FABRICATION OF NANOPORE WITH DIFFERENT PARAMETERS IN DEPTH AND DIAMETER ON SILICON SUBSTRATE USING ONE-STEP FOCUSED ION BEAM MILLING

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A thesis submitted in fulfilment of the
requirements for the award of the degree of
Master of Philosophy

Malaysia-Japan International Institute of Technology
Universiti Teknologi Malaysia

AUGUST 2022

DEDICATION

This thesis is wholeheartedly dedicated to my parents and siblings and also myself.
Thank you for always being there for me through thick and thin.

ACKNOWLEDGEMENT

First and foremost, I would like to thank Almighty Allah SWT for giving me the source of strength, blessings, good health and for keeping all circumstances in favor of me in order to complete this thesis successfully.

The biggest thank you and sincere appreciation to my supervisor, Dr. Hafizal bin Yahaya for helping me and providing conscientious guidance throughout this research. It was a great privilege and honor to study under his supervision. I am extending my heartfelt thanks to my fellow postgraduate students, colleagues and others who have directly or indirectly assisted on various occasions. I am grateful to all those with whom I have had the pleasure to work during the last two years of my study.

I'm also ineffably indebted to Malaysia-Japan International Institute of Technology for funding my research and for giving me the opportunity to pursue my postgraduate study. I also acknowledge the vast investment from Universiti Teknologi Malaysia in establishing high-end research facilities that also fulfilled my needs and other students.

Nobody has been more important in the pursuit of my study than the members of my family. My gratitude goes to my parents for their love, prayers and support in educating and preparing me for my future. Finally, to my siblings for their constant encouragement which immensely helped me whenever I felt like giving up.

ABSTRACT

Focused ion beam (FIB) technique uses a focused beam of ions to scan the surface of a specimen, analogous to the way scanning electron microscope utilizes electrons. Recent developments of FIB technology have emerged as one of the most advanced multifunctional platforms for nanofabrication. It is a versatile tool for material removal with high accuracy at nanoscale. As a consequence, FIB has become increasingly popular among researchers for fabrication of nanopore structures. Nanopores are generally categorized as biological nanopores and solid-state nanopores. Biological nanopores are highly reproducible but suffer from shortcomings such as fixed pore size, mechanical instability and operate within minimal ranges of pH and temperature. Solid-state nanopores, in contrast, are favorable due to their robustness, controllable morphology, and high stability in various environmental conditions. Hence, solid-state nanopores fabricated using FIB are the focus of this research. Fabricating nanopore on silicon substrate using FIB is doubtless challenging in order to achieve fine structure and tip diameter less than 50 nm. These problems can be rectified by utilizing high-quality nanopores with relatively small tip diameter, an appropriate pore shape and proper materials. The ability to control the diameter of nanopores across a range of dimensions is considered crucial for this research. A smooth and fine surface of nanopore is challenging to obtain as it requires the proper selection of FIB parameters. Therefore, operating the milling process with appropriate FIB parameter selection is essential to achieve successful FIB milling. An acceleration voltage of 30 kV and beam current of 18 pA were used throughout this experiment. It was found that milling from the outer to the inner direction produced a better nanopore structure with less redeposition and a smaller tip. The nanopores milled using one-step FIB milling on silicon substrate resembled a conical-shaped structure. The maximum width and depth were measured at the base and tip of the nanopores, respectively. The milling diameter of 800 nm and depth of 1500 nm were found to be successfully employed to fabricate nanopores with tip diameter of 49.5 nm on thick silicon substrate. A smaller tip diameter was obtained when the aspect ratio was more than 1. These findings suggest that lowering the upper base diameter and increasing the depth may reduce the bottom tip diameter. The overall trend demonstrated that as the aspect ratio increases or milling depth increases, the angle of the nanopore sidewall gets more gradual and the conical shape becomes more defined. Finally, a nanopore with tip diameter of 49.2 nm was also demonstrated on thin silicon substrate using optimal parameters. In conclusion, this study provides greater insight into the nanopore fabrication process using FIB. It may serve as an important study for application of DNA sequencing and more applications involving solid-state nanopores.

ABSTRAK

Teknik pancaran ion terfokus (FIB) menggunakan pancaran ion terfokus untuk mengimbas permukaan spesimen, sama seperti cara pengimbasan mikroskop elektron menggunakan elektron. Perkembangan terkini teknologi FIB telah muncul sebagai salah satu platform pelbagai fungsi yang paling maju untuk fabrikasi nano. Ia adalah alat serba boleh untuk penyingkiran bahan dengan ketepatan tinggi pada skala nano. Sebagai akibatnya, FIB telah menjadi semakin popular di kalangan penyelidik untuk fabrikasi struktur nanopori. Nanopori secara amnya boleh dikategorikan sebagai nanopori biologi dan nanopori bukan biologi. Nanopori biologi mempunyai kelebihan iaitu mudah dihasilkan semula tetapi mengalami kekurangan seperti saiz lubang yang tetap, ketidakstabilan mekanikal dan hanya boleh beroperasi pada julat pH dan suhu yang sangat terhad. Nanopori bukan biologi pula berbeza, ia mempunyai kelebihan kerana kekukuhannya, struktur yang boleh dikawal, dan kestabilan yang tinggi dalam pelbagai keadaan persekitaran. Oleh itu, nanopori bukan biologi yang direka menggunakan pancaran ion terfokus telah diperkenalkan dalam kajian ini. Fabrikasi nanopori pada substrat silikon menggunakan FIB sudah pasti mencabar untuk mencapai struktur halus dan diameter hujung kurang daripada 50 nm. Masalah ini dapat diperbetulkan dengan menggunakan nanopori berkualiti tinggi dengan diameter hujung yang agak kecil, bentuk liang yang sesuai dan bahan yang sesuai. Keupayaan untuk mengawal diameter nanopori merentasi pelbagai dimensi dianggap penting dalam kajian ini. Permukaan nanopori yang licin dan halus adalah mencabar untuk diperolehi kerana ia memerlukan pemilihan parameter FIB yang betul. Oleh itu, pemilihan parameter FIB yang sesuai adalah penting untuk menjayakan kisaran FIB. Voltan pecutan 30 kV dan arus 18 pA telah digunakan sepanjang eksperimen ini. Didapati bahawa kisaran dari arah luar ke dalam menghasilkan struktur nanopori yang lebih baik dengan kurang pemendapan semula dan hujung yang lebih kecil. Nanopori dihasilkan menggunakan kisaran FIB satu langkah pada substrat silikon menyerupai struktur berbentuk kon. Lebar maksimum dan kedalaman maksimum boleh diukur pada pangkal dan hujung nanopori. Diameter kisaran 800 nm dan kedalaman 1500 nm didapati berjaya digunakan untuk menghasilkan nanopori dengan diameter hujung 49.5 nm pada substrat silikon tebal. Diameter hujung bawah yang lebih kecil boleh diperolehi apabila nisbah aspek lebih daripada 1. Ini menunjukkan bahawa apabila nisbah aspek meningkat atau kedalaman kisaran meningkat, sudut dinding sisi nanopori bentuk kon menjadi lebih jelas. Akhir sekali, nanopori dengan diameter hujung 49.2 nm juga ditunjukkan pada substrat silikon nipis menggunakan parameter optimum yang ditemui dari objektif pertama. Kesimpulannya, kajian ini memberikan gambaran yang lebih mendalam tentang proses fabrikasi nanopori menggunakan FIB. Ia mungkin berfungsi sebagai kajian penting untuk aplikasi penjujukan DNA dan lebih banyak aplikasi yang melibatkan nanopori bukan biologi.

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LIST OF ABBREVIATIONS

AAO	-	Anodic aluminium oxide
Al	-	Aluminium
Al_2O_3	-	Aluminium oxide
Ar	-	Argon
Au	-	Gold
Be	-	Beryllium
BN	-	Boron nitride
EBL	-	Electron beam lithography
EBSD	-	Electron backscatter diffraction
EDS	-	Energy dispersive dpectroscopy
FEB	-	Focused electron beam
FESEM	-	Field emission scanning electron microscope
FIB	-	Focused ion beam
Ga	-	Gallium
GFISs	-	Gas field ionization sources
HfO_2		Hafnium (IV) oxide
HIM	-	Helium ion microscope
MPVI	-	Multilevel pulse-voltage injection
MoS_2	-	Molybdenum disulfide
MspA	-	Mycobacterium smegmatis porin A
Pa	-	Palladium
PI	-	Polymide
RIE	-	Reactive ion etching
SEM	-	Scanning electron microscope
ssDNA	-	Single-stranded deoxyribonucleic acid
SiC	-	Silicon carbide
SiN	-	Silicon nitride
Si_3N_4	-	Silicon nitride
TEM	-	Transmission electron microscope

LIST OF SYMBOLS

D	-	Diameter
d	-	Hole diameter
ΔI	-	Blockage amplitude
δt	-	Dwell time
F(h)	-	Flux density
F_0	-	Total number of atoms sputtered per unit length
F/F_0	-	Normalized redeposition rate
h	-	Depth
h/d	-	Normalized height
kV	-	Kilovolt
µm	-	Micrometer
nm	-	Nanometer
pA	-	Pico ampere
v_{track}	-	Track region
v_{bulk}	-	Non-track region
W	-	Tungsten
Z	-	Depth

CHAPTER 1

INTRODUCTION

1.1 Research Background

Focused ion beam is still relatively new but has emerged as one of the most advanced multifunctional platforms for nanofabrication. It is a versatile tool for material removal with high accuracy at nanoscale. FIB was specially recognized as an attractive tool for the fabrication of micro- and nanostructures with complex geometries and shapes. Therefore, FIB has become increasingly popular among researchers for fabrication of nanopore structures [1,2].

There are three basic operation modes of FIB which are imaging, milling and deposition as shown in Figure 1.1. During imaging, FIB system uses a focused beam of ions scanned over the surface of material. The bombardment results in electron emission which also allows imaging of the sample as shown in Figure 1.1(a). The electrons are collected on a biased detector for secondary electrons. Apart from secondary electrons, secondary ions also can be emitted which also can achieve functions such as ion beam imaging and spectral analysis.

Figure 1.1(b) shows the milling process or also known as material removal takes place when the ion beam is bombarded on the surface of the substrate. The surface atoms will be sputtered when they received enough energy than the surface binding energy. It is a direct write process through which any given pattern can be transferred onto a substrate through the impingement of energetic ions.

On the other hand, when the energy of incident ions is smaller than the surface binding energy, deposition can be accomplished by adding extrinsic conditions such as gas molecules as shown in Figure 1.1(c). Deposit materials are often given by an internal gas delivery system that uses a gas injection mechanism to expose a chemical component near the site of impact. Furthermore, if the ion is not backscattered off of the sample surface, it will eventually implanted in the solid material at a depth below the sample surface, changing the material's surface properties.

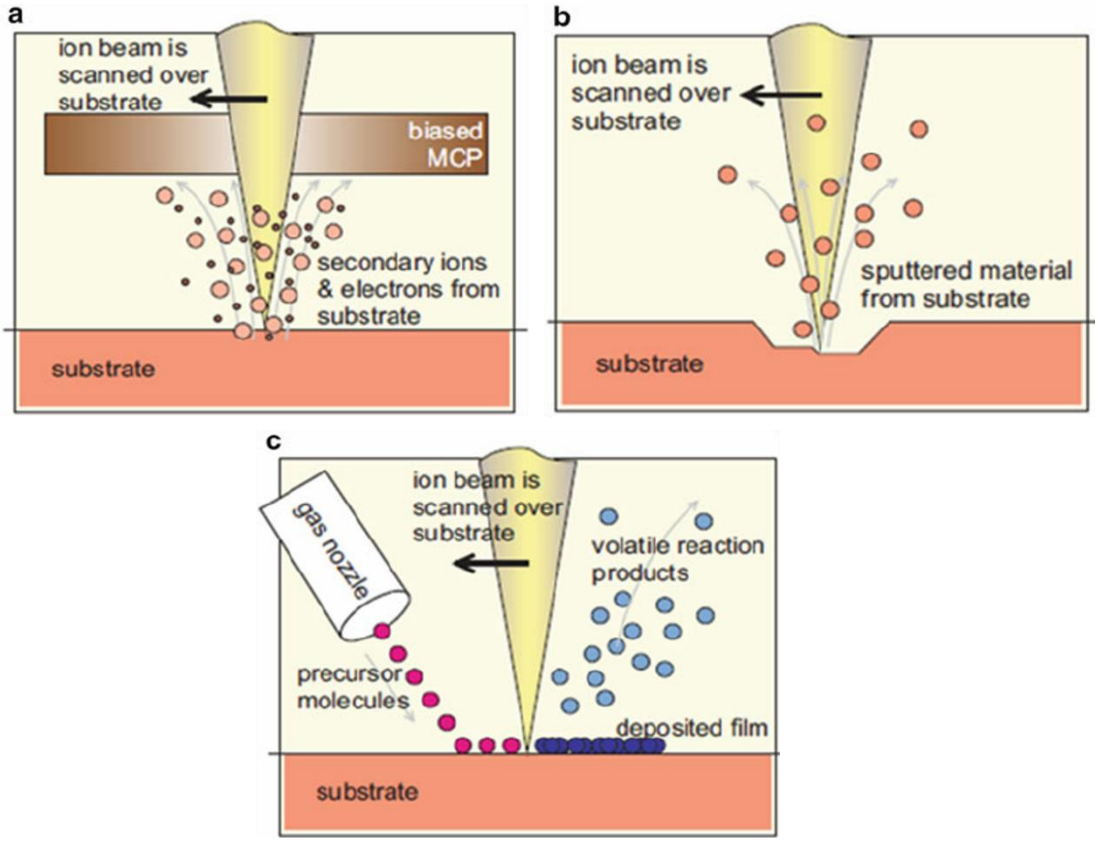

Figure 1.1 Principle of FIB (a) imaging, (b) milling, and (c) deposition [3]

Nanopore technology is a nanometer hole or channel embedded in a thin free-standing membrane structure to identify the potential change when charged molecules smaller than the nanopore pass through the hole. Nanopore technology has recently emerged as one of the most appealing options for detecting and sequencing single-molecule such as protein, deoxyribonucleic acid (DNA) and ribonucleic acid (RNA). It is considered to be low cost, high-throughput and faster method [4-6]. The central idea of nanopore technology is derived from Coulter counter’s principle. The idea was proposed independently by multiple research groups in the mid-1990s [7-9]. Generally, nanopores can be categorized as biological nanopores and solid-state nanopores.

Biological nanopores such as α-hemolysin, mycobacterium smegmatis porin (MspA) and bacteriophage phi29 motor are pore-forming proteins that self-assemble into lipid bilayer membranes. Biological nanopores are protein molecules that have been formed artificially or naturally through genetic engineering. α-hemolysin is the major cytotoxic agent secreted by the human pathogen Staphylococcus aureus, and it was the first bacterial ecotoxin to be recognized as a pore former. Meanwhile, MspA is an outer membrane porin isolated from mycobacterium smegmatis. It has a funnel shaped octameric channel pore with ~1.2 nm constriction and channel long of 0.6 nm channel long, allowing the movement of water-soluble molecules across the membranes. Phi29 is a motor channel from bacteriophage phi29 protein channel. The channel diameter of phi29 is larger than other protein channel, it possesses a channel with 3.6-6.0 nm wide which allows the translocation of larger molecule.

Biological nanopores are highly reproducible but suffer from shortcomings such as fixed pore size, mechanical instability and can operate over minimal ranges of pH and temperature. The limitations of biological nanopores have been addressed using solid-state nanopores created from artificially punctured pores in free standing membrane such as silicon, polymer and 2D membranes. Compared to biological nanopores, solid-state nanopores were advantageous due to their robustness, tunable shape, and excellent stability under various environmental conditions [9]. Furthermore, they are also readily integrated with on-chip electronics, nanofluidic or other nanodevices which can generate higher throughput.

Solid-state nanopores can be fabricated on various materials such as silicon, polymers, glass and single-layer membrane. Silicon is the most widely used material in semiconductor technology as it is compatible with most of the semiconductor processes, which are key components of various electronic devices. The subsequent processing and modification of the silicon-based nanopore are relatively easy to achieve. Silicon is inexpensive and durable, making it much easier to control thickness and pore size during nanopore fabrication. Compared to 2D materials such as graphene, which are mechanically less stable than thick membranes, their mechanical instability is more of a concern where they usually need a supporting structure.

Various approaches have been reported to fabricate solid-state nanopore. Current state of the art methods for nanopore fabrication include focused ion beam (FIB) milling, electron beam lithography (EBL), assisted reactive ion etching (RIE), focused electron beam (FEB) milling using a transmission electron microscope (TEM), ion track etching and dielectric breakdown [10-12]. For example, EBL-assisted RIE requires many steps such as resist spin-coating, patterning by EBL and nanopore formation by RIE. However, this method is tedious because it requires two equipment. The major drawback of this method is that it requires hours to form patterns at the wafer scale. EBL requires a large up-front capital investment due to expensive equipment compared to standard photolithography.

Another method such as dielectric breakdown is performed by applying transmembrane potential, which will generate an electric field and charges the interface with opposite ions. There will be a leakage of current through the membrane, traps are accumulated and finally pores are formed. However, it has poor controllability over the number of nanopores formed. It also lacks the capability for scalable production. Therefore, FIB milling has become one of the most desirable technologies for fabricating nanopores owing to its excellent resolution and ability to execute direct patterning [9].

In this study, solid-state nanopores were chosen and fabricated on silicon substrate using one-step FIB milling. One-step milling is a maskless direct milling method. During FIB milling, the scan method should be determined first based on the desired fabrication structure which is nanopore with conical-shaped and tip diameter less than 100 nm. Firstly, the milling direction was determined first either milling from outer to inner or inner to outer direction. Next, an appropriate ion beam current and other milling parameters such base diameter and depth were chosen to improve fabrication efficiency and nanopore structure. The nanopores were first fabricated on thick silicon substrate to find the optimal milling parameters which can fabricate nanopore with tip diameter less than 100 nm. Thick silicon substrate is the original silicon substrate with thickness of 500 µm without any pre-thinning process. Afterward, nanopore structure with the smallest tip diameter was fabricated on thin silicon substrate to study its repeatability. The thin silicon substrate is the silicon substrate that has been reduced in thickness until it matches the nanopore depth. FIB was also used to perform a cross-sectional and field emission scanning electron microscope (FESEM) to validate the nanopore in terms of morphology such as redeposition.

1.2 Problem Statement

Utilizing solid-state nanopores technology is preferable due to its long-read ability, high-throughput detection and allowing direct sensing. However, it presents significant challenges in terms of nanopore structure and fabrication process. It requires precise dimensional control of nanopores structure with nanometers precision in a solid membrane. In contrast, biological nanopores are built with a pore forming protein that self-assembles into lipid bilayer membranes, simplifying the operation. Therefore, establishing a reliably controllable nanopore fabrication method is very important.

It had been reported that EBL had successfully fabricated sub-10 nm nanopores [10]. Nevertheless, this approach is not desirable owing to their complicated and slow processing steps. Moreover, other techniques such as dielectric breakdown and ion-track etching are also not preferable as it yields pore size with poor distribution. Consequently, an alternative fabrication method using FIB direct milling technology has been explored. Fabrication of nanopores through FIB direct milling presents possibilities for controlling material modifications and patterning crucial dimensions in the nanometer range [12-16]. Compared to EBL, which involves additional processing steps, FIB milling is a one-step method that allows for direct writing and can aid the rapid fabrication of nanopores. Although FIB methods are highly versatile nanofabrication methods, some restrictions or negative secondary effects are connected with their use.

Attaining nanometre scale pore sizes is doubtless challenging. These problems require high-quality nanopores with relatively small pore size, an appropriate pore shape and proper materials. The ability to adjust the diameter of pores across a range of dimensions is considered crucial. Smooth and fine surface of nanopore is difficult to obtain as it requires the proper selection of FIB parameters. Therefore, the ability to operate the milling process with appropriate FIB parameter selection is essential to successful FIB direct milling. A suitable choice of parameters such as beam current, acceleration voltage, pattern size, and scan direction is required to achieve a structure with critical dimension and fine surface. Hence, it is essential to understand the fundamental phenomenon of the milling process.

Besides, the major effect of milling such as redeposition and surface roughness also important. Fabrication of high aspect ratio nanopores is known to be difficult due to the influence of redeposition. The redeposition effect can be minimized by FIB milling with a low current. The conical-shaped structure was chosen since it has been demonstrated experimentally to provide a significantly faster transport rate with less membrane resistance than an analogous cylindrical shape nanopore structure. The reliability and reproducibility of the nanopores also need to be considered. Our investigation involves relatively thick membranes. Besides, thick and thin membranes are included in determining whether high aspect ratio features can be fabricated reproducibly. Therefore, in this study, the milling strategy is very important to obtain nanopores with conical-shaped on silicon substrate using one-step FIB milling.

1.3 Significance of Research

This research will provide a better understanding on the fabrication of nanopores on silicon substrate using one-step FIB milling. FIB is a versatile tool for fabrication of pore with high accuracy at nanoscale. It is possible to fabricate a high aspect ratio nanopore by selecting proper milling parameters. Therefore, a fundamental insight on the effects of FIB milling parameters on the structure of nanopore can be obtained. The fundamental understanding and information gained from this study will help experimentalists to realize the nanopore fabrication process. This new finding can also support the ecologically sustainable development of nanotechnology field especially in Malaysia and other countries as well.

1.4 Research Objectives

The aim of this research is to fabricate nanopore on silicon substrate using one-step focused ion beam milling. There are three objectives and they are described as follows:

i. To evaluate the effects of milling parameters in terms of diameter and depth on nanopore structure fabricated on thick silicon substrate using one-step focused ion beam milling.

ii. To examine the fabrication of nanopore via optimal parameters on thin silicon substrate using one-step focused ion beam milling.

iii. To validate the nanopores morphology using field emission scanning electron microscope.

1.5 Research Scope

This study will focus on fabricating nanopores on silicon substrate using FIB. Dual-beam focused ion beam-field emission scanning electron microscope (FIB-FESEM) from Fei (Helios Nanolab G3 UC) was used to fabricate the nanopores. Firstly, nanopores were fabricated on thick silicon substrate to evaluate the effects of FIB parameters on the structure of nanopores. The bottom tip of the nanopore aimed at least 100 nm diameter. The FIB parameters such as beam current, voltage, pattern size (i.e., diameter and depth) and milling direction were controlled to optimize the nanopore structure. Next, the optimal parameters obtained from the previous method were used to fabricate nanopore on a thin silicon substrate. The silicon substrate was thinned out using hand lapping polisher and cross-section polisher. Besides, the morphology such as redeposition was also validated by using FESEM. The details of this scope were discussed in chapter 3.

1.6 Thesis Outline

This thesis consists of five chapters. Chapter 1 is an introductory part, which provides an overview of this research. It begins with research background of FIB and nanopore technology. Next, current problems associated with the fabrication of nanopores are also described. In addition, this chapter also states the significance of the research, the objectives and the scopes.

Following the introduction in chapter 1, the rest of the thesis is structured as follows. Chapter 2 is the literature review that explains the basic principles of nanopore technology. Besides that, recent advances related to biological and solid-state nanopores were also reviewed. This chapter also presents the fabrication process involving solid-state nanopores. Furthermore, the FIB and FESEM mechanisms were also explained.

Chapter 3 consists of the methodology used in this study. Research flowchart is presented consisting of all the scopes. This chapter elaborates on the experimental method for nanopore fabrication and structural validation process.

Chapter 4 discusses the fabrication approach we have explored to obtain nanopore with the smallest tip diameter on the silicon substrate using FIB. The fabrication process includes evaluation of FIB parameters and structural validation is discussed in this chapter. Lastly, chapter 5 summarizes and concludes the thesis. Future works and potential application were also included in this chapter.

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LIST OF PUBLICATION

1. **Sabili, S. N**., Yahaya, H., & Ahmad, A. (2020). The Effect of Depth on Fabrication of Nanopore using One-step Focused Ion Beam Milling for DNA Sequencing Application. In *2020 IEEE International Conference on Semiconductor Electronics (ICSE).*(pp. 93-95). IEEE, Kuala Lumpur, Malaysia. https://doi.org/10.1109/ICSE49846.2020.9166877