Supplementary Material (ESI) for Lab on a Chip


# ELECTRONIC SUPPLEMENTARY INFORMATION

## Microfluidic perfusion system for culturing and imaging yeast cell microarrays and rapidly exchanging media

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### FEM modeling

3D numerical simulation werre performed based on a finite element method using a commercial software (COMSOL Multiphysics 3.5a, USA). A computer (Dell Xeon) with 64 bit CPU and 26 GB RAM was used. The mass transfer was computed by solving a time dependent convection diffusion equation on the geometry represented in Fig.3a which represents a small portion of the setup yet is representative as diffusion only occurs in a vertical direction, and as the high flow resistance of the pores preclude flow across the membrane. The area used for the model is $4.35 \times 2.175\ \mu m^2$ . A series of holes 220 nm in diameter accounting for a porosity of 20% which is the maximum porosity for track-etched membrane and a thickness of 20 μm. The system was thus represented by one reservoir at the top that was 10 um high, and a reservoir at the bottom which was 3 μm deep, reflecting the gap within which the yeast cells are trapped. The diffusion coefficient was chosen as $D = 314\ \mu m^2/sec$. Two different conditions were calculated, (i) the diffusion of the chemical from top reservoir to the bottom reservoir and (ii) the diffusion of the chemical out of the bottom reservoir to the top one which is remaining empty. For (i) the initial concentration in the system is C = 0, and the boundary condition at the walls of the upper reservoir were $C = 1\ mol/m^3$ reflecting the fact that the large volume and flushing continuously replenish the volume continuously, whereas a symmetry condition was used for the bottom reservoir, *i.e.* the walls are impermeable representing the condition where the chemical diffuse simultaneously across the entire membrane. For part (ii) the initial condition in the reservoirs is $C = 1\ mol/m^3$ and symmetry conditions are used for the bottom reservoir, whereas for the top reservoir the boundary conditions are set to C = 0 reflecting the fact that the chemicals are quickly flushed away by the flow.

**Video S1.** This video shows the application and exchange of a series of blue and yellow dyes to a perfusion system made of the bottom and top coverslip that sandwich a mesh. On the right, a paper is contacting the mesh which protrudes beyond the coverslips, and wicks the liquid away. On the left, the mesh slightly protrudes, which allows distributing the liquid at the inlet. When the liquid is entirely drained from the inlet, the sandwiched mesh acts as a CRV, and prevents drainage of the fluid from the gap.

**Video S2.** Video made from images captured using a spinning-disk confocal microscope of yeast cells trapped in the microfluidic flow chamber.

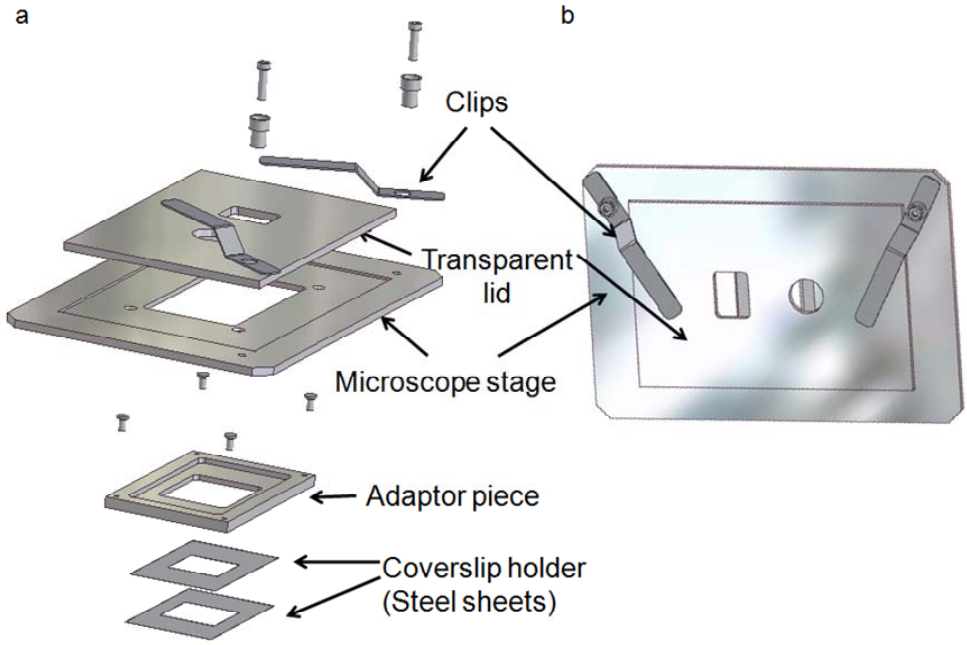


**Fig. S1.** (a) Exploded view of the microfluidic perfusion system and (b) top view with the sample inlet (right) and outlet (left).

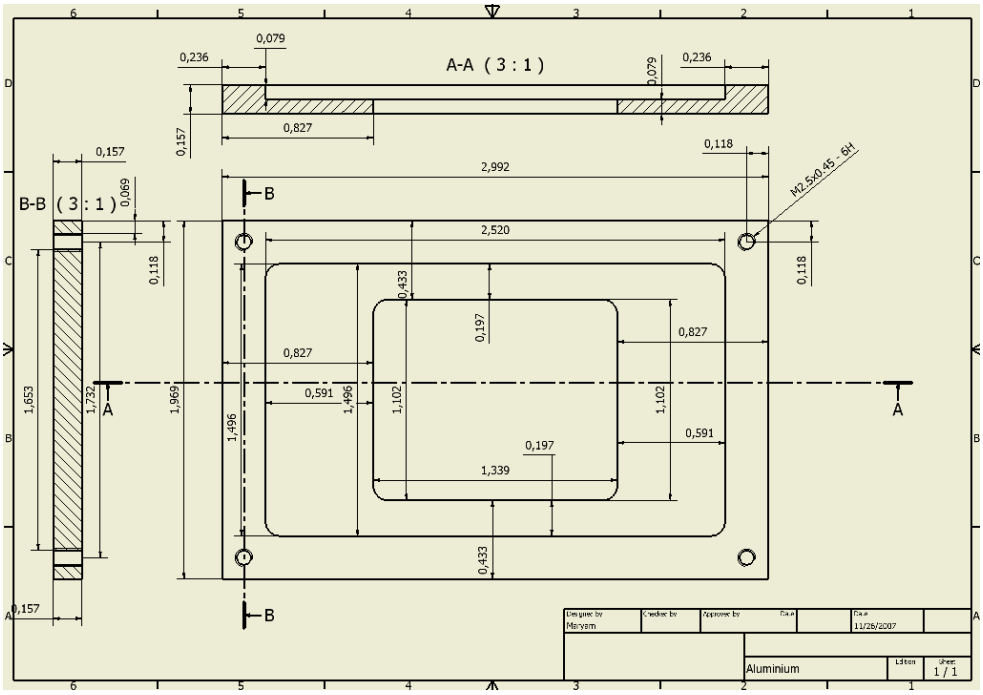


**Fig. S2.** Technical drawings of the Adaptor piece using the Autodesk program. Units are in inches.

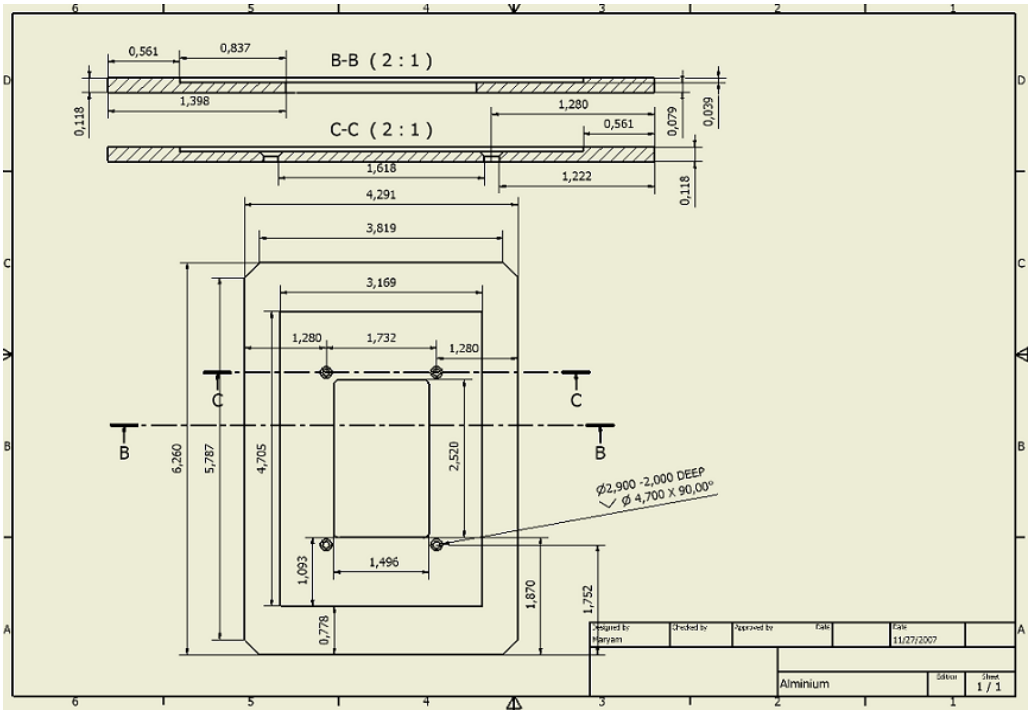


**Fig. S3.** Technical drawings of the Microscope stage insert using the Autodesk program. Units are in inches.

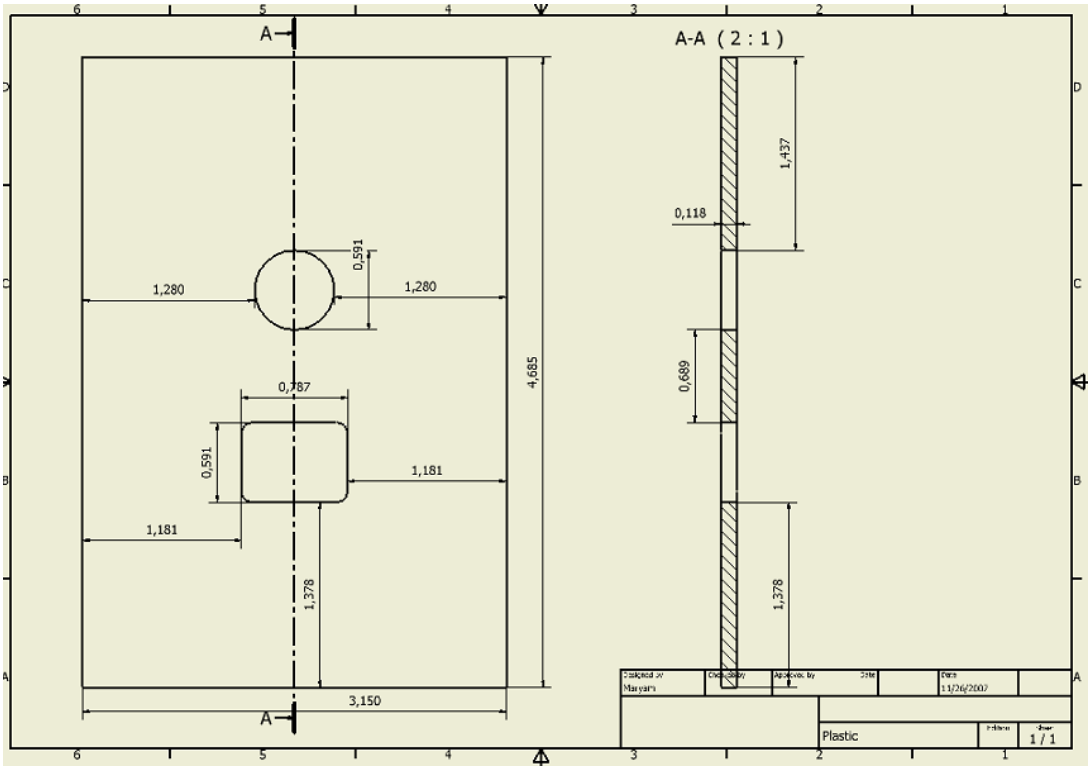


**Fig. S4.** Technical drawings of the Transparent lid using the Autodesk program. Units are in inches.

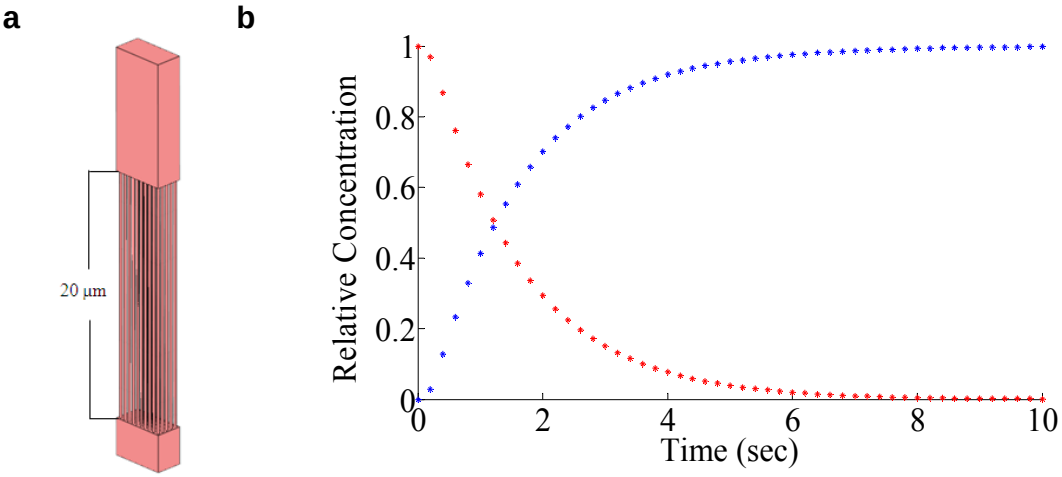


**Fig. S5**. Simulation of the diffusion of chemicals across a small section of the track-etched membrane. (a) Schematic of the track etched membrane with 20 µm long vertical holes, each 220 nm in diameter that collectively make up 20% of the surface area. The area modeled here is 4.35 × 2.175 $\mu m^2$, and the depth at the bottom is 3 µm corresponding the gap between membrane and glass. A10 µm high reservoir at the top represents the flow conduit with the mesh. For the simulation, the bottom reservoir is considered closed reflecting the symmetry and the no flow condition, whereas the boundaries of the top reservoir are reflecting the set concentration, C = 1 when simulating the Lat-A diffusion from the reservoir to the gap, and C = 0 for the diffusion of Lat-A out of the gap. (b) Relative concentration profiles at the center point of the 3 µm gap following addition of Lat-A to the top reservoir (blue diamonds) and after flushing of the top reservoir and diffusion of Lat-A out of the gap (red diamonds). A relative concentration of 0.5 is reached after less than 2 s in both cases, and saturation is visible after a few seconds only.

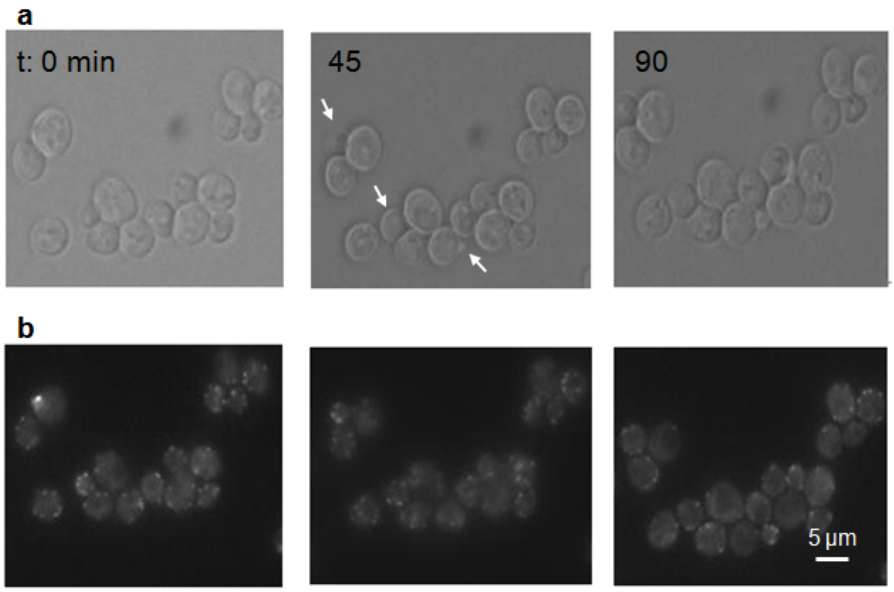


**Fig. S6.** Viability of printed Sac6-EGFP yeast cells assessed by time course microscopy of yeast cells after inkjet spotting (a) Differential interference contrast image of the cells and (c) fluorescence images of the same cells revealing the actin patches and bud formation, showing that the cells are growing.

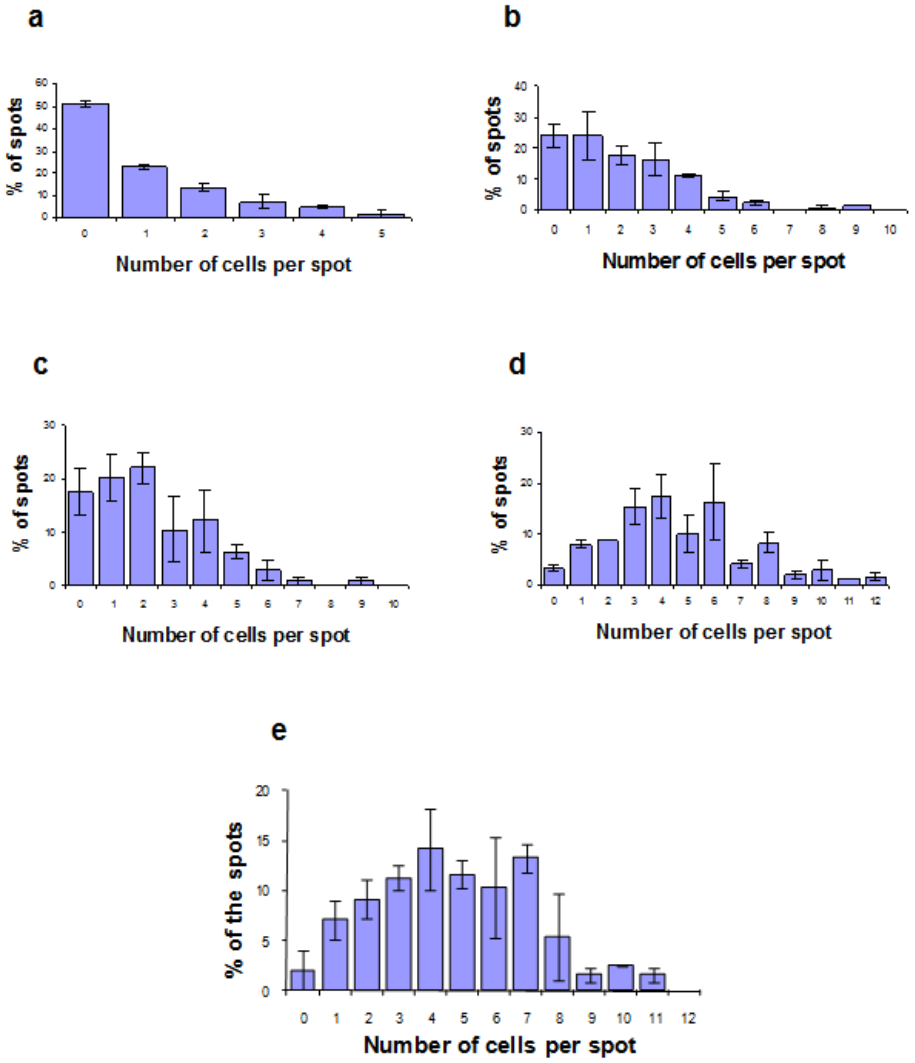


**Fig. S7.** The distribution of cells in the individual droplets are shown for five different densities. Optical Density of 0.2, 0.3, 0.4, 0.6 and 0.8 correspond to Fig. a,b,c,d and e respectively. ~ 200-400 pl of cell suspension was delivered. Error bars are SE.

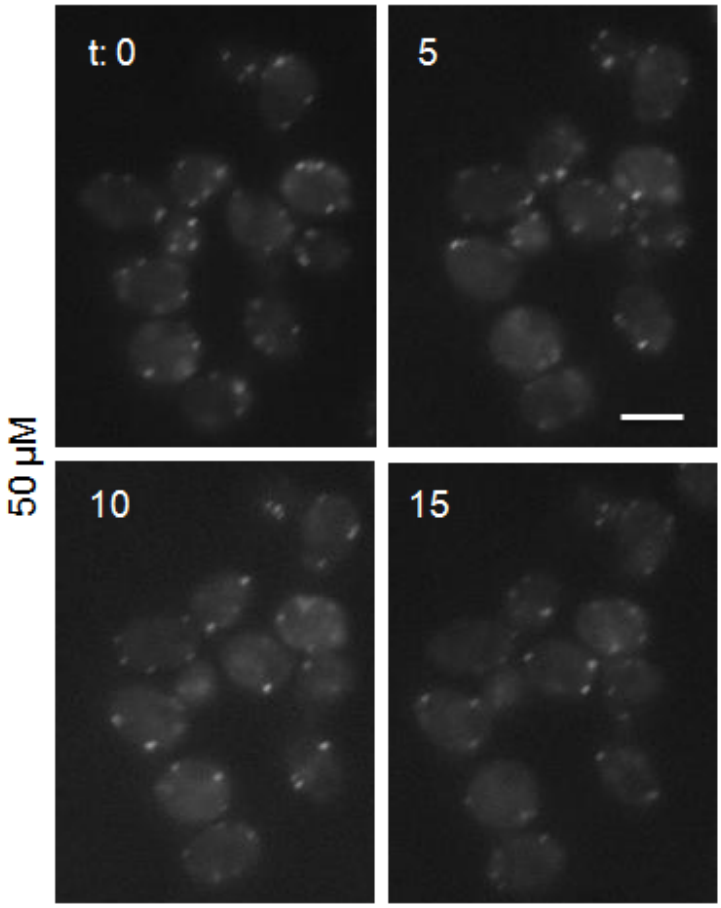


**Fig. S8.** Fluorescent micrographs of actin structures (arrows) in living yeast cells before, during and after exposure to 50 µM Lat-A. (a) Shows cells before (t:0), and after 5, 10 and 15 min of exposure to Lat-A. There is no significant difference visible on these images between cells exposed to 50 µM of Lat-A during 15 min. Scale bar is 5 µm.

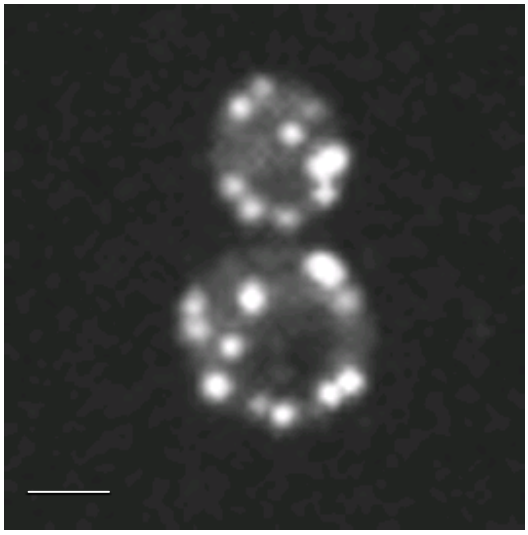

**Fig. S9.** Fluorescent micrograph of two yeast cells obtained using a spinning-disk confocal microscope with a 63× , 1.4 NA objective. This result confirms that the chamber may be adapted to a variety of different inverted microscopes, and illustrates the image quality that may be obtained in conjunction with the microfluidic flow chamber. See video S2 for more views. The scale bar is 2 μm.