SUPPORTING INFORMATION

Flexible Fabrication of Microarrays of Microwells

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List of contents:

1. Effect of solvents on microwell fabrication
2. Controlling cell binding number via well size
3. Cell types used for cell array generation
1. Effect of solvents on the microwell fabrication

The solvents used were acetophenone, ethyl acetate, toluene and 2:1 ratio of acetophenone/ethyl acetate. A significant difference in the shape and sizes of microwells was observed during fabrication using various solvents. A qualitative explanation for this difference can perhaps be advanced by considering some physical characteristics of these solvents, such as density ($\rho$), viscosity ($\eta$), surface tension ($\gamma$), boiling point (bp) and vapour pressures ($P_v$). The physical properties of acetophenone are ($\rho = 1.028$ g mL$^{-1}$, $\eta = 1.68$ m Pa s, $\gamma = 39.04$ mN m$^{-1}$, and bp = 220 °C) significantly higher (except vapour pressure, $P_v = 0.046$ kPa) than those of ethyl acetate ($\rho = 0.9003$ g mL$^{-1}$, $\eta = 0.425$ m Pa s, $\gamma = 23.39$ mN m$^{-1}$, $P_v = 12.6$ kPa, bp = 77 °C) and toluene ($\rho = 0.8669$ g mL$^{-1}$, $\eta = 0.560$ m Pa s, $\gamma = 27.93$ mN m$^{-1}$, $P_v = 3.79$ kPa and bp = 110 °C) (J. Brandrup, E.H. Immergut, and E. A. Grulke, *Polymer Handbook*, 4th ed., Wiley, New York (1999)). A large difference in the characteristic properties of the solvents is advantageous in microwell fabrication. The addition of ethyl acetate with acetophenone compensates the rate of evaporation of ethyl acetate, while modulating viscosity and local vapour pressure, resulting in a higher amount of solute being transported from the centre of the drop to the edge resulting in uniform microwell formation.

The two solvents (e.g. acetophenone and ethyl acetate), also have different parameters for polymer solution. Thus the first solvent has a high boiling temperature and has a low solubility for PS, and the second solvent has a low boiling point and displays high solubility of this polymer (discussed below). The dissolving potential of polystyrene in solvents can be explained by the difference of the solubility parameter ($\Delta \delta$) between the solvent and the polymer. The $\delta$ of acetophenone, ethyl acetate and toluene are 21.7, 18.6 and 18.2, respectively, and of polystyrene 18.6. It is well established (L.H. Sperling, *Introduction to Physical Polymer Science*, 2nd ed., Wiley, New York, Ch. 3 (1992)) that if the difference ($\Delta \delta$) between the solvent and polymer is $> 3.5$ MPa$^{1/2}$, the polymer will not dissolve in that solvent. The differences in the solubility parameters $\Delta \delta$ between acetophenone and polystyrene, ethyl acetate and polystyrene, and toluene and polystyrene are 3.1, 0.0 and 0.4, respectively. A mixture of two parts of acetophenone and one part of ethyl acetate will have a solubility parameter of 20.67 ($21.7 \times 2/3 + 18.6 \times 1/3$) with a $\Delta \delta$ of 2.1, indicating a higher dissolution of polystyrene in the mixture of acetophenone and ethyl acetate compared to that of acetophenone.
2. Controlling cell binding number via with well size

Cells were cultured on different sizes of the wells, as shown in Figure 1 and 2. Figure 1 (A-C) shows that the number of cells adhering in the well increased significantly with increasing well diameter and depth.

![Figure 1](image)

**Figure 1.** Microwells hosting a monolayer of K562 suspension cells. Composite digital image: Phase contrast and DAPI-staining.

![Figure 2](chart)

**Figure 2.** Cell numbers vs well diameter.

3. Cell types used for cell array generation

A number of cell types have been successfully applied to the arrays of wells. The list is not exhaustive but includes:

ES Cells (46 C): Generated by gene targeting of E14Tg2a cells (mouse neural progenitor, selected for Sox1 expression cell line, Whole Cell Lysate).
L929: Mouse fibroblast cell is a subclone of the parental strain L, established by W R Earle in 1940. The L strain was derived from normal subcutaneous areolar and adipose tissue of a 100 day old male C3H/An mouse.

B16F10: A mouse melanoma cell line (mouse skin cancer cells).

HeLa: Derived from a cervical carcinoma from a 31-year-old Negro patient. This was the first aneuploid line derived from human tissue maintained in continuous cell culture.