ASPEN

ASPEN 2017

The 7th International Conference of Asian Society for Precision Engineering and Nanotechnology Program

November 14 Tue.
- 17 Fri., 2017

COEX, Seoul, Korea

Organized by

KSPE Korean Society for Precision Engineering

Co-Organized by

中国机械工程学会 CHINESE MECHANICAL ENGINEERING SOCIETY

CPMS Chinese Precision Machine Society

JSPE 公益社団法人 精密工学会 The Japan Society for Precision Engineering

TSPE 台灣精密工程學會 Taiwan Society for Precision Engineering

Supported by

SEOUL METROPOLITAN GOVERNMENT

ASPEN

Technical Program Information

- Guest Speeches
- Oral Presentations
- Poster Presentations

Poster Presentations

Novemver 16 Thu.

Time	Paper ID	Title / Speaker / Affiliation

10. Molding and Forming Technology — Lobby

13:30~14:50	MFT-P-01	***Micro Injection Molding and Assembly of Parts in EDM Machine that was Used for Making Mold Cavities*** Kazuki Oshima(The University of Tokyo), Masanori Kunieda(The University of Tokyo)
	MFT-P-02	***Development of Micro-embossing Equipment for Precision Glass Optical Microstructures*** Lihua Li(The Hong Kong Polytechnic University), Kin Leung Chan(The Hong Kong Polytechnic University), Chang Yuen Chan(The Hong Kong Polytechnic University), Wan Bun Lee(The Hong Kong Polytechnic University)
	MFT-P-03	***Calculation of Oblique Cutting Shear Force Formula for Sheet Metal with Thickness Effect*** Yiwei Zhu(Guandong University of Technology), Qiusheng Yan(Guangdong University of Technology), Jiabin Lu(Guandong University of Technology), Biao Tang(Guandong University of Technology)

11. Nano/Bio Technology — Lobby

13:30~14:50	NBT-P-01	***Development of Portable Laser Lancet for Painless Blood Sampling*** Taeho Ha(Korea Institute of Machinery and Materials (KIMM)), Younggyu Kim(Jinyoung HNS)
	NBT-P-02	***Antibacterial Based on Magnetically-Controlled Silver-coated Fe_3O_4 Nanoparticles*** Ming Chang(Chung Yuan Christian University), Wei-Siou Lin(Chung Yuan Christian University), Weihao Xiao(Chung Yuan Christian University)
	NBT-P-03	***Aqueous Counter Collision Water Jet Machine for Nanomaterials Processing*** Lindong Zhai(Inha University), Jung Woong Kim(Inha University), Jungho Park(Inha University), Jaehwan Kim(Inha University)
	NBT-P-04	***Surface-textured Polyimide Cantilever for Cardiac Cell Study*** Seonyoung Lee(Chonnam National University), Dong-Su Kim(Chonnam National University), Dong-Weon Lee(Chonnam National University)
	NBT-P-05	***Biodegradable Smart Stent for Wireless Pressure Monitoring*** Jongsung Park(Chonnam National University), Ji-Kwan Kim(Gwangju University), Dong-Weon Lee(Chonnam National University)

Surface-textured Polyimide Cantilever for Cardiac Cell Study

Seonyoung Lee[1], Dong-Su Kim[2], Dong-Weon Lee[3,#]

1 School of Mechanical Engineering, Department of Biological Sciences, Chonnam National University, Korea
2 Graduate School of Mechanical Engineering, Chonnam National University, Korea
3 School of Mechanical Engineering, Chonnam National University, Korea
Corresponding Author / E-mail: mems@jnu.ac.kr

KEYWORDS:Functional cantilever, PI(Polyimide), Nano groove, Cardiomyocyte, Contraction force

In this study, we have developed the functional PI (Polyimide) cantilever with nano grooves on its top surface and measured the contractile force of cardiomyocytes. The functional PI cantilever is capable of aligning cardiomyocytes by forming a nano groove in the longitudinal direction on the top surface. By using a contraction force to the cells, it is possible to induce a mechanical deformation of the cantilever, and quantitatively measured. The patch clamp, which is a conventional electrophysiological method, measures the micro current at a single cell level; while the functional PI cantilevers, is a mechanic physiological method has the advantage that the behavior of cardiomyocytes cells in tissue level can be measured through mechanical deformation. The functional cantilever proposed in this study is fabricated by spin coating a liquid polyimide (dfcpi-301) on a PI substrate and then patterning the nano groove using a PDMS stamp (800 nm line and space). Using a shadow mask, the functional PI cantilever has been fabricated with dimensions of 6 mm × 2 mm × 16μm, and basic experiments on material properties are carried out. The resonance frequency of the functional PI cantilever measured at 124 Hz, and the calculated Young's modulus and the spring constant are found to be 6.5 GPa and 220 mN/m, respectively. The cell showed the maturity difference according to the substrate's Young's modulus and surface topology. Using the functional PI cantilever with the nano grooves, it can be used as a device for measuring the contractility of cardiomyocytes.

1. Introduction

Drugs are marketed after administrated a drug toxicity evaluation in the FDA. But, after marketing, many people are suffered from side effects that lead to arrhythmia or death[1]. The reason for this is the patch clamp, which is the only method used to evaluate drug toxicity, is less accurate. Patchclamp predicts the behavior of myocardial cells by electrophysiology, which measures the action potential by the entry and exit of ions (K^+, Na^+, Ca^{2+}). However, the accuracy of the drug toxicity evaluation is affected by the inaccurate phenomenon that the ion entry and the behavior of the myocardial cells are inconsistent. Therefore, in order to improve the accuracy of the drug toxicity evaluation, a method of quantitatively measuring the contractile force, which is the behavior of the myocardial cell, has been proposed. Among them are Micropost and Cantilever. Micropost [2] measures the contractility of single myocardial cells according to the degree of bowing of the column. However, there is still a limit to the difficulty of measuring tissue cells by measuring only single cells such as patch clamps. Therefore, Cantilever's measurement method has attracted attention. Cantilever has the advantage that it can measure tissue contraction force rather than single cell compared with existing patch clamp and Micropost. The method of measuring myocardial contractile force using cantilever is to culture cardiac cells on cantilevers and measure the displacement with the laser sensor. For the reason, myocardial cell growth is affected by the material and surface structure of the cantilever. The surface structure is groove patterned micro and nano structure. This structure helps the cardiomyocytes to grow in one direction. The sarcomere length of cardiomyocytes cultured by the groove patterning shows that are increased by 13% higher compared to non-groove pattern. These results indicate the cells have more matured. Also, cantilever displacement is increased. Because, the cardiomyocytes are aligned and contracted in one direction. However, in spite of studies suggesting that the myocardial cell is more mature in nanostructure[3], Nano-structures had never been applied to the cantilevers to the drug toxicity assessment.

In this study, PI(polyimide)-cantilever with nano groove patterning was produced and the contractility of myocardial cells was measured. Polyimide is a biocompatible material that enhances cell metabolism in vitro [4] and has the advantage of making nano structures by simple PDMS stamping technique.

2. Material and methods

2.1 Design and fabrication of PI-cantilevers

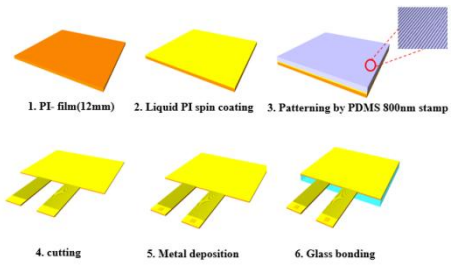

Fig.1 Process flow ofPI(polyimide)-cantilever

The fabrication method of PI - cantilever is shown in Fig.1. First coat a 12μm polyimide film with 4μm thick liquid polyimide (dfcpi-301). Then nano grooves are patterned on polyimide (dfcpi-301) using a PDMS stamp (800 nm line and space). After baking, the cantilever is cut and a metal reflector (Al, 100 nm) is deposited using a shadow mask. The completed cantilever is shown in Fig. 2, and the size is 6 mm x 2 mm x 1 6 μm. The basic experiments on material properties of the cantilever were performed. The resonant frequency of the cantilever was measured at 124 Hz. The Young's modulus calculated using the resonant frequency was 6.5 GPa and the spring constant was 220 mN/m. The contact angle is 73 degrees for the non-groove pattern, 84 degrees for the 800 nm groove pattern, and the cell culture petridish is 65 degrees. This result implies that the contact angle of Polyimide is suitable to grow when compared to Cell culture petridish..

2.2contractile force of cardiac cell measurement

The basal experiment of cardiomyocytes growth was performed according to the surface structure, and maturation scales were Sarcomerelength and α-actinin protein expression[5]. Sarcomerelength of cardiomyocyteswas measured by fluorescence staining and α-actinin protein expression was measured by Western blot.The NRVM (Neonatal Rat Ventricular Myocyte) isolation and cultured in a non-groove, 800 nm groove pattern..Fig. 2 shows that800nm groove surface shows arranged cardiac cell, while non-grooved surface shows cardiac cell is not. In addition, sarcomerelength of 800nm grooved surface(1.7 μm) is longer than non-grooved(1.5 μm) surface. α–actinin's protein expression of 800nm grooved surface is higher than non-groove. These results indicate that the muscle is a more developed mature cardiomyocytes. In order to quantitatively show the difference in the behavior of the non-groove and 800 nm grooves, cardiomyocyteswere cultured on the cantilever through NRVM isolation. After the incubation, the myocardial contractile force was measured on the 3rd to 10th day through the laser displacement sensor, and the myocardial contractility was maximized on the 9th day. The maximum displacement of the Cantilever was 25μm for the non-grooves and 70μm for the 800 nm grooves, which showed a displacement of 2.8 times greater than the 800 nm groove pattern. This means that the cells are more mature and aligned in the 800 nm groove.

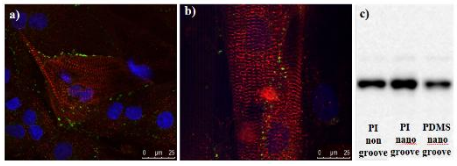

Fig 2. The sarcomere length of the cardiac cells cultivated on the PI and protein amount. a) is non-groove PI, b) 800nm groove. The cardiac cells of b) have been align.c) alpha-actinin protein amount. PI-nano groove showed the most protein.

3. Conclusions

In this study, 800nano-groove functional PI-cantilevers were fabricated and the contractile force of myocardial cells was expressed as mechanical strain. Polyimide has the advantage of patterning the nanometer groove with a simple PDMS stamping method and it has proved that the contact angle, the sacomerelength and the alpha-actinine expression of the myocardial cell were superior when compared with the conventional cantilever material PDMS. If hiPSC-CMS is cultured in the proposed PI-cantilever, it can be grown into mature myocardial cells to measure contractility. This functional PI-cantilever is a new paradigm for assessing drug toxicity, expecting it to be used as a drug toxicity evaluator to improve the accuracy of drug toxicity assessment in that it can directly measure the contractility of the heart.

ACKNOWLEDGEMENT

This study was supported by International Collaborative R&D Program through KIAT grant funded by the MOTIE(N0000894) and National Research Foundation of Korea(NRF) grant funded by the Korea government(MSIP)(No.2015R1A4A1041746 and 2015R1A2A2A05001405)

REFERENCES

1. Boudou, T. et al., "A Microfabricated Platform to Measure and Manipulate the Mechanics of Engineered Cardiac Microtissues," Tissue Engineering Part A, Vol. 18(9-10), pp. 910-919, 2011.
2. Rodriguez, M. L. et al., "Measuring the Contractile Forces of Human Induced Pluripotent Stem Cell-Derived Cardiomyocytes With Arrays of Microposts," Journal of Biomechanical Engineering, Vol. 136(5), pp. 051005-051005-051010, 2014.
3. Martínez, E. et al., "Effects of artificial micro- and nano-structured surfaces on cell behaviour," Annals of Anatomy - AnatomischerAnzeiger, Vol. 191(1), pp. 126-135, 2009.
4. Raynaud, F. et al., "Pharmacological properties of the PI3 kinase (PI3K) inhibitors PI-540 and PI-620," Cancer Research, Vol. 68, pp. LB-149-LB-149, 2008.
5. Kim, J. et al., "Designing nanotopographical density of extracellular matrix for controlled morphology and function of human mesenchymal stem cells," Vol. 3, p. 3552, 2013.