

BIOCOMPATIBILITY STUDY USING MICROELECTRONIC MATERIALS TO GROW AND DIRECT TUMOR CELLS

A. Finn, J. Alderman & J. Schweizer

National Microelectronic Research Centre, NMRC, Cork, Ireland,

INTRODUCTION: Over the last 10 years, the ability to grow mammalian cells on microelectronic materials has led to the development of a whole range of biosensors that can measure cellular function. The growth control of cell colonies is important for optimizing cellular recording by forcing the cells to grow on a measuring electrode or gate of a field-effect transistor. The most common materials used for culturing of cells are Silicon Dioxide, Silicon Nitride, Gold, Platinum and Polyimide. Many researchers use Gold or Platinum as a recording electrode for neural electrical studies with silicon nitride as a passivation layer (Borkholder et al, 1998, Wilkinson et al, 1990 and Offenhaeusser et al, 2000). These materials have been shown to be biocompatible with cardiac myocytes. Some researchers (Matsuzawa et al, 1993, Britland et al, 1993) successfully constructed simple neural networks on photo-lithographically patterned substrates. The cells can be confined using a combination of grooves in the substrate and by the selective adsorption of various adhesive proteins such as polylysine and laminin to the substrate.

In this paper, the growth of tumor cells on various microelectronic surfaces will be described as well as efforts to direct the growth of these cells on suitably patterned substrates.

METHODS: Silicon wafers with silicon dioxide, silicon nitride, polyimide, titanium nitride, gold and platinum layers were diced into 8x8mm² pieces and cleaned in acetone, IPA and DI water before baking them dry in an oven. They were then placed into 10mm diameter culture plates prior to cell seeding. For the directed growth, a special template was designed containing lines and grids of various sizes and spacings (Figure 1). The line-widths used were 3, 5, 10, 20 and 30 μm with spacings of 5, 10, and 50 μm . Five of each line-type was drawn and the dimensions of the entire array was 4mm². The above patterns were then dry etched into oxide and nitride layers by stopping on an underlying nitride and oxide layer respectively. We then attempted to increase (and decrease) the adhesion properties of these surfaces using a Silanization treatment. Thus, one wafer was coated with Amino-silane (APTES) to make the surface hydrophilic while another was coated with HMDS vapor to make the surface

hydrophobic. The substrates were then placed into 10mm² culture well plates. Prior to cell culture, all substrates were sterilized by exposure to a UV-light source. Finally the culture medium, containing approximately 10⁵ DU-145 adherent prostate carcinoma cells was put into each well and the cells were kept in an incubator for two days before examination under a standard reflection microscope at x10 magnification.

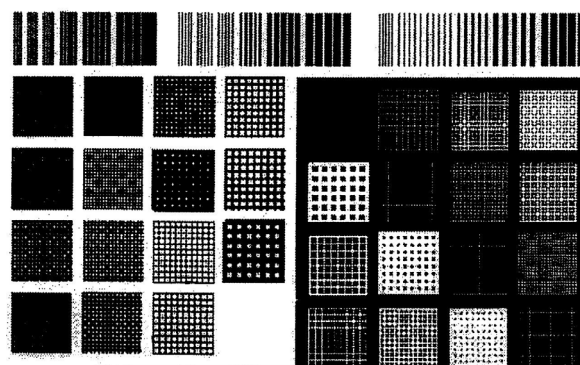


Fig 1: Design template used in selective growth studies.

RESULTS: The results of this study are shown in Figures 1 and 2. Figure 1 shows DU145 tumor cells cultured on a range of microelectronic substrates for 2 days in-vitro. Silicon dioxide, Silicon nitride, Polyimide, Gold and Platinum all resulted in a cell densities and morphologies very similar to the same cells cultured on glass slides. Titanium Nitride, however, produced poor cell densities and morphologies with cell growth being patchy and sparse. This material appears to be unsuitable for culturing of this cell type at least. Figure 3 shows the results obtained after growing these cells on, both treated and untreated, patterned substrates. Confinement varied from poor to moderate and only grooves greater than 10 μm were able to confine these cells. The silanized surfaces were very different in that the APTES-treated surface produced moderate confinement but the HMDS-treated surface produced very poor cell growth with hardly any confinement either to the surface or to the grooves.

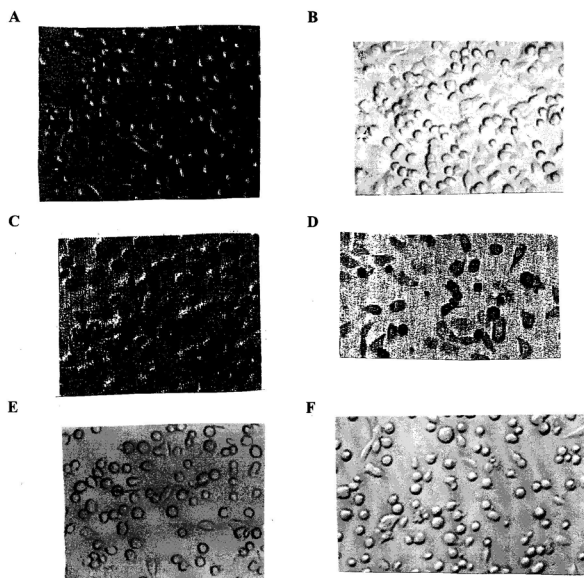


Fig 2: Growth of DU145 tumor cells on various microelectronic materials. (A) silicon dioxide (B) silicon nitride (C) polyimide (D) titanium Nitride (E) platinum and (F) gold at 10^5 cells/ml $\times 10$

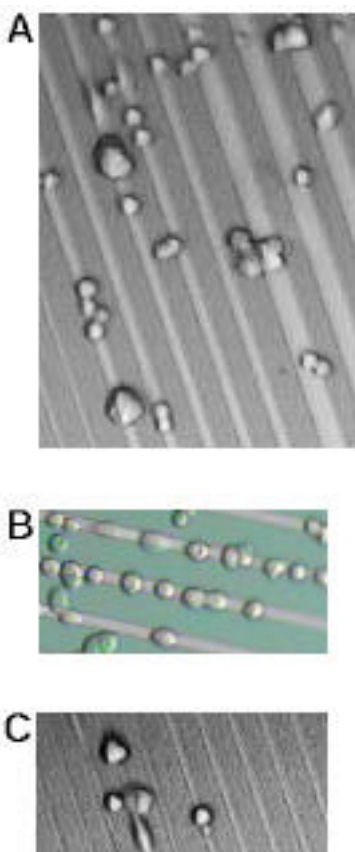


Fig 3: Selective DU145 tumor cell growth on different surfaces. (A) 20um wide untreated silicon dioxide tracks (B) 20um wide silicon dioxide tracks pre-treated with ADTES and (C) 10um wide silicon dioxide tracks pre-treated with HMDS. X20

DISCUSSION & CONCLUSIONS: These results show that tumor cells can be successfully grown on a range of microelectronic surfaces and show no significant differences compared to these cells grown on glass slides. Selective growth of these cells was attempted with some success. The condition of the substrate surface is an important factor in determining selective cell growth. Future work will involve more interesting cell types e.g. neural cells, which have processes that can be guided and directed, using suitably patterned and treated surfaces, and thus the construction of simple neural networks should be possible.

ACKNOWLEDGEMENTS: We would like to thank the staff at NMRC central Fabrication in the design and fabrication of the structures. We are very grateful to Dr T.Cotter for the use of his cell culture facility.

REFERENCES: ¹ S.Britland et al, (1993) *Growth cone guidance and neuron morphology on micropatterned laminin surfaces*, J.Cell Science **105**, 203-212 ² M. Matsuzawa et al, (1993) *The Containment and growth of neuroblastoma cells on chemically modified and patterned substrates*, J.Neuroscience methods **50**, 253-260 ³M. Matsuzawa et al, (1996) *Chemically modifying glass surfaces to study substratum-guided neurite outgrowth in culture*, J.Neuroscience methods **69**, 189-196.