Microfluidic Cell Culture of Bacterial and Mammalian Culture for miniaturized therapeutic examination.

Marie Keays[1,2], Patrick A. Kiely[2], Tara Dalton[1]
Stokes Institute, University of Limerick, Limerick, Ireland[1] Department of Life Sciences, and Materials and Surface Science Institute, University of Limerick, Limerick, Ireland[2].
Marie.keays@ul.ie
Keywords: Biomedical Engineering; Biology & Life Sciences

Abstract

The realisation of personalised medicine has been the target of many multidisciplinary scientific teams. The goal to develop a system that can individualise point of care diagnostics has the potential to revolutionise medical treatment. Currently, drug discovery uses cell culture techniques as a test to monitor the cells reactions to drugs. However, the requirement of sample containers such as dishes or flasks, limit culture methods in volume and automation. With a need to understand and diagnose disease and infection at a cellular level there is a requirement to undertake cell culture in multi-experiment, cost effective and automated environment. This has encouraged the development of a microfluidic droplet cell culture system. The proposed system will create droplet cultures (500-2500nl), incubate and image the cultures on one platform. The culture droplets are created in a nano-litre range and multiple unique bioreactor droplets can be prepared and analysed on one system making this a time and cost effective instrument as the droplets as one current 10ml flask culture is equivalent to 20,000 individual bioreactor cultures on this system. The culture droplets can be mixed with any drug of interest and the reaction monitored on the platform or taken for further analysis. This system can benefit research as both mammalian and bacterial culture can be established on the system making this a powerful instrument in understanding cells.

1. Introduction

Microfluidics is the manipulation of fluids under one millilitre this has been a method of the development of complex experimentation in a high-throughput platforms. Microfluidic droplets can act as chemical bio reactors where a stable environment for biological and chemical samples. It will allow the miniaturization of cell culture techniques that can be applied in personalised medicine and pharmaceutical drug discovery on a high throughput, low cost platform. The scale of the microfluidic droplets will encourage a microenvironment closer to the cells in vivo environment with mammalian cells being used 10-20um in diameter. Three dimensional models will allow for this in the form of multi-cellular spheroids. Figure 1 demonstrates the proposed system which will comprise of a microfluidic droplet typically 500nl in volume this will contain the cells and culture media. It will be contained in PTFE tubing which is gas permeable to CO_2, allowing for the diffusion of gasses in and out of the droplet. A layer of oil will form a thin barrier around the droplet preventing contamination. Each droplet is essentially an individual bioreactor. Bioreactor droplets can have unique properties making it possible to do multiple different tests on one platform.

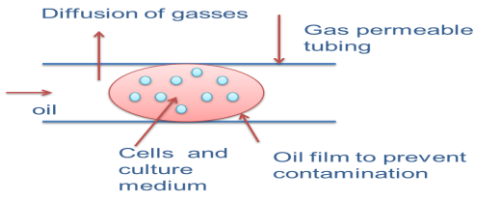

Figure 1. Schematic of microfluidic droplet in t ubing.

5. Results and Discussion

To validate the instrument developed a number of tests were performed where an approved drug was tested against the droplet cultures. Initially after the validation of growth of bacteria Ampicillin and Kanamycin mixed as droplets on the system and the blocking of growth of *E.coli* is seen immediately fig 2 where a control of lysogene broth(lb) remained constant over 8 hours and *E.coli* without drugs continues growing.

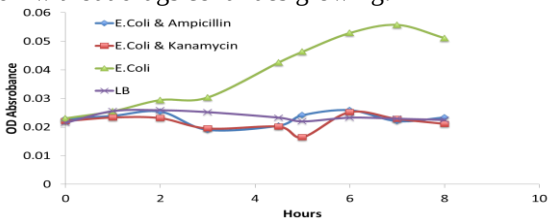

Figure 2. E.coli droplets mixed with Ampicillin and Kanamcyin

Similar tests were undertaken for BT474 breast cancer cells where, when tested against an approved treatments Lapatinib there is a change in gene expression found by O'Neill et al. A study under the same conditions on the microscale instrument has found to have the same change in gene expression. This proves that the instrument has sufficient in treating cells at a miniaturized level and can be used in advancing biological understanding of cells.

8. References

[1] References: F. O'Neill, F.S,Madden, S.T.,Ahern,M., Clynes,J.Crown,P.Doolan,R.O'Connor. *''Gene expression changes as markers of early lapatinib response in a panel of breast cancer cell lines''*, Molecular Cancer (2012) 11(41)