Apparatus and Methods for Dual-Module Automated High-Throughput Microinjections of Adherent Cells

Health & Wellness

Biomedical and Genetic Engineering

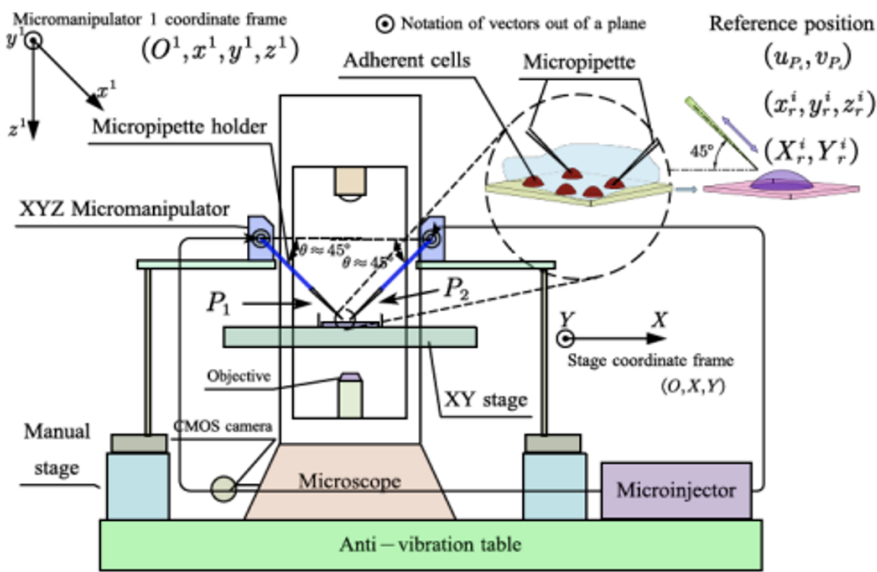Computer/AI/Data Processing and Information Technology

Testing Instruments

Fig.1 The schematic diagram of the automated microinjection system for adherent cells.

Fig.2 The sample of an optimal injection path of two micropipettes injecting 29 cells under 40× magnification.

IP Status
Patent filed

Technology Readiness Level (TRL) ❓ 5

Inventor(s)

Prof. SUN Dong

Dr. PAN Fei

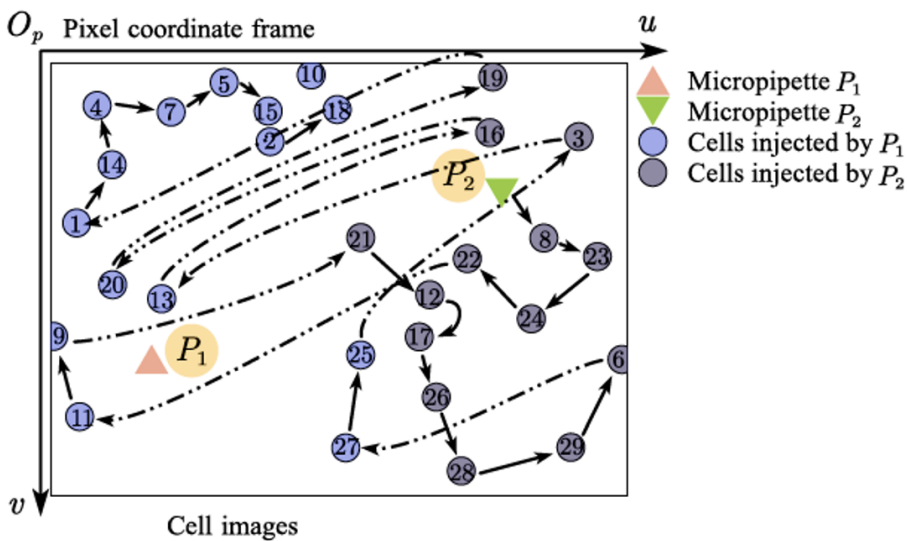**Dr. CHEN Shuxun**

Miss JIAO Yang

Enquiry: kto@cityu.edu.hk

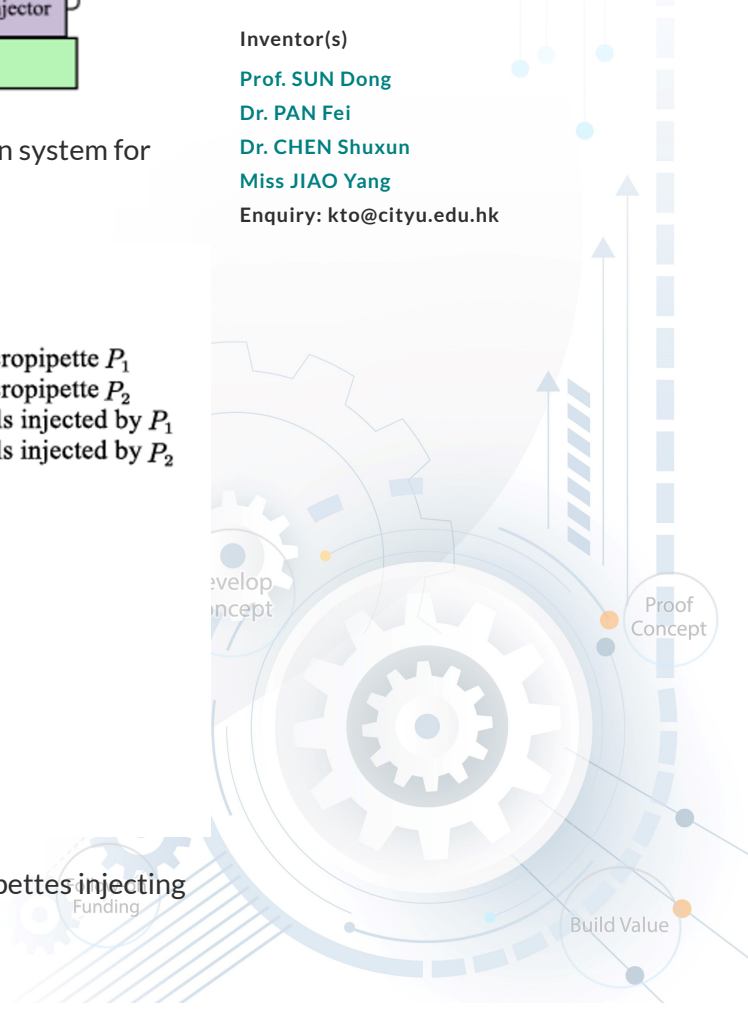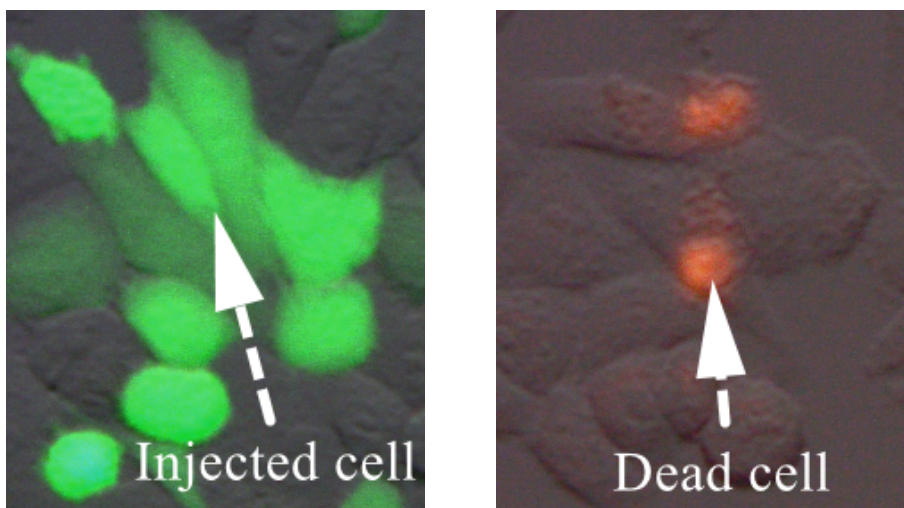Fig.3 The image of cells successfully injected with fluorescein isothiocyanate (Green) and dead cells stained by propidium iodide (Red).

Opportunity

Every cell in the body is protected by a membrane to separate the interior of the cell from outside environment, and to govern the permeability of materials into and out of the membrane. It has been a challenge to deliver useful but membrane-impermeable materials, e.g. dye for visualization, DNA, RNA, or proteins, into living adherent cells that grow on a surface, because of the barrier created by the membrane.

Microinjection is a practical technique that can introduce very low quantity of membrane-impermeable materials, even down to femtoliters, into a single live cell with the use of micropipette, which is tiny laboratory equipment. However, a significant challenge for the developed microinjection systems is the low-throughput that only limited number of injection experiments can be performed within a certain period of time. The problem cannot be solved by simply running the existing injection experiment for a longer period to obtain a sufficiently larger number of injected cells, since the cells in the experimental platform can only maintain its activity within a limited period.

This invention provides an efficient apparatus and method for high-throughput automated microinjection to handle utmost amount of cells within a designated period of time.

Technology

In a first aspect, this invention utilized two motorized micro-robotic arms (dual-module micromanipulators), where each is equipped with a micropipette, on the same cell handling platform to inject cells cooperatively to improve productivity.

While it is challenging and critical to visualize live cells without addition of colored chemicals to stain the live cells, particularly for those injected cells with further biomedical applications, a second aspect of this invention envisaged deep learning for analysis of cell images for cell detection, and involved deep learning algorithms which can produce better results when
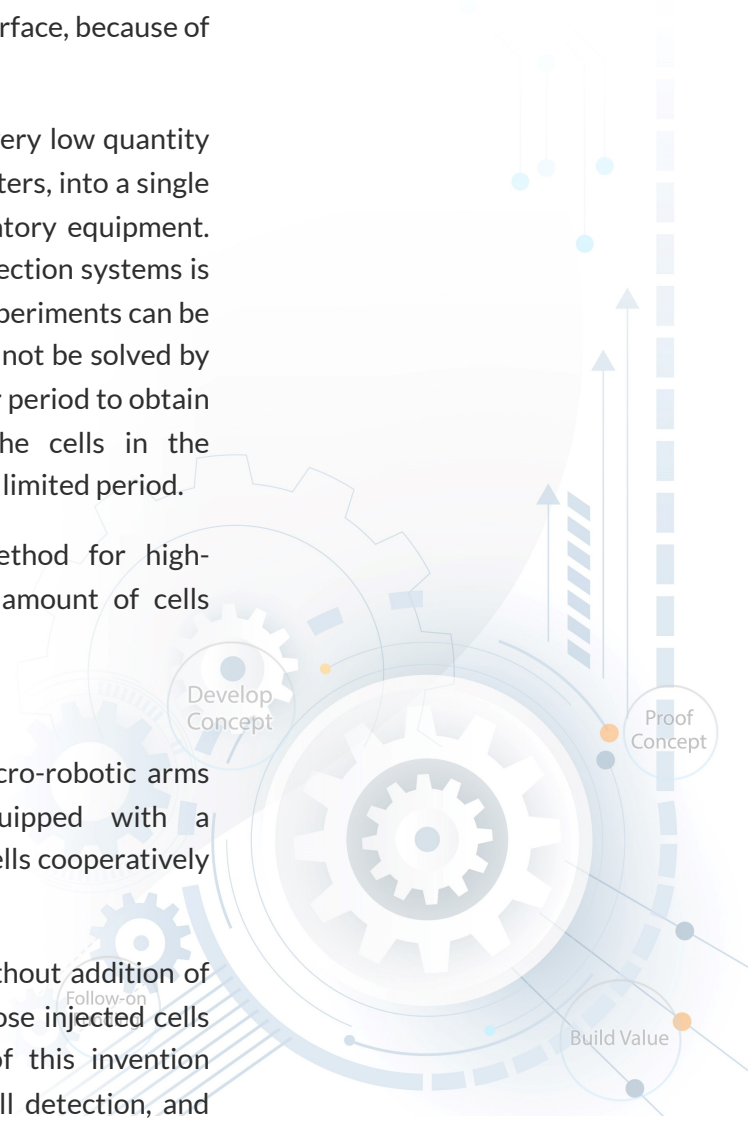
they are trained with more and more training data. Meanwhile, guiding both micropipettes to inject only nearby cells is advantageous to save time, the selection of cells for injection and optimization of the injection path was automated to reduce manual intervention and therefore improve the processing efficiency.

As a demonstration, a fluorescent dye, FITC, was injected into MC3T3 fibroblast cells (connective tissue cells derived from mouse bone) to evaluate the success and survival rates. FITC is a chemical that emit light only in live cell but not in dead cell. Also, FITC is impermeable to cell membranes and dissolves quickly in the medium, so only successfully injected live cells were counted for evaluation purposes. Experimental results demonstrated that the developed microinjection system can process about 4,000 cells per hour at a success rate of about 60.3% and maintain a high survival rate of about 82%. Predictably, the throughput will increase further when more robotic micromanipulators are employed.

Advantages

- Highly automated

- Simple operation

- High-throughput

- Deep learning technology for detection of unstained adherent cells accurately

Applications

- Perform highly precise gene knock-in and knock-out research

- Ideal cell transfection method to produce genetically modified cells

- Biomedical applications