE-SEM)

*Resolution 1,1 nm*

### ❑ Focused Ion Beam (FIB)

*Resolution 7 nm – Ga<sup>+</sup> Ion Energy 5 ÷ 30 KeV*

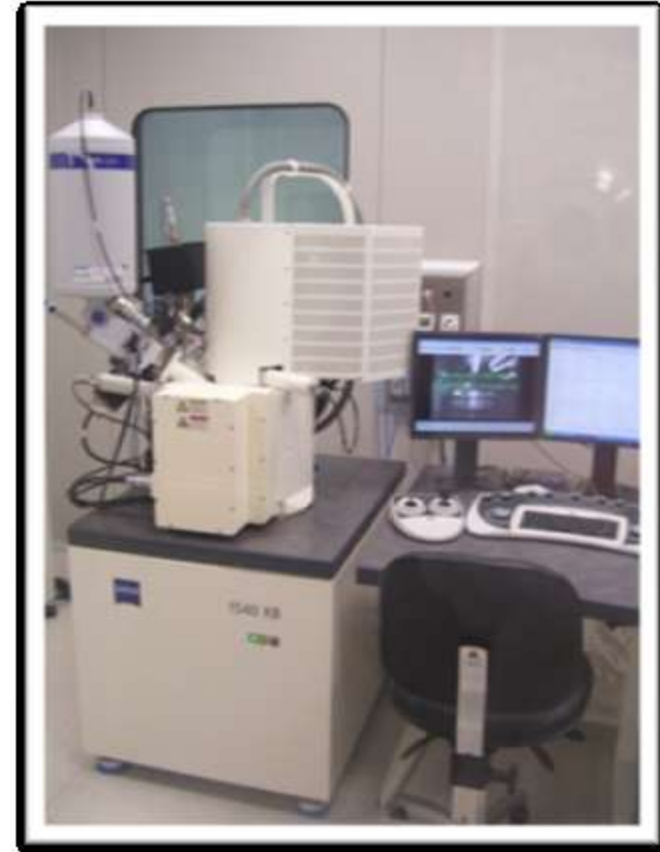
### ❑ Gas Injection System (GIS)

5 gas: Pt – W – Insulator – Water - F

### ❑ EDS Microanalysis

Energy resolution **127 eV** at Mn K<sub>α</sub>

### ❑ STEM



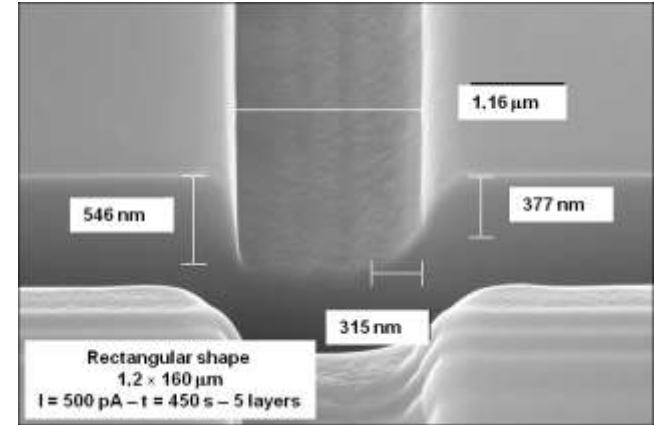
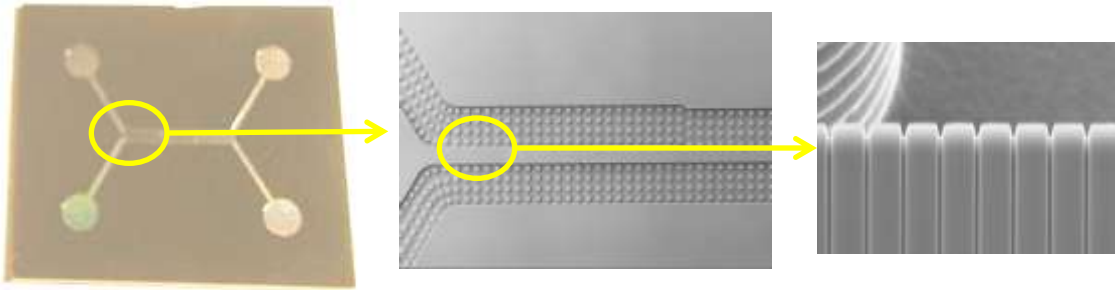


# NanoMolecular Sieves

E. Angeli, G. Firpo, et al, *Lab Chip*, 2011,11, 2625-2629

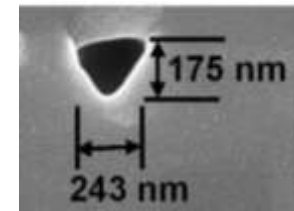
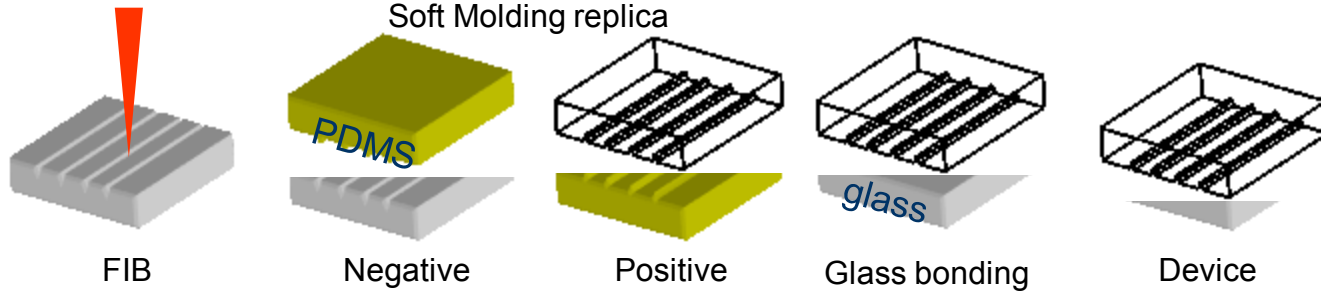


## CHANNELS FABRICATION



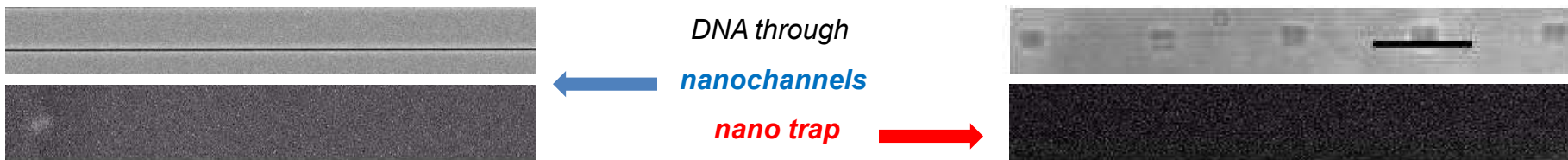
MASTER CROSS SECTIONS

## DEVICE FABRICATION – SOFT MOLDING

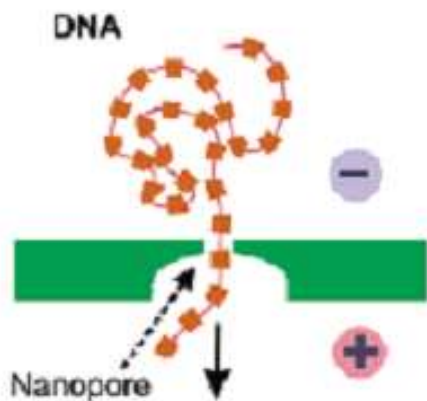


DEVICE CROSS SECTIONS

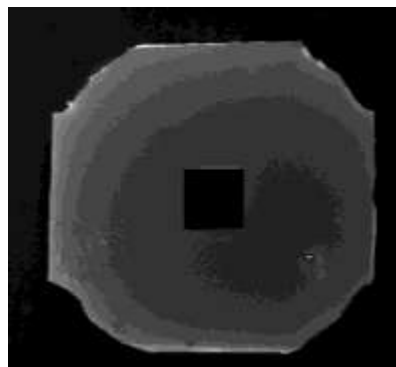
## FIRST RESULTS



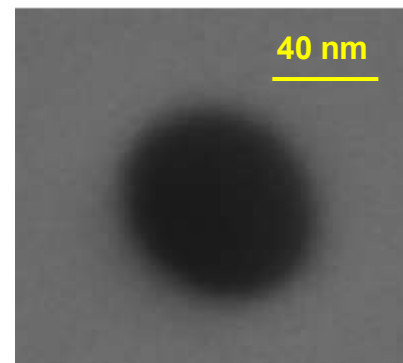
## TRANSLOCATION EXPERIMENT



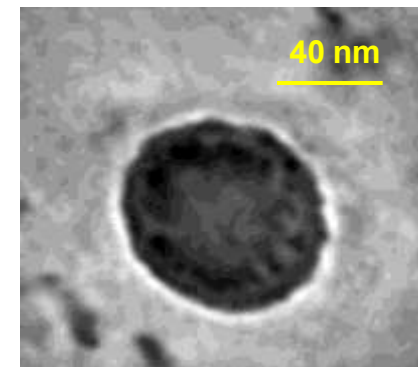
## NANOPORE FABRICATION



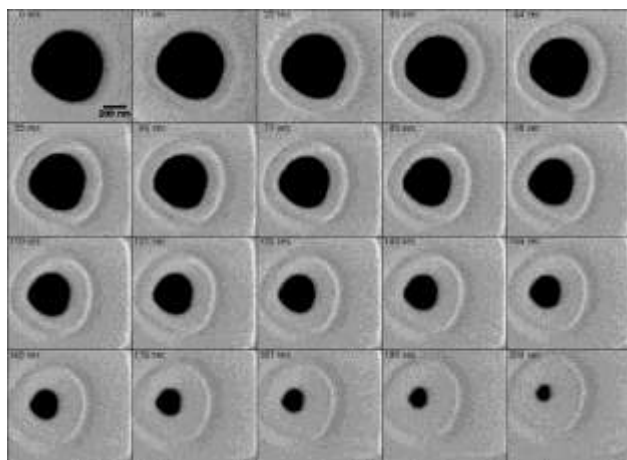
Si<sub>3</sub>N<sub>4</sub> membrane  
100 nm - thick



FIB spot  
1 pA @ 30 keV - t = 1 s  
∅ 30 ÷ 40 nm



Pore Functionalization



SEM rastering @ 200 V – WD 2 mm

## AN OPEN QUESTION . . .

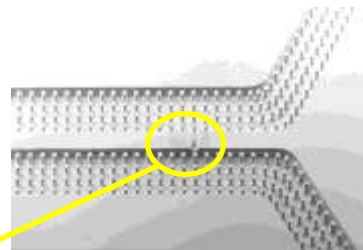
Reduce the pore size is possible with  
SEM rastering

- ✓ Si<sub>3</sub>N<sub>4</sub> pore shrinkage ?  
(*Appl. Phys. Lett.* **90**, 163102 (2007))
- ✓ Carbon deposition?

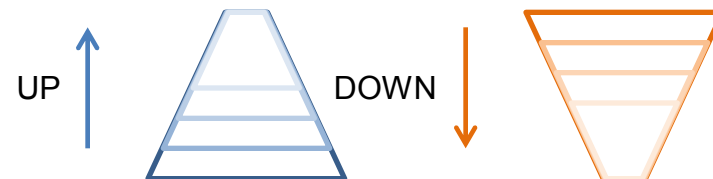
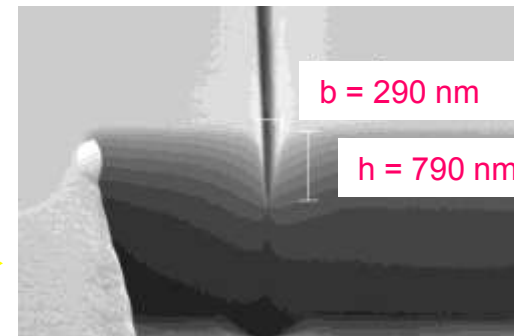
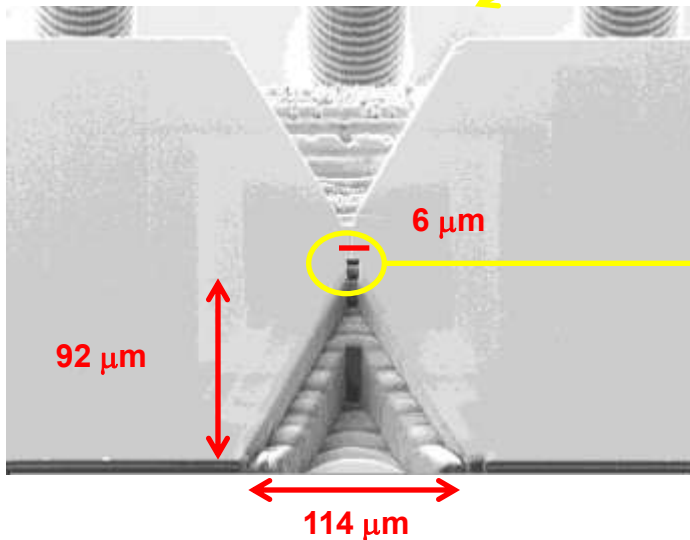
## POLYMERIC NANOCHANNEL DEVICE

*A LOW NOISE alternative to nanopore technology to DNA detection*

FIB is used to fabricate two facing trapezoidal excavations (the depth decreases gradually from 20  $\mu\text{m}$  to 2  $\mu\text{m}$ ) to reduce the distance between the two microchannels till 15  $\mu\text{m}$



The microchannels are connected by a FIB drilled nanochannel, with length  $L = 15 \mu\text{m}$





# Nano Patch-Clamp

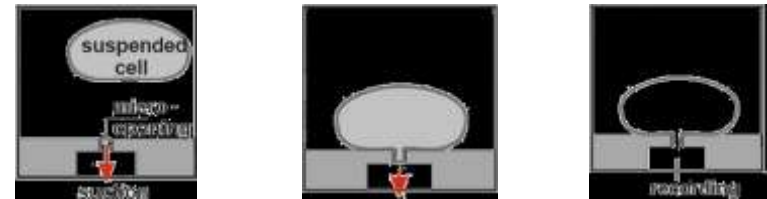


## CLASSICAL PATCH CLAMP



Performing with glass pipette with tip end diameter in the range of 15 micron

## PLANAR PATCH CLAMP



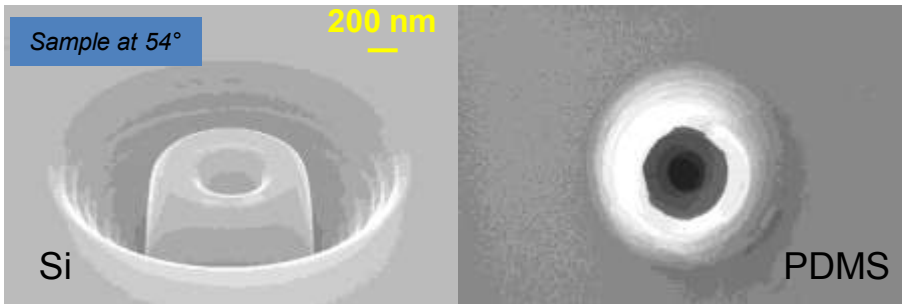
The glass pipette is replaced by a microopening in a planar substrate

## FABRICATION OF PLANAR SUBSTRATE

With **air moulding technique**: from silicon master to PDMS biocompatible polymer substrate

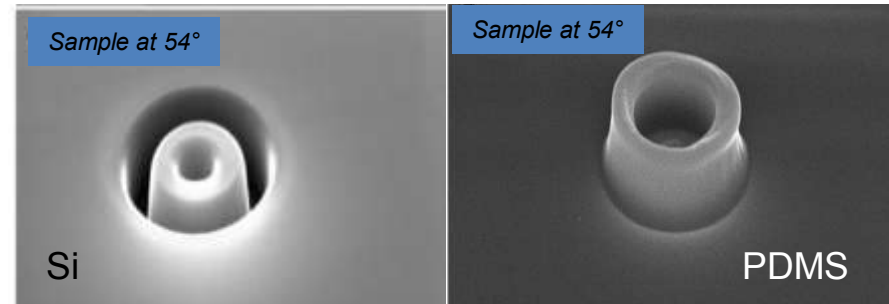
J Eur J Physiol **449** p.56 (2005)

FIB has been used to prototype masters with different morphologies for the creation of polymeric micro-nozzle pipettes similar to the glass pipette traditionally used for patch clamp.



Silicon master with conical morphology and 500 nm pore

Air moulding replica



Silicon master with cylindrical morphology and 500 nm pore

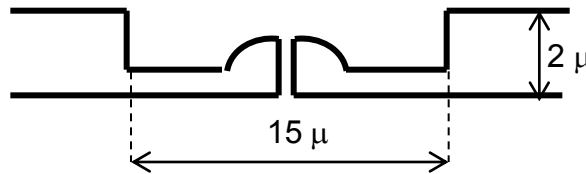
Air moulding replica



# FIB Artefacts



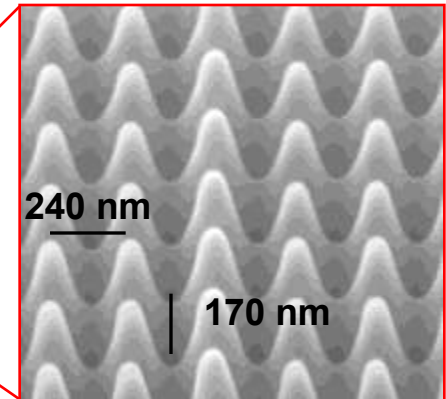
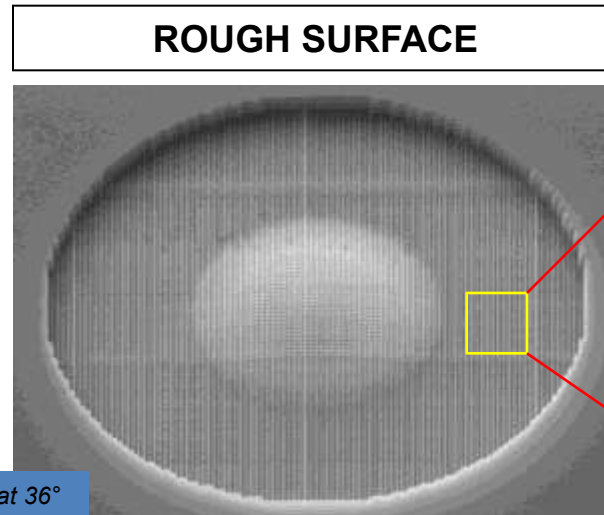
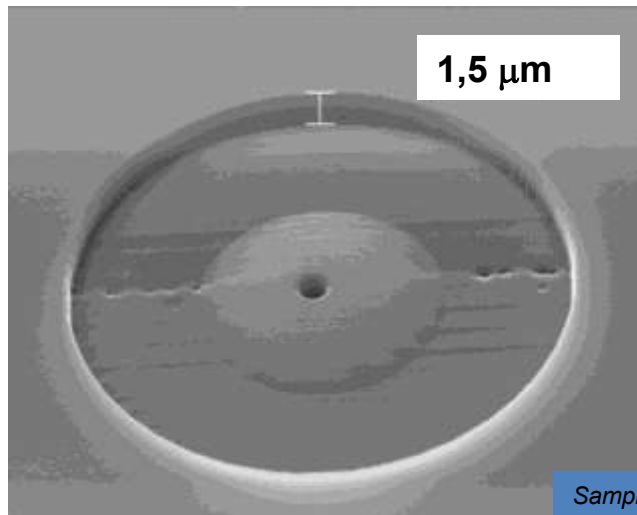
Another type of profile for master morphology for planar patch clamp.



We create a txt file containing a matrix 2D with dwell time such that to realize the profile.



Load txt file on FeatureMill SW of the FIB



TOO ROUGH FOR CELL ADHESION  
**BUT**  
FAST METHOD TO FABRICATE HYDROPHOBIC SURFACES

**FIB parameters**

I = 500 pA  
T = 4500 s

**pixel size 130 nm  
(M = 2800)**



I = 500 pA  
T = 4500 s

**pixel size 243 nm  
(M = 1500)**



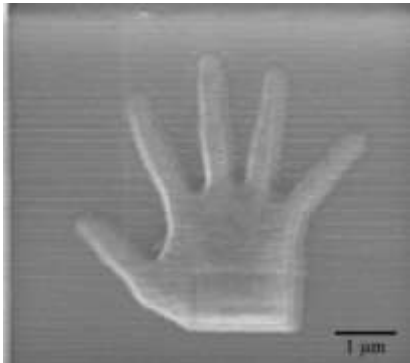


# 3D Reconstruction

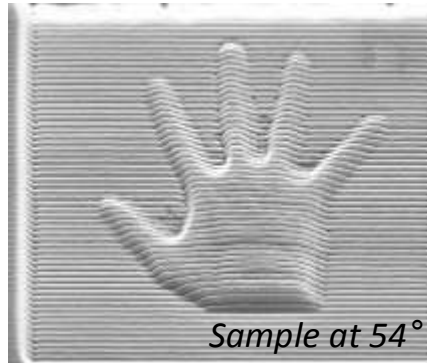
L. Repetto, G. Firpo et al, *Ultramicroscopy* 2009, **109**, 1338–1342



## GEOMETRICAL CHARACTERIZATION OF 3D STRUCTURES by FIB-SEM



Scaled copy of hand obtained by optical profilometry milled on silicon



Milling equispaced parallel lines 40 nm wide, 15 nm deep, 100 nm spaced 1 pA, 30 keV, normal incidence L = 10 μm t = 5 s, Total time 5 min.

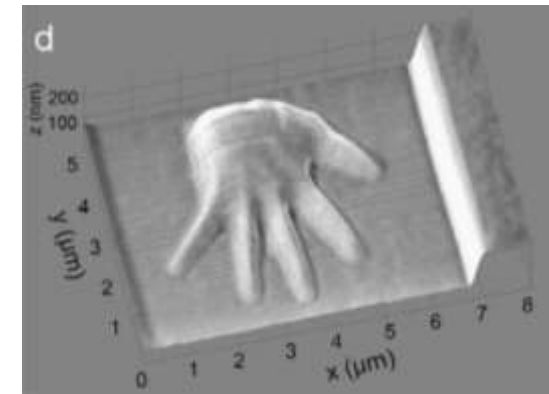
The grooves appear deformed



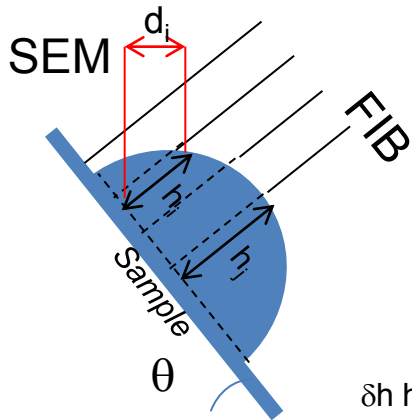
Extraction of geometry of the lines



SW



### PRINCIPLE OF OPERATION



Taking into account tilt correction of SEM view:

$$h_k(x) = \frac{d_k(x)}{\tan\theta} \quad (1)$$

$$\delta h = \frac{\Delta}{\tan\theta}$$

$\delta h$  height resolution -  $\Delta$  pixel size



Topography retrieved by applying Eq. (1) to the pattern of lines. The pixel size of the starting SEM image was 10 nm, and according to Eq. (2) this implies a nominal height resolution of 7nm

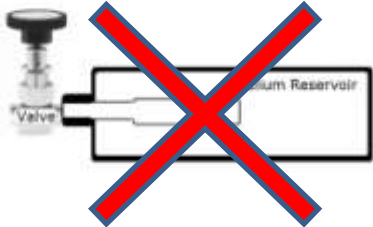




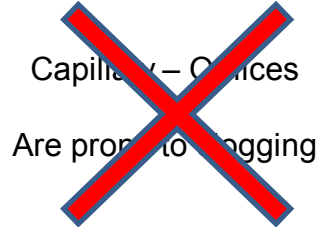
# Leak detection



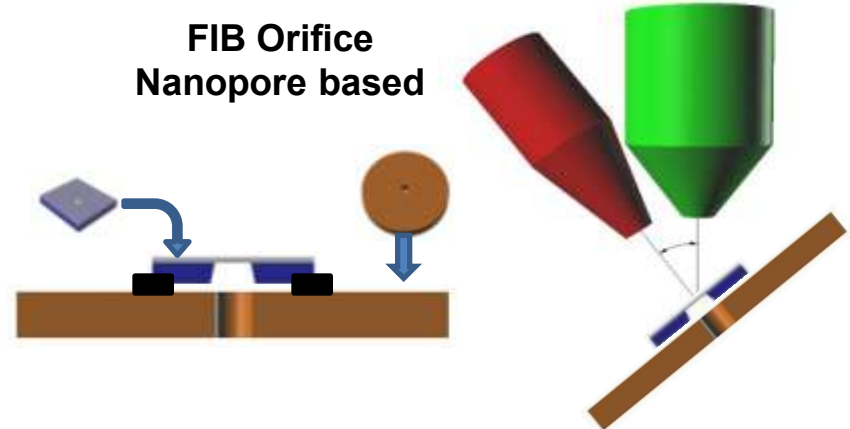
## PERMEATION LEAK



## PHYSICAL LEAK



## FIB Orifice Nanopore based



$d$  = orifice diameter -  $\lambda$  = mean free path gas particles

$\lambda$ (helium) = 170 nm @ 1 atm

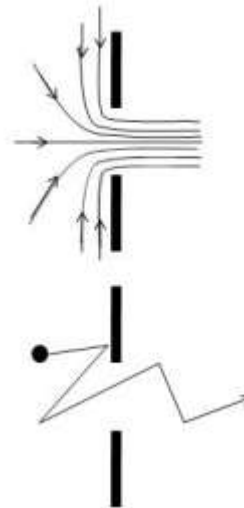
IF  $d < 200$  nm  
(easy and fast with FIB)



WORK IN MOLECULAR FLOW  
REGIME UNTIL ATMOSPHERIC  
PRESSURE



STABLE WITH RESPECT TO  
THE CLOGGING



$d \gg \lambda$   
(viscous flow)

$d \leq \lambda$   
(molecular flow)

Patent: TO2008A000683 - PCT IB2009/054039  
Licensed to Agilent in 2009

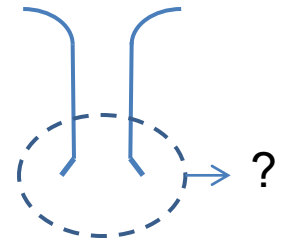
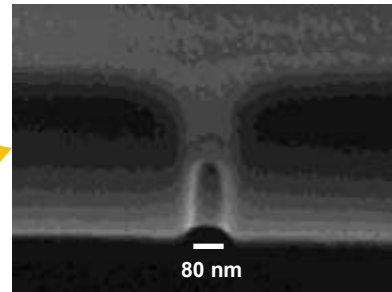
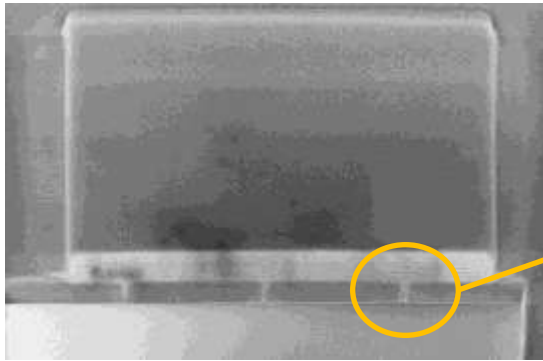


# Leak detection

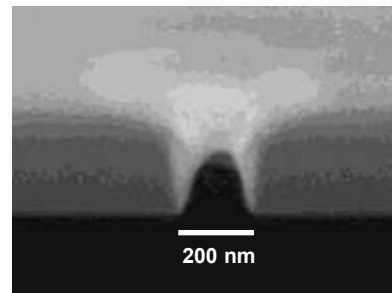


## ORIFICE PROFILES

How to obtain the profiles



FIB nano holes at PTB, Germany



**EMRP**  
European Metrology Research Programme  
# Programme of EURAMET  
The EMRP is jointly funded by the EMRP participating countries  
within EURAMET and the European Union

**Vacuum metrology for production environments**

[← Fastest opening IPIV gate valve of the world tested at PTB](#) 20/09/2012 17:08 Page 3

**Nano holes prove successful**

Nano holes of about 100 nm diameter were produced by focused ion beam (FIB) at the University of Genova. The flow through such elements is molecular up to atmospheric pressure and therefore completely predictable for any gas. Investigations at PTB showed that the experimental flow rate agreed to the calculated one from simulations fully within the uncertainties.

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The research leading to these results has received funding from the European Union on the basis of Decision No 812/2006/EC.  
L&E updated: 2011-07-26

It's a real shape  
or FIB artefact  
due to milling?



See mini-poster session