Template: PASCA Application Table (including instructions to fill in the table in green)

PASCA Table: Title of experiment

Prepration of cell solution	SCM Protocol	Cell Stimulation Protocol	Incubation	Cell Analysis	Scientific Questions/ Observations
Describe the preparation of the cell solution to be supplied to the SCM. Specify at least: ◆ Buffer ◆ Cell type ◆ Concentration ◆ Labelling or staining protocols ◆ Environmental conditions, like temperature, humidity, sterile etc. ◆ Specific requirements	Describe how the cell solution should be manipulated in terms of sorting, detection system (optical and/or impedance), spatial distribution on the substrate, numbers of cells to be dispensed etc. Use drawings, if appropriate. Specify at least: ◆ Substrate ◆ Spatial cell arrangement ◆ # cells/spot ◆ Detection system	Describe how the cells should be stimulated after dispensing. Cite scientific references or product manuals, if appropriate Specify at least: ◆ Equipment (pipettes, transfection equipment etc.) ◆ Stimulation process ◆ Reagent or kits to be used ◆ Specific requirements	Describe how the cells should be incubated after stimulation and prior to cell analysis. Cite scientific references or product manuals, if appropriate Specify at least: ◆ Incubation time ◆ Environmental conditions ◆ Controls to be performed during incubation (e.g. inspection) ◆ Specific requirements	Describe how the cells should be analysed after incubation. Cite scientific references or product manuals, if appropriate Specify at least: ◆ Analysis method ◆ Equipment ◆ Preparation process, if any (e.g. labelling) ◆ Specific requirements	Describe which scientific questions should be answered by the experiment. Which results should be observed in particular? How can these be obtained? Cite scientific references, if appropriate

Author:	Peter Koltay (IMTEK)	Date:	11.2.2011	Revision:	1.1

PASCA Table Example: Single cell viability study for *S.Cerevisiae* / Azmi Yusof (IMTEK)

Prepration of cell solution	SCM Protocol	Cell Stimulation Protocol	Incubation	Cell Analysis	Scientific Questions/ Observations
- Buffer: physiologic saline solution - Cell type: Fresh & homogenous yeast suspension (size; 3 – 5 µm) - Cell's density: 10^4 to 10^6 cells/ml. - Staining: Aliquot 1 ml buffer with Trypan blue - Perform cell counting & statistical viability check with hemocytometer (100%)	- Substrate: Sterilized culture dish - Culture media: Yeast glucose agar with chloramphenicol (YGC), sterilized. - Feed 1 ml suspension into sterile reservoir of SCM - Add 1 µM fluorecein into suspension (1:1) - Print 10 x 5 spots with single cell per spot (pitch 500µm). Repeat 10 x - Use camera with fluores. filter for cell detection - Droplet volume: 100pl (piezo parameters: ds = 8 µm; s = 70 µm/ms ; us = 1 µm/mm)	N.A.	- Optical inspection under microscope prior to incubation - Save pictures of single cells on culture dish for subsequent use - Determine deposition efficiency (Count # of single cells and relate to # of dispensed droplets = 5 x10) - Incubate for 72 h at 25°C - Inspection every 12 hours	- Optical inspection under microscope - Save pictures of cells colonies - Determine cell survival rate (Count # of colonies and relate to # of deposited single cells)	1. Survival rates are to be used as indicative tool to determine physiological effects from mechanical stimulation (shear stress, heat from illuminations, ect...) induces on cells during SCM process, if any. 2. Are there morphological differences of SCM seeded single cell colonies compared to randomly seeded single cell colonies?

Author:	Azmi Yusof (IMTEK)	Date:	30.9.2010	Revision:	1.0