Test of BIOMIC optoelectronic chips V1 for diagnostic application

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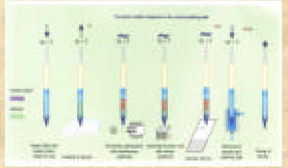
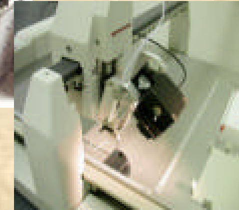
The project **BIOMIC (A BIOANALYTICAL MICROSYSTEM BASED ON AN OPTICAL MICROCHIP)** foresees the fabrication of monolithic optoelectronical circuits using standard silicon technology. This chips are functionalized with protein and DNA recognition elements, and integrated in a system consisting of mechanical microfluidic modules and electronic components. The complete microsystem is tested against specific multianalyte protein and DNA assays.

The choice of the application of the chip for the diagnosis of **Acute Myocardial Infarction (AMI)** has been done since cardiovascular diseases are the most lethal diseases in western world and the interest in developing fast and sensitive tests based on specific markers is very high. At this stage of the project some first version (V1) chips have been tested for the detection of myoglobin.

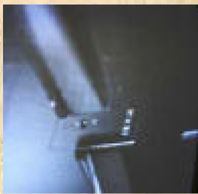
Myoglobin is expressed in cardiac and skeletal muscle so it is not cardiac-specific, but it appears in patients blood 1-3 hours after the onset of sympoms (peak within 8-12h) so it is the earliest marker and its predictive value in case of negative result is 100%.





The nanoplotter station: GeSim NP12

The deposition of the protein recognition molecules on the optoelectronic chip fibers has been performed by means of a nanoplotter of the german firm GeSim.The instrument is equipped with a *nanotip* and a *picotip* that are micromachined silicon/glass micropipettes able to dispense droplets of about 0.4nl and 0.1nl, respectively. The operating principles of the pipettes is based on a piezoelectric element. Droplet size is influenced by both sample properties (viscosity, density and surface tension) and pipet parameters. A stroboscopic light and a videocamera provide an image of the droplets and allow the optimisation of the pipette parameters.



Deposition on V1 chips

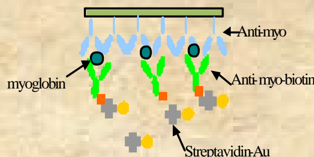
Fine positioning of the pipetting head and continuous visual control of the deposition have been achieved with a micro videocamera mounted on the nanoplotter head. A program for the nanoplotter has been written which will begin successive fibers alternatively from left to right and viceversa in order to avoid coalescence of the initial large part of the deposition.

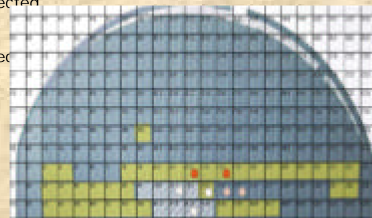
Patterning of fibers

300µm antimyoglobin signal drop expected
500µm unrelated antibody negative control
700µm antimyoglobin signal drop expec
900µm unrelated antibody negative control

Myoglobin immunoassay scheme

The detection assay is based on a colloidal gold labelling which has been shown to produce a drop in the photocurrent measured across the chip fibers.





Practically all the wafer has been deposited but, after microscopy inspection, the best deposited chips have been identified and they are highlighted in yellow in the picture (some other good ones are highlighted in dashed grey, while the remaining ones had some defects).

The red, pink and white spots identify the packaged chips that have been sent to TB for the measurements and are repoted in the table below.

Packaged V1 chip with the microfluidic module realized in St Ingbert (IBMT)

The microfluidic channels have been connected to a peristaltic pump with Tygon tubings of internal diameter 0.25 mm; the liquid flow rate was about 20 µl per minute under suction.

Steps after packaging:

1. Measurement of initial photocurrent with the electronic control board
2. Myoglobin solution (10′ in flowing and 20′ in static condition, followed by wash 10′ in flow)
3. Biotinylated antimyoglobin (then wash)
4. Streptavidin colloidal gold conjugated (then wash)
5. Chips have then been dried in vacuum for 15′ and the final photocurrent has been measured with the electronic control board

chip no.	fiber µm	leakage	delta	delta after reaction	drop %
	900	ns	ns	no fluidic	
	700	658	1479	no fluidic	
J13	500	347	1434	no fluidic	
	900	411	ns	ns	
	700	ns	ns	ns	
J14	500	329	2320	2603	12.20
	900	247	ns	584	
	700	320	539	584	8.35
J15	500	173	1078	1023	-5.10
	900	173	895	damaged	
	700	256	685	damaged	
J11	500	119	1342	damaged	
	900	256	1242	1242	0.00
	700	320	1224	749	-38.81
I12	500	201	2155	1991	-7.61
	900	320	ns	ns	
	700	173	1105	827	-25.16
I14	500	265	1279	1300	1.64
	900	247	356	ns	
	700	310	566	ns	
K12	500	164	749	ns	

Good results came from devices I12 and I14, they had good electrical contact for functionalized 700µm fibre that showed a good signal drop, as reported in the table. These two chips are marked above with a red spot and it must be noted that they were previously identified as good deposited ones (highlighted in yellow). The chips marked with a white spot could not be measured for fluidic problems (J13 and J11) or electronic problems (K12 had too low initial photocurrent). The chips marked with a pink spot (J14 and J15) were measured but did not give interesting results in terms of signal drop as could be expected, since they had previously been identified as not good at the deposition step.

The results presented here have demonstrated the complete functioning of the chip in principle. The V2 chips will contain 9 fibers and will be used to test three different cardiac markers; myoglobin, troponin complex and creatine kinase MB.

Acknowledgements

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These results are the sum of a long chain of steps which have taken place at the different partner laboratories. So we greatly acknowledge the work of all BIOMIC partner and take the opportunity to thank all of them for the pleasant and fruitful collaboration.