MICROSCOPIC ENCLOSURES MODELLING DESIGNATED FOR BIOMEDICAL APPLICATIONS OF CELLULAR MANIPULATION TYPE IN LIQUID ENVIRONMENT

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Abstract: *Cellular manipulation and single cell manipulation especially, created a strong focalization of biomedical researching world around of this direction. Today we can find out a great diversity of manipulation methods, but our researching collective conceived an innovative one, that involved a special designing session for obtaining an optimal enclosure. The manufacturing process needs a simple, robust and economical technology. In this work we present a technical solution that includes all remembered attributes. Finally we present the experimental model achieved following the concept idea.*

Keywords: *bio-manipulation, cell, enclosure, fabrication technology*

1. INTRODUCTION

Many types of technical system are used within micromanipulation domain, materializing different physical principles such as magnetic or electric field, or mechanical or optical techniques, MEMS [1]. The biomedical laboratories use these kind of systems for developing complex studies regarding cells communication and migration, or other localized phenomena. In present, using the above remembered principles and techniques, are fabricated and validated a lot of robotic systems that have the capacity to execute in automatic way cell manipulation [2].

Related to the bio manipulation, it exists a specific preoccupation for separation of cells from a tissue, using unconventional methods like pressure jet into working micro fluidic networks [3,4].

Biological liquid environments are indispensable in cell manipulation systems. They need by a adequate enclosures. If the capillarity is a part of the system, the specific dimensions of different areas of the enclosures will be small and very small. In the present case it was conceived a particular enclosure, designed for storing a microlitric liquid quantity.

To obtaining this enclosure we put in scene an economical method that need just two main elements: ultrathin metallic sheets and laser ablation developed with high precision equipment. The work accent was centered on the technologic fabrication method.

2. LASER ABLATION

The technologic equipment designed for cutting of ultrathin metallic sheets is "Laser ablation system". In the below text are presented the technical details regarding this kind of system and that refers to main specific parameters for this equipment (Figure 1). The system is so built that the optics, subsystems of control and displacements are integrated into a special housing, with granite table, anti-vibration subsystem and laser protection class 1. This system has incorporated a laser type Diode Pumped Solid State (DPSS). This type of laser is invisible, able to execute bores and cutting in thin materials like metals, ceramic and semiconductors. For a good contour tracking the system has a numerical control for axes of displacement on X, Y and Z.

Fig. 1. Laser ablation system

Laser specifications:
- laser DPSS with emission in 532 nm;

- laser power is 8 W;
- the system generates pulses with duration in the range 5-50 ns. This duration can be constant or programmable in the remembered range;
- The frequency of pulses train in the range 1-150 kHz;
- The stability of the pulse energy is less than 3%;
- The maximum divergence of fascicle less than 2 mrad;
- Motorized axes for X, Y and Z.

In the table 1 are shown the basic technical characteristics of the table positioning subsystem related to laser beam:

Table 1 - Accuracy of XYZ positioning subsystem

Axis	Z	X	Y
Displacement (mm)	100	200	200
Resolution (μm)	0.5	0.25	0.25
Repeatability (μm)	±1	±0.75	±0.75
Accuracy (μm)	±6	±2	±2
Max. speed (mm/s)	100	250	250

3. METALLIC SHEETS

In commercial space we regularly find industrial products presented in sheet form with biocompatibility feature and a very small thickness (less than 1 millimeter). In the present case, we considered a stainless steel material as 316L. In the next presentation are shown few important characteristics of this biocompatible material. A spectra-chemical test (using a method conforms to STAT 11464-80 standard) revealed for sample (taken conform to SR EN SO 14284-2003) the next results (Table 2):

Table 2 - Spectra-chemical results for 316L

Sample	Results of spectra-chemical analyze (%)	
Stainless steel 316L	C	0.036
	Mn	1.00
	Si	0.38
	S	0.002
	P	0.036
	Cr	16.45
	Ni	10.95
	Cu	0.38
	Mo	2.09
	V	0.060
	Ti	<0.01
	Co	0.16

4. THE CONCEPT OF ENCLOSURE

The metallic sheets are characterized by a constant thickness and that permits to make a very simple calculus for volume of enclosure. To obtain an enclosure reduces itself to one thing: defining of enclosure surface. A single enclosures supposes at least to layers materialized through cutting of sheets. This cutting of sheets must be made conform with the need of the application and this supposes an precise equation to define this geometrical cutting way. In the figure 2 we see the cavity obtained through cutting of sheet.

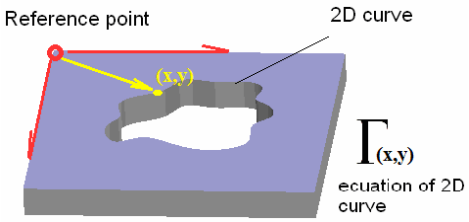

Fig. 2. A cavity obtained by cutting of sheet

The curve Γ(x,y) supposes a modeling what creates an adaptation and matching with the needs of technical application that will includes this kind of piece. Having the mathematical definition of this curve, it can be easily adapt this geometry to laser equipment for automated cutting. The laser spot will describe the curve Γ(x,y) and in this way it will create the effective cutting. In the present case it was imposed the obtaining of microlitric volume for fabricated the enclosure. The used sheet had 200 micrometers thickness.

4. DEFINING OF ENCLOSURE DIMENSIONS, 3D DESING AND NUMBER OF LAYERS

A first request for the enclosure features it imposed to fabricate a channel with a given width, respectively 50 micrometers. The idea of channel, it limits the minimum number of layers to three (base, channel, cover).

We established the next definition dimensions for enclosure:

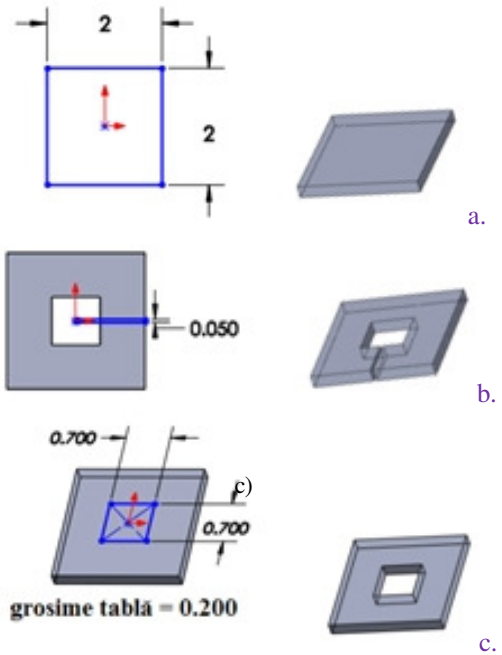

Fig. 3. The design for enclosure layers

- The entire box that includes the enclosure: 2 mm x 2 mm x 0.6 mm;
- Enclosure: 0.7 mm x 0.7 mm x 0.4 mm;

In figure 3 are shown the three layers: a) the basic layer; b) the middle layer that contains the channel; c) the cover layer.
The square section was chosen, and the final obtaining of enclosure supposes an overlap of those three layers

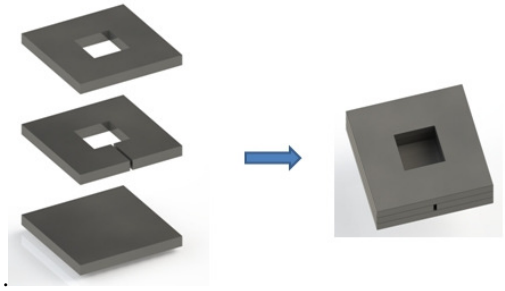

Fig. 4. The virtual assembly of the enclosure

Observe that it was obtained o square section enclosure and a channel designated for outside communication in the working system.

5. FABRICATION OF LAYER-ELEMENTS AND THE ASSEMBLY OF ENCLOSURE

The element was fabricated using the described above laser equipment. This equipment belongs to Institute of Multidisciplinary and Technological Scientific Research, Laboratory C04 "Physical and structural characterization of matter". In figure 5 is shown a capture of equipment screen, during the cutting process of one component (one layer).

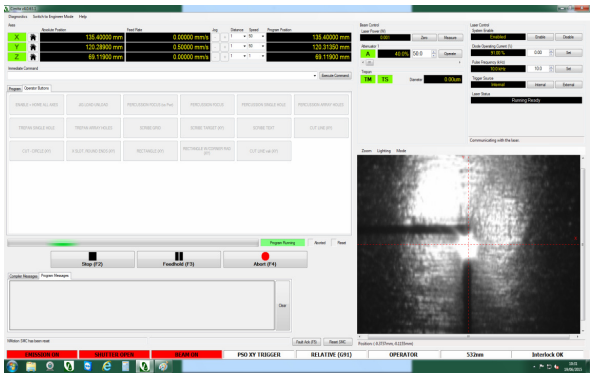

Fig. 5. The laser cutting process

After cutting processes were obtained the three layer elements shown in figure 6.

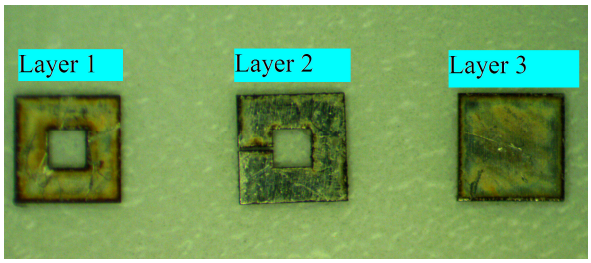

Fig. 6. Layer-elements

The micro assembly process was developed on MARYLIN-F131B microscopic system, with 45 zoom (figure 7), in the Laboratory 22 of the same institute.

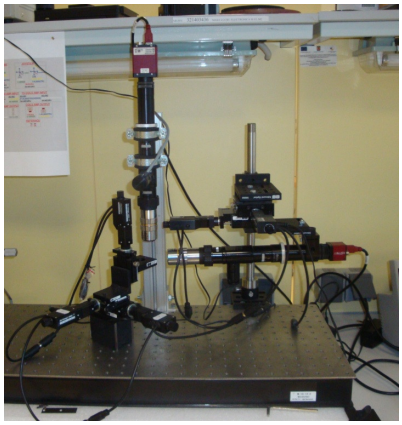

Fig. 7. MARYLIN-F131B microscopic system

The final result of micro assembly process is presented in figure 8.

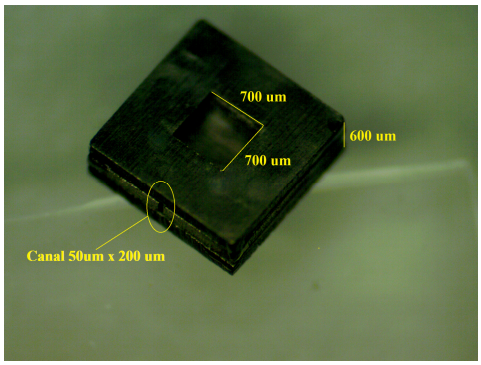

Fig. 8. The microlitric enclosure

6. CONCLUSIONS

The engineering of microsystems is founded on traditional principles of engineering and the research in micro-engineering includes the designing, fabrication of components and manipulation on micro scale of involved objects. The designed enclosure can be included in domain of manipulation and nondistructive insulation of cells, helping to be built a complex system dedicated for sorting cells, without changes of biological morfology. The sorting process can be achieved using microrobots and microfluidic network with very small chanels that assure the specific of sorting migration.The enclosure represents a first step into conceiving of complex sorting systems for biological cells.

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