

Thesis progression

Thesis title: Comparison of SERS Performance for Standard Molecules and Clinical Specimens.

Thesis progression title: Introduction to Comparison of SERS Performance for Standard Molecules and Clinical Specimens.

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1. Introduction

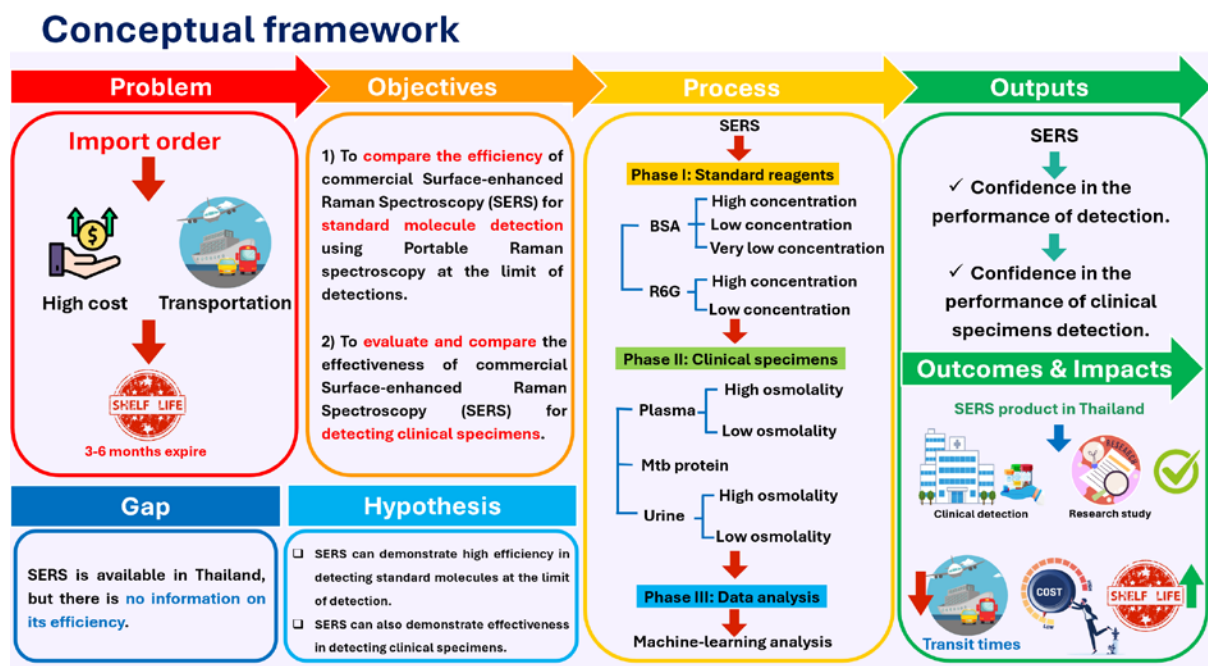
Surface-enhanced Raman spectroscopy (SERS) is a powerful analytical technique for molecule detection that can investigate conformational changes in complex biological molecules with enhanced Raman scattering, allowing for the detection of molecules even at low concentrations. The enhancement of Raman scattering involves two mechanisms: typically, strong electromagnetic enhancement and chemical enhancement (Aitekenov et al., 2022; Eiamchai et al., 2024). Recently, there have been many achievements in the research, publication, and application of SERS in various fields, including molecular biology, biomedicine, and environmental science (Sultangaziyev et al., 2022). Hopefully, the SERS method will become a promising tool for future routine clinical tests due to its speed, cost-effectiveness, and high accuracy.

Since the development of this technique by Fleischmann et al. in 1974, SERS has been demonstrated in many research publications, particularly in medical research, including studies on cancer (Issatayeva et al., 2024; Joseph et al., 2018), renal disease (Aitekenov et al., 2022; Kukkar et al., 2023), liver disease, dengue fever (Chen et al., 2019), Alzheimer's disease (D'Andrea et al., 2023), and tuberculosis (Botta et al., 2018; Crawford et al., 2017; Eiamchai et al., 2024; Kaewseekhao et al., 2020) with high accuracy. Despite these achievements, only a few SERS substrates have been commercialized. However, SERS has not yet been

successfully adopted as a standard clinical diagnostic tool due to limitations such as the need to import SERS substrates from abroad in some countries, leading to high costs, long transportation times, and a short shelf-life of 3-6 months.

In Thailand, SERS substrates are available, but there is no information on their efficiency. Therefore, SERS chips must be evaluated in terms of sensitivity, specificity, accuracy, and other factors related to detection performance to overcome the limitations of current SERS substrates. This study aims to compare the efficiency of five commercial SERS substrates using standard molecules and clinical specimen detection with Portable Raman spectroscopy.

2. Conceptual framework



3. Objectives

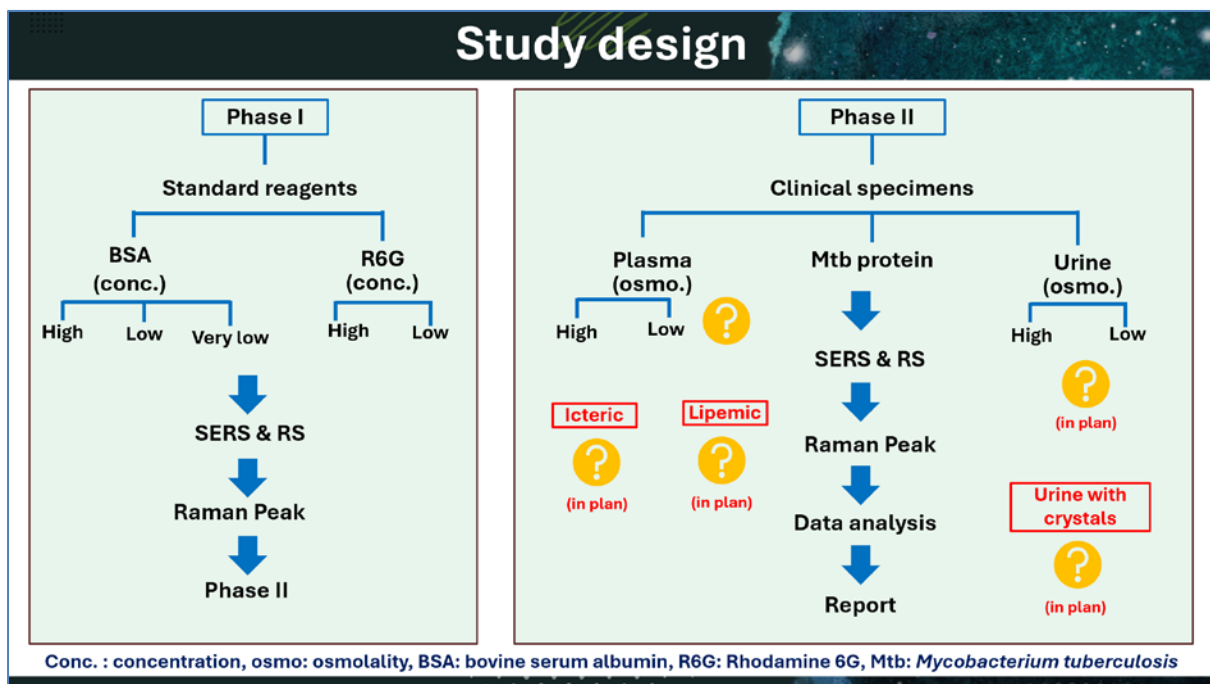
3.1 To compare the efficiency of commercial Surface-enhanced Raman Spectroscopy (SERS) for standard molecule detection using Portable Raman spectroscopy at the limit of detections.

3.2 To evaluate and compare the effectiveness of commercially available Surface-enhanced Raman Spectroscopy (SERS) for detecting clinical specimens.

4. Hypothesis

- 4.1 Commercial Surface-enhanced Raman Spectroscopy (SERS) can demonstrate high efficiency in detecting standard molecules at the limit of detections.
- 4.2 Commercial Surface-enhanced Raman Spectroscopy (SERS) can demonstrate effectiveness in detecting clinical specimens.

5. Study design



6. Materials and methods

- 6.1 Comparison of commercial SERS for standard molecule detections.

To compare the efficiency of SERS substrates for standard molecule detection at the limit of detection, SERS measurements will be performed using a portable Raman mapping spectrometer (NECTEC, TH). Two types of standard reagents, Bovine Serum Albumin (BSA) and Rhodamine 6G (R6G), will be used. BSA will be tested at high (1 $\mu\text{g}/\mu\text{l}$), low (1 $\text{ng}/\mu\text{l}$), and very low (1 $\text{pg}/\mu\text{l}$) concentrations. R6G will be tested at 10^{-2} M (47.902 $\text{ng}/\mu\text{l}$) for high concentration and 10^{-5} M (47.902 $\text{pg}/\mu\text{l}$) for low concentration. Then, 2.0 μl of each reagent

will be individually dropped onto the SERS chips and left to air-dry for 10 minutes. A Raman spectrum will be collected from a central region on each SERS chip using a 785 nm wavelength laser with 25 mW laser power and a 10-second exposure time. For each sample, the spectral data will be taken from 49 points covering a grid area of 7 x 7 with 15 µm steps.

6.2 Comparison of commercial SERS for clinical specimen detections.

To evaluate and compare the efficiency of SERS substrates for clinical specimen detection, SERS measurements will be performed using a portable Raman mapping spectrometer (NECTEC, TH). Plasma samples with high and low osmolarity, Mtb (H37Ra) protein, and urine samples with high and low osmolarity will be used. Then, 2.0 µl of each reagent will be individually dropped onto the SERS chips and left to air-dry for 10 minutes. A Raman spectrum will be collected from a central region on each SERS chip using a 785 nm wavelength laser with 25 mW laser power and a 10-second exposure time. For each sample, the spectral data will be taken from 49 points covering a grid area of 7 x 7 with 15 µm steps.

7. Progression

In this progression, I have not yet started the laboratory section. However, I have completed several preparatory steps for this thesis. These include passing Biomedical Research Ethics training, Biosafety and Biosecurity for BSL-II training, establishing direct contact with SERS commercial suppliers to request quotes for SERS chips, studying SERS fabrication techniques, participating in the conference "Spectroscopic AI: Health, Agriculture, and Food Science Applications," and completing training on the Portable Raman Mapping System. For the next steps, I will focus on proposal examinations, obtaining ethical clearance (EC), and funding requirements.

8. Thesis plan

Activities	2024										2025			
	4	5	6	7	8	9	10	11	12	1	2	3	4	
Literature review and planning	■	■	■	■	■	■	■	■	■	■				
Proposal writing	■	■	■	■										
Proposal examinations					■	■	■							
EC and funding requirement					■	■	■							
RS and SERS parameter setting up						■	■	■	■					
Sample measuring						■	■	■	■					
Data analysis						■	■	■	■					
Manuscript preparation										■	■	■		
Thesis defense examinations and publish research												■	■	

9. References

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