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# Etching Time on Structural and Electrical Properties of Porous Silicon SERS Substrates for Non-Invasive Dengue-NS1 Detection

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**Abstract:** Surface Enhanced Raman Spectroscopy (SERS) is a sensitive and specific analytical technique which has been explored in many applications, including disease detection. However, SERS performance is highly dependent on type of SERS substrate. This work is aimed to develop a SERS substrate that is sensitive to an early dengue virus biomarker known as Dengue virus nonstructural 1 (DENV-NS1) protein from saliva of infected patients. The new SERS substrate will allow non-invasive and rapid detection method for Dengue as early as day one of infection. Early detection of infection within the first five days is crucial to monitoring patients to help in reducing the fatality rate. Here, the electrochemical etching technique is employed to fabricate porous silicon (pSi) with variation in structural features to serve as the SERS substrate base. Variation in surface structural and electrical properties of pSi with etching time is recorded. Structural surface properties of the samples are investigated using the Field Emission Scanning Electron Microscope (FESEM) and energy-dispersive X-ray spectroscopy (EDX). While the electrical properties are observed through I-V, resistivity and conductivity curve. From FESEM images, micro size cross-shaped porous structures are observed to have formed. Top-view reveals micro-size cross-shaped structures, while triangle-shaped structures from the cross-sectional view. The size of the structure formed increases with the etching time. Based on the structural and electrical properties an etching time between 20 to 28 minutes is found optimal for producing more uniform surface structures.

**Keywords:** Porous Silicon (pSi), Surface-Enhanced Raman Spectroscopy (SERS), fabrication, raman, Nonstructural 1 (NS1), Dengue virus (DENV)

## 1. Introduction

SERS is an analytical tool with high sensitivity and specificity for detection of analytes [1]-[5]. Specific information on molecules and molecules adsorbed on nanostructured metallic surface are viable through peak enhancement of the Raman spectrum. The nanostructured metallic surface, known as SERS substrate, is responsible for enhancing the Raman scattering of the adsorbed molecules. Sensitivity performance of SERS is highly dependent on its substrate, especially in the application of biomarker sensing [1], [3].

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The enhancement of Raman scattering is associated with the highly localized regions of intense local electromagnetic field caused by surface plasmon resonance, which can be regulated by nanostructure of SERS substrates [6]. Another important parameter that has significant influence on the SERS substrate performance is the electrical conductivity property of the solid platform. Materials with higher conductivity exhibit better Raman intensity [7]. In this study, porous silicon (pSi) is chosen to be the solid platform of SERS substrate to be developed for detection of the DENV-NS1 protein.

Porous silicon (pSi) is prepared on crystalline Si and has excellent structure that has been extensively studied for biological sensing tools. It is commonly used to develop label-free, low-cost optical biosensor development [4], [8]-[10]. pSi structure offers attractive morphological and chemical properties, which assume its optical properties. The pSi features have been adapted in many applications in the field of electronics, such as biomedical, solar cells and sensors [8]. For SERS, the main advantage of pSi is the high surface area produced by the etching process, which can increase Raman intensity [4], [11], [12]. The surface structure formation depends on the preparation conditions.

Fabrication of pSi is the first step in producing an efficient and affordable SERS substrate for detection of infectious diseases such as dengue virus (DENV). Dengue is one of the viral infection diseases which is dangerous and lethal. Unlike bacterial infection diseases which can be subdued by antibiotic treatment, the recovery of viral infection diseases depends on antiviral medication and vaccine. Until now, there is no vaccine yet for dengue [13], [14]. Hence, early detection is essential, which leads to the needs for an effective early biomarker sensor. Nonstructural protein 1 (NS1) has recently been accepted as an early biomarker for dengue infection. It is also reported to present in saliva, giving hope to the development of an early, non-invasive diagnostic method for DENV. However, the concentration of NS1 protein in saliva is far less than that in blood, thus making its detection more challenging.

It is worth noting that the pSi structure formation process is fully compatible with the current semiconductor processing technology. The electrical characterization is one of the crucial aspects that needs consideration in the pSi structure layer development. The effect after tuning the variables shows the electrical behavior of the substrate development [15]. The electrical behavior of pSi is extremely sensitive to its surface characterization and composition, and the presence of biomolecules on its surface [15]. The integration of pSi in electrical biosensors results in improved sensitivity. Moreover, its particular surface structure and large specific surface is highly attractive for the nucleation of bio-ceramic apatite in simulated body fluids, presenting a catalytic environment for protein nucleation [15].

This paper aims to examine the surface structural and electrical properties of pSi in variation with the electrochemical etching time. Section 2 explains the overall framework in fabricating the SERS substrate for dengue detection. Section 3 elaborates on methods to characterize the surface structural and electrical properties of pSi. Section 4 discusses findings from the characterization.

## 2. Fabrication Framework for DENV-NS1 SERS Substrate

Fig. 1 illustrates the conceptual model of SERS substrate to be developed for detection of DENV-NS1 protein. It consists of pSi coated with silver nanoparticles (AgNPs). *n*-type (100) Si wafer doped with phosphorous is used as the base of substrate. The electrochemical etching technique is first applied for the porous structure fabrication process. Next, the pSi is coated with AgNPs to cover the porous structure. Then, the AgNPs coated pSi is functionalized with Dengue antibody to detect DENV-NS1 protein.

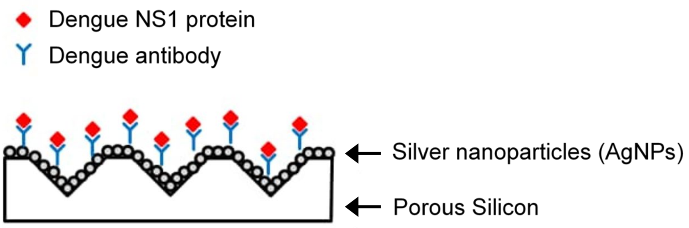



**Fig. 1 - Conceptual model of DENV-NS1 protein SERS substrate**

## 3. Methodology

Fabrication of pSi is the first step in the development of SERS substrate for detection of DENV-NS1. In this study, electrochemical technique is used to fabricate the pSi. Amongst parameters of etching process, such as etching time, current density, types and concentration of chemicals, this paper focuses only on the effect of etching time on the structural and electrical properties of the pSi.

Fig. 2 shows the overall methodology for this study. It starts with wafer preparation and ensues with fabrication of pSi structure using electrochemical technique [16]. To investigate the effect of etching time on the structural and electrical properties, the etching time is varied from 20 to 30 minutes at an increment of 2 minutes. The structural and electrical properties of the fabricated pSi samples are then characterized.

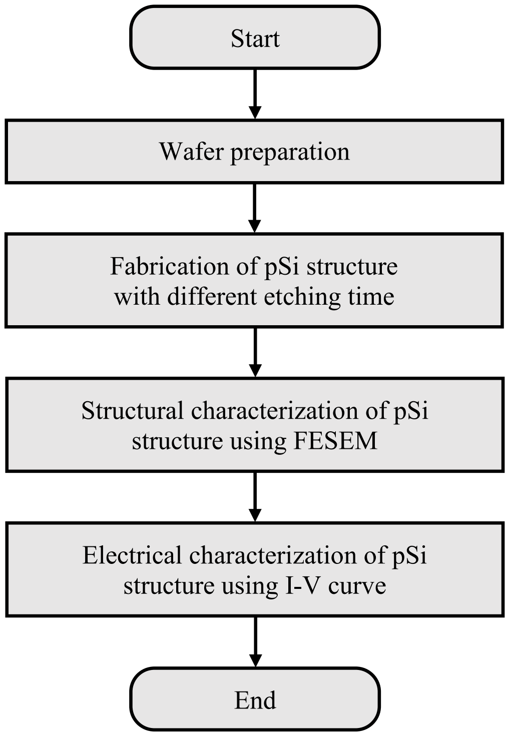



**Fig. 2-Methodology flow for development of SERS substrate for detection of DENV-NS1**

## 3.1 Structural Characterization of pSi

The surface morphology and topography of the pSi samples was characterized by the field emission scanning electron microscopy (FESEM) (Model: JEOL JSM 7401F). The NanoScope Analysis software was used to analyze the surface roughness and the estimated pore depth of the samples with scan area of 10 µm × 10 µm [16].

The pSi structure surface is the first layer for SERS substrate development. It is crucial to analyze the structure layer formation to determine the optimal SERS substrate. The FESEM images shows the porous structure produced by the process, as etching time varies.

## 3.2 Electrical Characterization of pSi

In addition to the nanoscale topography with enhanced activity, the SERS substrate is expected to exhibit excellent conductive property for rapid and in-situ detection [17]. In this study, the electrical characterization of the pSi samples was conducted using Keithley 4200-SCS PK2 Parameter Analyzer. The voltage was swept from -10 V to +10 V and applied to the pSi structure to produce the I-V curve graph. The I-V curve enables greater insight into the electrical properties of the pSi samples and their operation.

# 4. Results and Discussions

## 4.1 Effect of etching time variation on structural properties of pSi

Fig. 3 and Fig. 4 show the FESEM images of the pSi samples at magnification of 5000X, from top and cross-section views, respectively. From the top view, cross-shaped structures pSi are observed to have formed from the fabrication process. Meanwhile, the cross-sectional view observes triangular opening structures. As shown in Fig. 3 (a)-(e), size of the cross-shaped structures increases as etching time increases from 20 to 28 minutes. Increase in size is also found from measurement of triangular opening from the cross-sectional views in Fig. 4 (a)-(e). However, as the etching time is increased more than 30 min, a different structure appears as in Fig. 3 (f). Some of the cross-shaped structures start to connect with neighboring cross-shaped structures to produce rougher pSi surface. The rougher surface is evidenced from the cross-sectional image in Fig. 4 (f). Owing to the interconnection of structures, it is difficult to place the measurement cursor. Thus, the average size of the cross-shaped structure at 30 min is not measurable. Table 1 summarizes the size of the cross-shaped and triangular-opening, as well as depth structures of pSi.

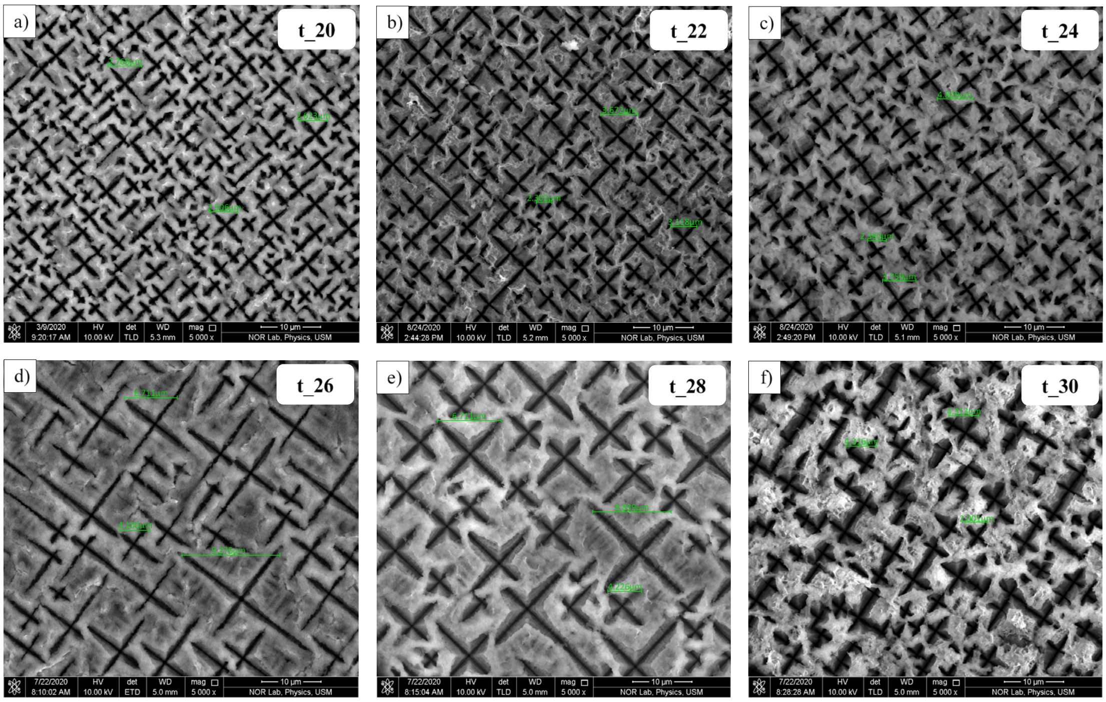



**Fig. 3 - FESEM Characterization for optimal SERS structure surface from top view at 5000× magnification by setting various timing for the etching process (a) 20 minutes, (b) 22 minutes, (c) 24 minutes, (d) 26 minutes, (e) 28 minutes and (f) 30 minutes**

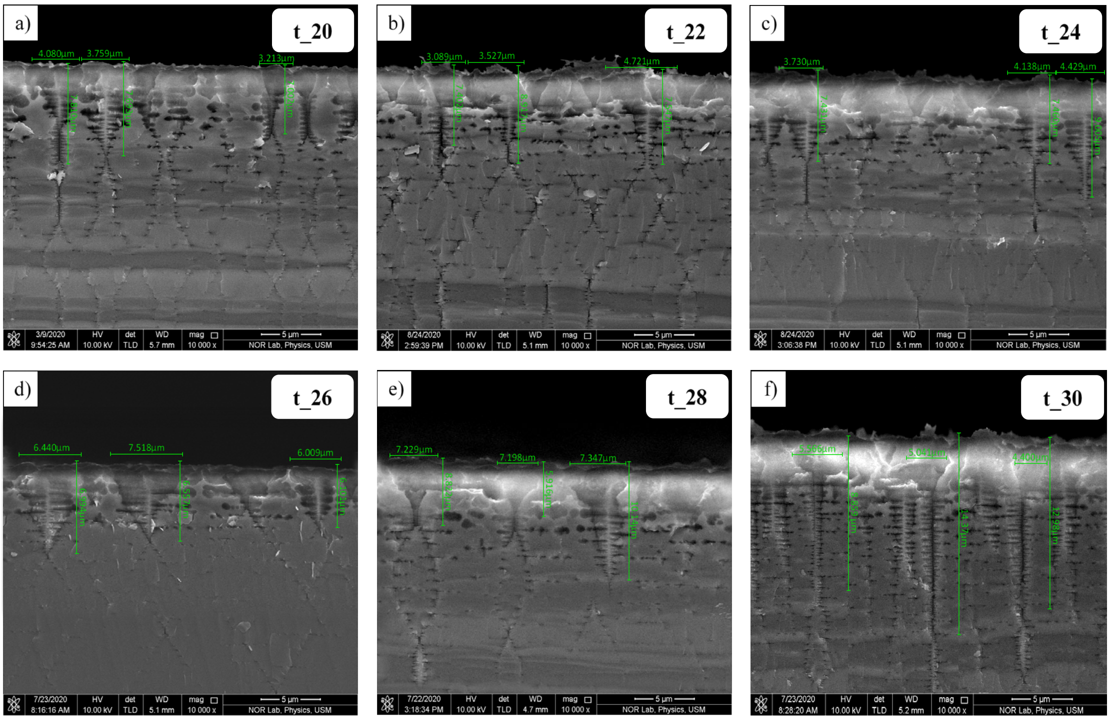


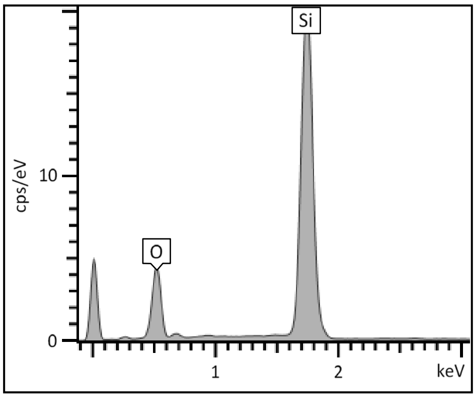

**Fig. 4-FESEM Characterization for optimal SERS structure surface from cross-section view at 10000× magnification by setting various timing for the etching process (a) 20 minutes; (b) 22 minutes; (c) 24 minutes; (d) 26 minutes; (e) 28 minutes; and (f) 30 minutes**

**Table 1 - Size of Cross-Shaped pSi for Different Etching Time**

| Etching time (min) | Top-view Average size (µm) | Cross sectional-view Average size (µm) | |
|---|---|---|---|
| | | Triangle opening | Depth |
| 20 | 2.632 | 3.684 | 7.502 |
| 22 | 3.031 | 3.779 | 7.955 |
| 24 | 3.643 | 4.099 | 7.965 |
| 26 | 6.696 | 6.656 | 6.551 |
| 28 | 6.592 | 7.258 | 8.293 |
| 30 | NA | 5.002 | 12.070 |

With reference to Table 1, as etching time increases from 20 to 28 minutes, a significant increment can be observed on the size of the cross-shaped structures and triangular opening. The size of the cross-shaped structures increases from about 2.5 to 6.5 µm (~160%) and that of the triangular opening increases from about 3.5 to 7.2 µm (~106%). However, variation in depth is less significant (~10.7%).

(a)




(b)

| Element | Atomic % | Wt% | Wt% Sigma |
|---|---|---|---|
| O | 33.7 | 22.45 | 0.45 |
| Si | 66.3 | 77.55 | 0.45 |
| Total: | 100 | 100 | |

**Fig. 5 - EDX Characterization of pSi structure. (a) the spectrum confirms the elemental composition of material existing in the porous structure; (b) the percentage of the atomic and weight for materials exist in the porous structure**

Fig. 5 displays the EDX characterization for pSi structure. The spectrum in Fig. 5 (a) illustrates the elemental composition of pSi structure. The pSi structure consists of silicon (Si) and Oxygen (O). Si is the original chemical element, while O is produced from the electrochemical etching process. Table b in Fig. 5 shows the atomic and weight percentage of the materials. The atomic percentage of O and Si is 33.7% and 66.3%, while the weight percentage is 22.45% and 77.55% respectively. For different etching time, almost similar composition is observed.

## 4.2 Electrical Properties of Psi for Different Etching Time

Electrical characterization in I-V curve of materials is important for materials selection and processing during the design of a component or structure. Fig. 6 (a) shows the I-V curve graphs of the pSi samples at different duration of etching time. The etching time starts with 20 minutes is labeled as t-20. The other etching time is labeled as t-22, t-24, t-26, t-28 and t-30. With reference to the figure, all the samples exhibit exponential increase in current for voltage from 0 to 10 V, resembling the property of a diode. This is expected since pSi is a semiconductor material. It is also observed, as etching time increases from 20 to 28 minutes, the graphs show incremental trend on the current values starting from 20 μA to 200 μA. t-28 sample gives the highest current (~200.29 μA) compared to other samples. Interestingly, lower current values are observed for t-30 sample, where some of the cross-shaped porous structures start to connect with neighbouring cross-shaped structures as illustrated in Fig. 3 (f).

Fig 6 (b) depicts the conductivity and resistivity versus different etching time setting. Conductivity is reciprocal of resistivity as displayed. All the pSi samples behave like semiconductor materials, where conductivity increases while the resistivity decreases with etching time, from 20 to 28 minutes. However, as the etching time increases more than 30 minutes, the conductivity performance is slightly reduced. Higher conductivity is expected as the porosity of pSi increases with etching time duration. Higher porosity means larger surface area that allows more free electrons to produce higher current from the n-type silicon doped with phosphorus dopant. The reduction in conductivity performance for t-30 might be due to the uneven structures produced, as discussed in the previous section.

Fig. 6 (c) illustrates the sensitivity property of the pSi structure with different etching time setting. The sensitivity values are extracted from I-V curve at 10V for every sample. The slope, *m*, is calculated from the best-fit line through all the etching time samples. The slope of current versus etching time is ~16.72 μA/min. The graph shows sensitivity for all samples increasing linearly with etching time.

From Fig. 6 (c), t-20 is selected as optimal etching time for fabrication process. It is located nearest to the best fit line, making it the most sensitive. Also, at this etching time, the triangle opening is smaller compared to the other samples (see Table 1). Another choice is t-22 which is located second nearest to the best fit line. t-24, t-26 and t-30 situates away from the best fit line and hence are not suitable. It can be concluded from the sensitivity data, the optimal etching time to produce porous structure ranges from 20 to 22min, obtained base on the minimal distance from the line of best-fit, which is close to the slope.

## 5. Conclusion

With reference to Fig. 3, Fig. 4, Fig. 5 and Fig. 6, the effects of etching time on pSi as SERS substrates for non-invasive dengue detection, in term of structural and electrical characterization are analyzed. The etching time contributes to the porosity of Silicon material structure. From electrical characterization, etching time equal and less than 28 minutes is found to produce exponentially increasing I-V graphs and cross-shaped porous with more uniform surface structures. This supports our statement that etching time of 28 min and below is more suitable to produce cross-shaped porous with a more uniform porous structure [17]. Amongst all, etching time of 28 minutes yields the highest current value. Fig. 3 (e) and Fig. 4 (e) present the optimal pSi structure for SERS substrate of our application. The range limits of etching time for pSi structure production are also identified experimentally. The range of 20 to 28 minutes of etching time setting is found optimal for production of pSi structure for SERS substrate or biosensors. For future work, the pSi structure coated with AgNPs will be tested with suitable Raman probe to investigate the Raman peak intensity based on the substrate structure; enhancement for different structure layers in SERS substrate will also be investigated.

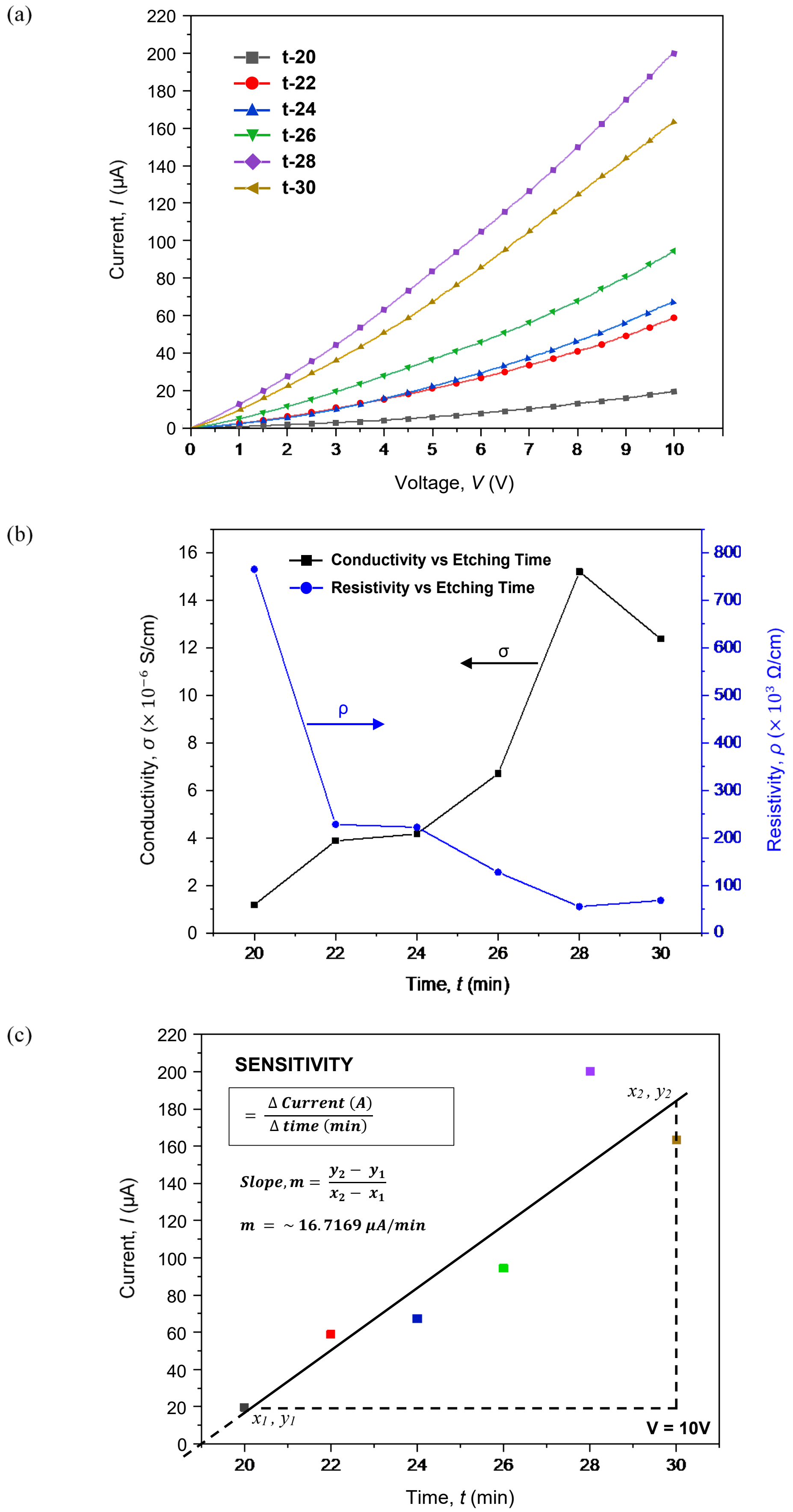



**Fig. 6 - Electrical Characterization for pSi structure. (a) I-V curve of the pSi structure with different etching time setting; (b) Conductivity and resistivity properties versus etching time; (c) Sensitivity property of pSi with different etching times**

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