Magnetic Biosensors and Biomedical Devices

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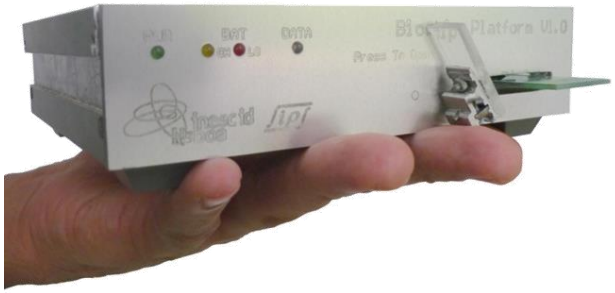
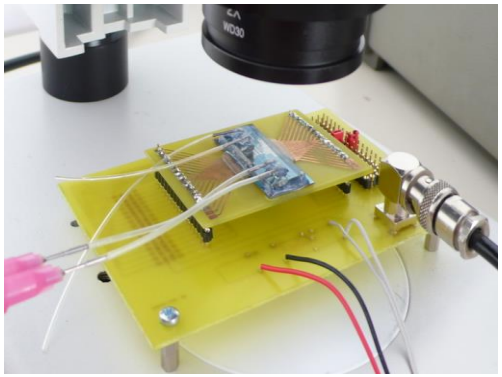
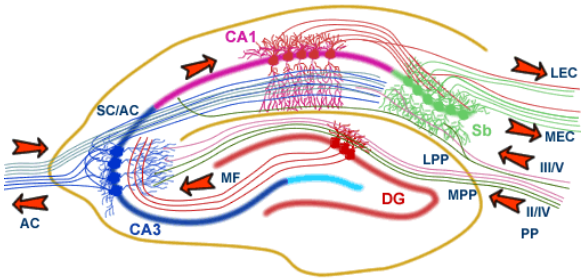
biochips



Magnetic separation



neuroelectronics



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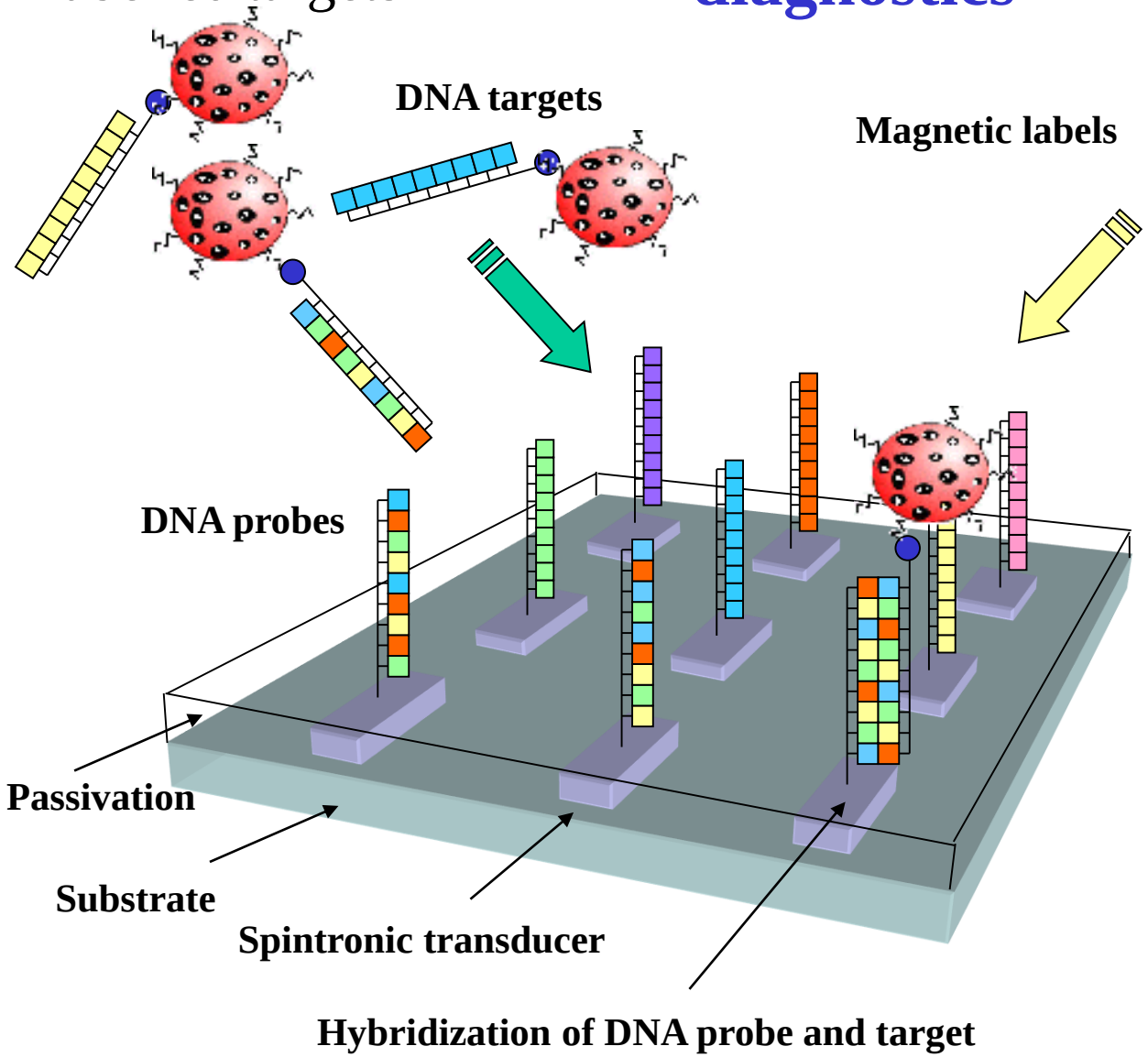
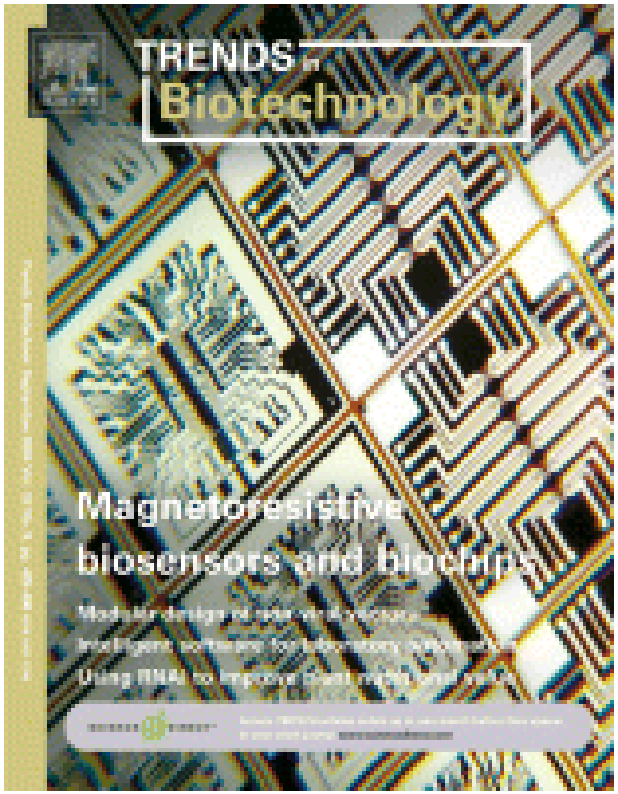
JACA 2013

1-Scalable magnetoresistive biochips for point of care diagnostics



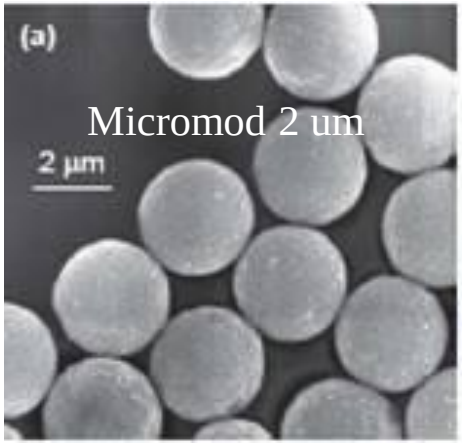
Hybridize first
Label after

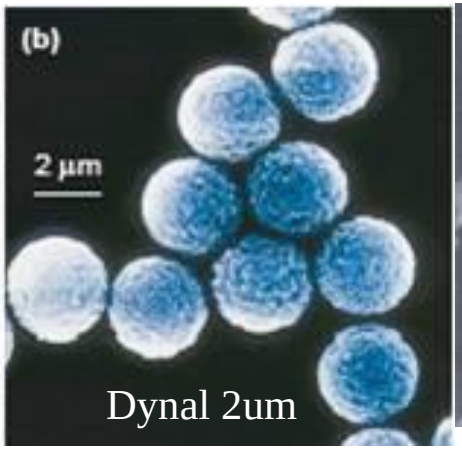


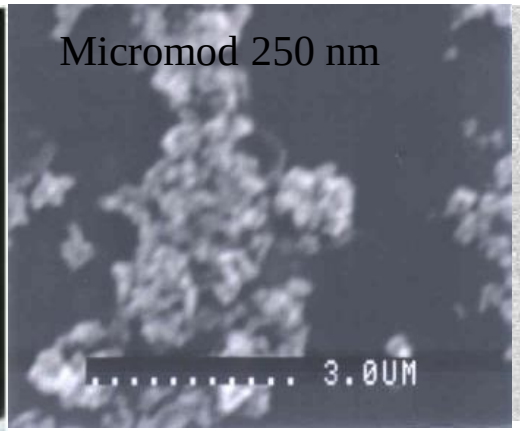
Trends in Biotechnology, August 2004

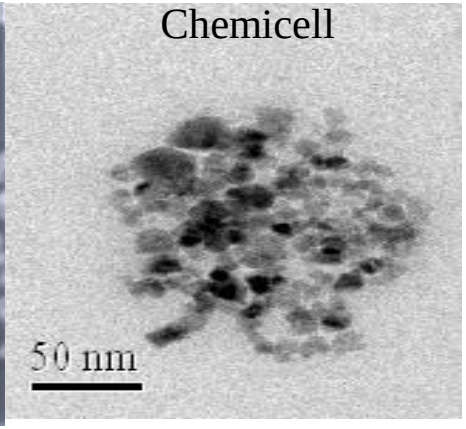
1 a) magnetic labels

Particles typically used in bio-separation







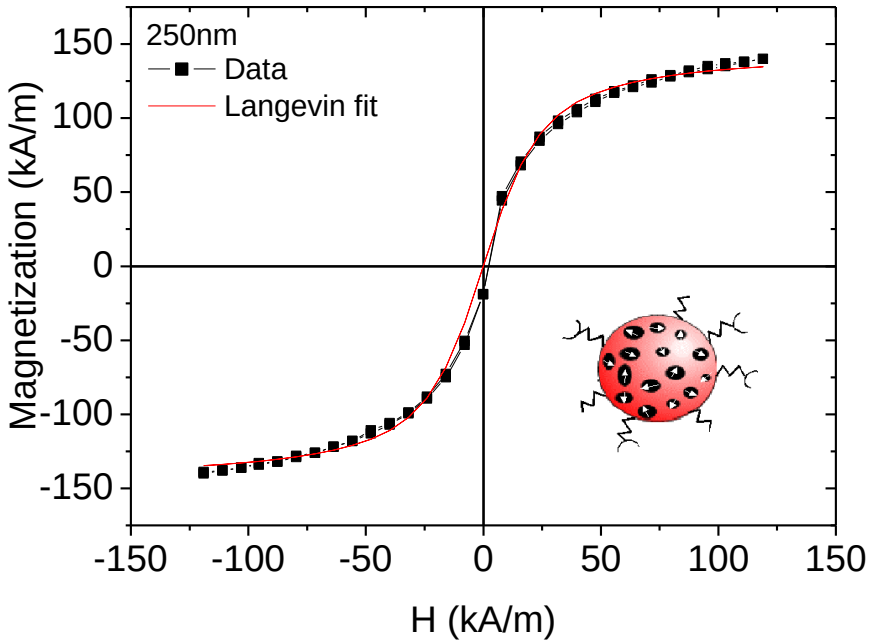


Micromod, Germany http://www.micromod.de

Dynal Biotech, Norway http://www.invitrogen.com

ADVANTAGES

- Non remanent magnetic moment (avoid clustering)
- High saturation moment (high signals)
- Small (avoid steric hindrance)
- Stable



Lowest fields to be detected

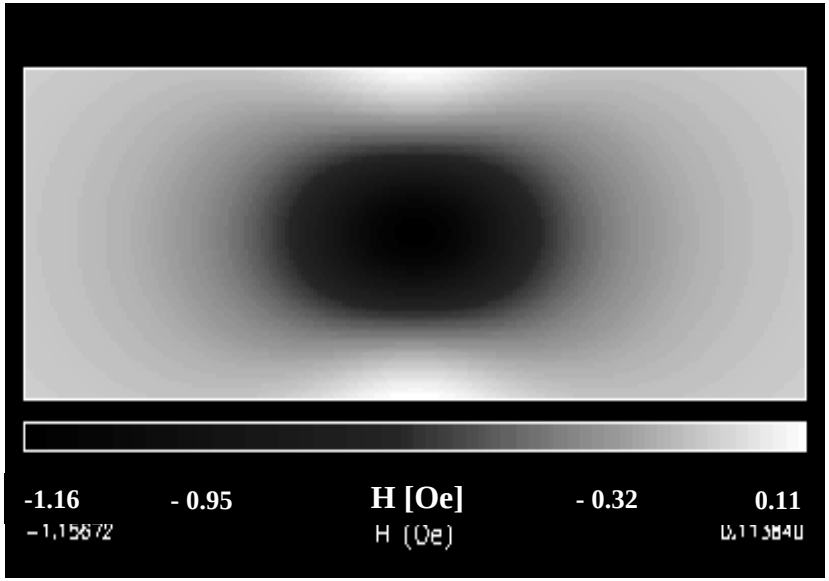
For a 50nm FeOx particle:

χ=0.7, H=1.2kA/m, d=0.2um

B^{max}=4μT

But for a 6μm^2 sensor,

B_{aver} = 1 nT

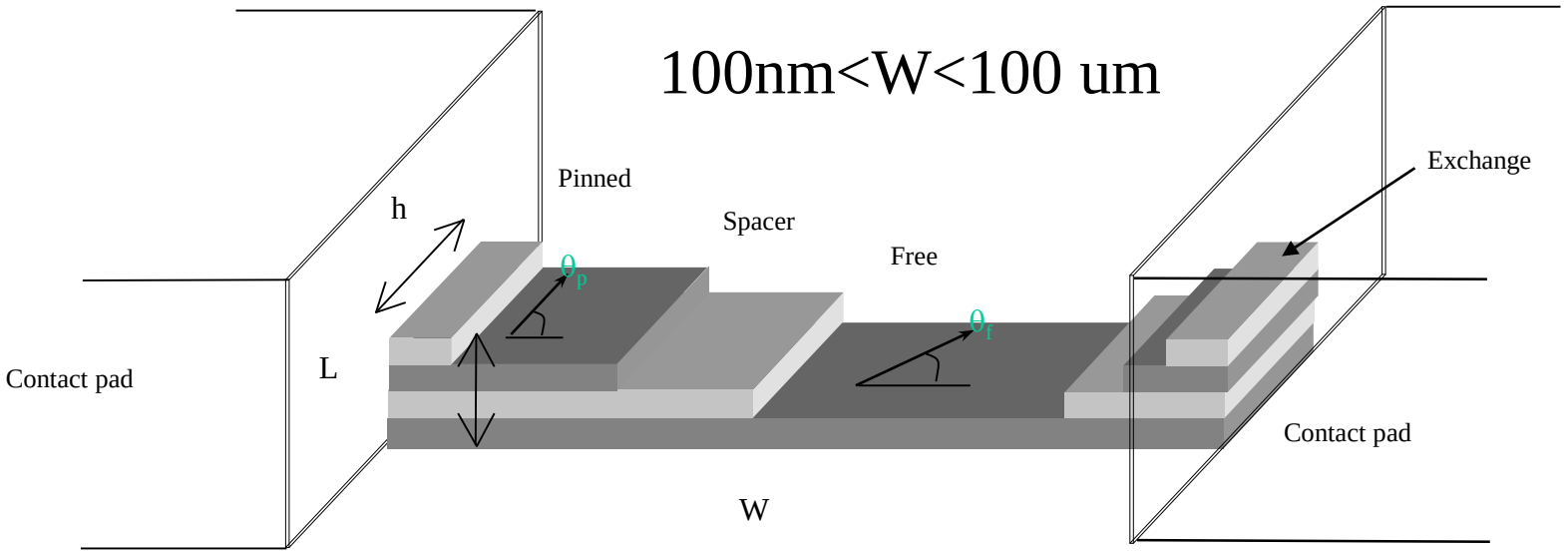


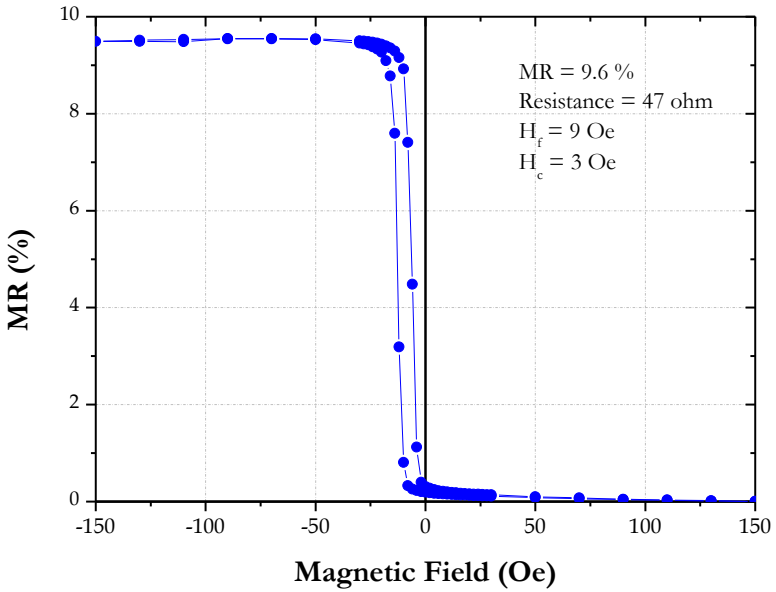
HOW TO DETECT THESE FIELDS?

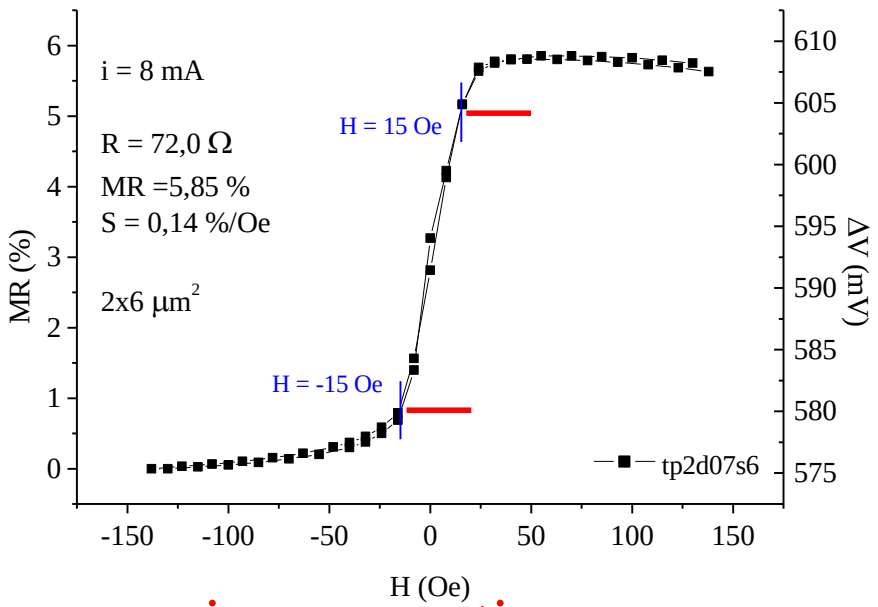
Magneto encephalography, B 10fT (SQUIDS, SVs+SC FG)

Magneto cardiography, B few pT

THE SPIN VALVE SENSOR





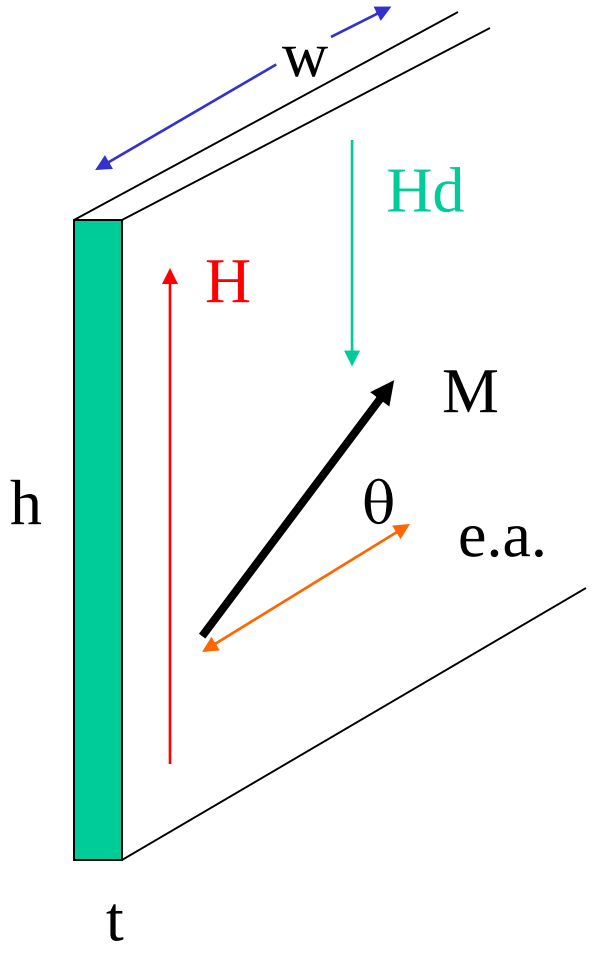


See annex for microfab + transfer curve micromagnetics

How to make a SV or MTJ sensor?

1-Control the magnetics of the thin free layer:

Magnetic Energy of a semi-infinite thin film **(w>>h,t)**



$$E/V=-\mu_o H.M + K \sin^2\theta - \tfrac{1}{2} \mu_o H_d.M$$

Where $Hd_y = -M_y N_y$

Minimizing energy:

$$\sin\theta = \mu_o H M_s / 2 K_{eff}$$

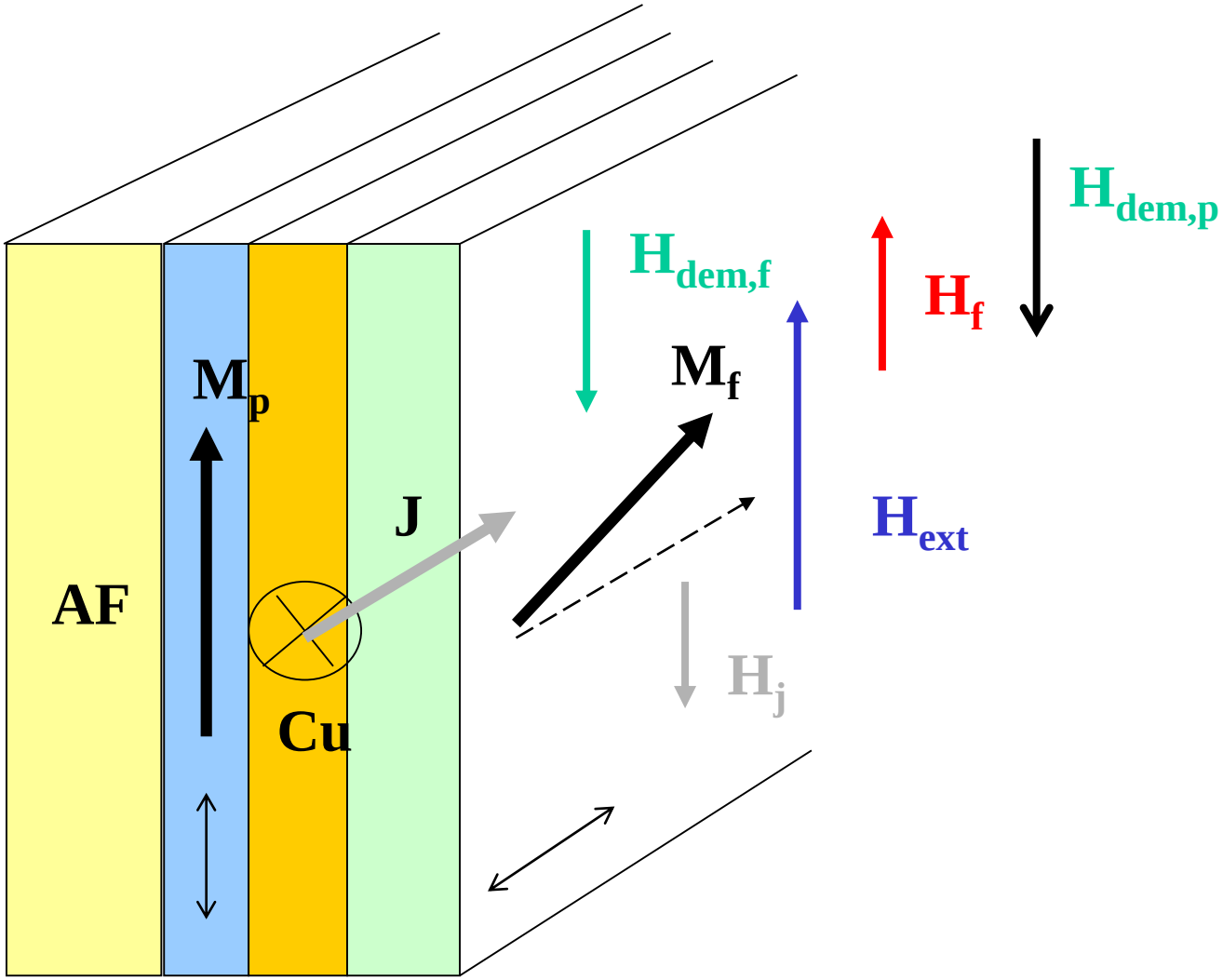
$$K_{eff} = K + \tfrac{1}{2} \mu_o M_s^2 N_y$$

$$Ny \cong (t/\pi h) \ln [(h-\Delta)/\Delta]$$

B.D.Cullity(1972) Introduction to Magnetic Materials, A.W, MA

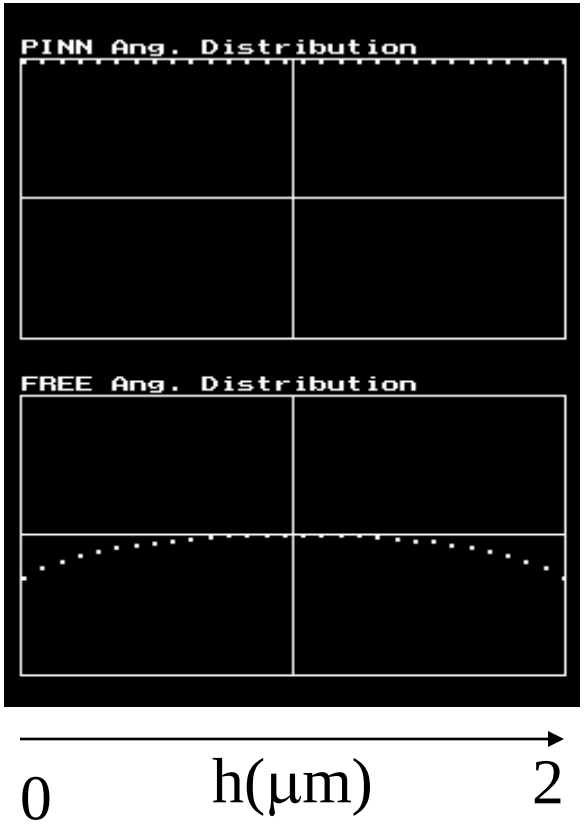
Theory Magnetic Recording, N.Bertram, p.172

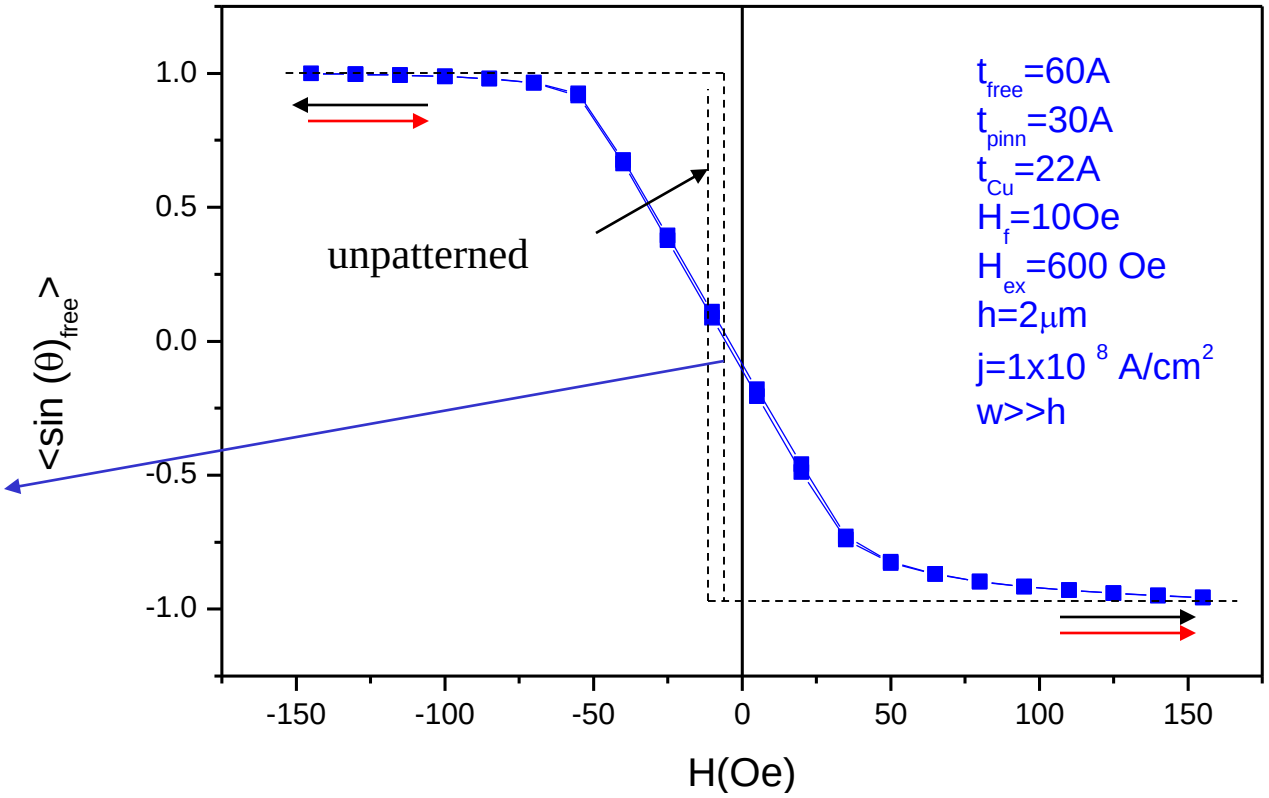
Spin Valve or MTJ Sensor: biasing



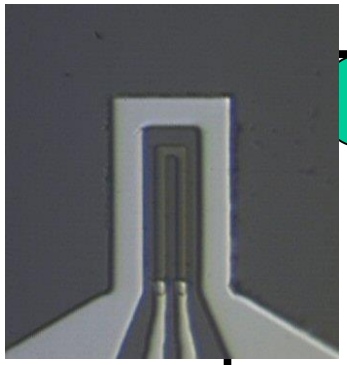
$$\sin\theta_f = \mu_o \left[H_{ext} - H_j - H_{dem,p} + H_f\right] M_s / 2 K_{eff}$$

Micromagnetic simulation for SV sensor





1-g) How to chose the best sensor? Noise spectrum



U shaped SVs, (80x2μm²) ——— data

For 1 mA sense current: - - - · fit

- 1/f knee at 50 kHz
- thermal noise: 0.2 nT/√Hz (linear regime)
- 30 Hz: 13 nT/√Hz (corresponds to 250 nV)

γ ~1; α ~1
R ~ 650 Ω
Nc ~ 1.8 E+11 noise carriers

0.1 mA sense current

1 nT/sqrt (Hz)

Calculated thermal background

$S_V(V^2/Hz)$

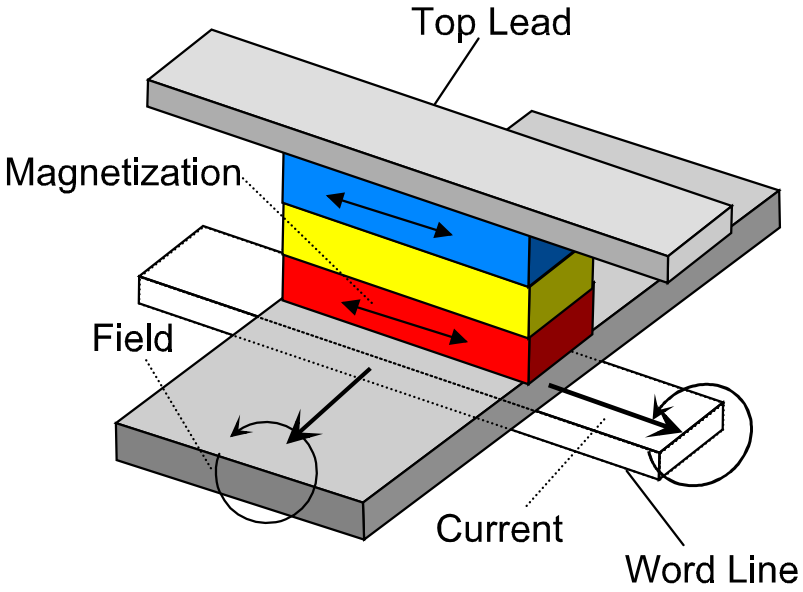
10^{-16}

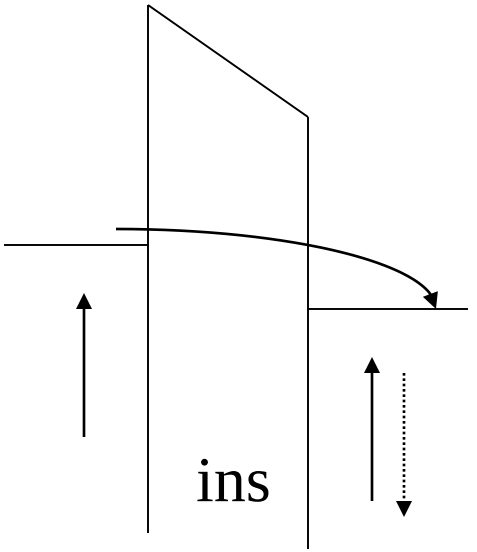
10^{-17}

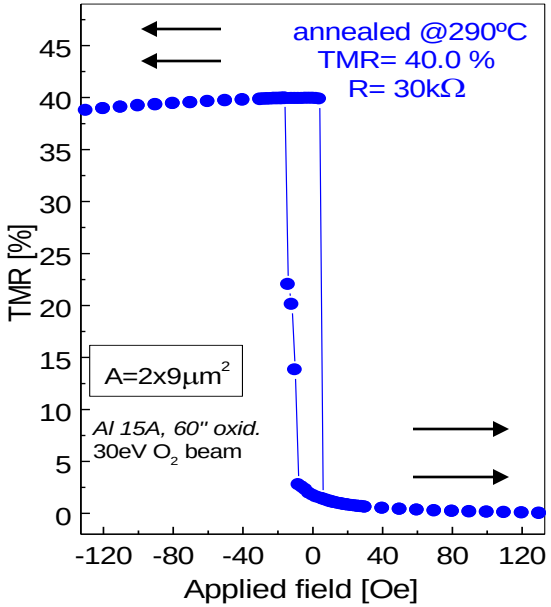
0.1 1 10

Frequency (kHz)

The Magnetic Tunnel Junction







$$TMR=2P_1P_2/1+P_1P_2$$

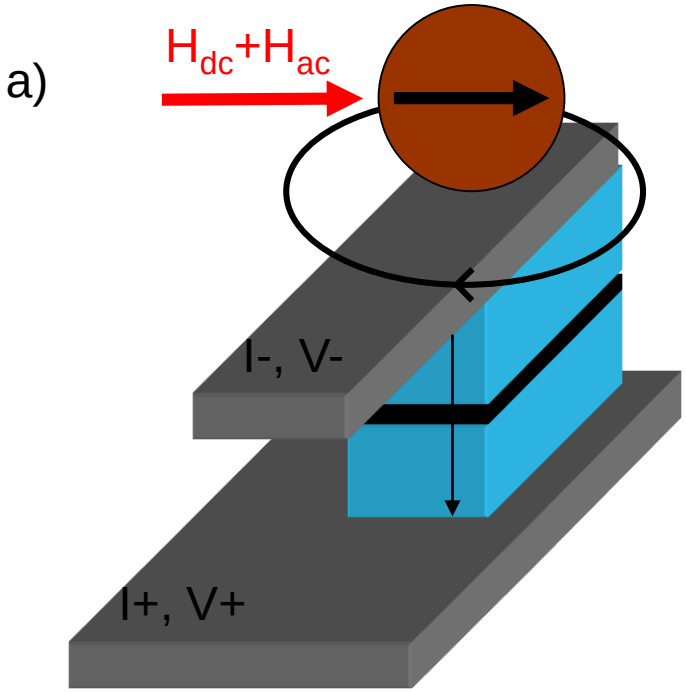
$$P=[D_{\uparrow}(\varepsilon_F)-D_{\downarrow}(\varepsilon_F)]/\ [D_{\uparrow}(\varepsilon_F)+D_{\downarrow}(\varepsilon_F)]$$

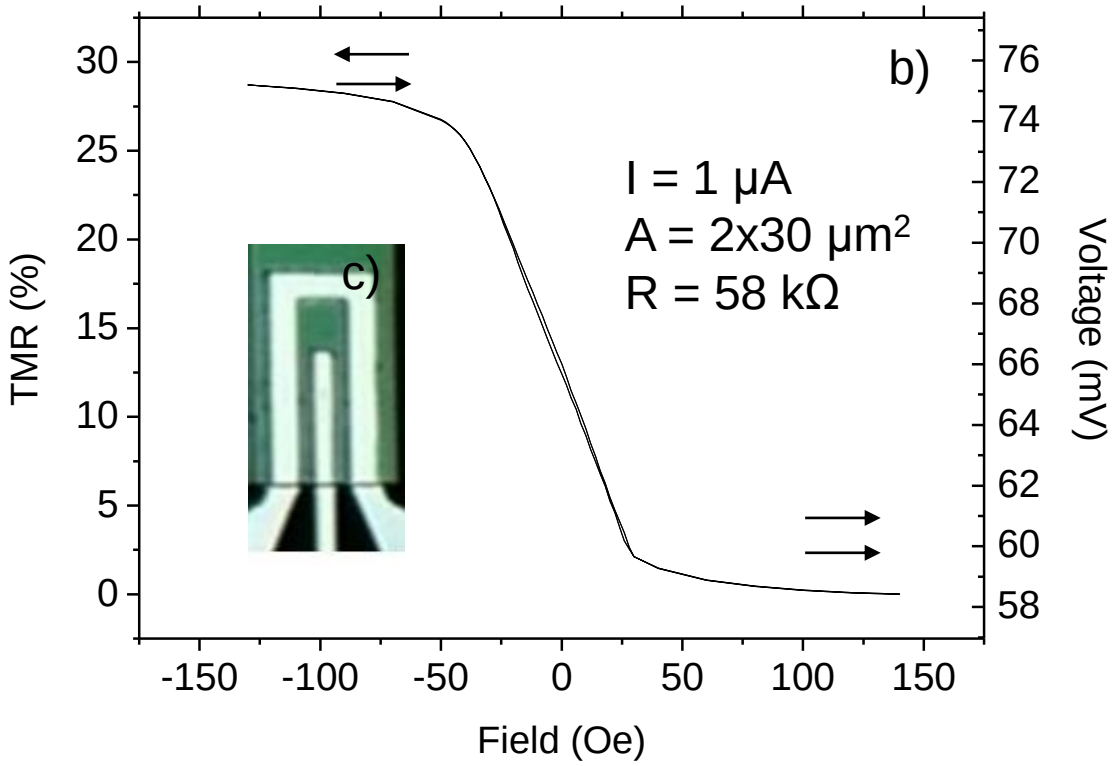
P	%
CoFe	55
half metal	100

Increasing sensitivity I

AlO_x based Magnetic Tunnel Junction sensor transfer curve

Ta30A/NiFe30A/MnIr60A/CoFe25A/Ru8A/CoFeB35A/AlOx10/CoFeB30/Ta30



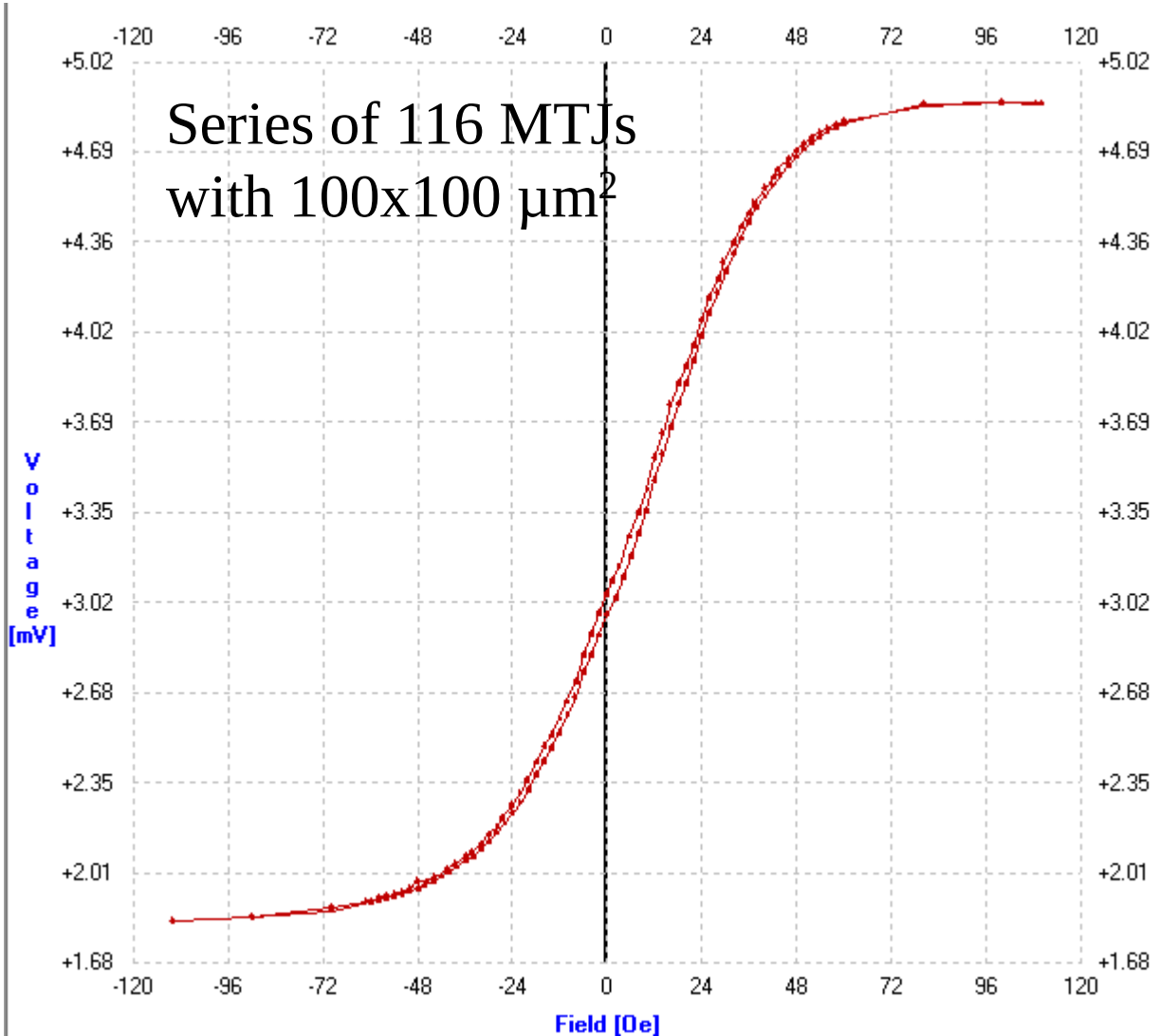


See annex for microfab + transfer curve micromagnetics

IMAGIC stacks : Generation 3

Transport results

TJ933 - [5 Ta / 25 CuN]x6 / 5 **Ta** / 5 **Ru** / 20 IrMn / 2 $CoFe_{30}$ / 0.85 **Ru** / 2.6 $CoFe_{40}B_{20}$ / **MgO** [22 kOhm um2] / 2 $CoFe_{40}B_{20}$ / 0.21 **Ta** / 4 NiFe / 0.20 Ru / 6 IrMn / 2 **Ru** / 5 **Ta** / 10 **Ru**



TMR = 166.38 %

H_f=8.94 Oe

$H_{c\ measured}$= -0.89 Oe (real

RxA min : 29387.04 Ω µm

RxA max : 78282.26 Ω µm

Ibias : +5.365910 mA

V_{out}(H=0)=3.0V

Sensitivity at H=0 :

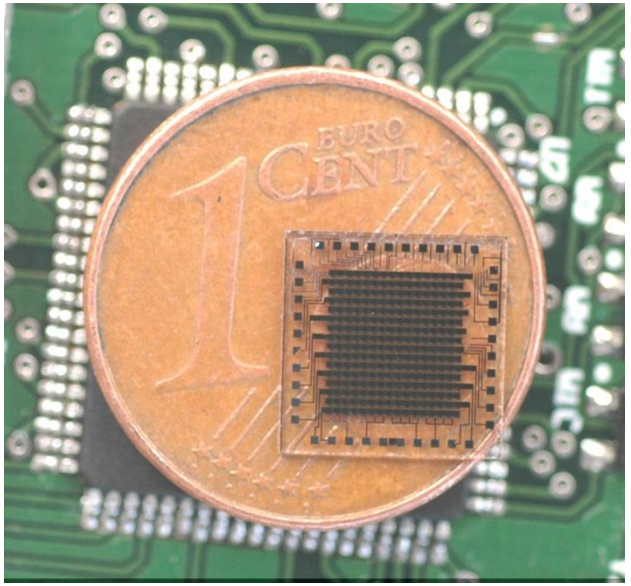
19.83

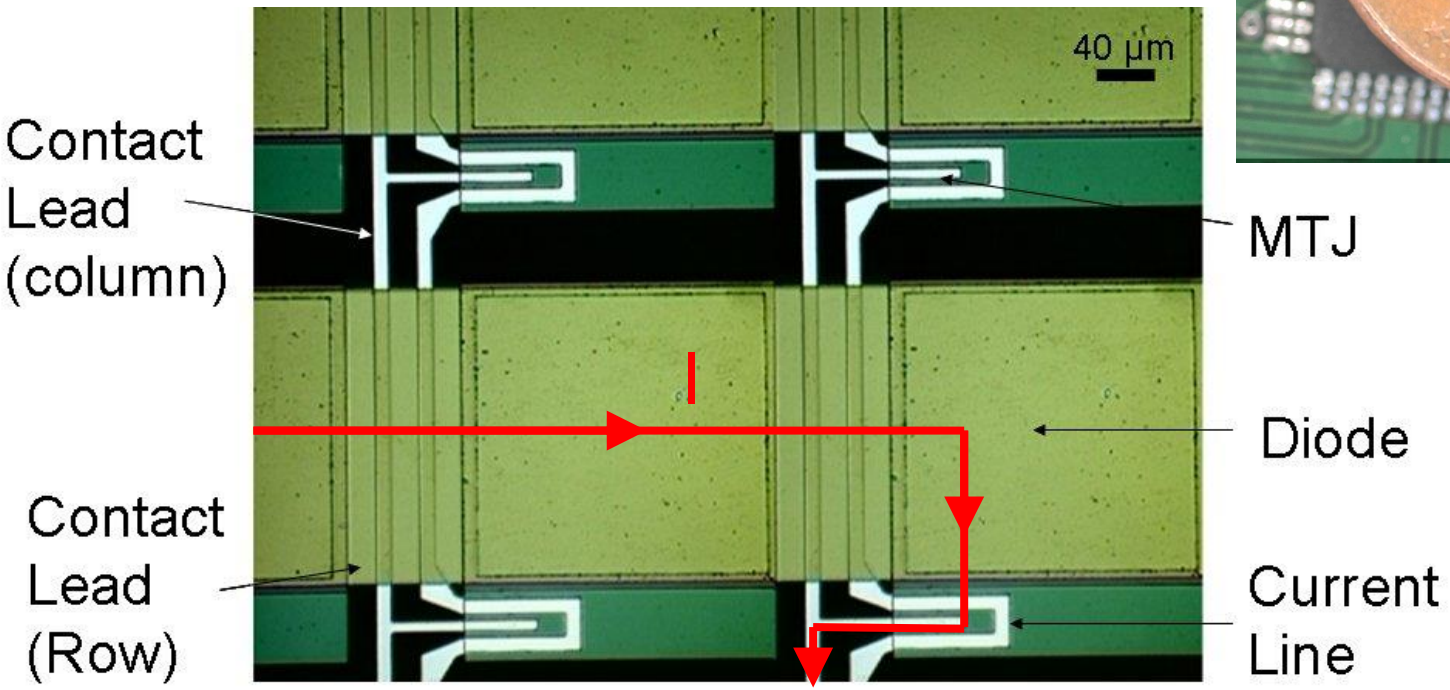
mV/V/Oe

198.3 V/V/T

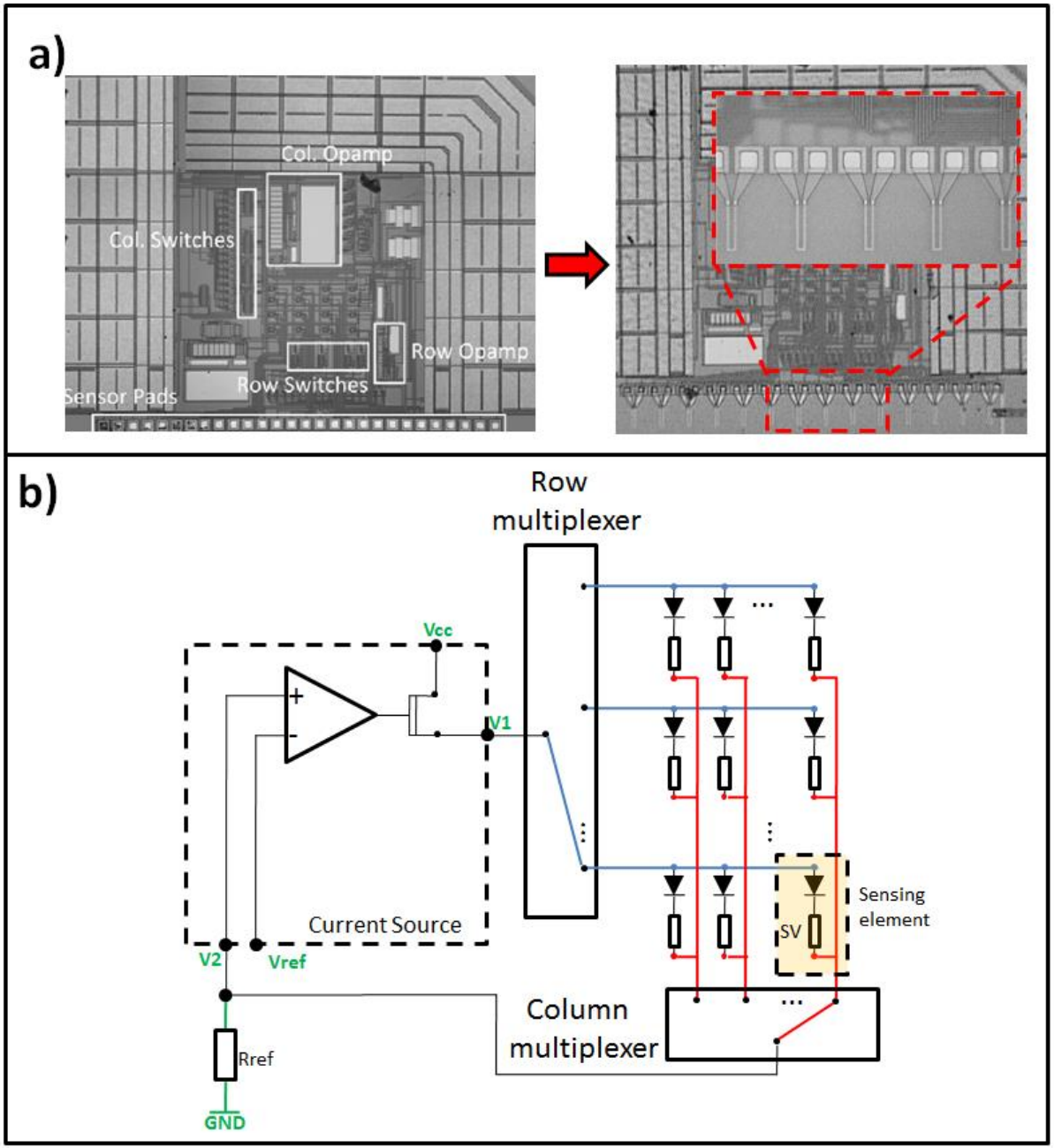
Matrix architecture - Scalability

- A 16x16 element matrix has been fabricated



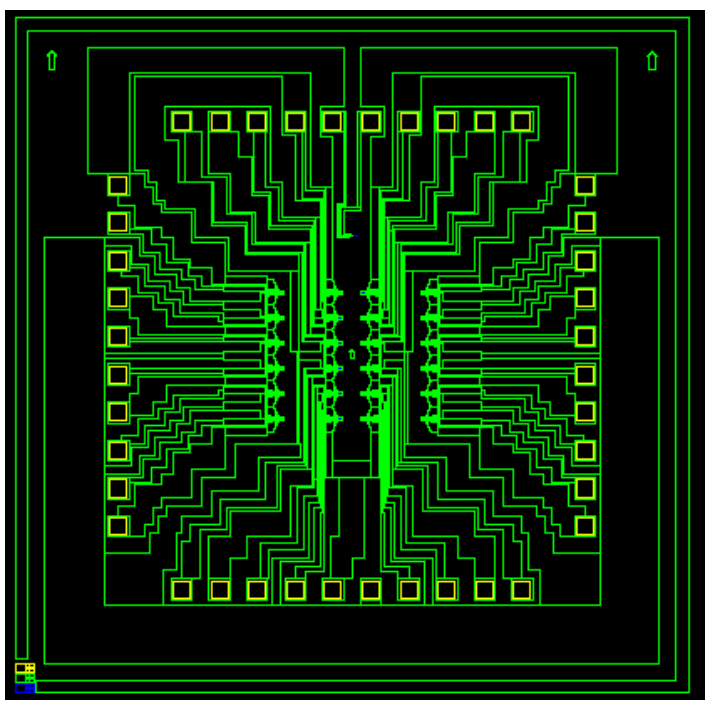


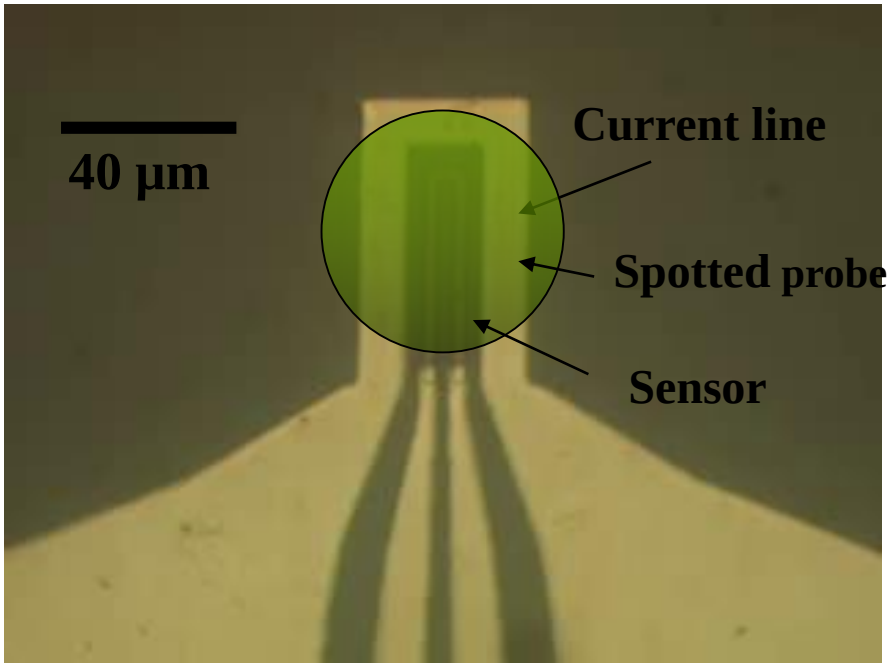
Integration of Magnetoresistive Biochips on a CMOS circuit.



Dec 2011

INESC-MN's 3rd generation MR biochip





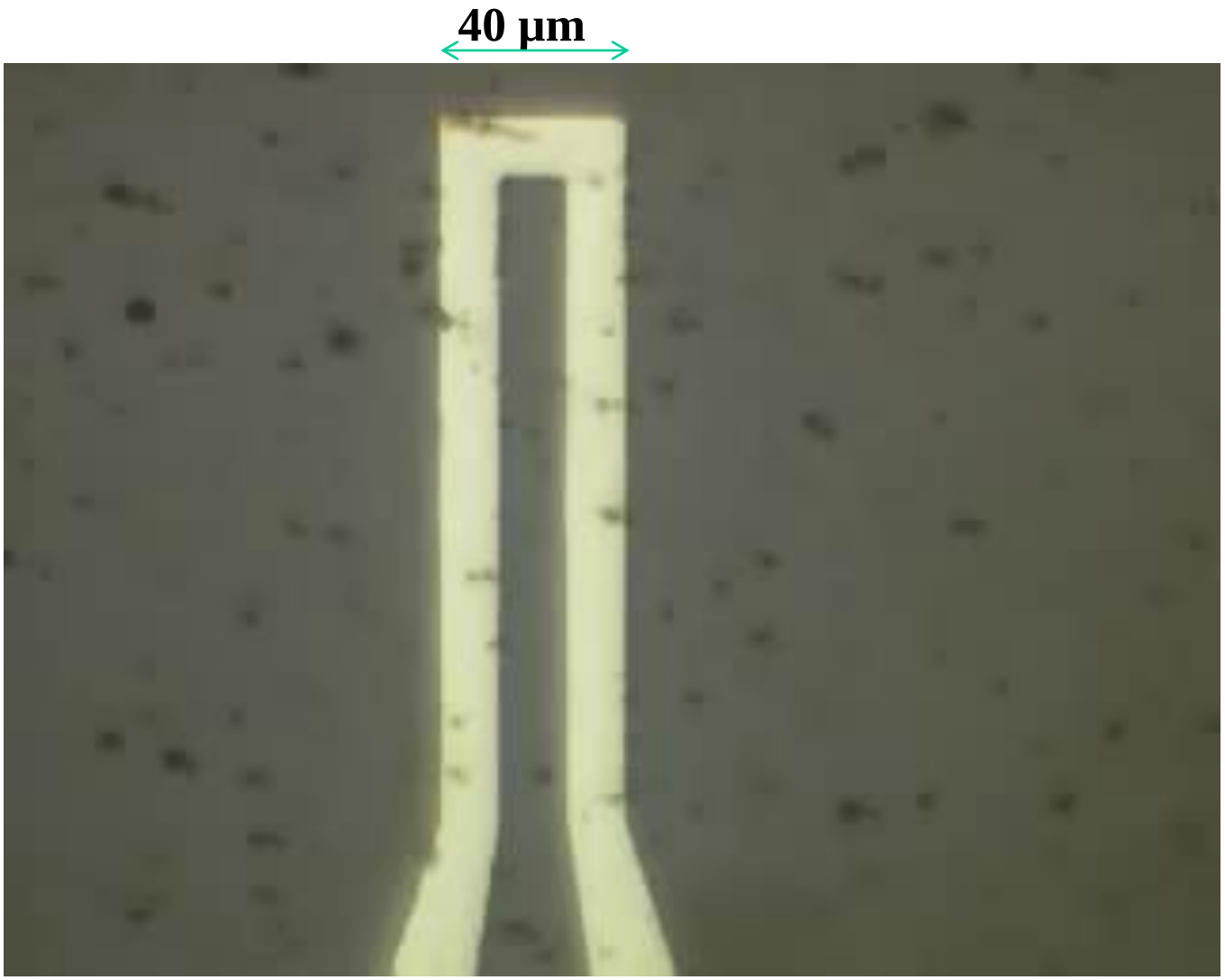
- 24 sensing units:
  - 1 U-shaped spin-valve sensor (2.5 µm × 80 µm)
  - 1 U-shaped current line (l = 50 µm; w = 10 µm; s = 17 µm)
- 1 single sensor (2.5 µm × 80 µm)

→
- **Higher dynamic range**
- **Higher biological sensitivity**
- **Need to focus labels in large areas (1000-2000 µm²)**

←

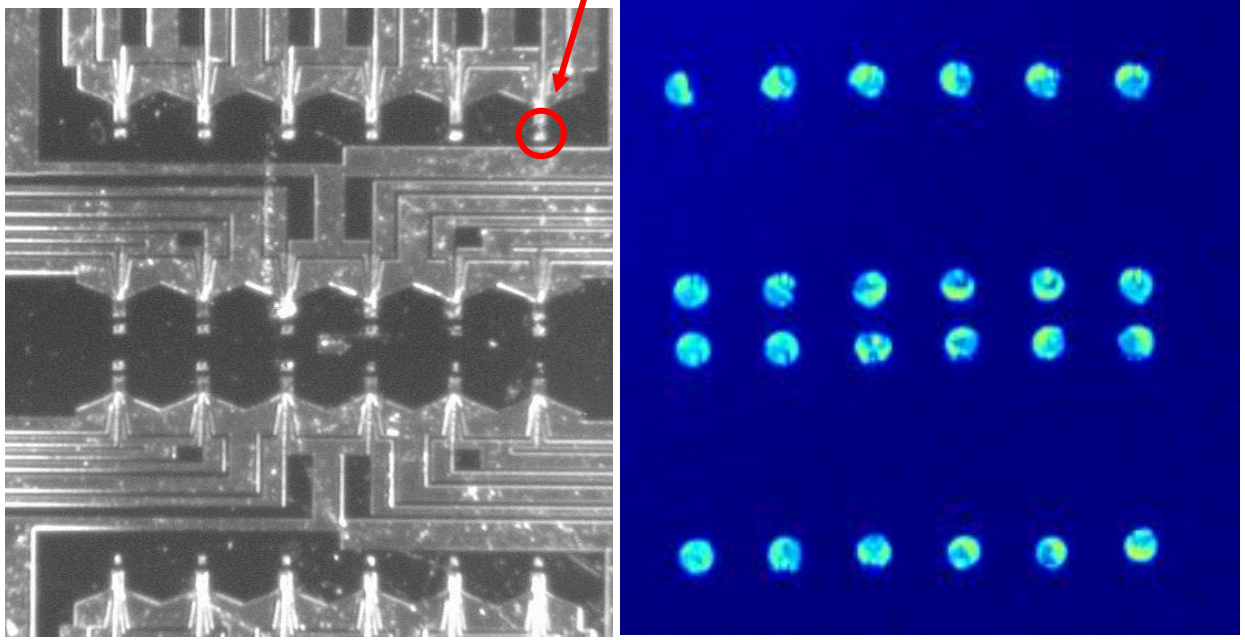
Appl.Phys.Lett., vol.87, pp.013901, 2005

1-e) target arraying over immobilized probes: 250nm beads and magnetically assysted hybridization

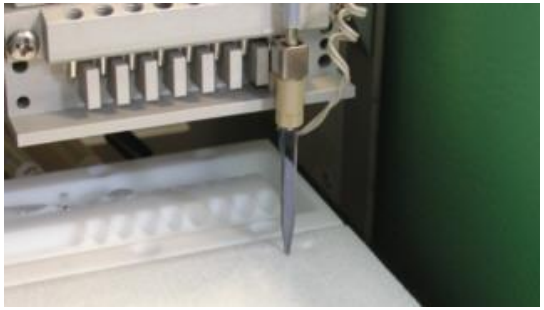


1-d) Spotting biological targets on the biosensing platform

Spotting site



1 μM Oligo solution, Cy5 labeled
200 pL droplets



Gesim spotter

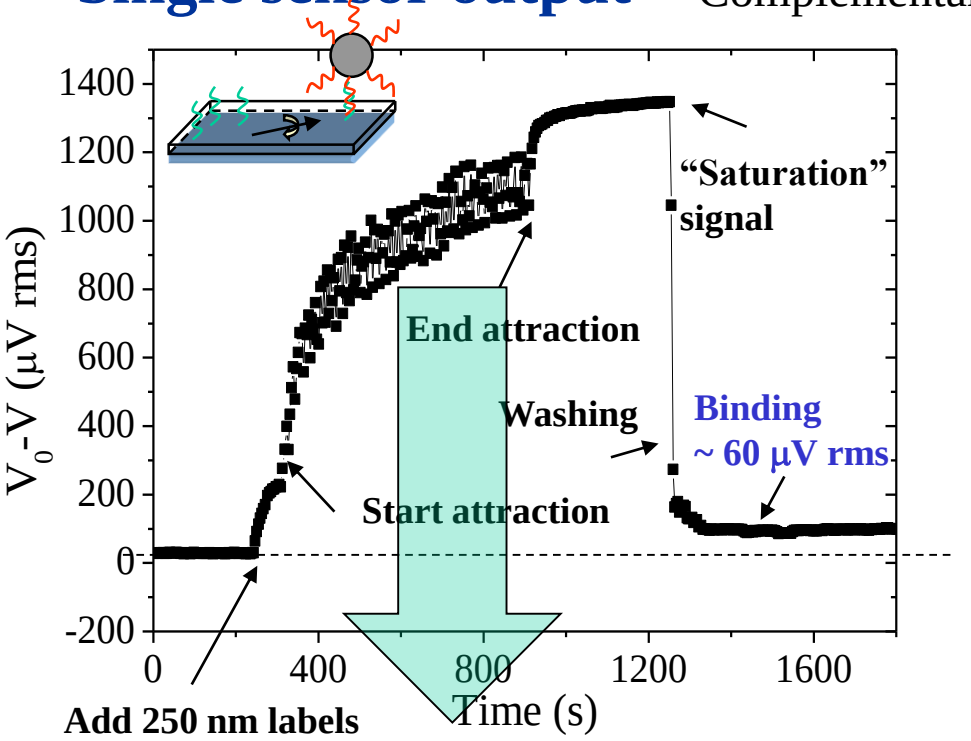
Disposable biochip

Biomolecular recognition experiments

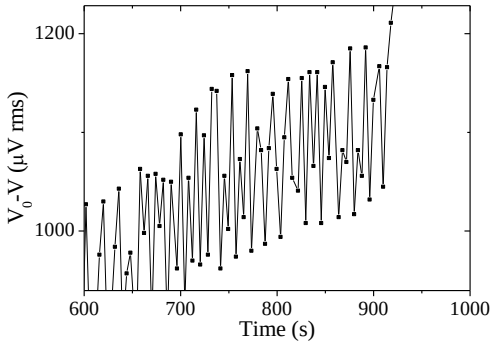
Cystic Fibrosis Related DNA hybridization

Single sensor output

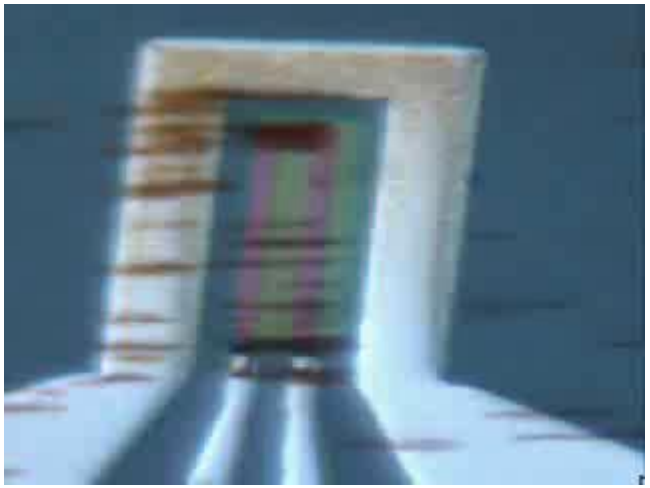
Complementary target

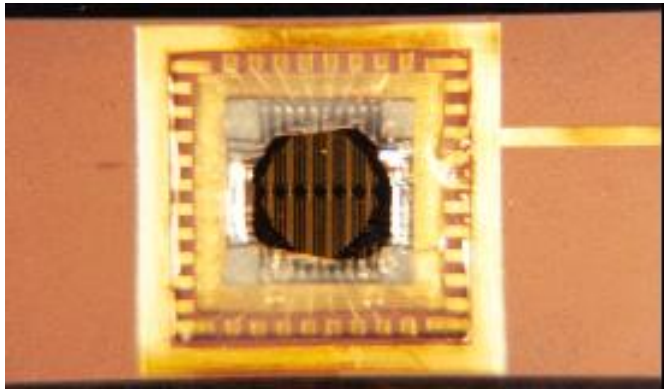


Add 250 nm labels

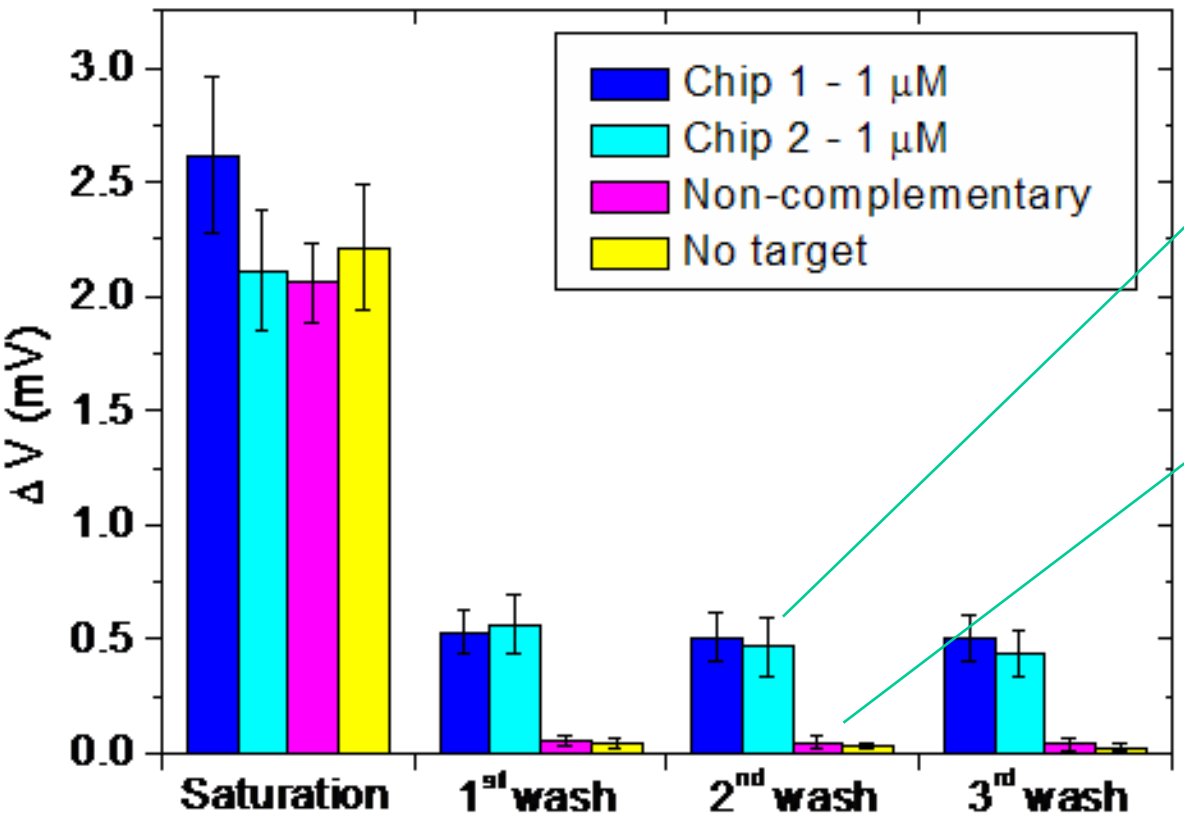


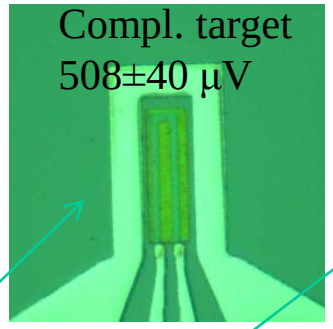
Dynamic hybridization at work

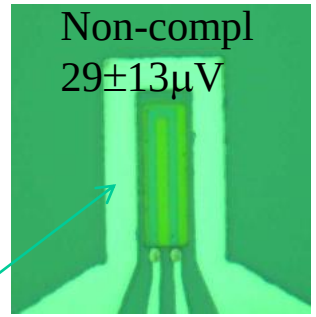




Detection of 20mer DNA Hybridization probe on Au







•Values are an average of the signal of 16 sensors.

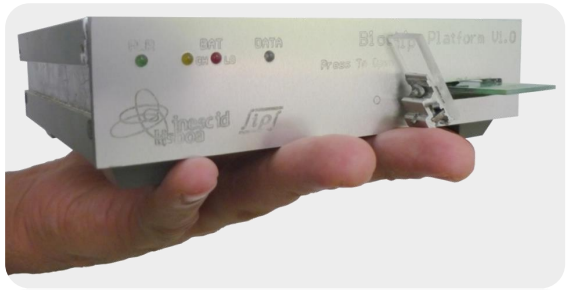
• Reproducibility using different chips for the same target solution.

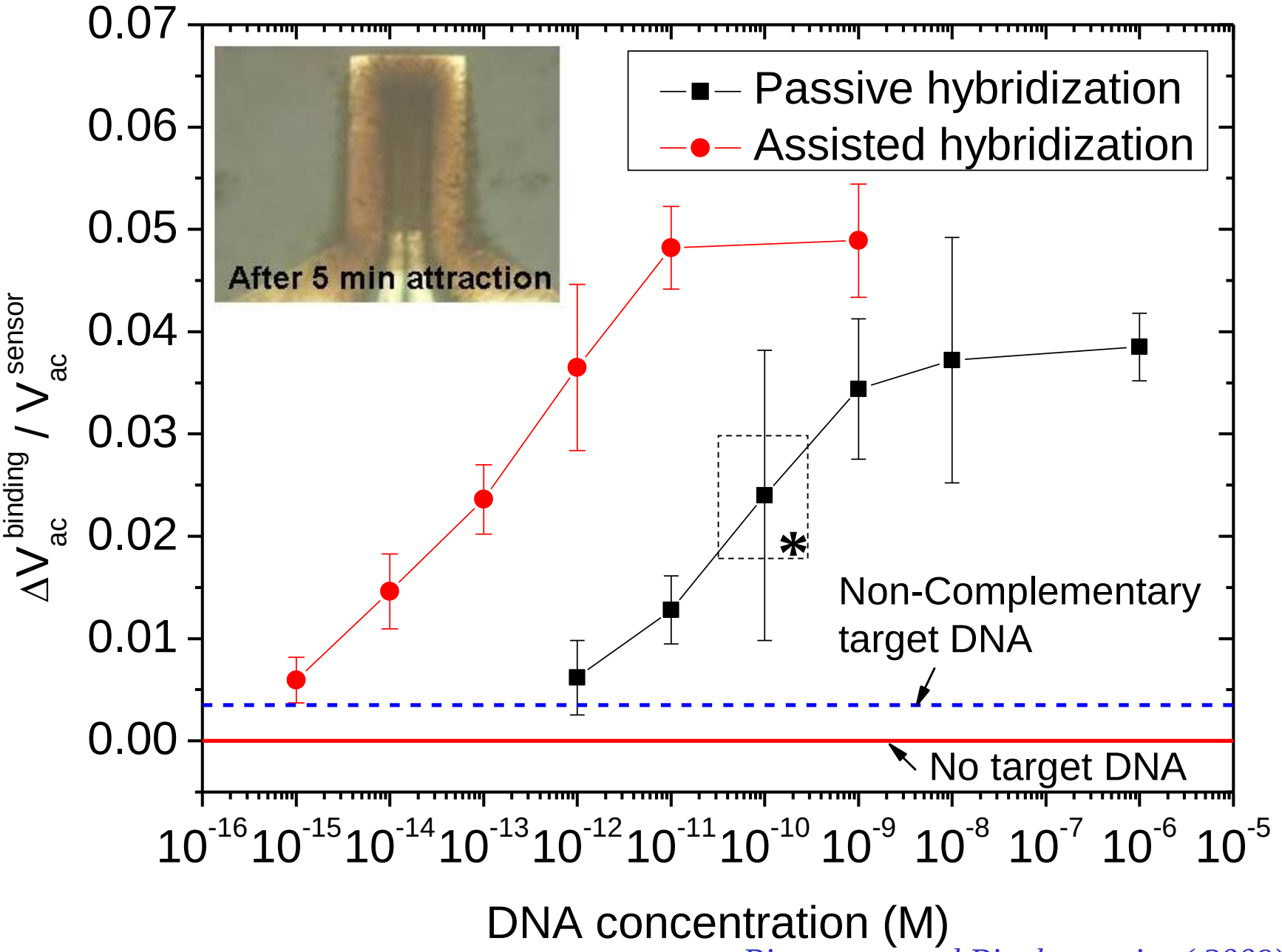
• Higher than 15:1 ratio between complementary and non-complementary targets.

• Reduced non-specific recognition.

AIP Conf. Proceed, vol. 1025, pp.150-175, 2008

POC Diagnostic Platform
INESC MN
INESC ID



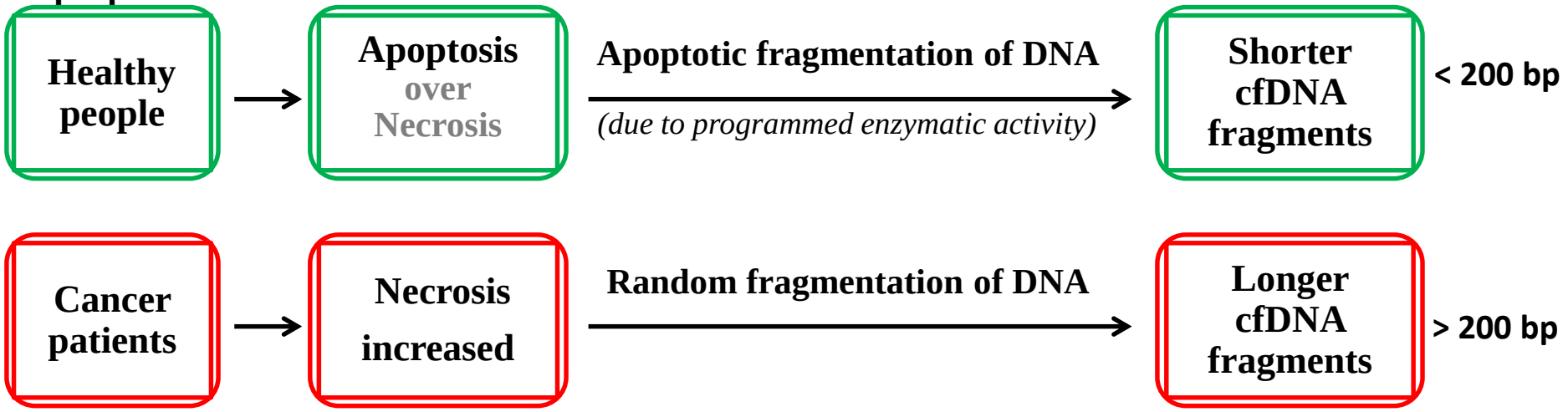


Biosensors and Bioelectronics (2009)

Application: Cell-free DNA as cancer biomarker?

- **Cell-free DNA:** DNA that can be found outside of cells in blood circulation. Results mainly from dying cells (apoptosis or necrosis)

- **The cfDNA found in cancer patients is qualitatively different from what is found in healthy people:**



Integrity index:ratio $\frac{\textbf{ALU247}\ \textit{(longer cfDNA fragment)}}{\textbf{ALU115}\ \textit{(shorter cfDNA fragment)}}$

Universal cancer biomarker in therapy follow-up ?

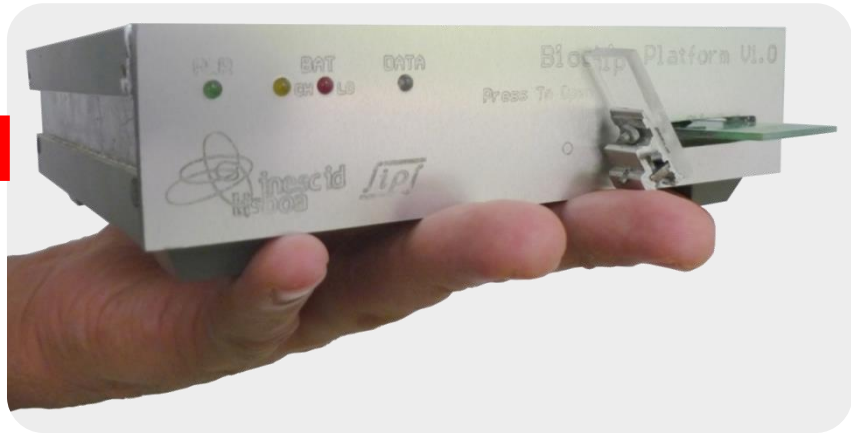
Blood finger-prick

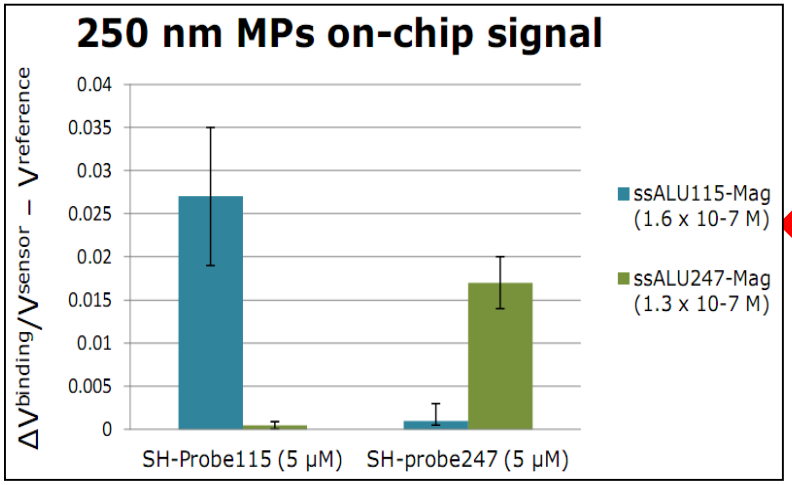
Sample preparation step

separation of plasma from blood cells

Plasma injected in the detection chip

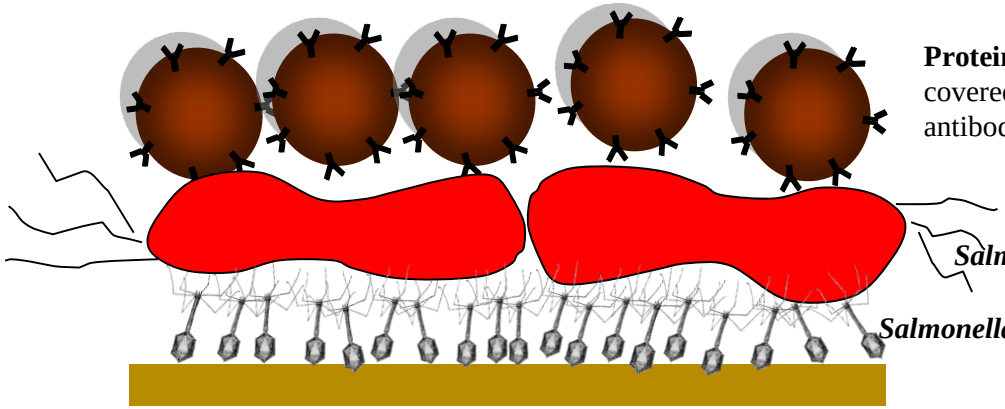
Measurement of the chip





Also immuni assay biochips

Heterogeneous "sandwich-assay" concept

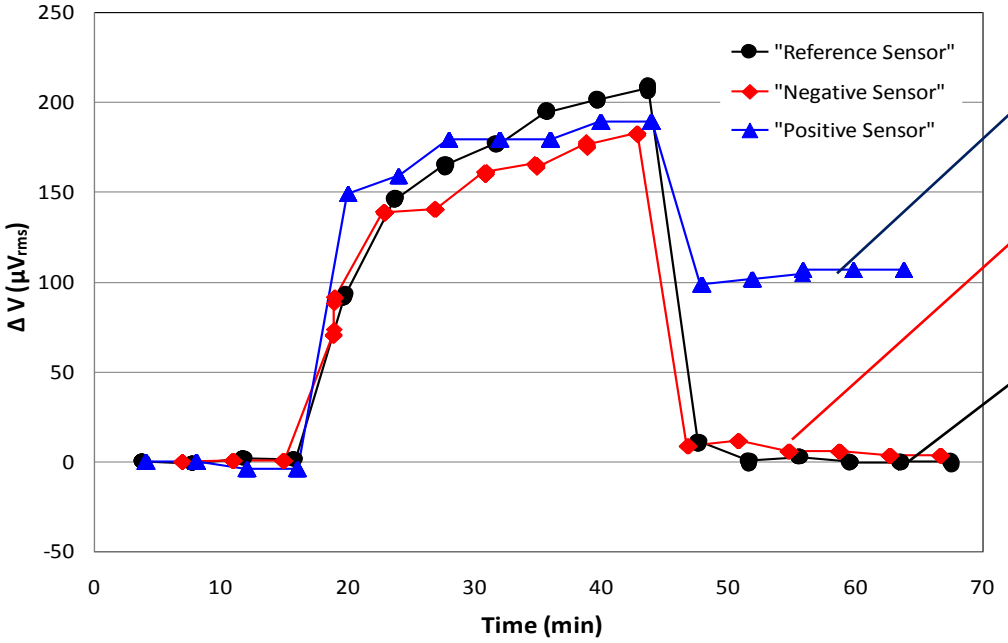


Protein A MNPs 250 nm covered with anti-*Salmonella* antibody

***Salmonella* Enteritidis**

***Salmonella* Bacterio(phage) PVP-SE1**

The signal obtained…



Specific bacteriophage for Salmonella enteritidis

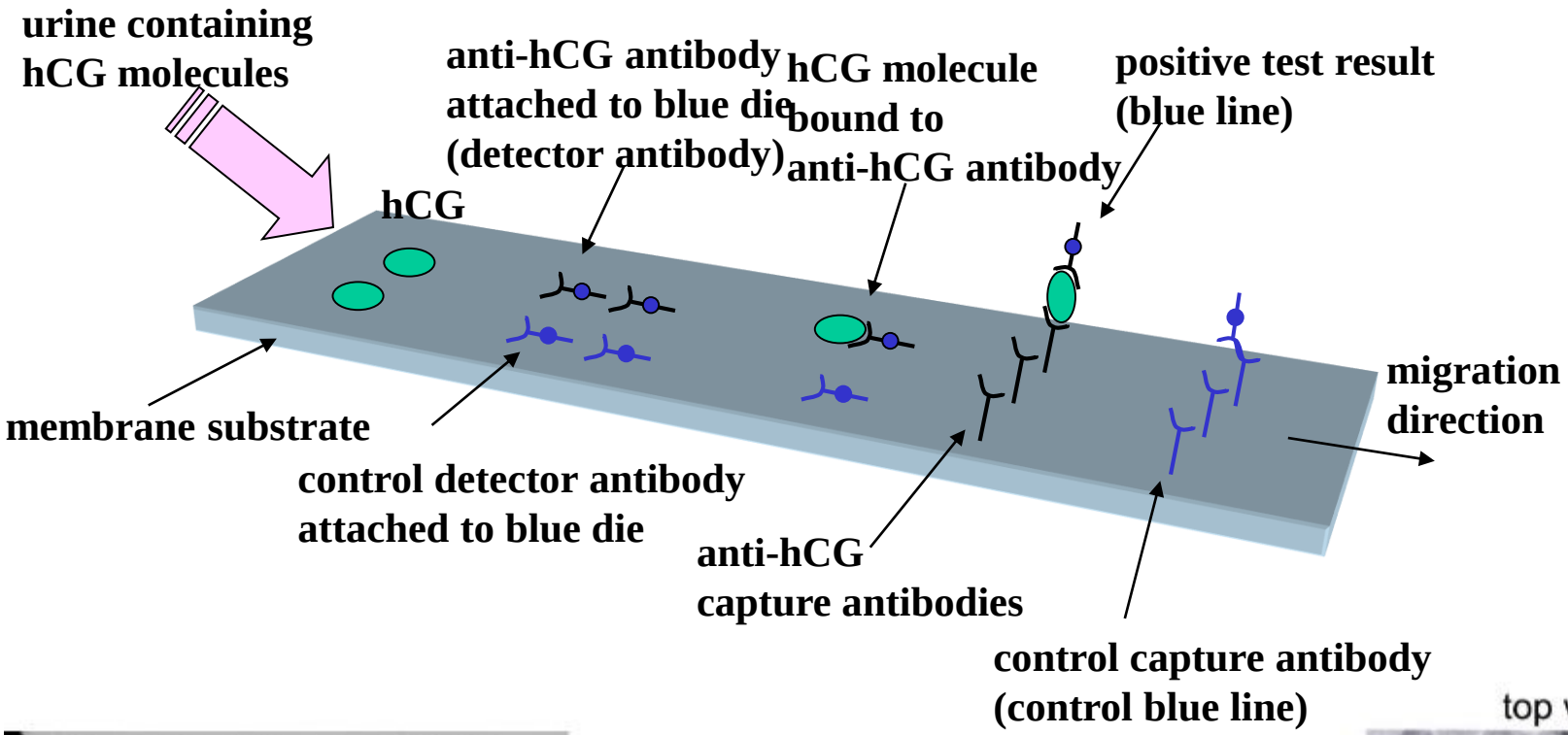
Unspecific bacteriophage

No bacteriophage

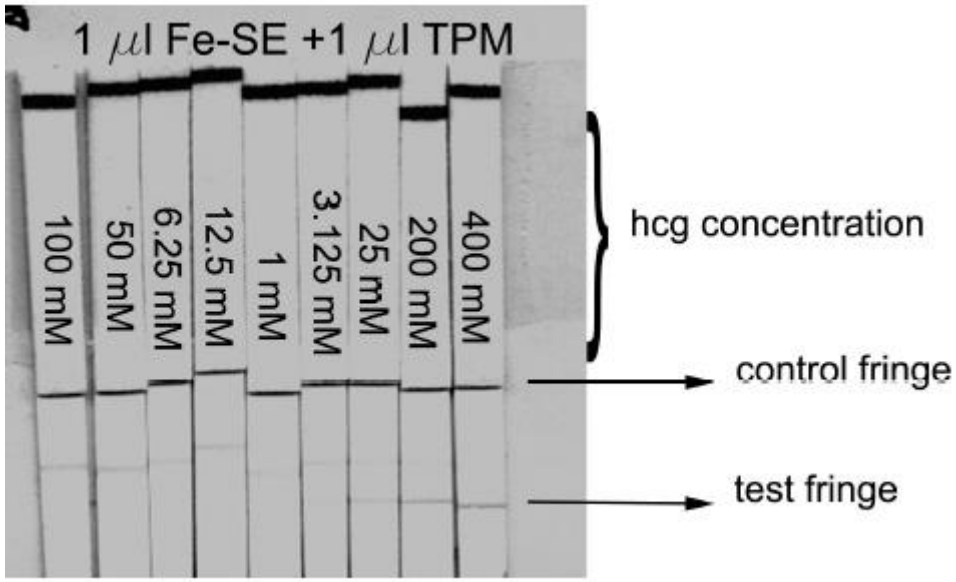
Affinity of the specific phage was about 20 times higher (average absolute value of 100 μV against 5 μV).

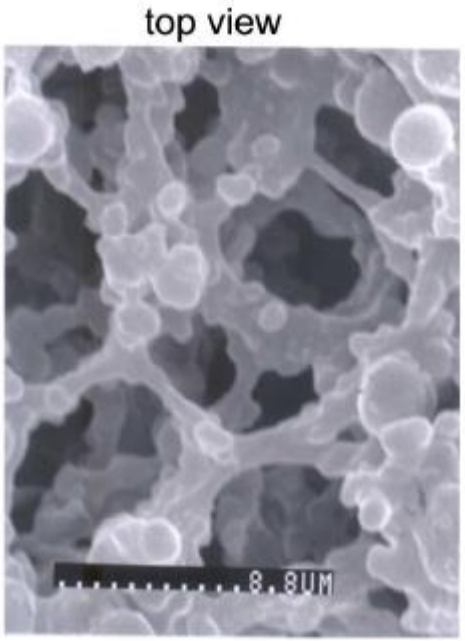
Elisabete Fernandes, PhD Student
Contact E-mail: elisabete.fernandes@deb.uminho.pt

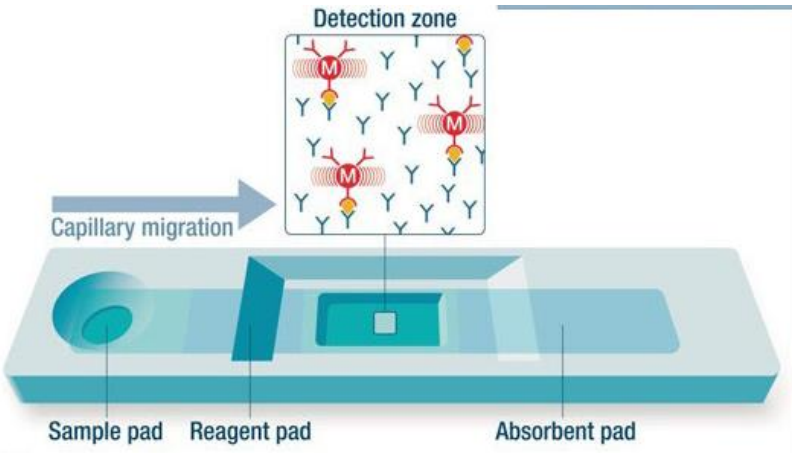
1-j) Immunocromatographic tests: also a target for MR biochips



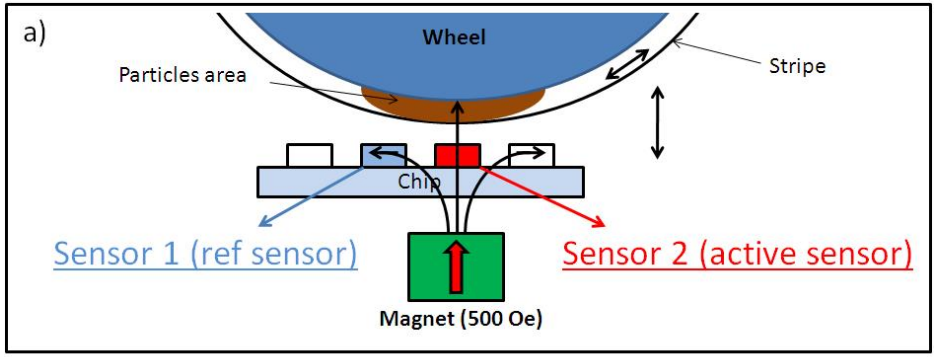
With
R.Ibarra+
de Teresa
D.Serrate
Zaragossa

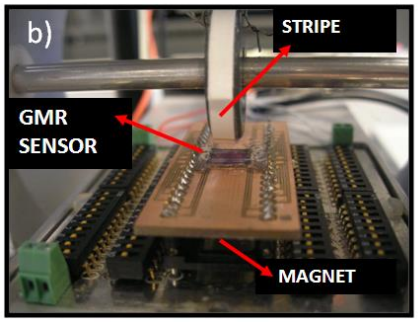


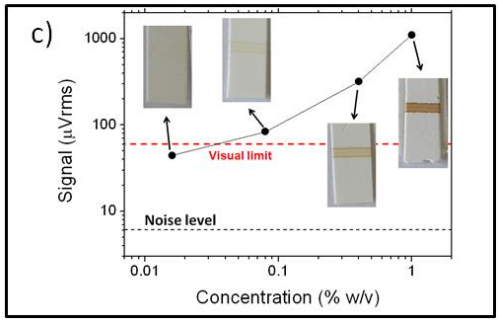




I)





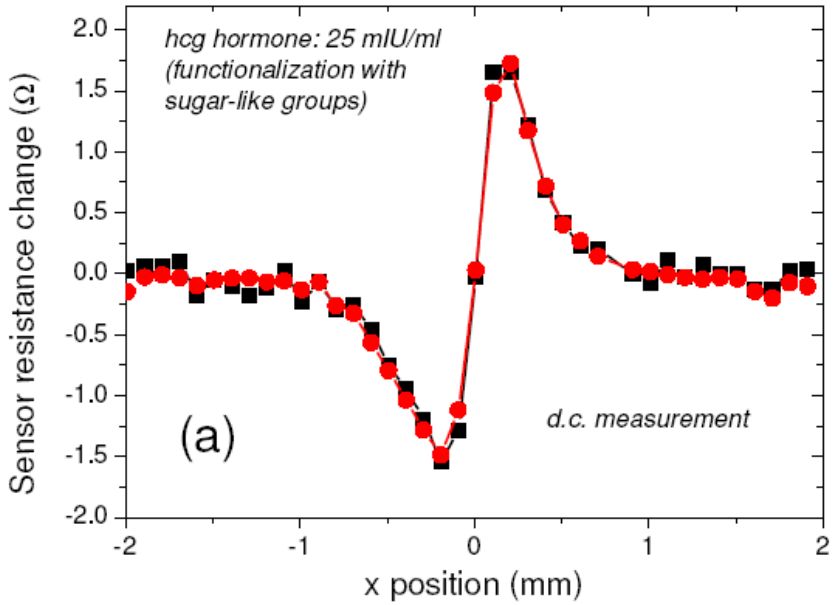


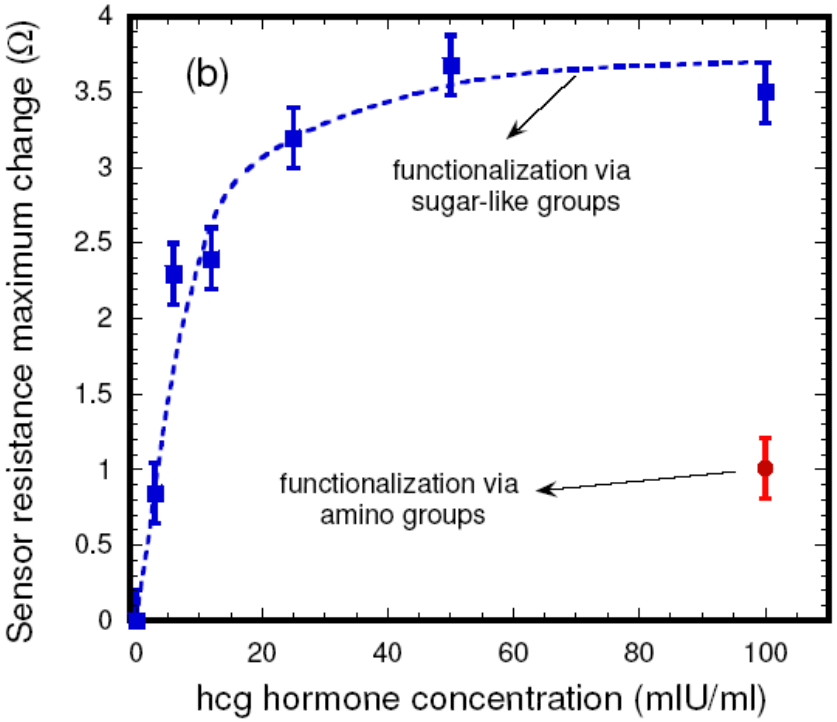
Lateral Flow Membrane

Labels the analyte of interest with magnetic nanoparticules and captured by antibodies in the detection zone.

II)

No moving parts.....

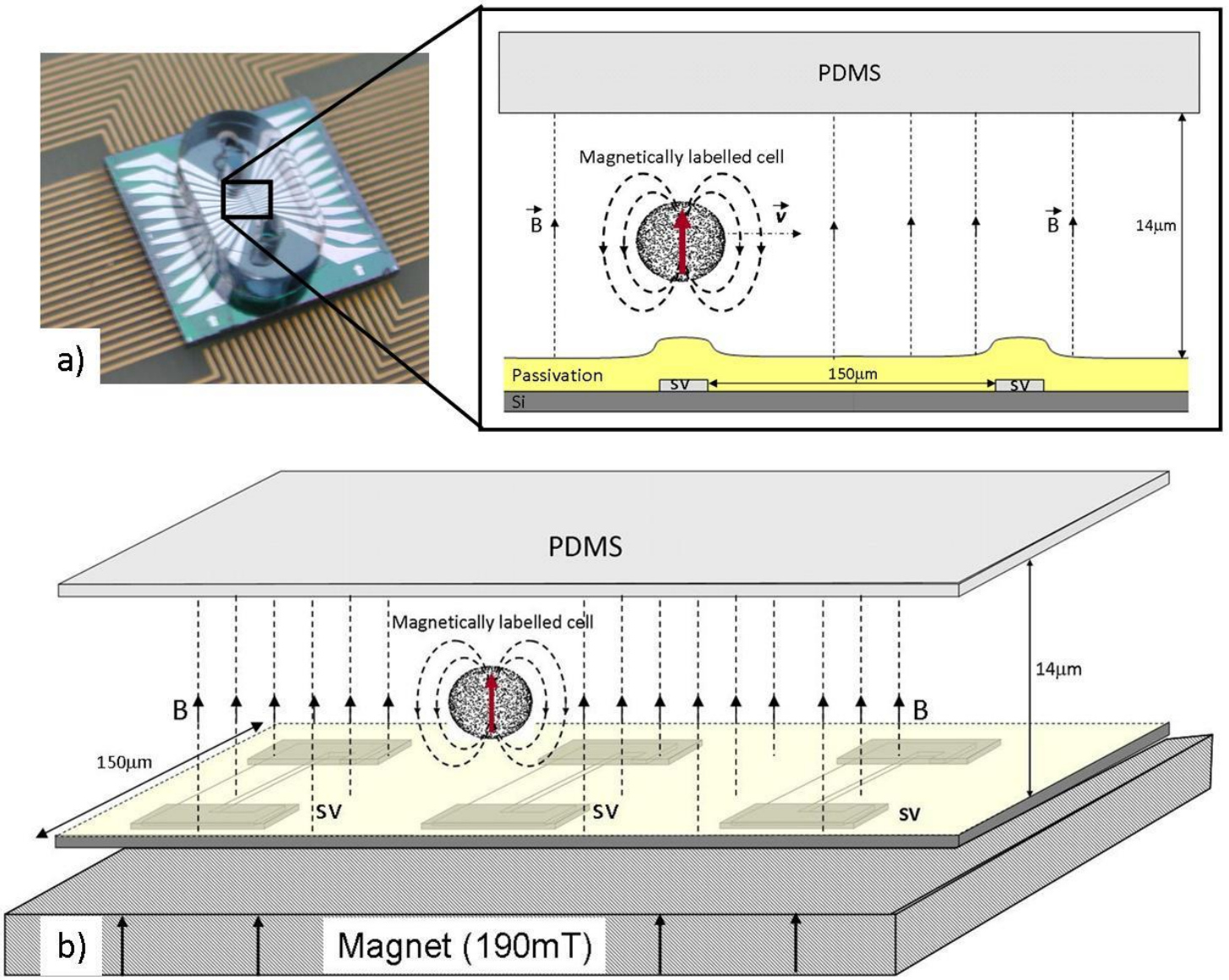


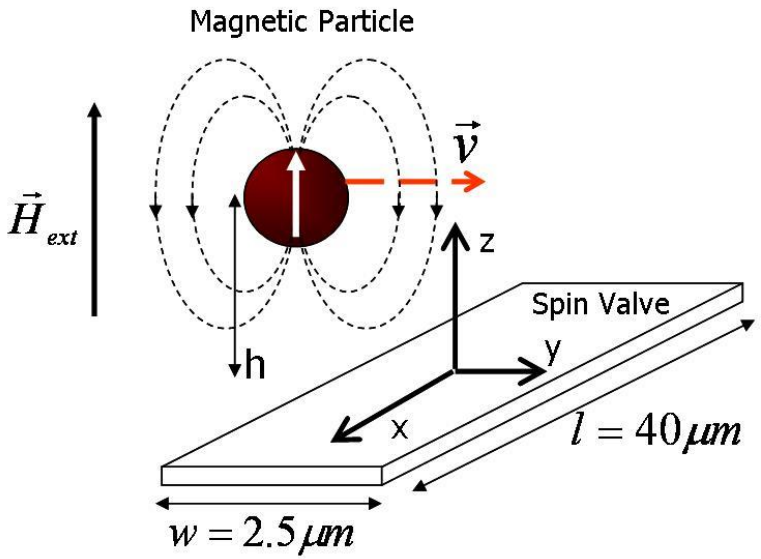


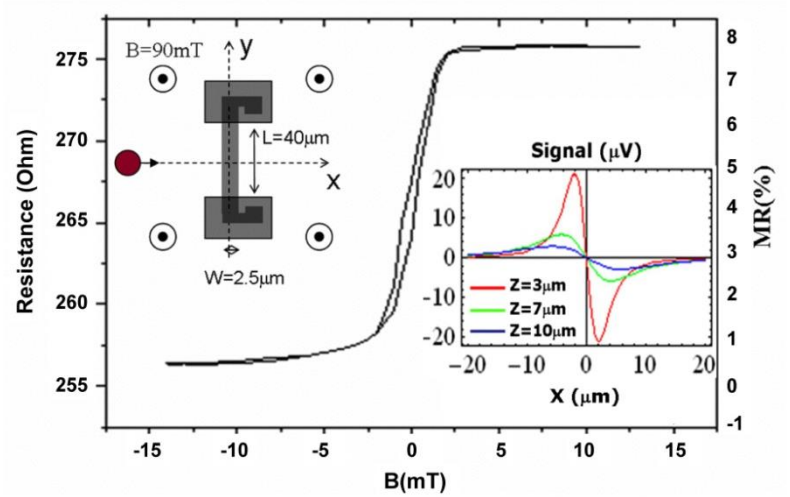
Biosensors and Bioelectronics, 2012

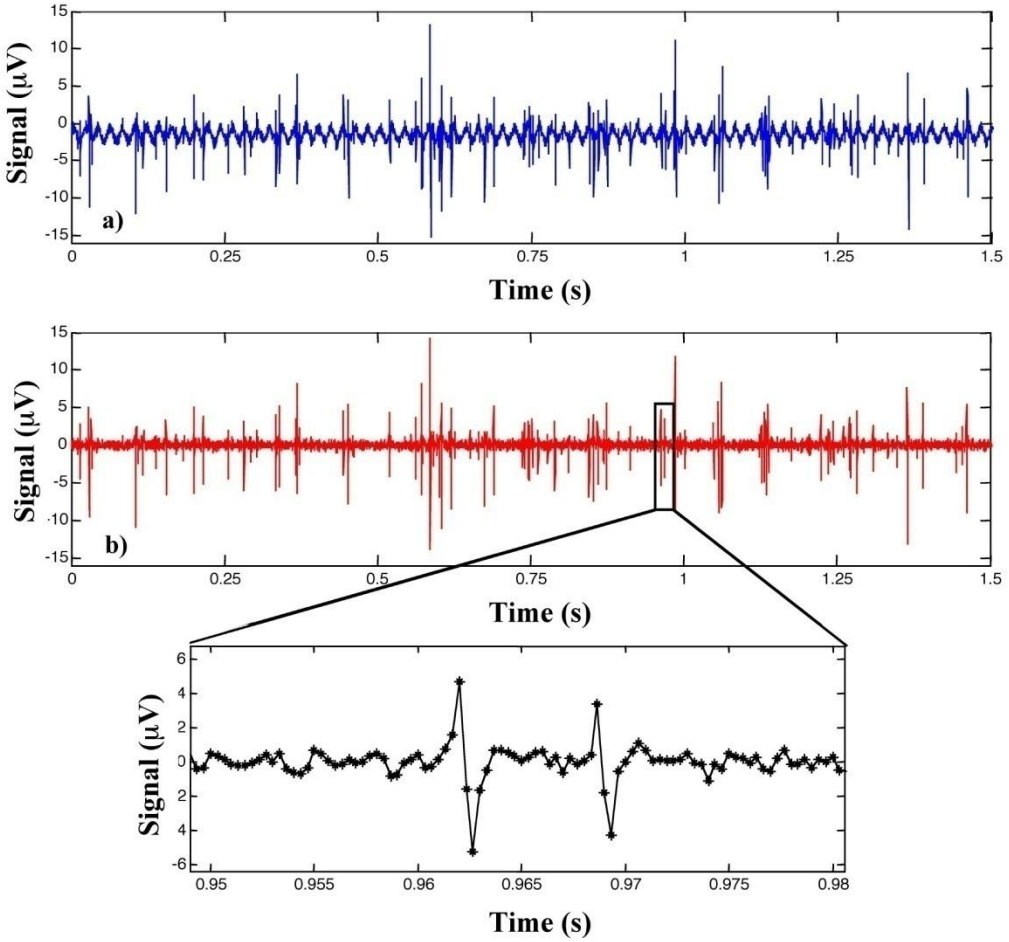
Zaragossa, INESC MN

Detecting labelled cells in flow





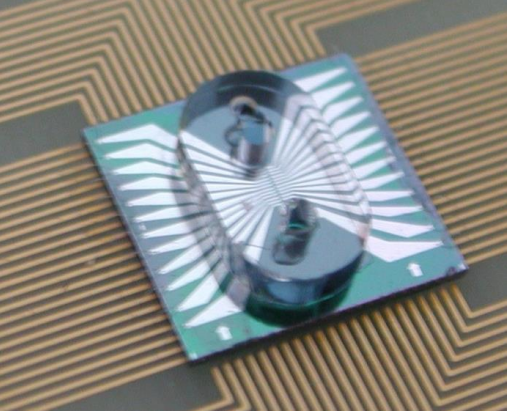


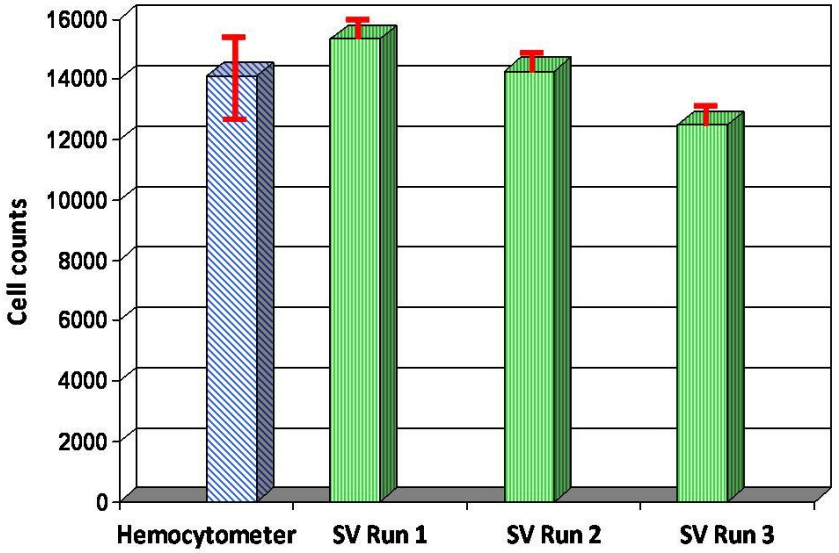


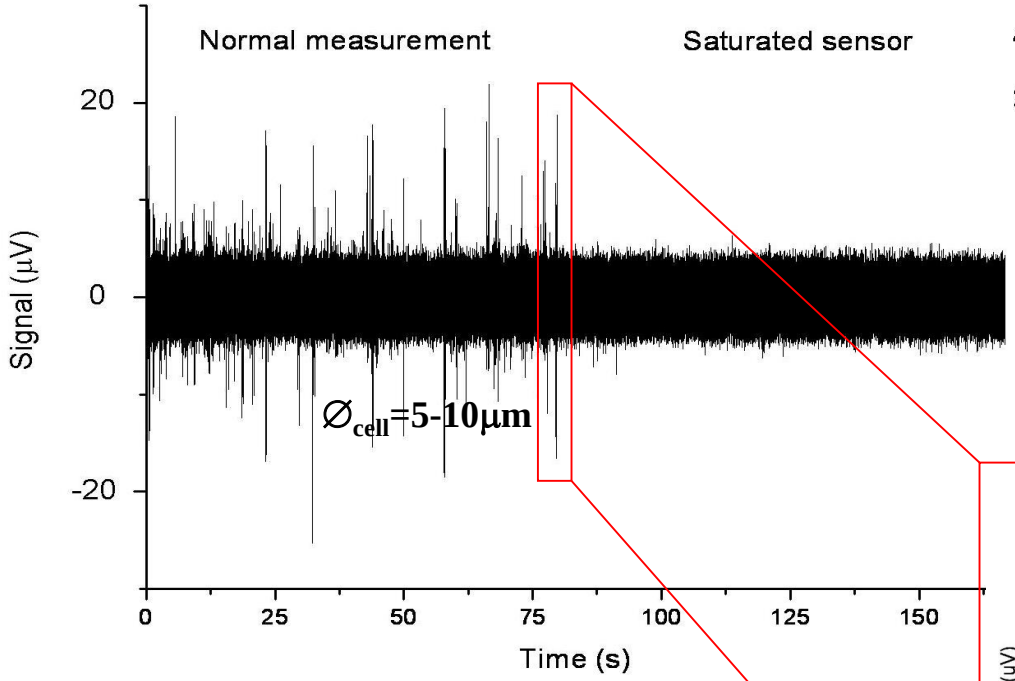
With C Fermon, and M Pannetier

APL 2009

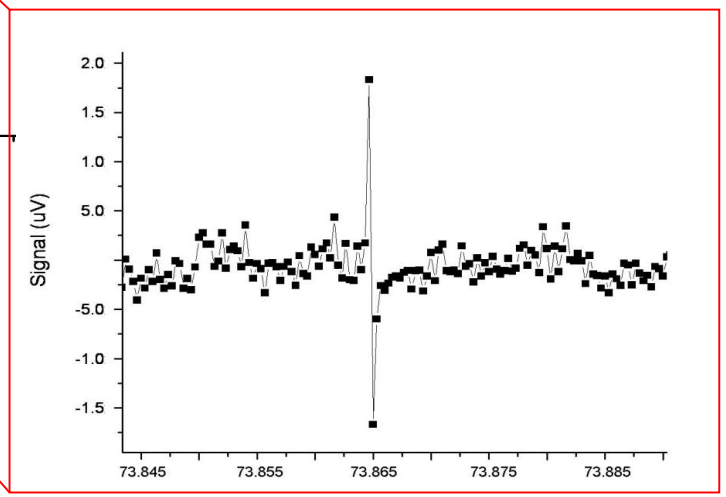
Cell detection – Kg1a cells







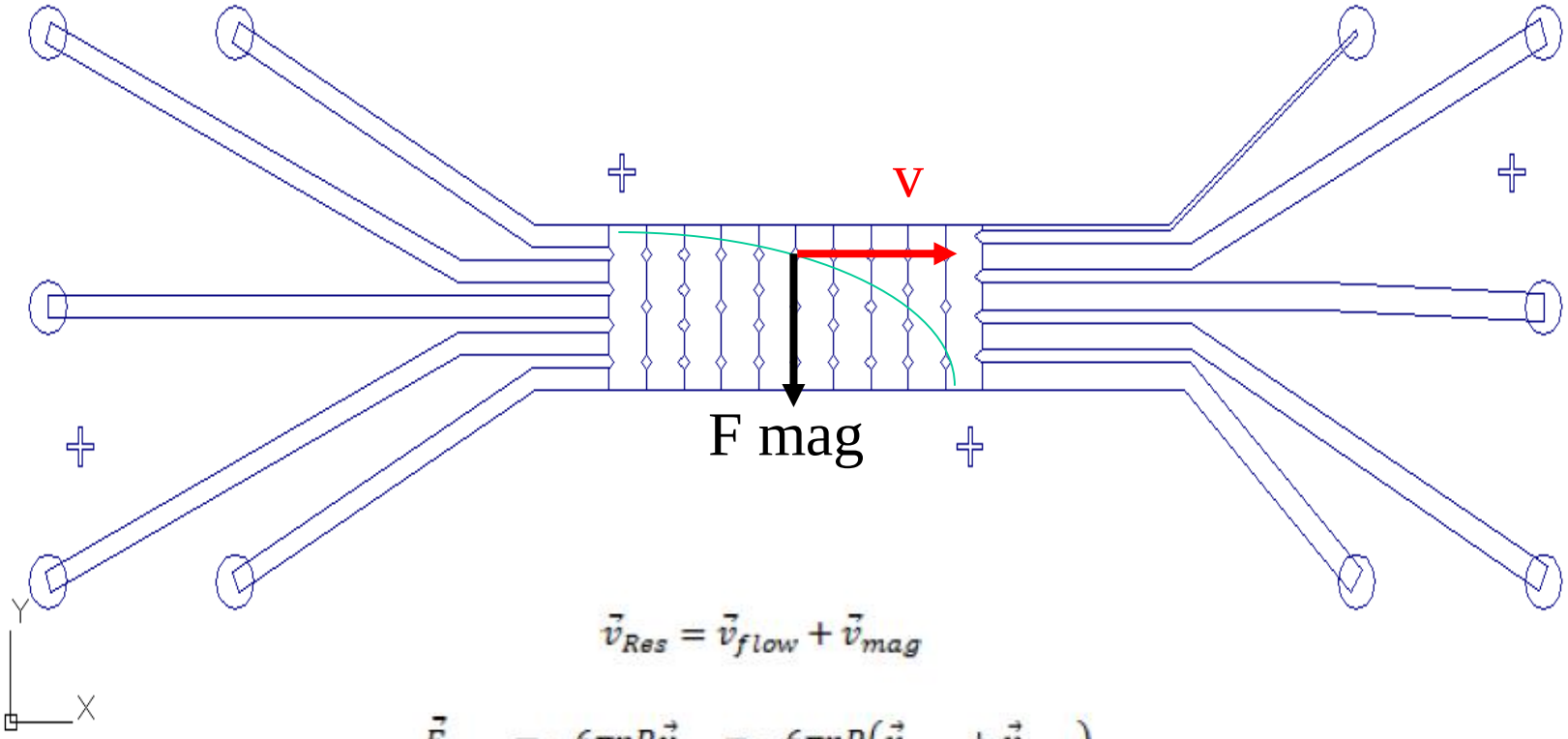
Lab on Chip (2011)



Cells marked with 50nm FeOx particles

Mgnetophoresis: using magnetic beads to separate biological entities

$$F_{mag} = \nabla (m.B)$$



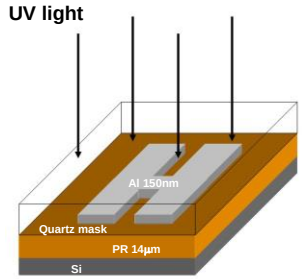
$$\vec{v}_{Res} = \vec{v}_{flow} + \vec{v}_{mag}$$

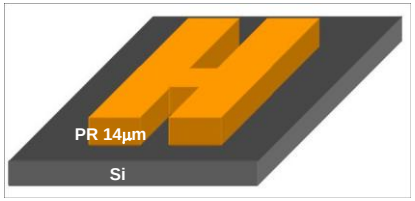
$$\vec{F}_{drag} = -6\pi\eta R\vec{v}_{res} = -6\pi\eta R(\vec{v}_{flow} + \vec{v}_{mag})$$

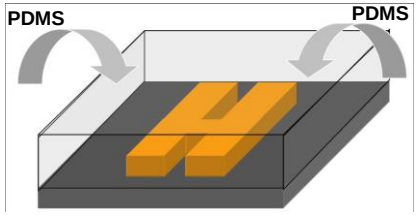
$$v_{mag} = \frac{F_{mag}}{F_{dragx}} = \frac{\mu_0 m}{6\pi\eta R}\nabla\vec{H}$$

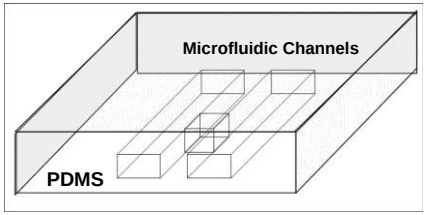
NANODEM FP7 (2012-2015)

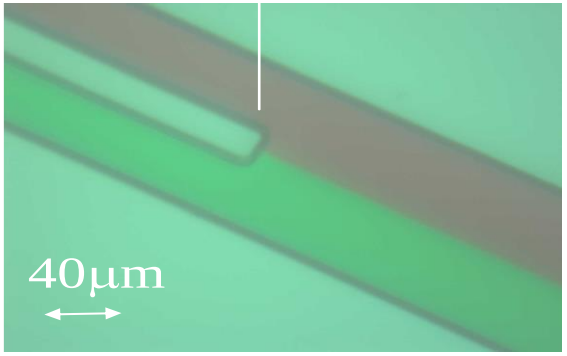
MICROFLUIDICS PROCESS











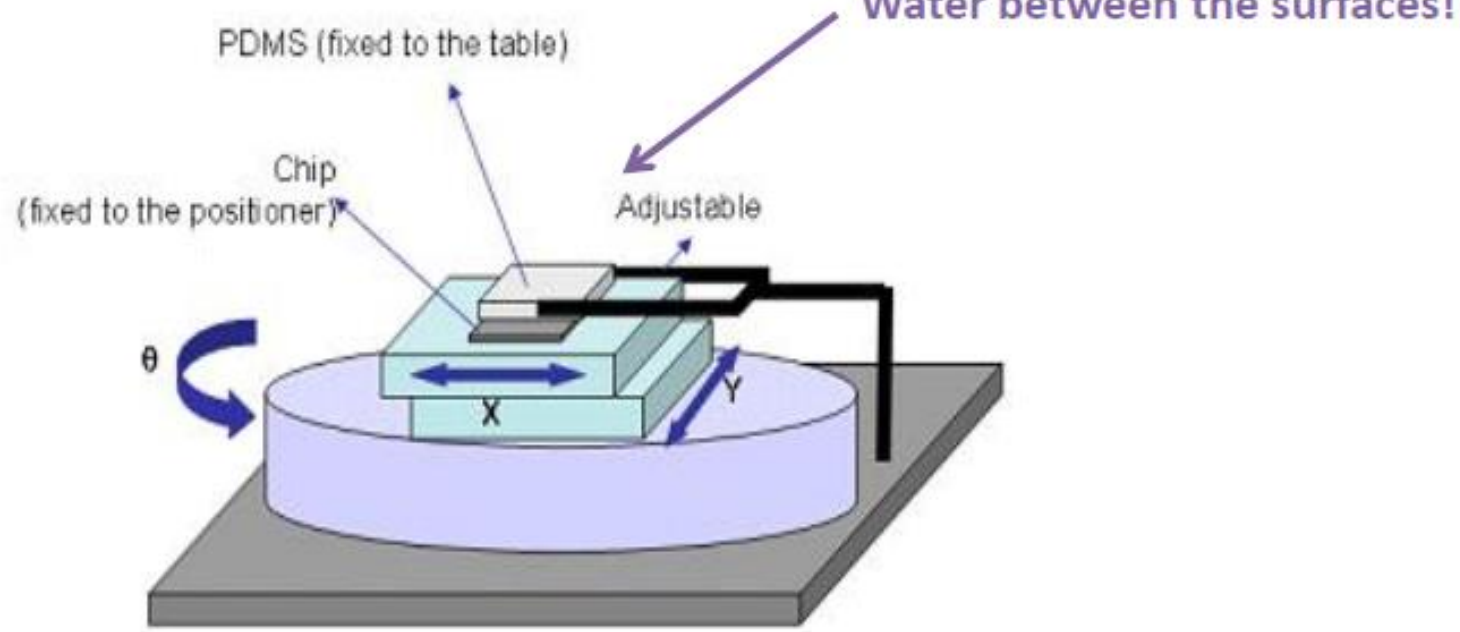
The fluidic component is made of PDMS using a standard micro molding technique.

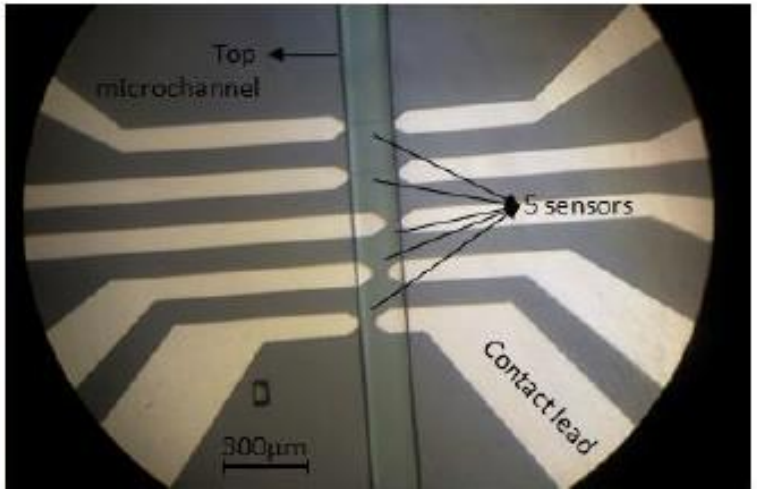
A master made of photoresist was patterned with UV lithography to define a counter mold of PDMS

Irreversible bonding between PDMS surface and the passivation surface of the chip: SiO_2

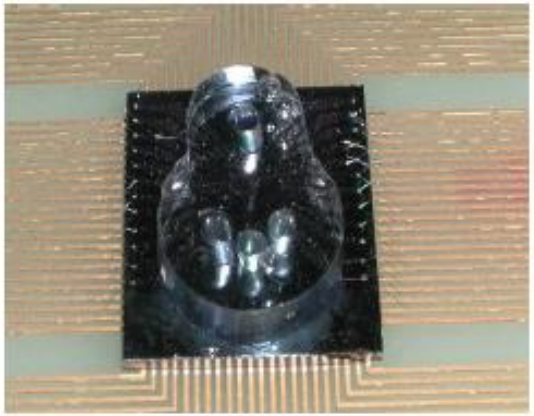
This picture shows a perfectly sealed system without leaks and with laminar flow.

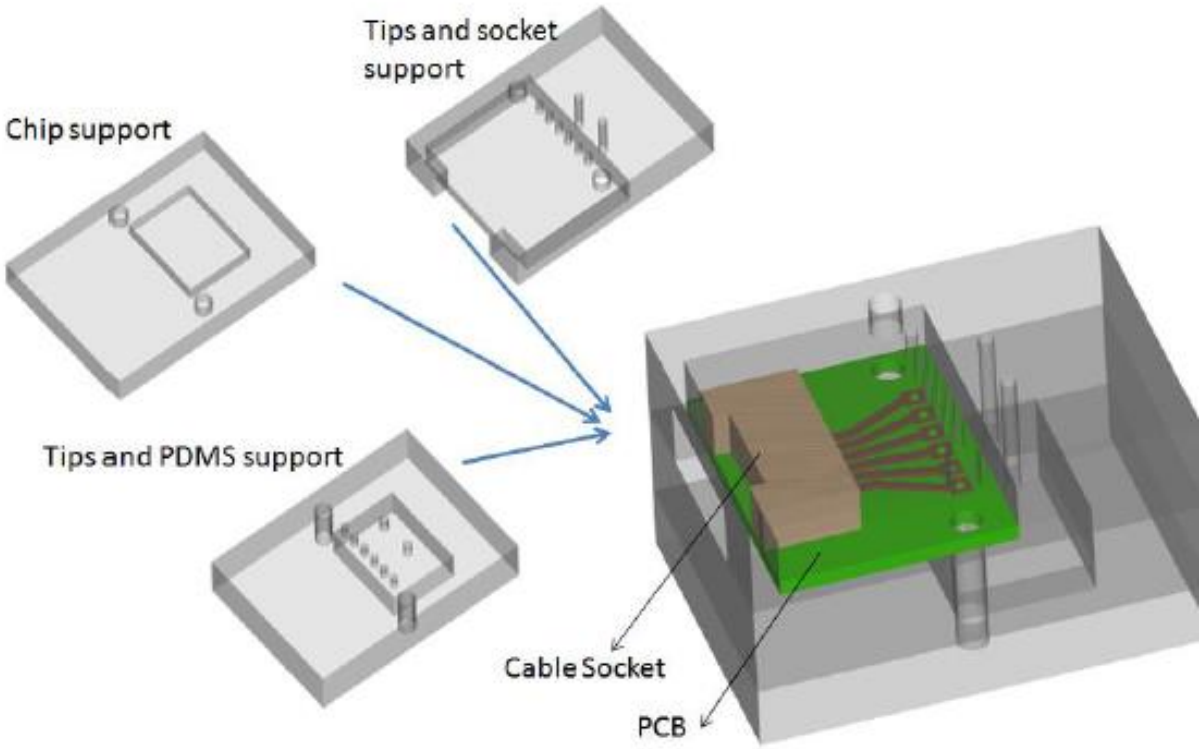
Alignment


Water between the surfaces!
PDMS (fixed to the table)
Chip
(fixed to the positioner)
Adjustable
θ
X
Y


Top
microchannel
5 sensors
Contact lead
D
300μm

Wire bonding or packaging

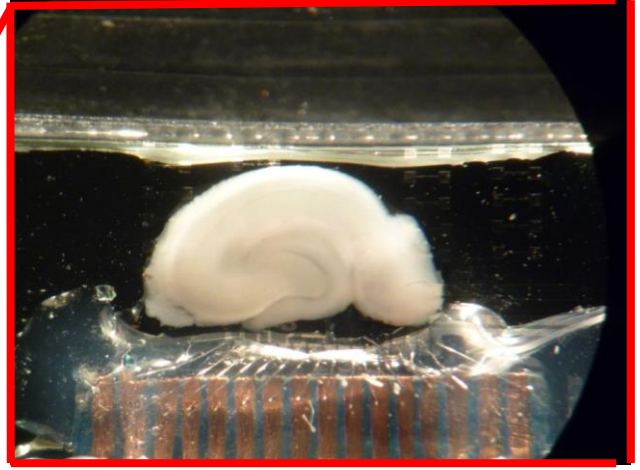


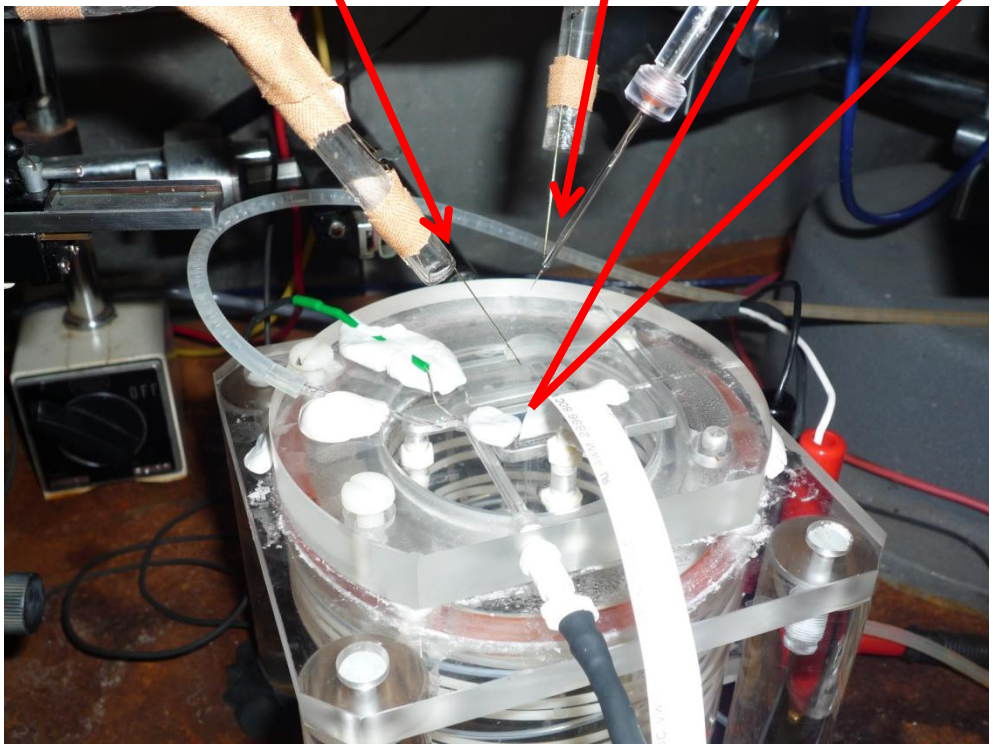


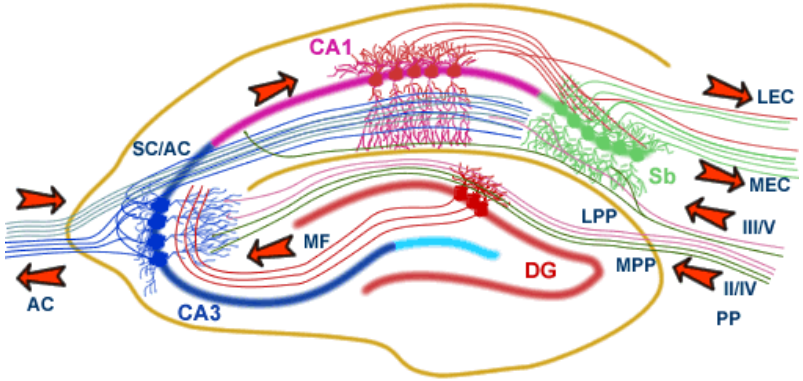
Synaptic current monitoring with high
Spatial resolution (with A.Sebastiao, IMM, V.Santos, ICVS)

Stimulation electrode

Recording electrode





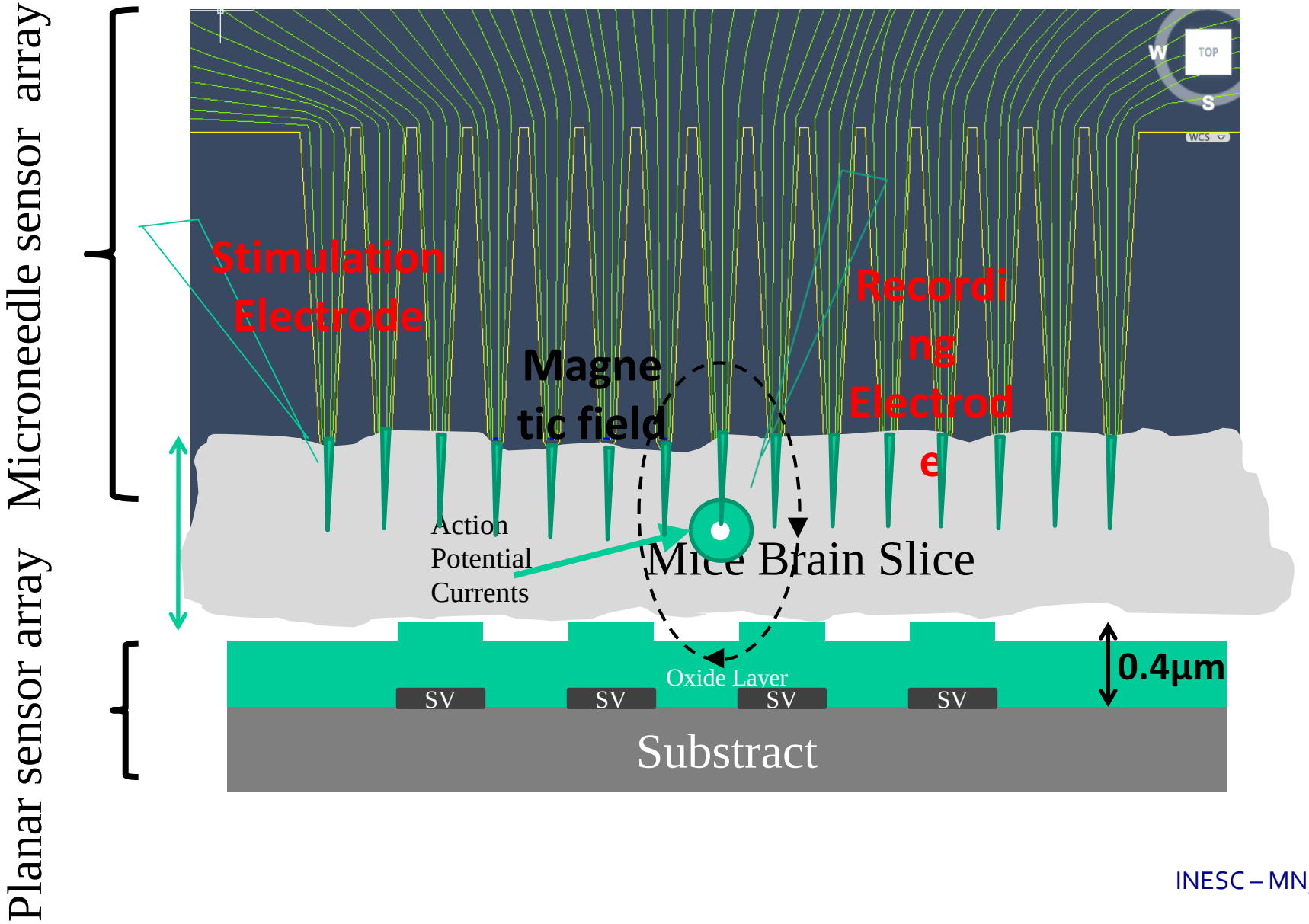


INESC MN and IMM

Rat hippocampus

MAGNETRODES, FP7 (2013-2016)

Si Needles with MR sensors or planar electrode array of MR sensors



Characterization

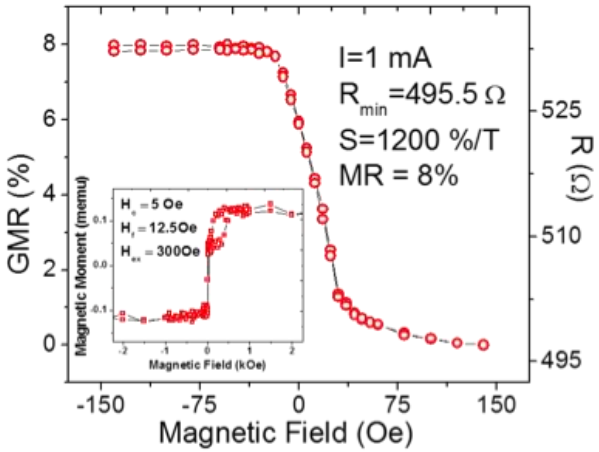
Spin Valve Sensor

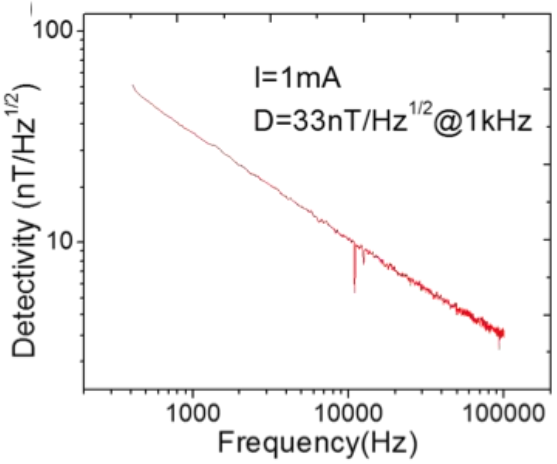
MR=8.14%;

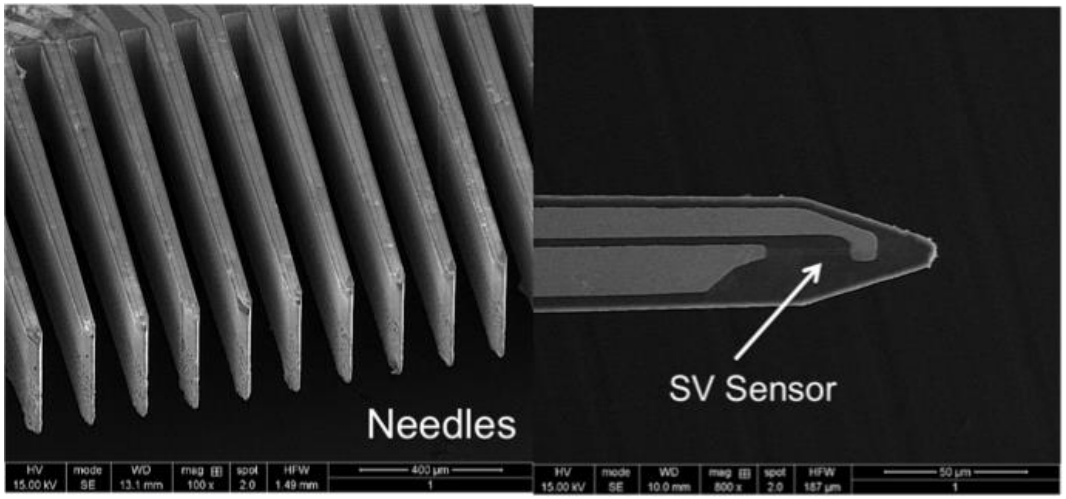
Hc=4.1 Oe;

Hf=17.8 Oe

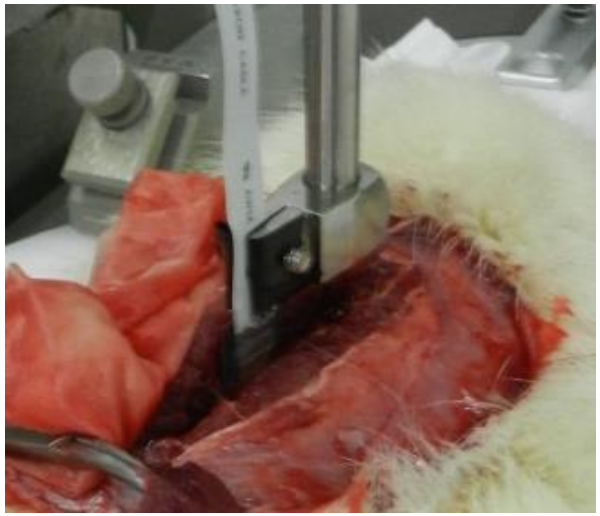
Ta	5nm
IrMn	8nm
CoFe	2.3nm
Cu	2.2nm
CoFe	2.3nm
NiFe	3.5nm
Ta	5nm



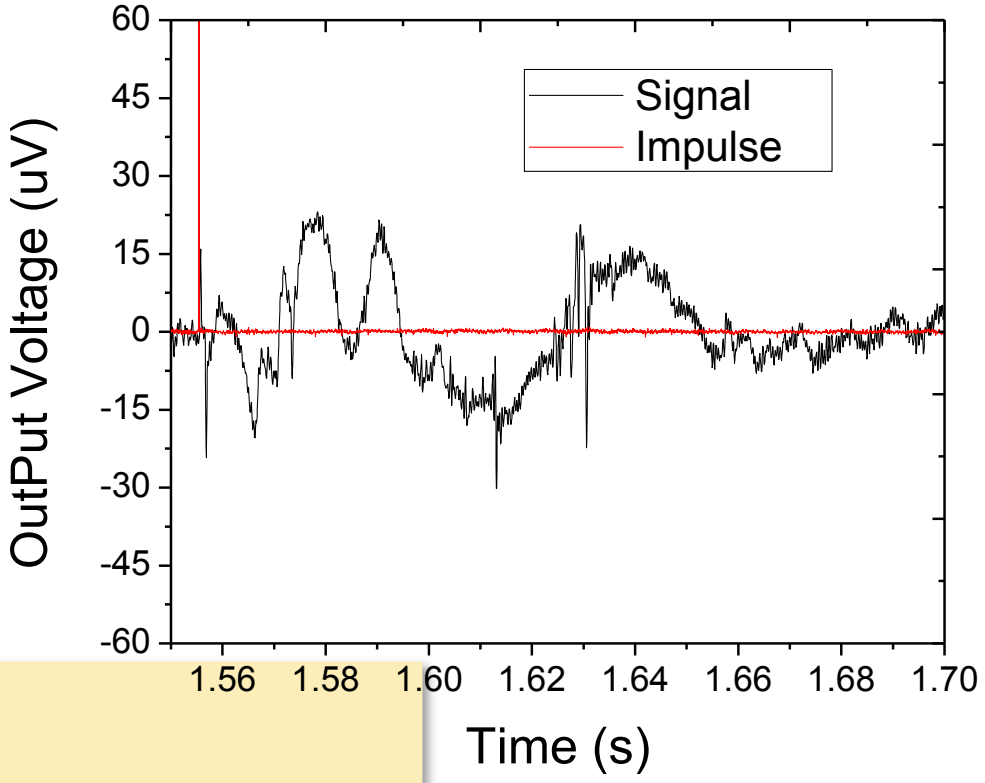




Results – MTJ response



In Vivo - Spinal Cord



The MTJ sensor readout:

- 20µV amplitude

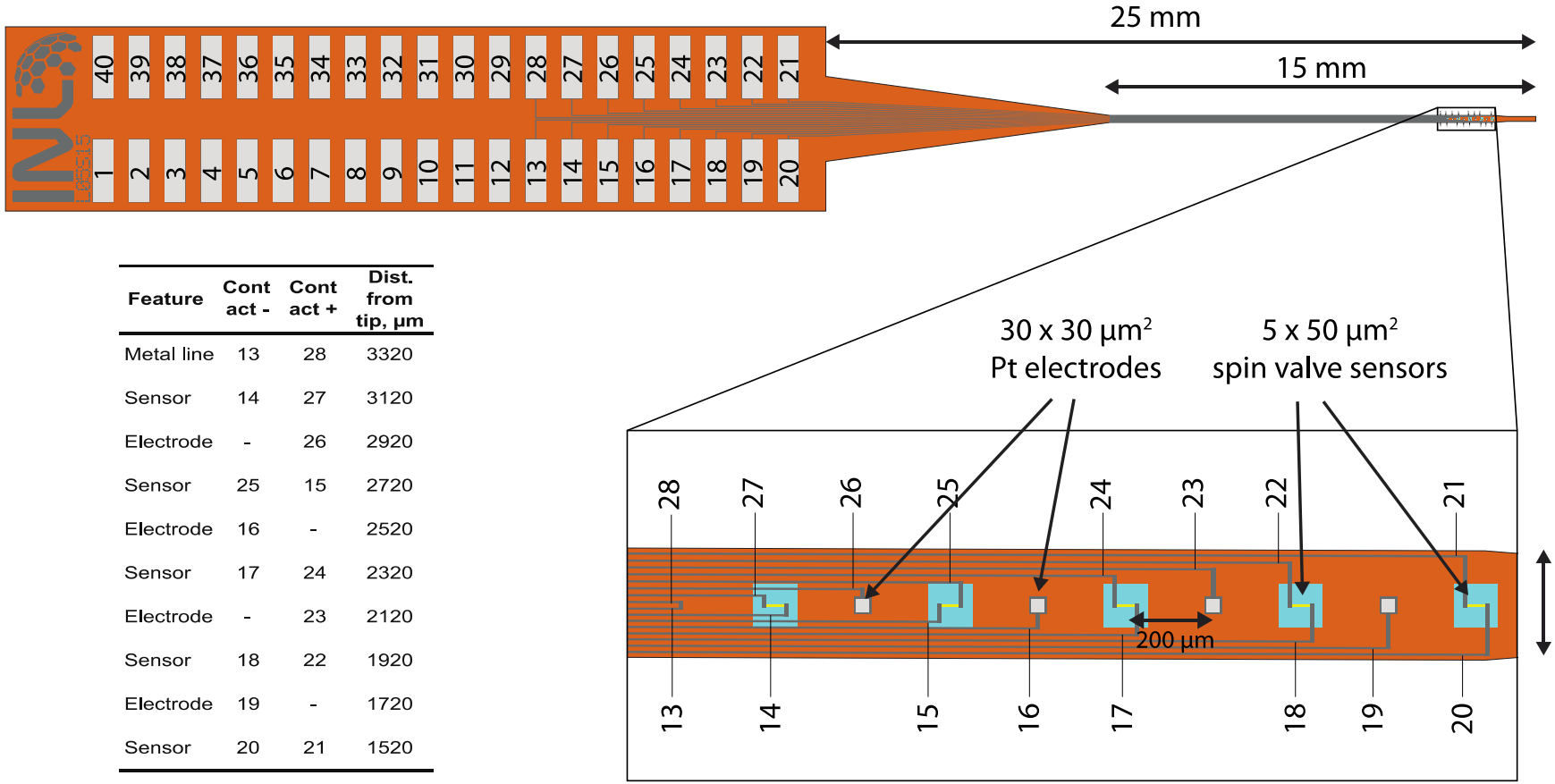
- 20uV amplitude signal corresponds to a magnetic field of about 3µT. (too strong ?? Evidence of capacitive coupling through ionic medium) VERIFIED Expect few hundred pT maximum

- Type of signal expected

Going to flexible probes for in vivo experiments

Probe Layout and Pinout

Pads size/pitch designed for Harwin 2-row, 40-way vertical connector

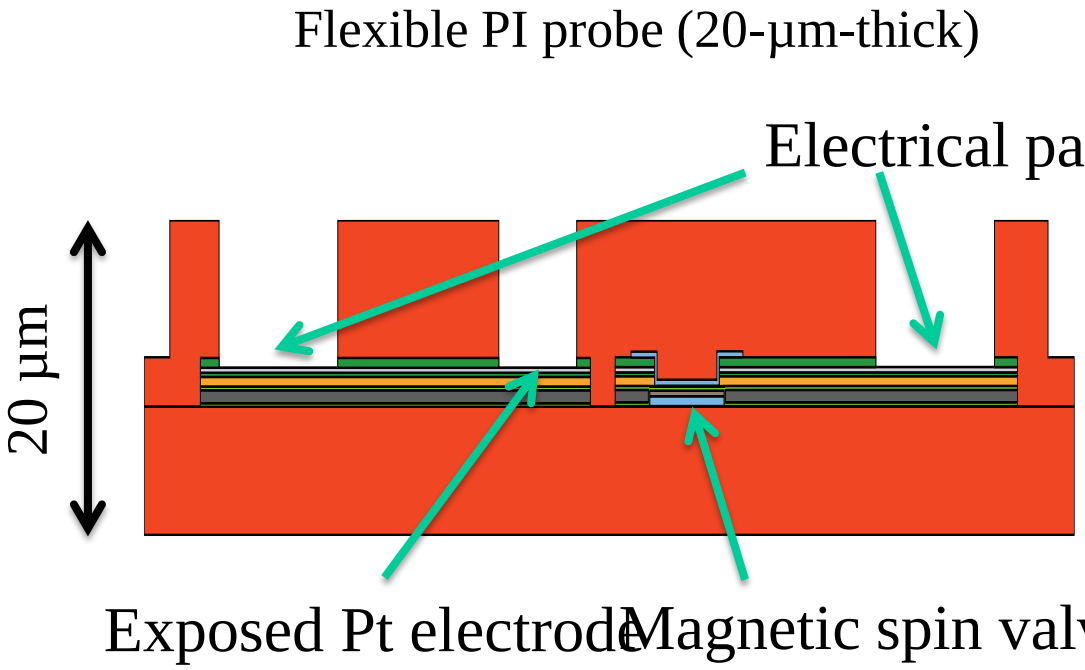


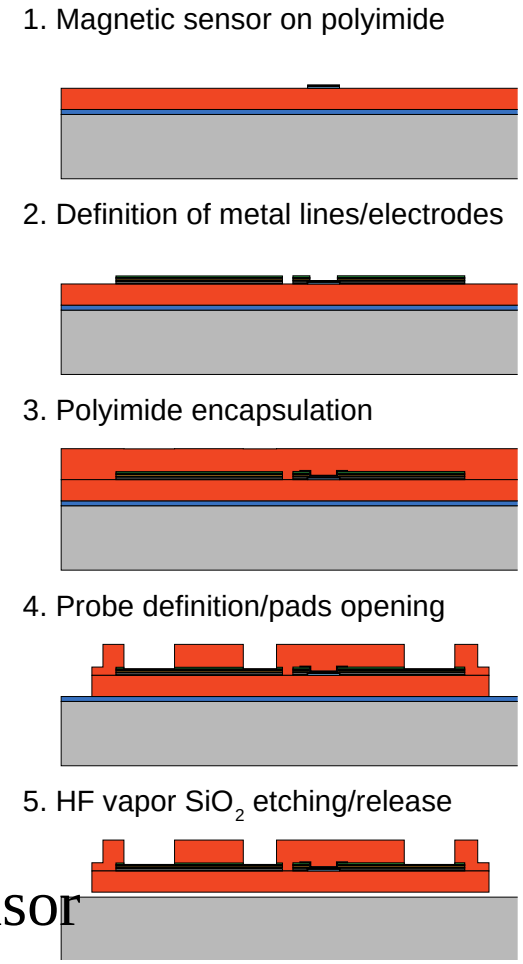
Feature	Cont act -	Cont act +	Dist. from tip, µm
Metal line	13	28	3320
Sensor	14	27	3120
Electrode	-	26	2920
Sensor	25	15	2720
Electrode	16	-	2520
Sensor	17	24	2320
Electrode	-	23	2120
Sensor	18	22	1920
Electrode	19	-	1720
Sensor	20	21	1520

J.Gaspar, J.Noh, INL

Fabrication Process

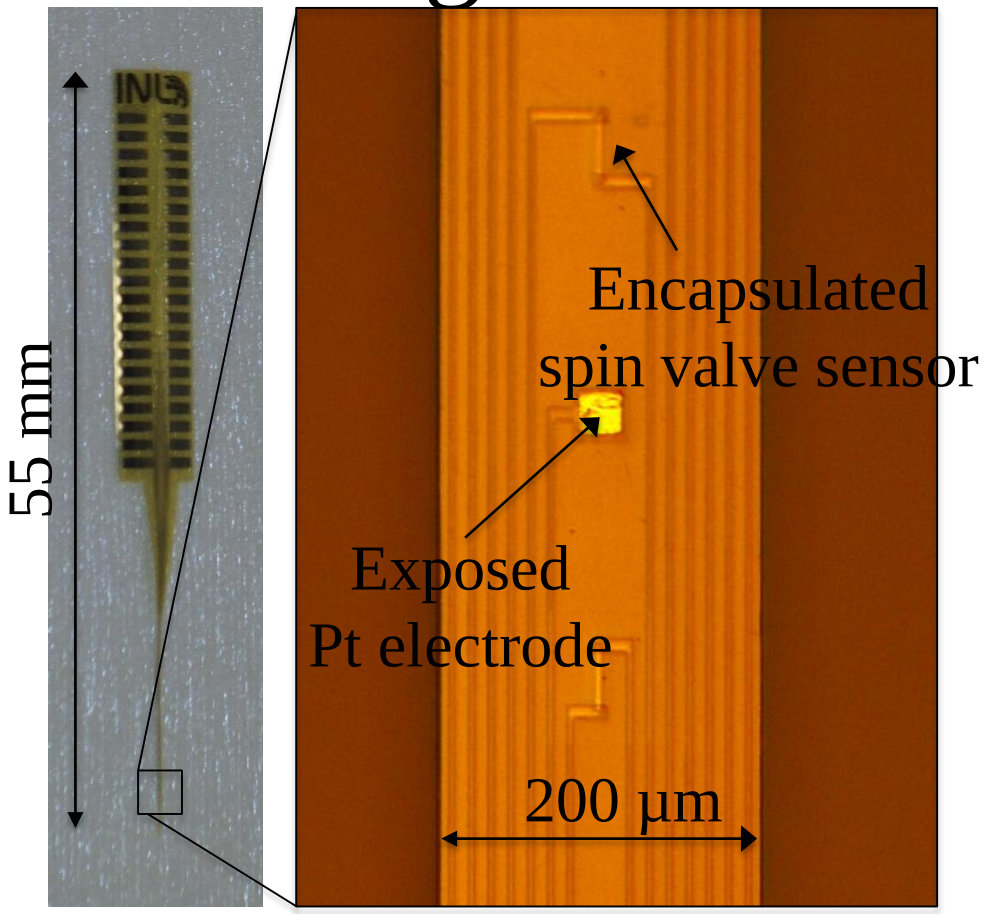
- Encapsulated magnetic sensors/exposed Pt electrodes

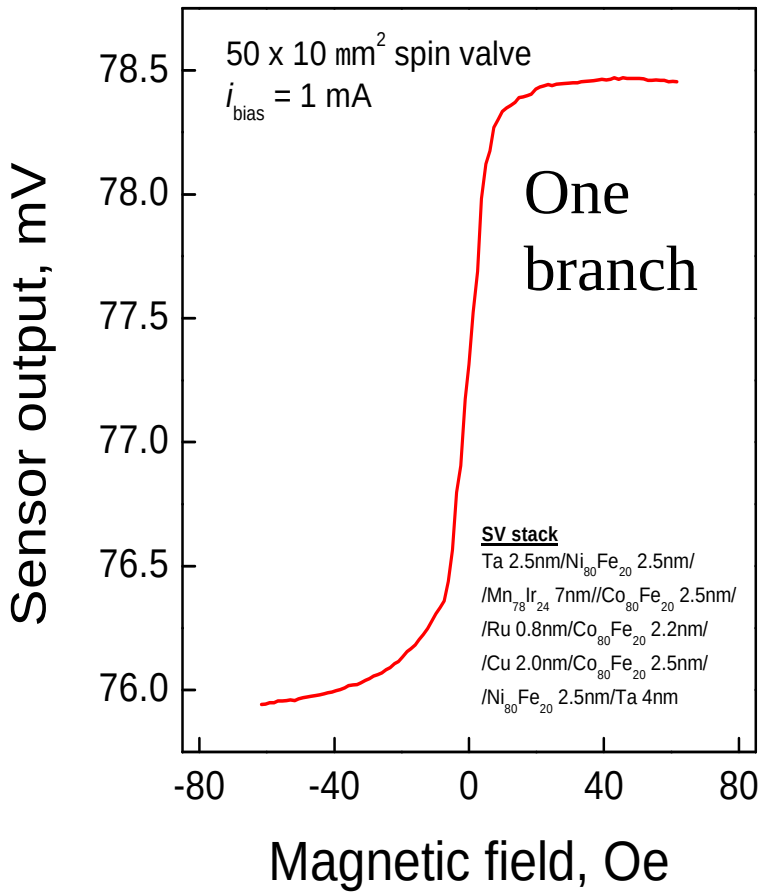




J.Gaspar, J.Noh, INL

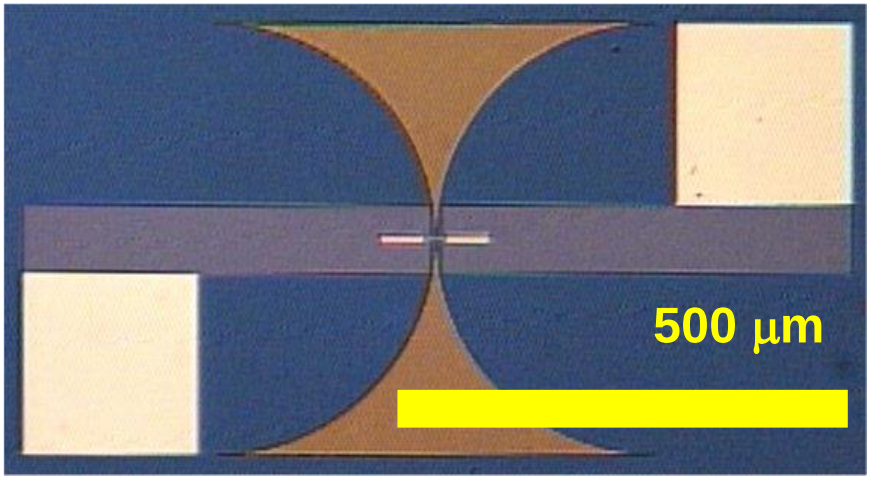
Magnetoresistive Behavior

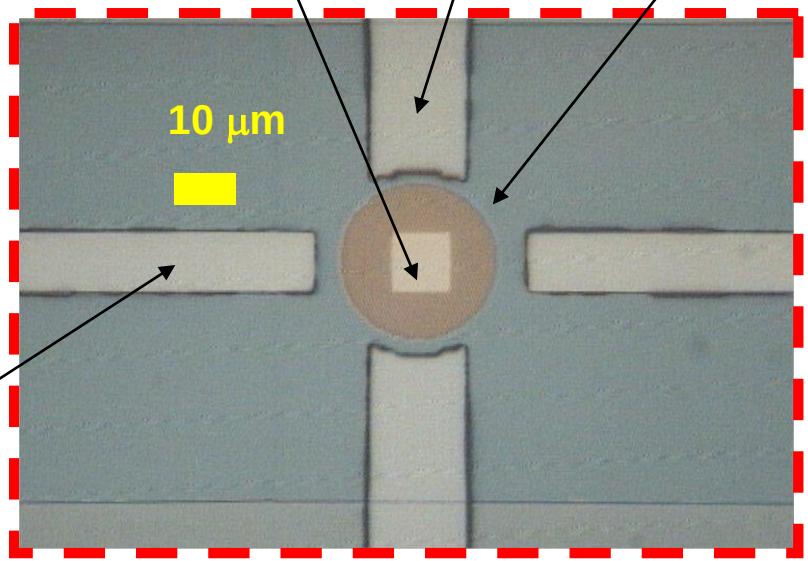




J.Gaspar, J.Noh, INL
S.Cardoso, INESC MN

Incresasing sensitivity III:
Hybrid MTJ+flux guide structures: towards pT detection at 1Hz

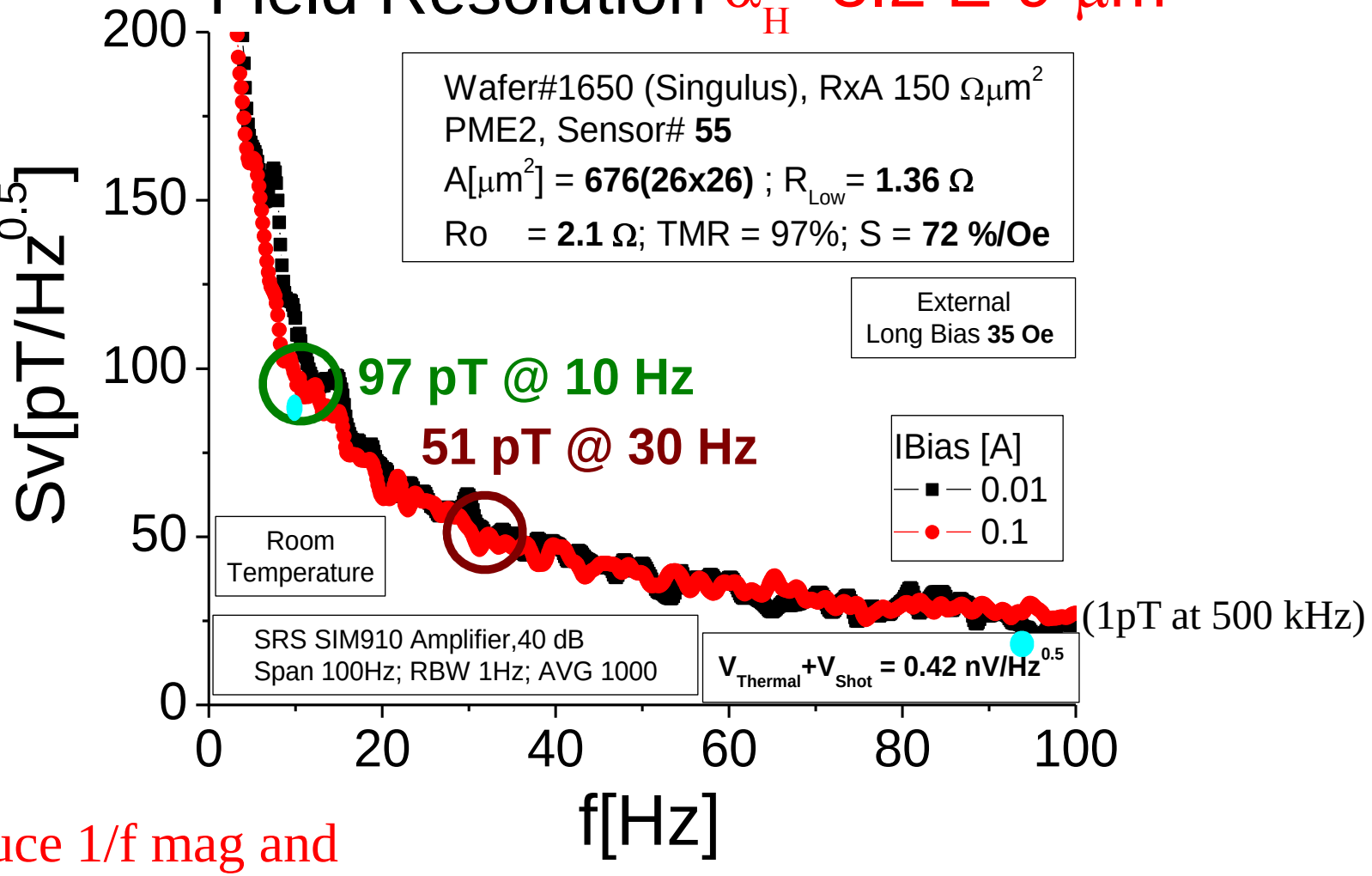




Goal: increase volume of free layer-reduce magnetic 1/f noise
increase junction area-decrease barrier 1/f noise
increase sensitivity: flux guides + MgO MTJ

Noise characteristics

Field Resolution α_H=3.2 E-9 μm^2



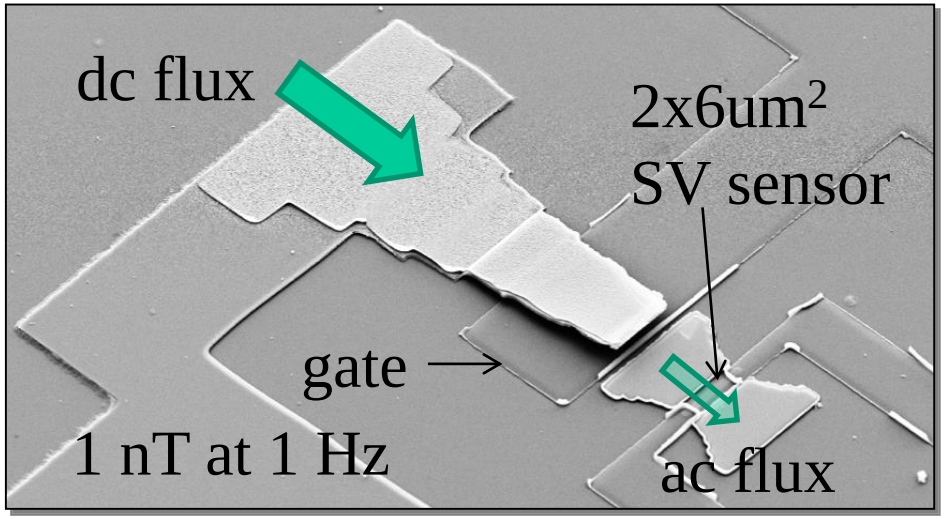
Need reduce 1/f mag and white mag noise

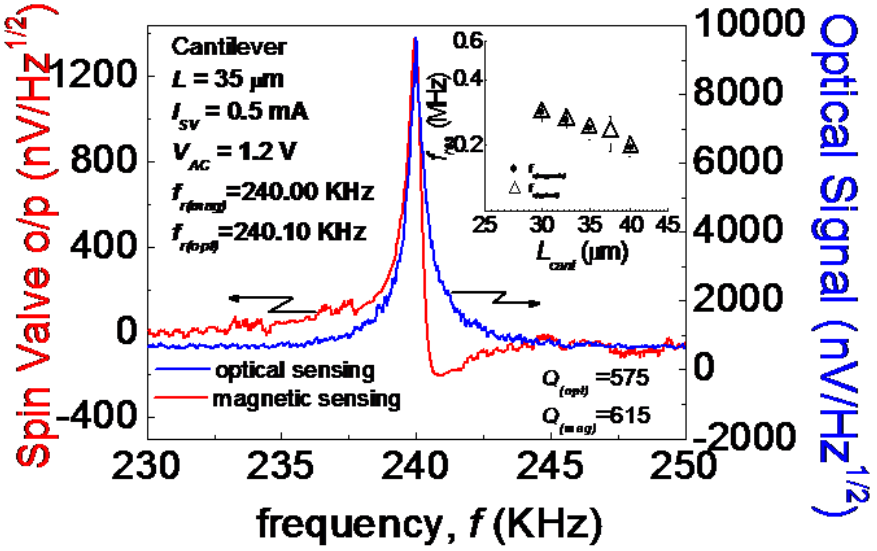
Appl. Phys.Lett., 91, 102504, August 2007

pT DC Magnetic Field Detection

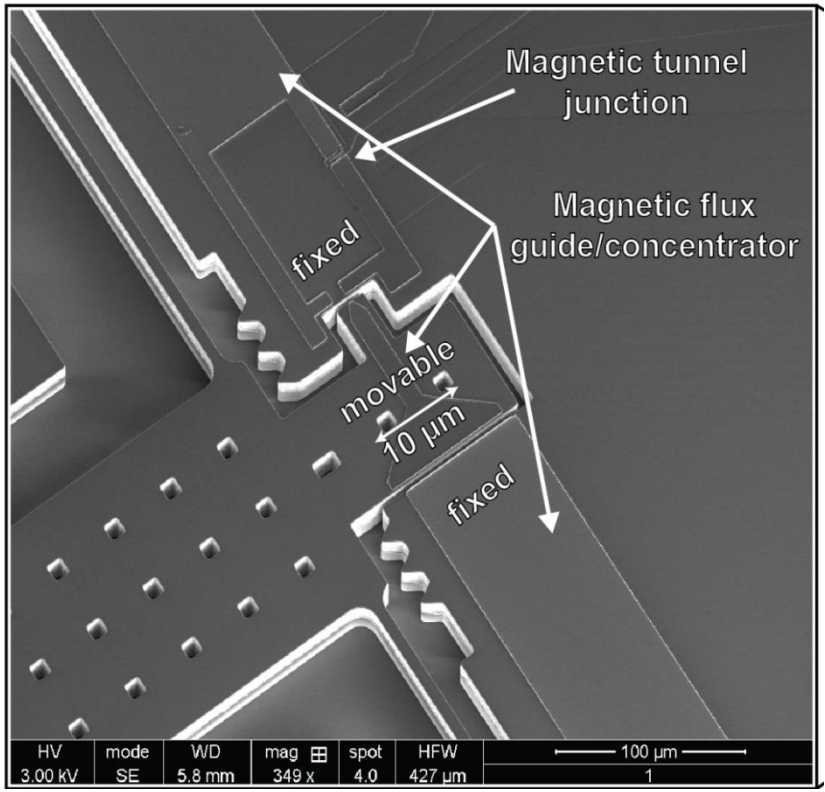
Hybrid SpinMEMS; DC to AC flux transformer

2006-2010, hybrid MR / thin film MEMS



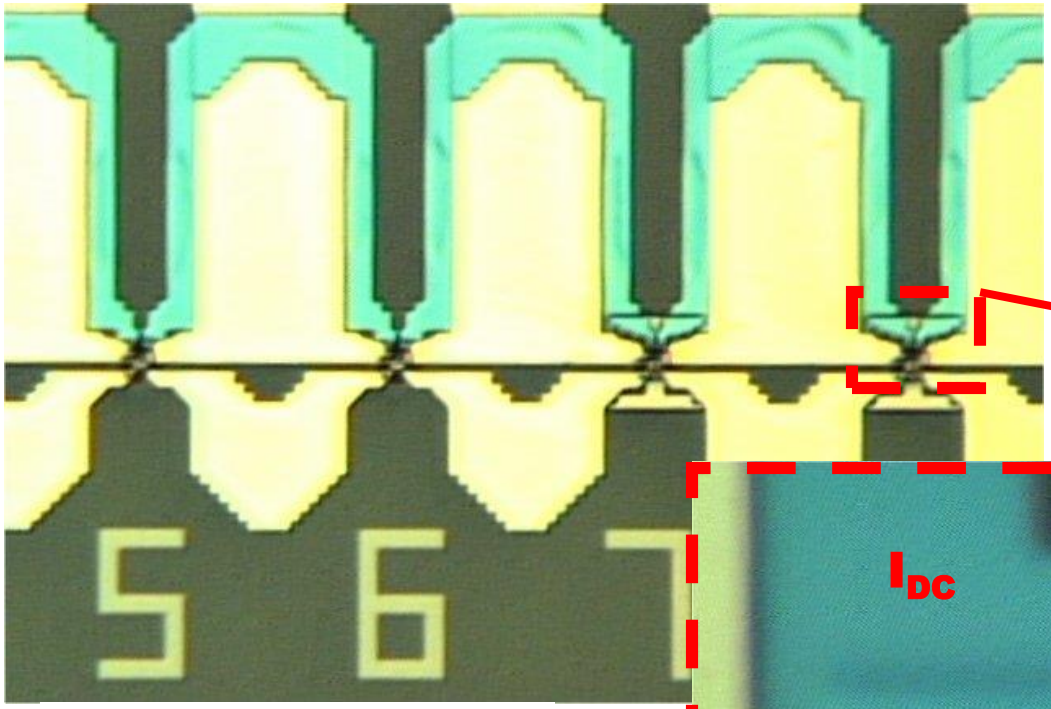


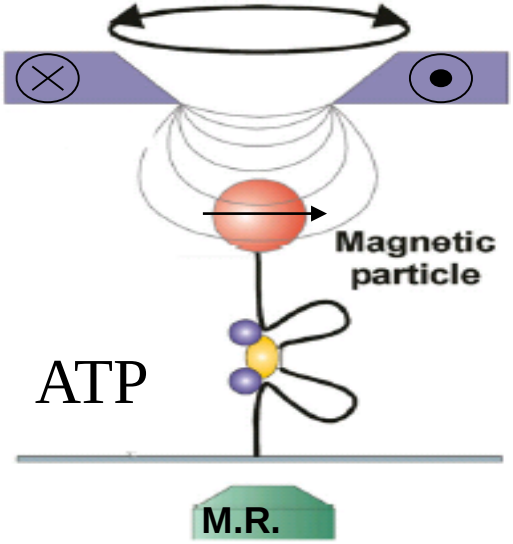
2011-onwards,
also bulk MEMS, INL and UC Berkeley/Davis

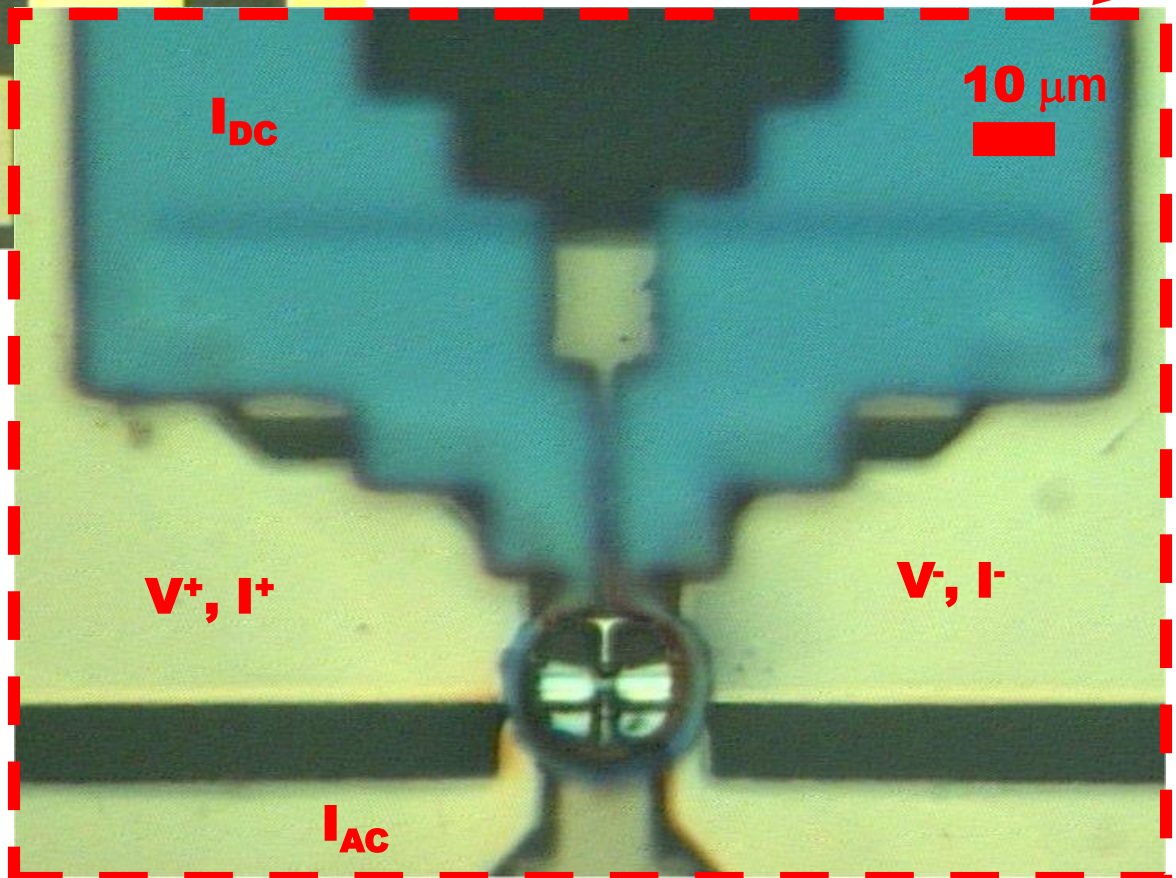


Magnetic Tweezer, DNA manipulation, DNA translocation

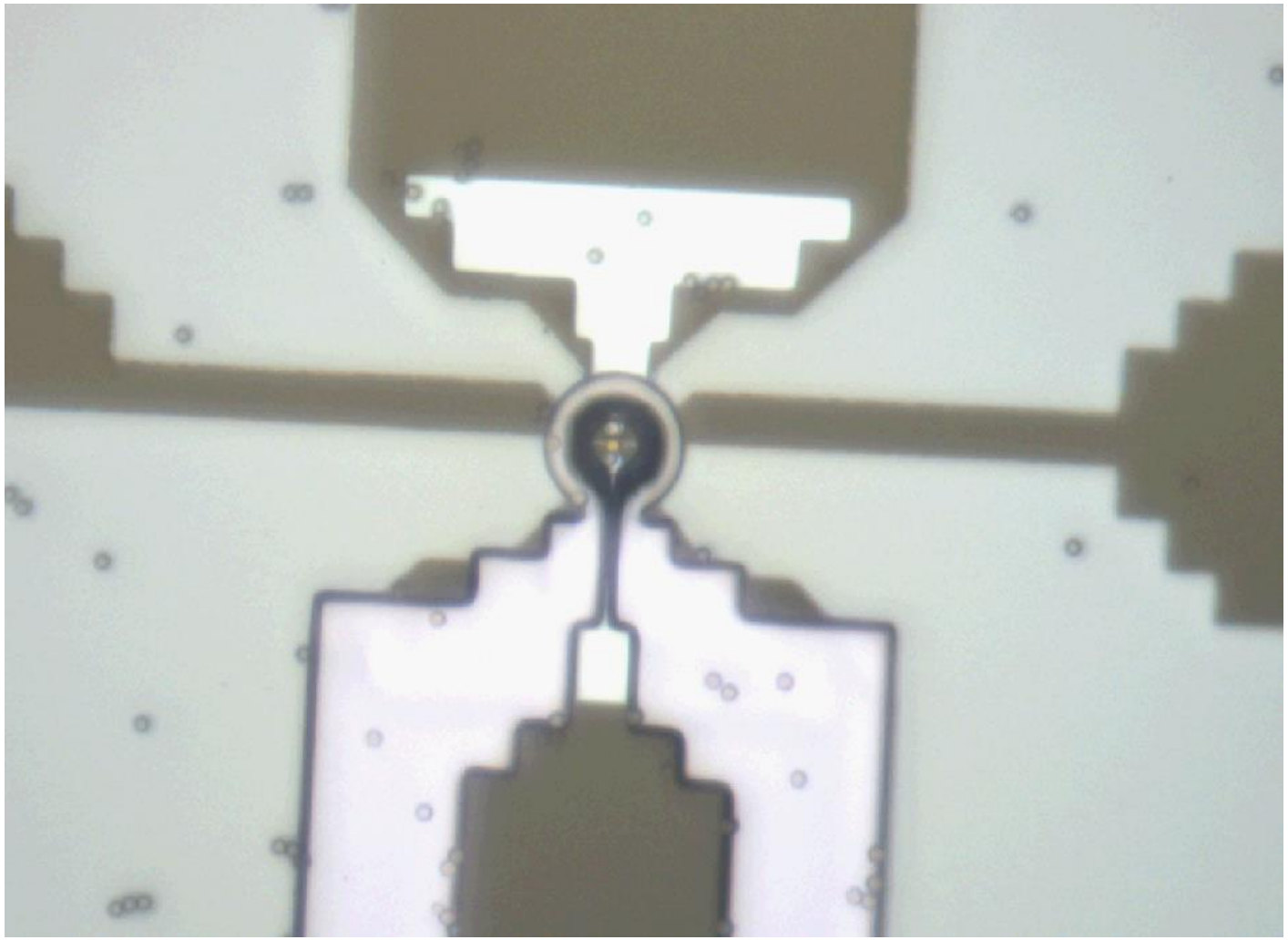
FP6 BioMolSwitch







Optical μScope – Vertical Switch



I_{MT}

0 mA

45 mA

Achieved:
60nm resolution

Inesc MN research group

INL

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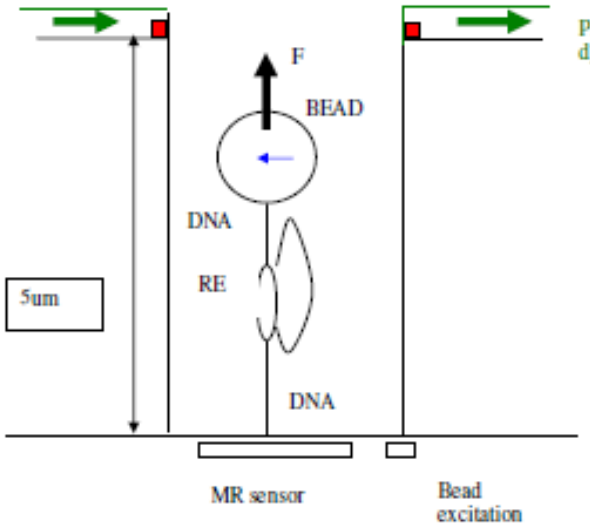
Obrigado!

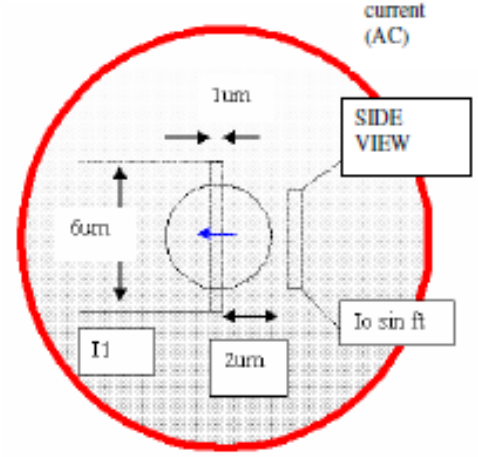
ANNEX 1

3 problems from the nanoelectronics course at IST-Lisbon

Problem 1

"Dimensioning a magnetic tweezer and a sensor to monitor DNA translocation using an restriction enzyme molecular motor"
Consider Fig.1





a) The Magnetic Tweezer. Calculate the force Fz that the 1um radius bead (susceptibility X=0.5, for H< 2kA/m) will be subject to, created by the field Bz caused by the current loop (I = 50mA, R=10um). The loop is placed on a oxide mesa, 5um thick. The bead attaches to a DNA strand, 2um long.

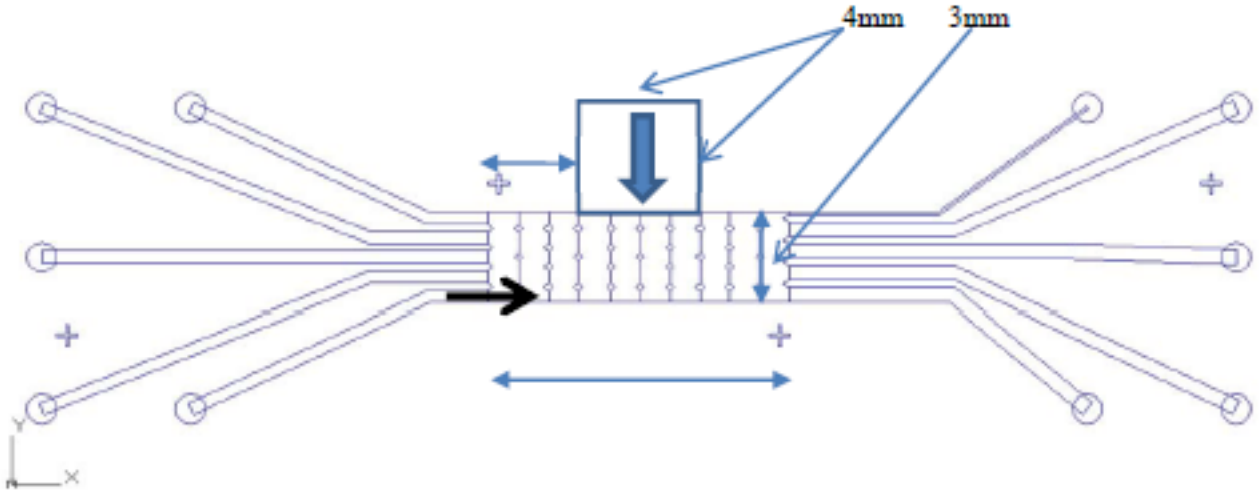
b) Now redesign the magnetic tweezer using a thin , magnetized CoPt film , 100nm thick (Mr=400kA/m), placed on top of the 5um thick oxide. For the field calculation, consider only the magnet extremities at the border of the pit, and use a 2D approach (magnet width >> magnet thickness). Calculate the force created by this magnet on the bead.
Hn = -(σ/2 π) (θ2-θ1) ; Ht= -(σ/2 π) ln (r2/r1) (see class, where σ is the magnetic surface charge M.n).

c) Now the bead is magnetized by an AC field created by a current wire on the substrate (I = 40mA sin 100 Hz t) Assuming the susceptibility given above, calculate the rms voltage in the magnetoresistive sensor created by the bead, at its initial separation. The magnetoresistive sensor is a spin valve, with a maximum MR=10%, a resistance per square R=200hm/sq, dimensions (6um by 2 um), with a DC bias current of 10mA, and a linear range of +-1kA/m. Calculate the spatial sensitivity (del V/del Z) around the initial bead position.

Problem 3: a biomolecular separation lab on chip platform (scheduled handing in time: October 16th 2012)

Fig.1 shows a top view of a microfluidic chamber used for biomolecular separation. The chamber is 14 um high, 10mm long and 3mm wide. Cells labeled with magnetic nanoparticles enter from the left bottom inlet, with a horizontal velocity of 1, 5 and 10 mm/s (fluid velocity) and are subject to the magnetic force created by a permanent magnet placed as indicated in the figure (Mr = 1100 kA/m, magnet dimensions 4x4mm2). Calculate the cell trajectory neglecting gravity and impulsion forces (only the Stokes force $\mathbf{F}_{drag} = 6\,\pi\,R\,\mu(v_{fluid} - v_{bead})$ and the magnetic force $\mathbf{F}_b = \mathbf{grad}\,(\mathbf{m}.\mathbf{B})$ will affect the bead motion, where R_b=1.0um, and μ = 0.001 Pa s). Assume that the cell coated with magnetic nanoparticles acts as a macro moment with saturation magnetization Ms=57 kA/m, for H > 500 kA/m (magnetic susceptibility χ = 0.25 for H < Hs = 500 kA/m), and moment m=MsVb. Calculate the bead trajectory for a bead that enters at the bottom for different horizontal fluid velocities of 1,5 and 10 mm/s.

Note: in practice assume that for a time intervals Δt the bead almost immediately reaches the terminal speed given where Fmag +F drag =0 and use this terminal speed to calculate the motion within this time interval. Take into account both horizontal and vertical field gradients created by the magnet on the bead. For the horizontal movement take also into account the magnetic force along x.

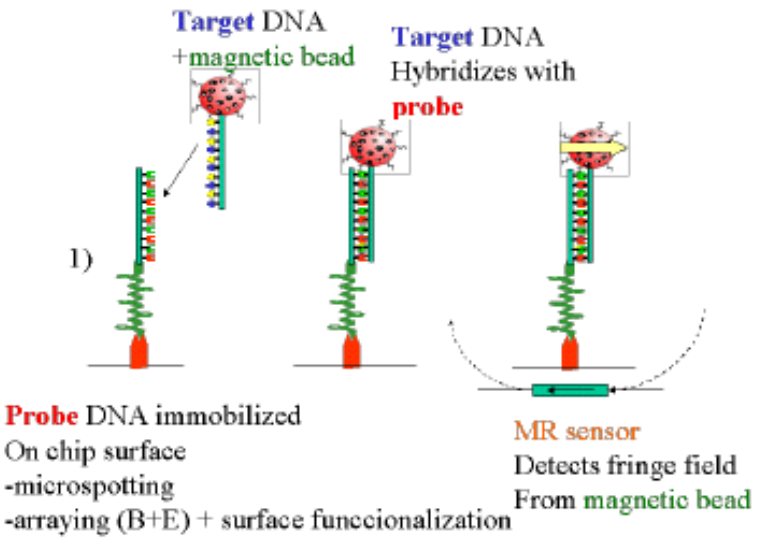


Problem 4: A magnetoresistive based biochip for biomolecular recognition detection.
(to be handed in Tuesday October 30th)

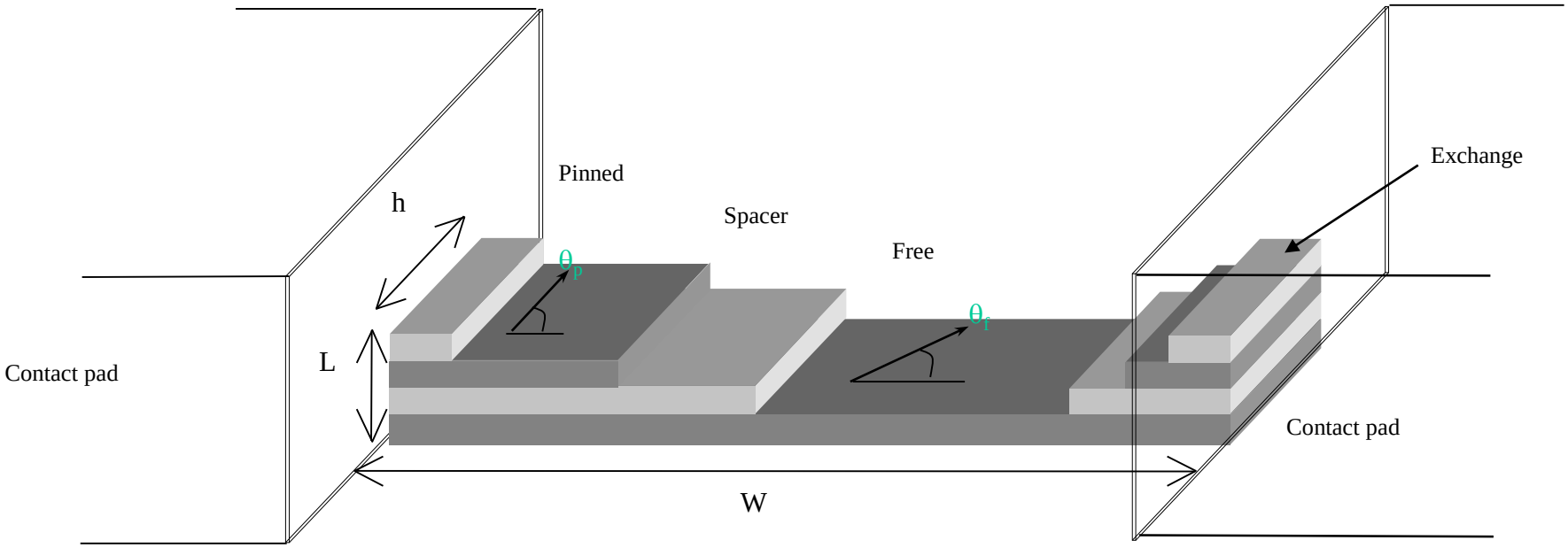
In a biomolecular recognition chip, probes are immobilized on a substrate, and labeled targets are arrayed over the probes, When complementarity exists between the probe and target, the target biomolecule becomes immobilized on the substrate, and the label can be detected by an integrated transducer. A magnetoresistive biochip uses a magnetoresistive detector to measure the fringe field coming from magnetic labels attached to biomolecular targets, that are specifically bound to immobilized probes. In this problem consider that target DNA is labeled with magnetic nanoparticles (magnetite based) with a diameter of 100nm and a magnetic susceptibility at low frequency (< 1 kHz) of 1. The labels are magnetized by a transverse AC field of 1kA/m (amplitude) at 800 Hz created by integrated Helmoltz coils. The label fringe field is detected with a linearized spin valve sensor with dimensions 2μm x 80 μm, with MR = 10%, H_k^{eff} = 1.5kA/m, Rs= 1kOhm, Is=2mA. The spin valve sensor is passivated by a AlOx layer 400nm thick.

a) Calculate the response of the sensor to a single bead fringe field, assuming the bead is placed over the center of the sensor. Assume the bead center to sensor free layer separation to be 400nm.
b) Knowing that the sensor noise level at 800 Hz is 50nV/sqrt(Hz) calculate the minimum number of these labels that can be detected by this sensor. Assume that a frequency bandwidth of 100 Hz is used for the measurement, and that the beads are located approximately near the center line of the sensor (away from the edges).

$\Delta V(SV) = MR \cdot Rs \cdot I \cdot \langle Hs \rangle / 2H_k^{eff}$



ANNEX II: The Spin Valve Sensor



$$\Delta V = \tfrac{1}{2} (\Delta R/R).I.Rsq.(W/h) \langle 1-\cos(\theta_f-\theta_p)\rangle$$

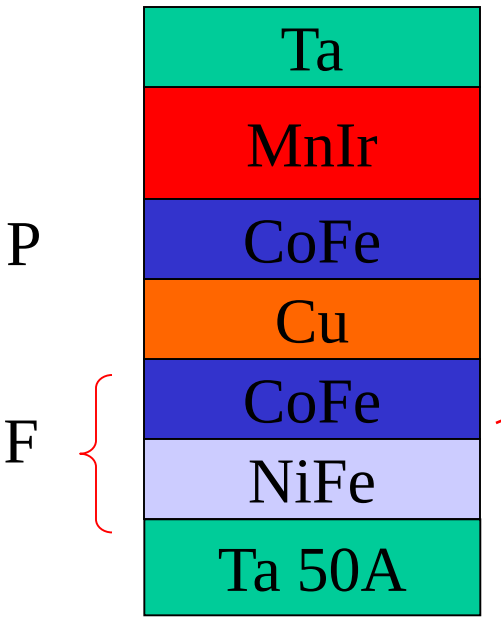
1-C.Tsang, R.E.Fontana, T.Lin, D.E.Heim, V.S.Speriosu, B.A.Gurney, and M.L.Williams, IEEE Trans.Magn., 30, 3801 (1994).

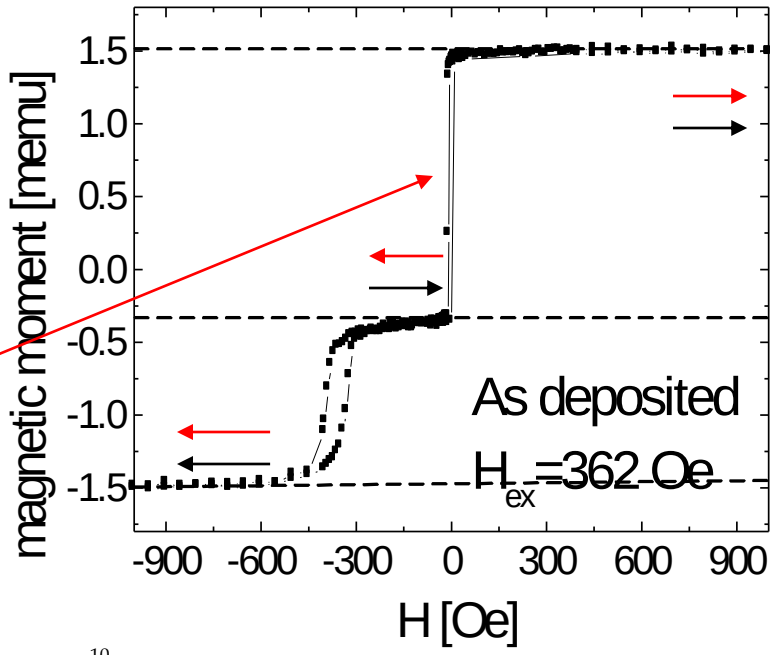
3- B.Dieny, V.S.Speriosu, S.S.Parkin, B.A.Gurney, D.R.Wilhoit, and D.Mauri, Phys.Rev.B, 43, 1297(1991).

4- D.E.Heim, R.E.Fontana, C.Tsang, V.S.Speriosu, B.A.Gurney, and M.L.Williams, IEEE Trans.Magn.., 30, 316 (1994); P.P.Freitas, J.L.Leal, L.V.Melo, N.J.Oliveira, L.Rodrigues, and A.T.Sousa, Appl.Phys.Lett., 65, 493 (1994);

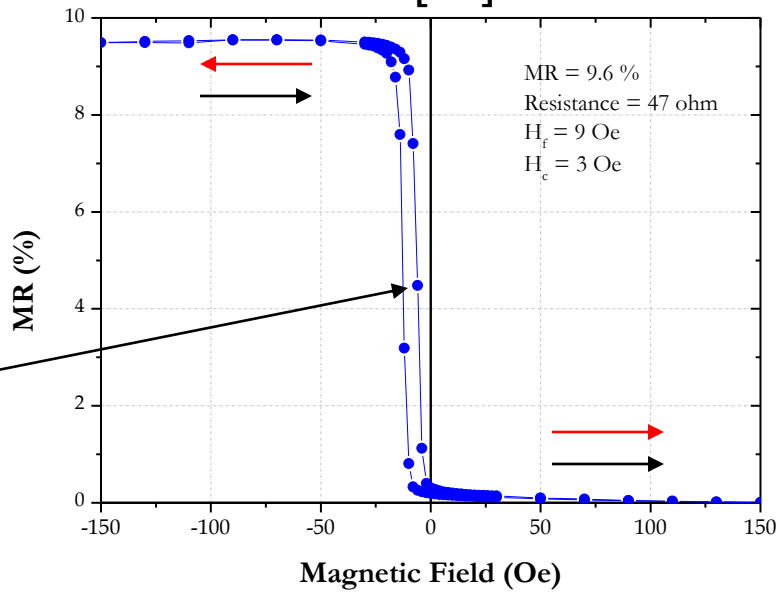
J.L.Leal, N.J.Oliveira, L.Rodrigues, A.T.Sousa, and P.P.Freitas, IEEE Trans.Magn., 30, 3031(1994).

Spin Valve sensors-magnetic response



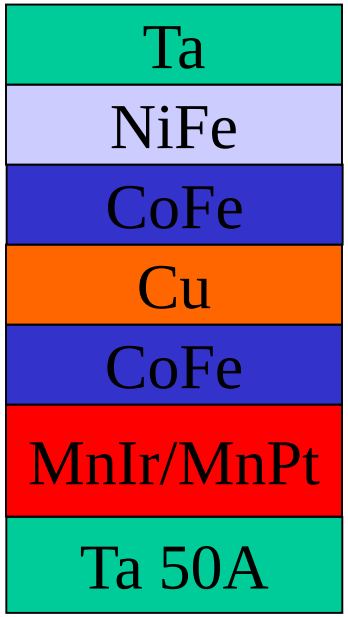


Neel Coupling



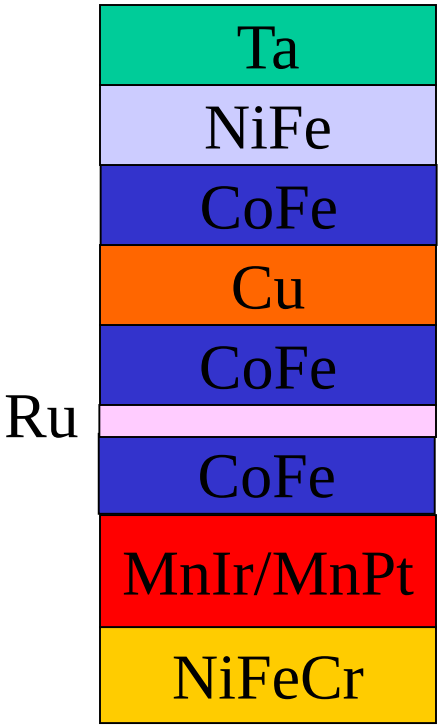
SV materials

Basic stack(94-97)



8%<MR<10%
Hc < 2Oe
Hf < 10 Oe
Hex > 600 Oe(MnIr)
> 1000 Oe (MnPt)

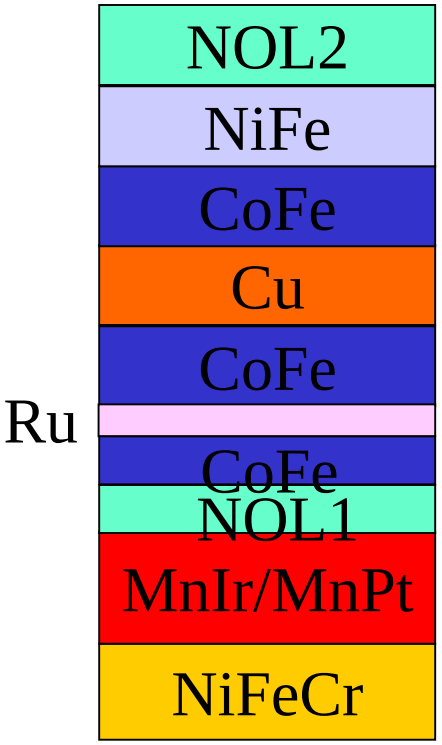
SAF+NiFeCr(97-02)



10%<MR<15%

Hex > 3000 Oe

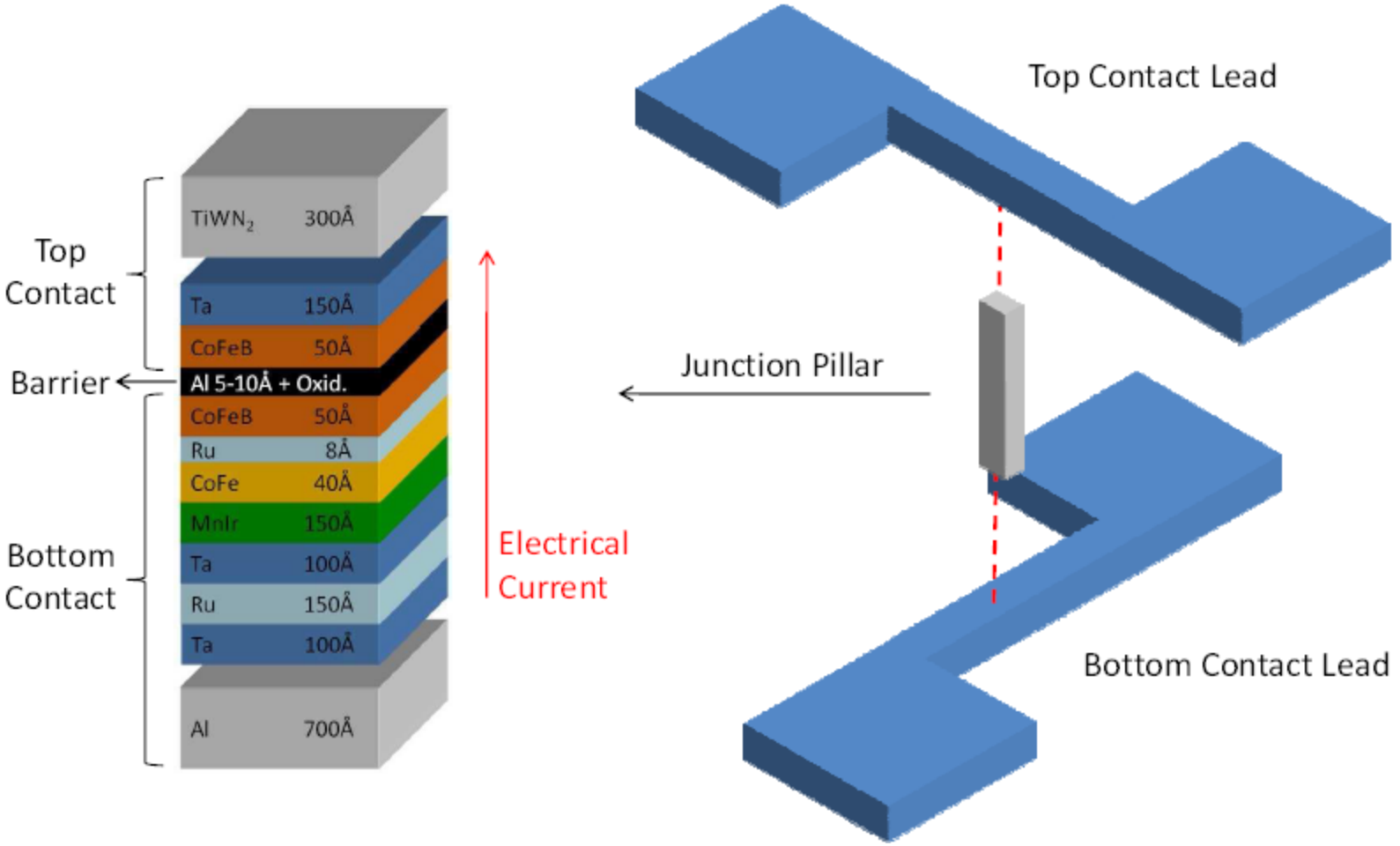
Specular SAF(01-)



14%<MR<16(20)%

ANNEX 3: MR DEVICE MICROFABRICATION PROCES

Current-perpendicular-to-plane (CPP) device fabrication

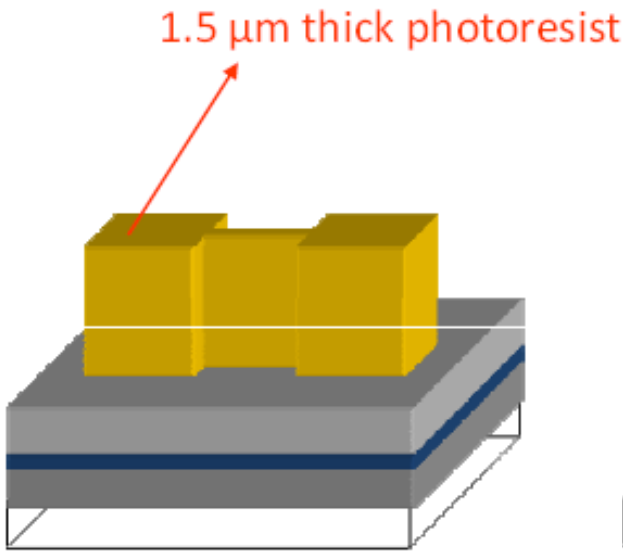


Microfabrication process

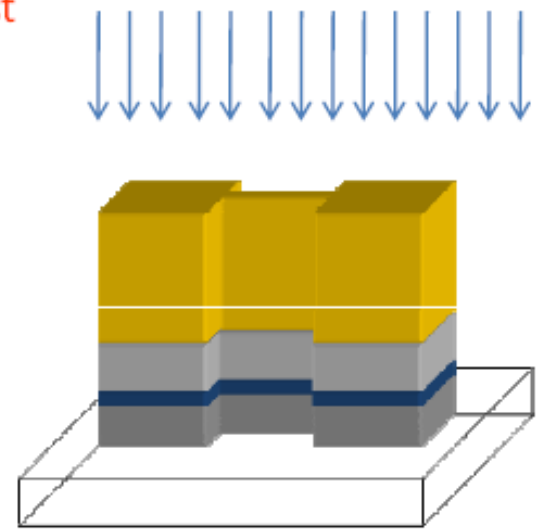
1) Deposition of the MTJ Stack

The complete stack is ~1800Å thick

2) 1st. Lithography



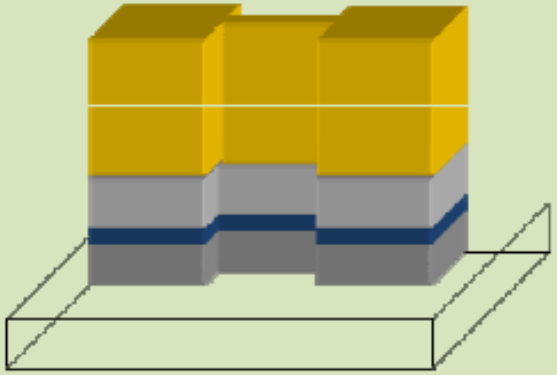
3) Ion Beam Milling



Stop point is signaled by the transparency of the substrate

4) Resist Strip

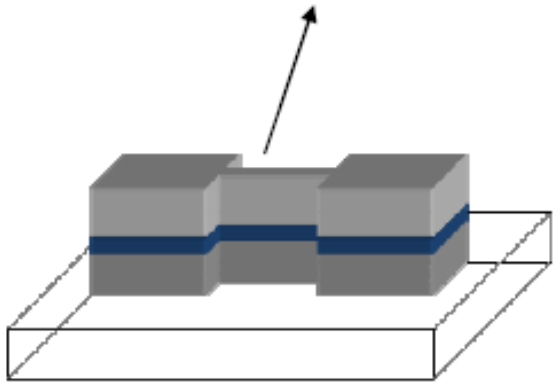
Resist Striper



Microstrip 2001 (Fuji) is used to remove the remaining photo-resist

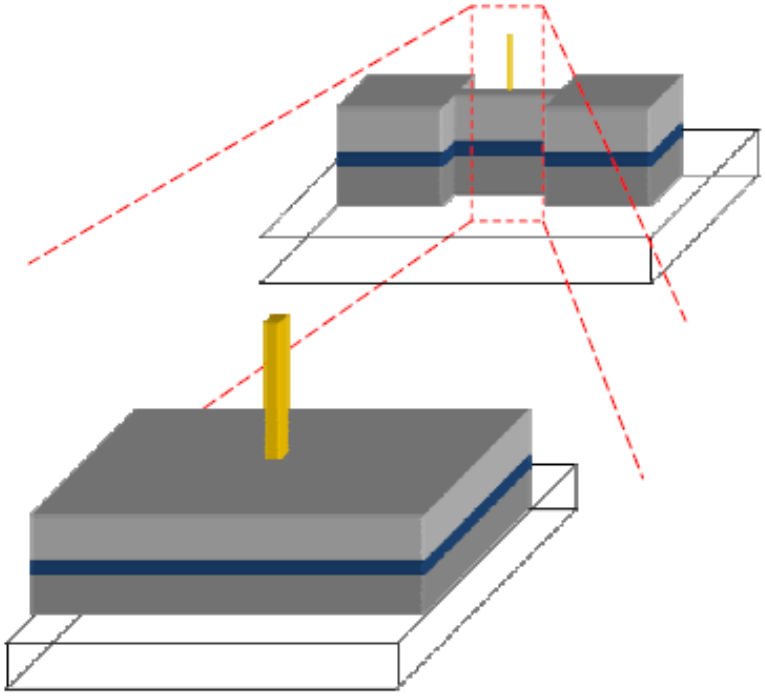
~2 hours in a hot bath (65° C) + Ultrasounds

750µm x 50µm



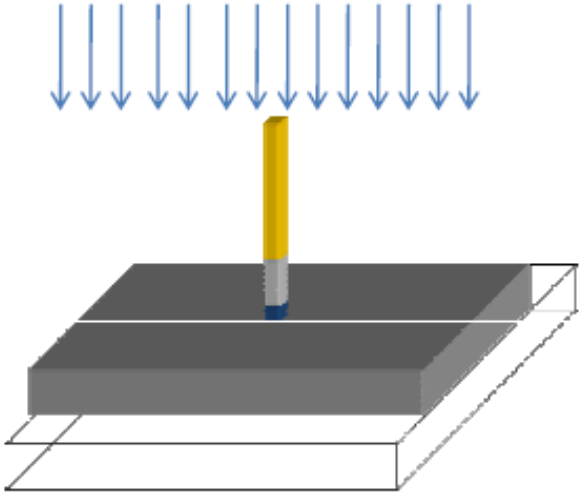
At this point, the shape of what will become the bottom contact lead is defined.

5) 2nd. Lithography : Junction Pillar Definition

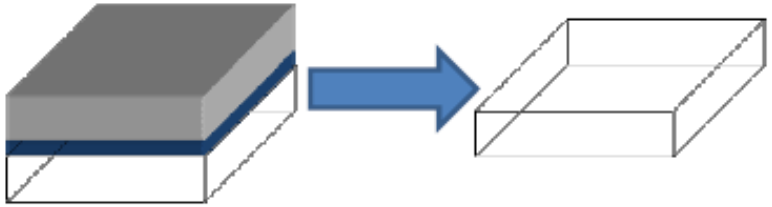


Minimum Junction Area : 1x1 µm²

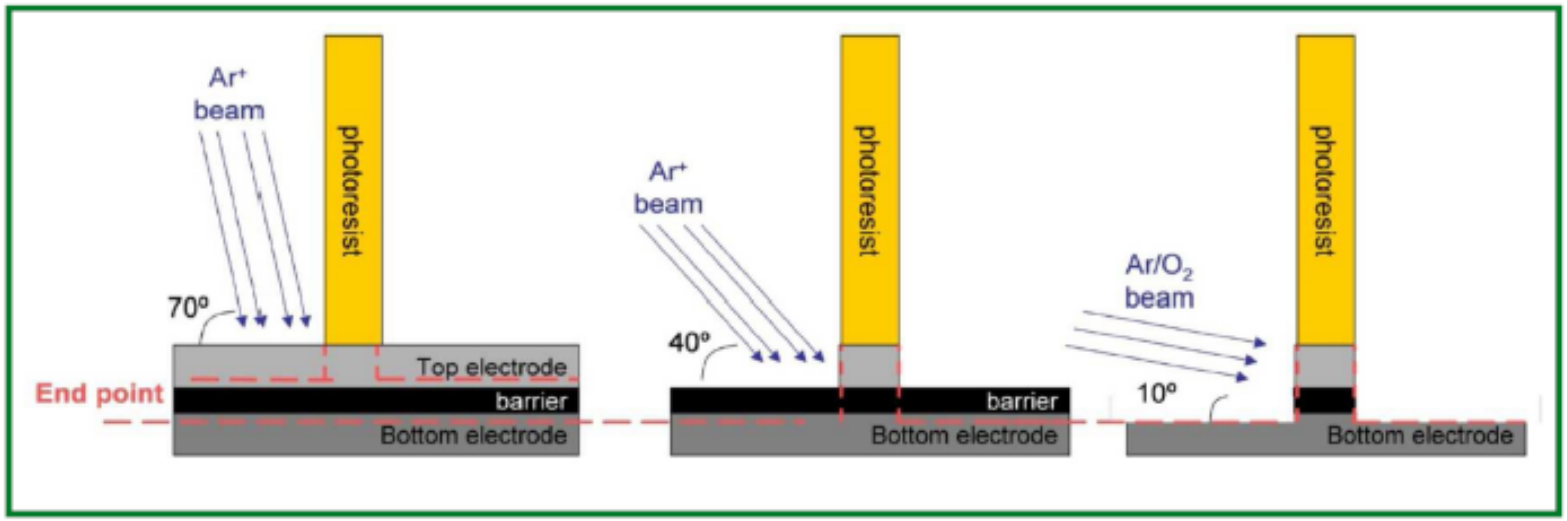
6) Junction Pillar Etching



Stop point must be after the barrier and before the substrate. Calibration samples are used to monitor the etching stop point.



6) Junction Pillar Etching



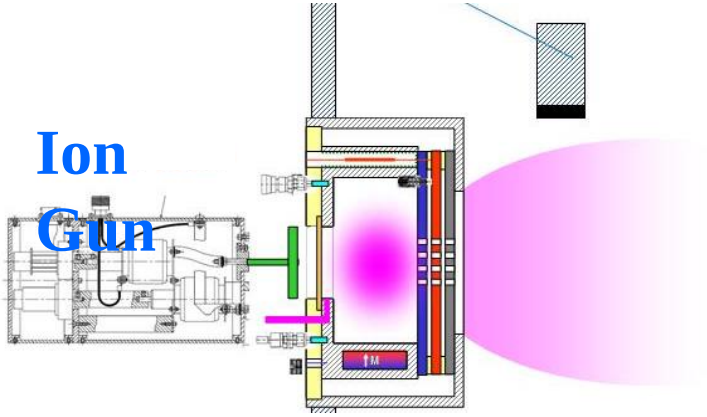
Early Etching Stage :
Large incident angle reduces shadow effects, but results in heavy redeposition

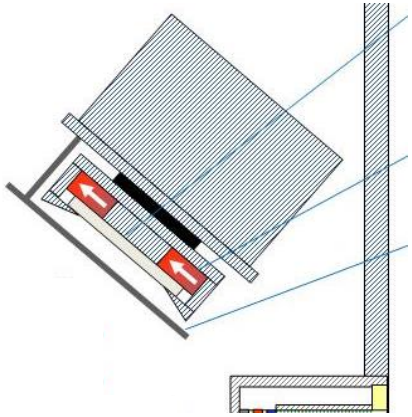
At the level of the barrier:
Shallow incident angle increases the etching in the sidewalls of the pillar, reducing the amount of redeposited material

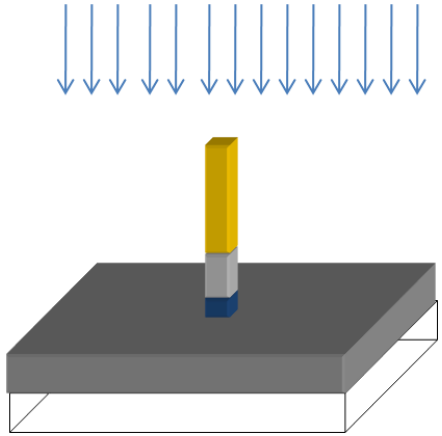
Final oxidation step:
Any material deposited in the sidewalls of the junction is oxidized, becoming an insulator.

Critical Step #1 : Ion Milling of a NanoPillar

Etch Stop Point Detection







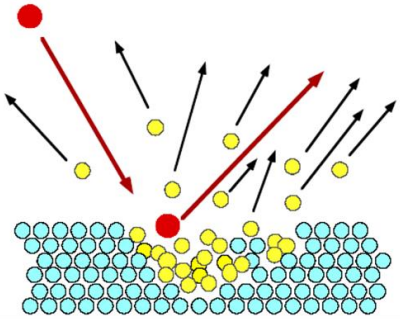
Key Parameters :

RF Power : 190W

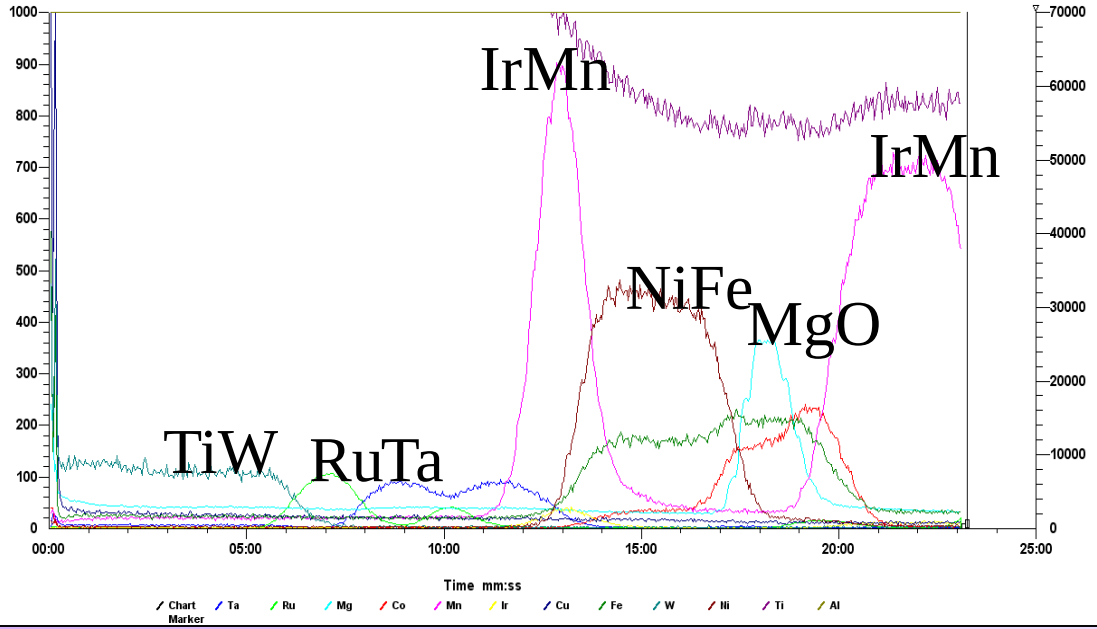
V+: +150V

V- : -1500V

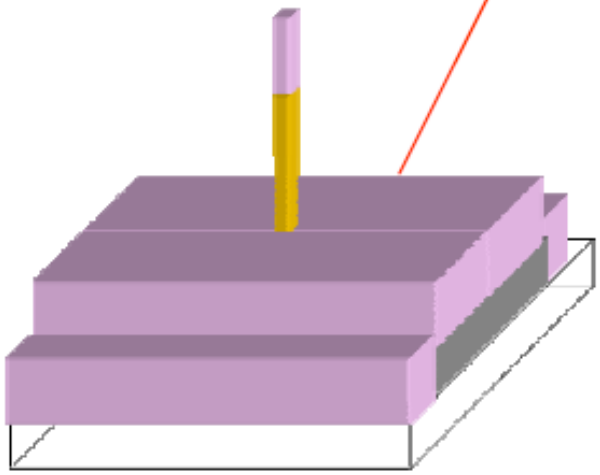
I+ : 184mA



TiW	15nm
Ru	7nm
Ta	10nm
Ru	7nm
Ta	10nm
IrMn	5.5 nm
NiFe	16 nm
Ta	0.21nm
CoFeB	2.6nm
MgO	~ 1.2nm
CoFeB	2.6nm
Ru	0.85nm
CoFe	2.0nm
IrMn	20 nm
Ru	5nm
Ta	5nm
Ru	15nm
Ta	5nm
Ru	15nm
Ta	5nm



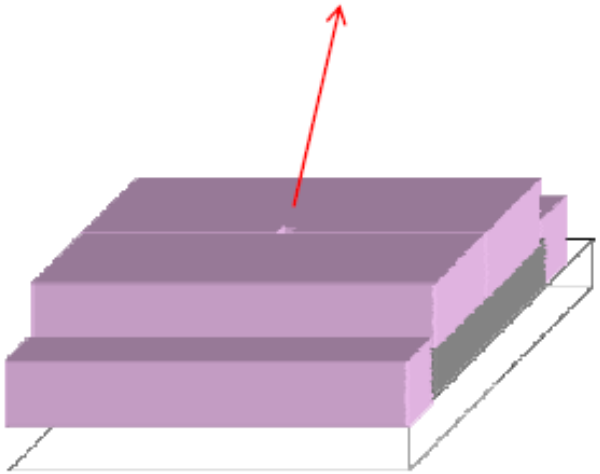
7) Insulating Oxide



Al_2O_3 or SiO_2
(>800Å)

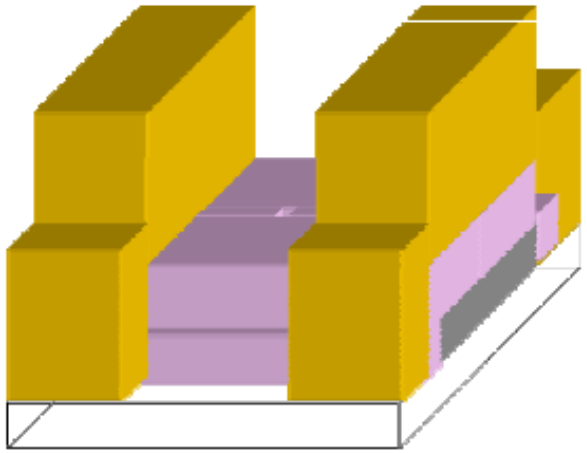
~1 day in a hot bath
(65° C) + Ultrasounds

Only connection to the top electrode



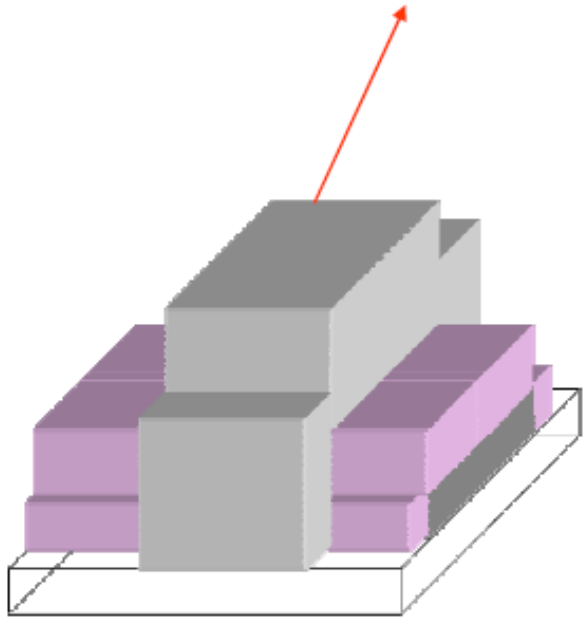
Via opening can be seen visually in an optical microscope.

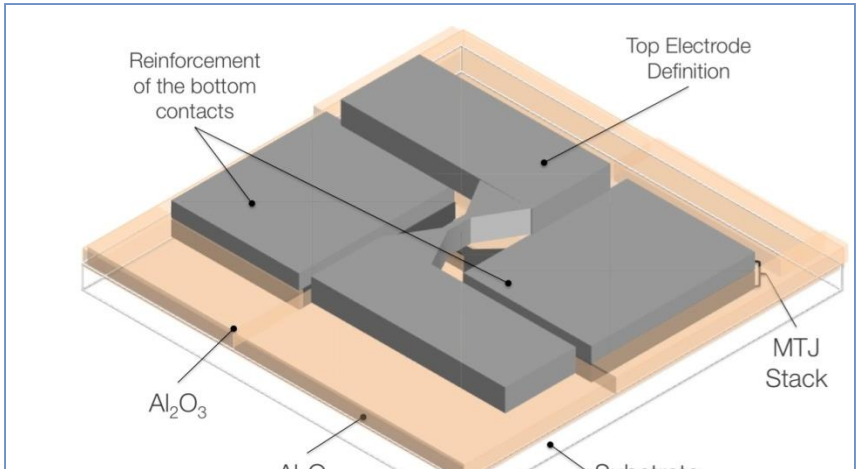
9) 3rd. Lithography : Top Lead Definition



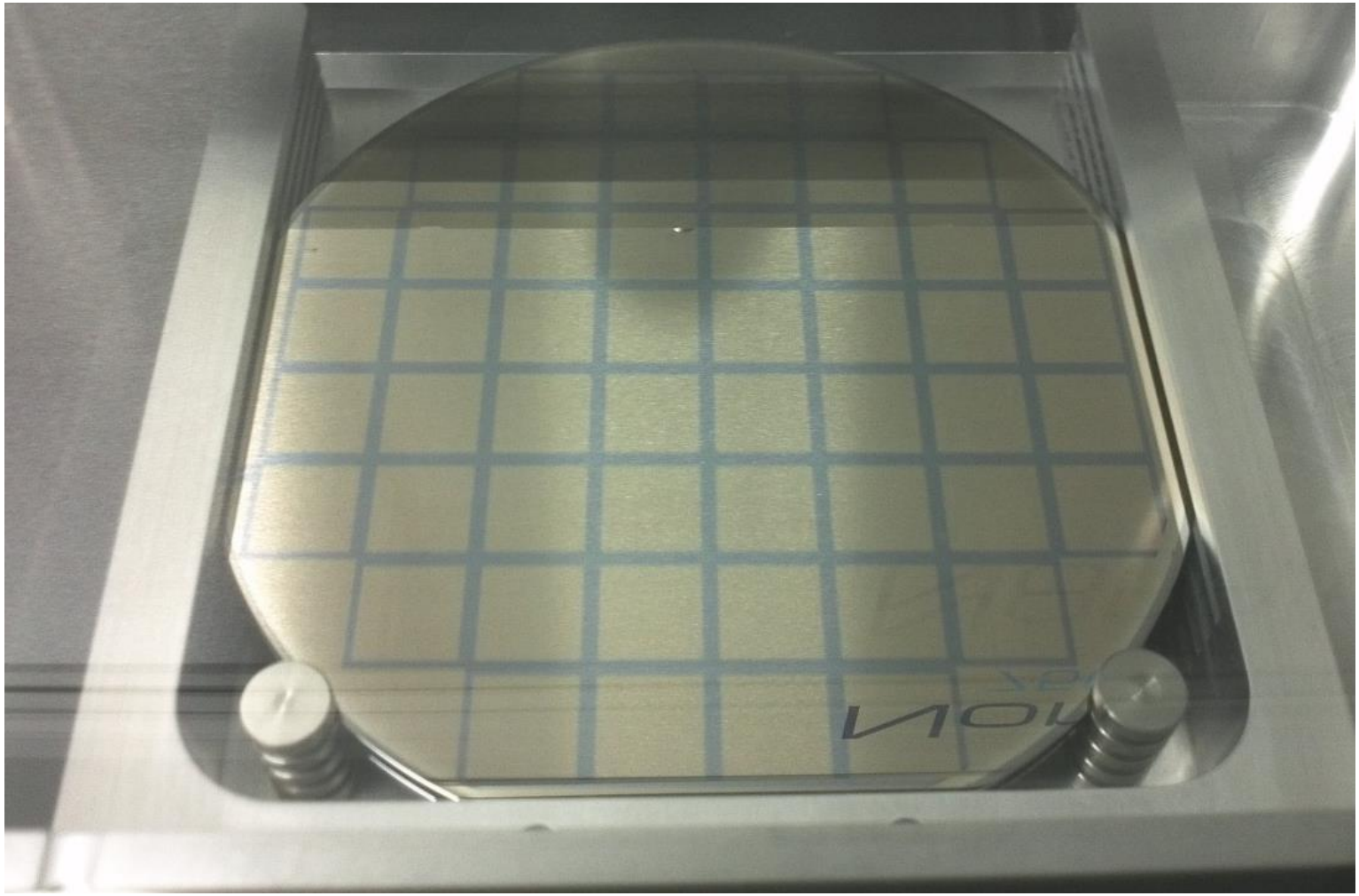
10) Top Lead Definition : Metal deposition + Lift-off

Al (3000Å) + $TiWN_2$ (150Å)





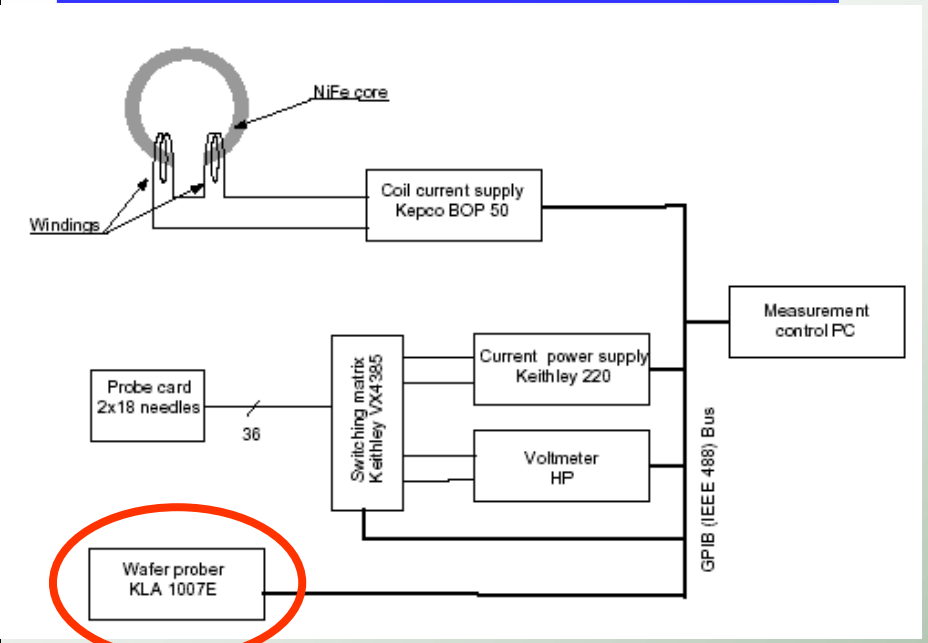
Up to 70,000 sensor devices in a 200mm diameter wafer



Tunnel Junctions Characterization
Automatic Measurement of Transport Properties

INESC-MN

Automatic Measurement Setup



Fully Automatic Measurement of magneto-transport properties :

- Resistance
- Magnetoresistance Transfer Curve
- Current-Voltage Characteristic
- MR Bias Voltage Dependence
- Breakdown Voltage
- Current Induced Switching

Integrated Data Analysis Software

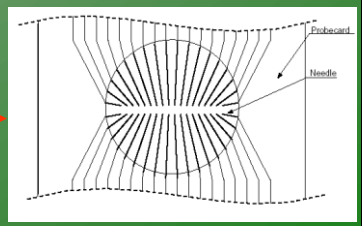
6" Wafers measurement capability (2 or 4 contacts)

KLA 3700

Probe Station

Probe Card

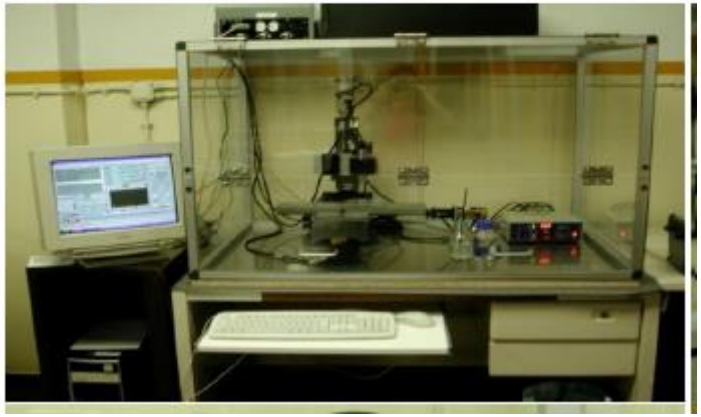
36 Kelvin Needles



ANNEX 4 Microfluidic PDMS processing

Microfluidic process development:

- Macro mold (PMMA)

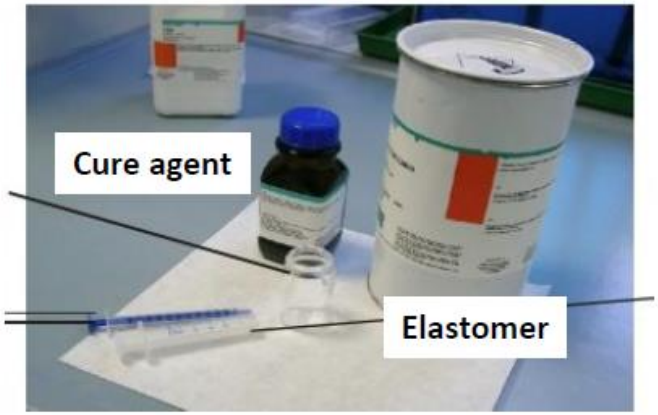


Milling machine

Consumables

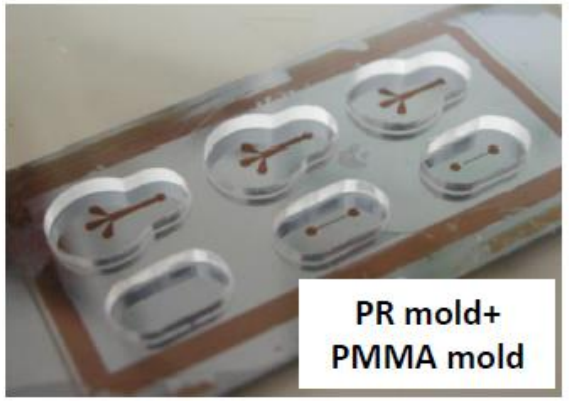
- PDMS mold

Mixture 1:10 ratio



Degasification step

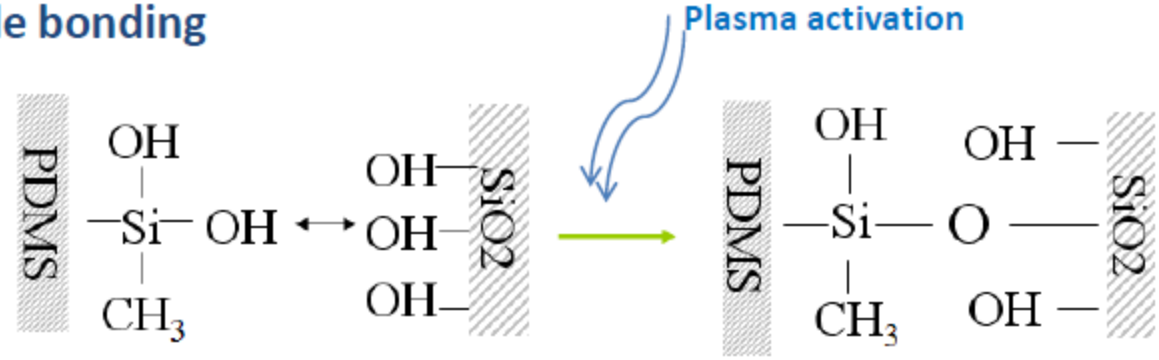
Injection into the mold



Irreversible surface bonding - II

SiO2 - PDMS

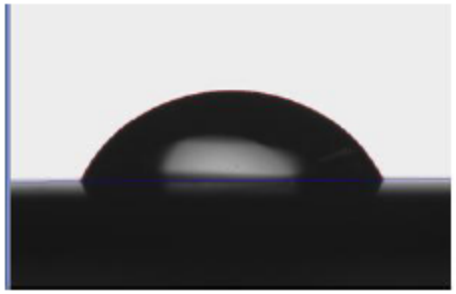
- Irreversible bonding



Pre-cleaning protocols (SiO2)

Machine	Plasma Conditions	Time	Contact Angle
LAM Rainbow	408 mW/cm², 275mTorr, 20 sccm O_2 (oxygen plasma)	150 seconds	<5º
UVO cleaner	28mW/cm², 5mm separation from the UV light (ozone plasma)	30min+2min exhaustion step	<5º

Note: the contact angle for SiO2 not cleaned is 72.2 ± 0.20



UVO Cleaner, Jelight