Fabrication of SERS Substrates for Molecular Detection

Summer Internship Report

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Om Tambe

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CHAPTER 1 INTRODUCTION

The use of non-permitted colours on food items is a growing menace faced by Indian consumers. Artificial colours can also be used to enhance the colour tone of the old stock of vegetables and pulses and make them look fresh. Such synthetic colourants have even been detected in packed foods and beverages [1]. Surface Enhanced Raman Spectroscopy (SERS) is an analytical technique that is well suited for point of use chemical analysis. Surface Enhanced Raman Spectroscopy (SERS) is a capable technique for rapid, on field detection of Raman spectra of molecules. The vibrational signature encoded in the Raman spectra can be used as a unique identifier of molecular structure. A plasmonic nanostructured substrate is required for amplification, by several orders of magnitude, of the weak Raman signal (almost exclusively emitted from the adsorbed molecular layer closest to the surface), and such 'surface-enhancement' renders SERS capable of detecting the presence of a single molecule. Raman Spectroscopy is concerned with the scattered radiation from a sample. Scattering refers to a change in the direction of light as it propagates through a medium. The power of SERS lies in its ability to identify chemical species and obtain structural information in a wide variety of fields including polymer and materials science, biochemistry and biosensing, catalysis, and electrochemistry. SERS is a highly sensitive and selective technique for use in the detection of biological samples. The implanted sensors are highly accurate and consistent, while also able to accurately measure low concentrations of blood glucose. The fabrication of SERS active substrates is a new method for the detection of toxic substances, narcotics, industrial toxic chemicals, biological analytes, and the analysis of other biological and non-biological species at low concentrations. Metals such as silver (Ag), gold (Au), copper (Cu), or aluminium (Al) are known to have excellent thermal and electrical conductivity due to the presence of a 'sea' of free conduction electrons that move around in a background of the positive ions ensuring the overall charge neutrality. The collective behaviour of these free conduction electrons is modelled as a single entity and denoted as plasma.

The goal of this research was to develop optimal parameter for the use of Au, Ag, Au-Ag (core-shell) SERS substrates for onsite detection of adulterants from food items. This reports implies to identify the detection of two commonly used adulterants, namely 'Metanil Yellow (MY)' from the surface of pulses, and 'Malachite Green (MG)' from the skin of vegetables using a portable Raman Spectrometer from Snowy Range Instruments (SnRI) with varying concentrations and

time of deposition nanoparticles as well as dye used. The long-term specific objectives were the detection of synthetic colourants on SERS substrate (fabricated with optimal parameters) used in the adulteration of eatables using a portable Raman Spectrometer.

CHAPTER 2.1 RAMAN SPECTROSCOPY AND SERS: OVERIVEW

Raman spectroscopy is a rapid, non-destructive vibrational spectroscopy used for identification and quantification of chemical composition. Raman spectroscopy is based on scattering; on the basis of light and matter interaction, scattering can be categorized into 2 parts: Elastic scattering & Inelastic scattering. Elastic scattering is also called as Rayleigh scattering. Inelastic scattering/ Raman scattering is further divided into Stokes and Anti-Stokes scattering.

When radiation having an energy **h.υ** is incident on a matter/ sample it is considered that the incident photons undergo collisions with the molecules. When the collision is elastic, the photons will go deflected unchanged i.e. when hit by a photon after excitation it returns to it ground state, then the scattering is called as Rayleigh scattering. But it is also possible that during the collision, energy is exchanged between the photon & molecule, and as a result the molecule can gain or lose energy ΔE, $\Delta E = h.\upsilon_{vib}$.

Scattering can be considered as an excitation to a virtual state lower in energy than a real electronic state. When the molecule gains energy ΔE, the photons will be scattered with energy, $h.\upsilon - h.\upsilon_{vib}$, then, the scattering is known as Stokes scattering and if the molecule lose energy ΔE, the scattered photon will have energy, $h.\upsilon + h.\upsilon_{vib}$ then, the scattering is known as Anti - Stokes scattering.

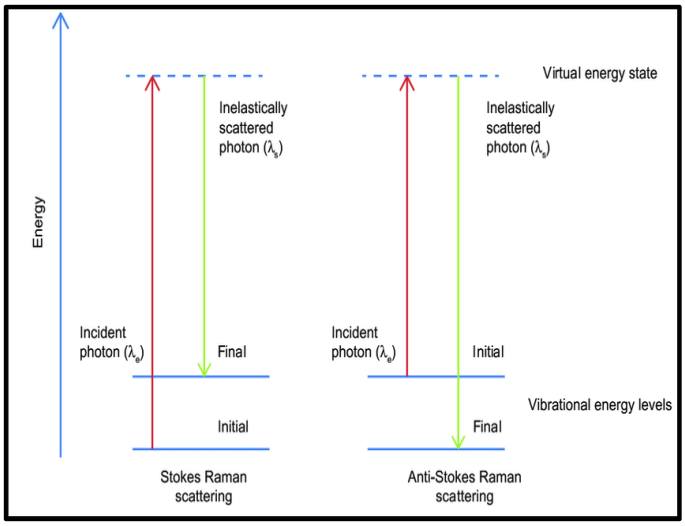


Fig.2.1.1: Schematic showing the difference between Stokes and anti-Stokes

Energy level: anti-Stokes > Rayleigh > Stokes

Lambda: Stokes > Rayleigh > anti-Stokes

A plasmonic nanostructured substrate is required for amplification, by several orders of magnitude, of the weak Raman signal (almost exclusively emitted from the adsorbed molecular layer closest to the surface), and such 'surface-enhancement' renders SERS capable of detecting the presence of a single molecule.

'Surface-enhancement' renders SERS capable of detecting the presence of a single molecule.

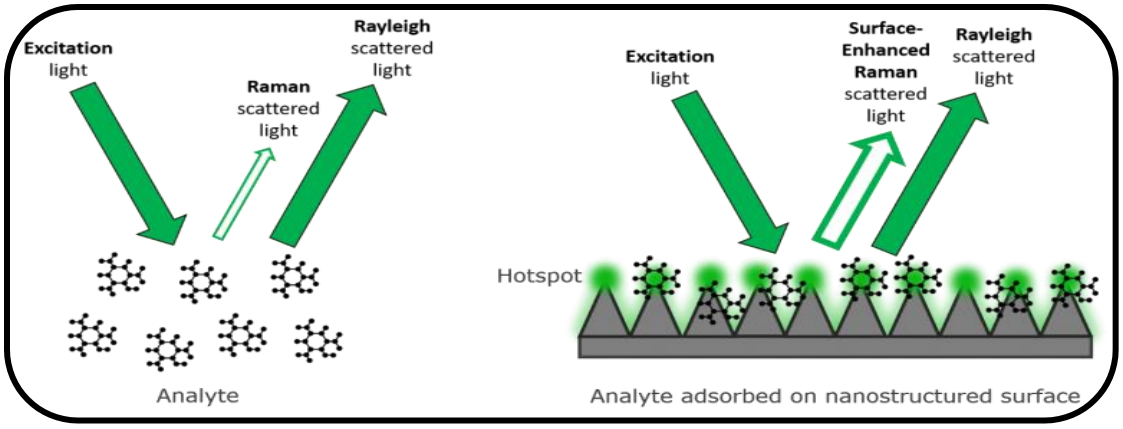


Fig.2.1.2: Laser-Analyte-Nanostructure interaction

Image Ref: https://sway.office.com/XtgAoh8F5QewSEFL*

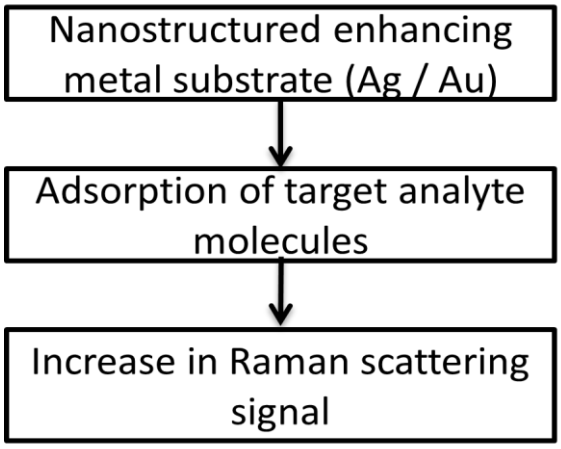


Fig.2.1.3: Schematics of SERS signal enhancement

The surface features of nanoparticles has lead to improved enhancement of Raman scattering signal of target molecules due to the increased number of sharp surface ability possessing multiple localized surface plasmon resonances (LSPR). Sometimes intuitive approach of increasing the number of nanoparticles to increase the signal intensity may prevent molecules from adsorbing on plasmonic nanoparticles because of nanoparticle aggregation, limiting the ability for the molecule's signal to be enhanced. SERS signal will be strongly affected by the presence of mono- or multi-layers of the analyte molecule in the sample and by the method of analyte deposition. However, if sample preparation is performed identically and dye concentration is limited to provide for sub-monolayer coverage.

A decrease in nanoparticle concentration decreases the SERS activity and complex morphologies increases SERS activity. Few applications of SERS are Pathogen detection; in forensics; in detection of metabolites in bodily fluids e.g. glucose, urea etc., and manufacturing quality check of food substances.

2.2. Factors affecting SERS

Parameters affecting SERS Enhancement

1. Geometrical shape: anisotropic metallic nanoparticles with complex shapes (e.g., hexagonal, triangular, cubical, star shape), could be beneficial due to the more intense near-fields at the sharp edges. Anisotropic nanoparticles are non-spherical structures (e.g., prisms, rods, cubes) which have shape-dependent chemical and physical properties and can be utilized in important applications spanning catalysis to sensing to optics.
2. Particle size: When the particles are too small, both the actual conductivity and the scattering properties are reduced, which expectedly reduces the SERS efficiency. As the particle size increases, the SERS effect increases since they act as larger scattering centres.
3. Interparticle distance: The closer the particles are to each other, the higher the specific surface coverage of their active surfaces is, and the surface density of hot-spots (number of areas with increased near field intensities due to coupled plasmons between the particles) is also higher, which altogether results in an improved SERS efficiency.
4. Excitation wavelength: the wavelength of the excitation source should match the plasmonic properties (LSPR excitation range) of the substrate as close as possible. The LSPR absorbance wavelength shifts to red with larger nanoparticle size and smaller interparticle distance (due to the plasmonic coupling). If the excitation wavelength is far from the plasmon excitation of the substrate, the SERS enhancement would be reduced.

The effect of material type, particle size, interparticle distance (which altogether determine the plasmonic properties of the substrates) and the excitation wavelength is directly dependent on SERS enhancement. The closer the particles are to each other, the higher the specific surface coverage of their active surfaces is, and the surface density of hot-spots (number of areas with increased near field intensities due to coupled plasmons between the particles) is also higher.

CHAPTER 3 PREPARATION OF PAPER-BASED AQUEOUS SILVER SERS SUBSTRATES

3.1. Introduction

Metallic nanomaterials have dimensions in size range of 1 – 100 nm and they exhibit several superior and fascinating properties as compared to bulk making them promising candidates for commercial applications. Nanometric particles or nanowires have attracted great interest in various fields of engineering due to its well known properties like having large surface to volume ratio, have size and shape tunable optical properties. A simple way of synthesizing silver nano structures called the Print-Expose-Develop technique.

Materials

Silver nitrate, potassium bromide, potassium iodide, potassium chloride, metol, hydroquinone, sodium sulphite, anhydrous borax salt were of AR grade. DI water was used in the experiment. Printing paper – A4 size was used as a substrate for inkjet printing. HP (1200 series) deskjet printer with 802 cartridges was used for printing precursor salt solutions.

Preparation of Salt Solution

DI water was used to prepare all the solutions. The molar ratio of KX to $AgNO_3$ printed was 2 : 1, to ensure complete conversion to AgX on the paper. For the results reported here, a bromide : iodide :: 95 : 5 wt% composition was used as the halide (X) source, as addition of iodide ions enhances photosensitivity. Typically, 4 M KX and 2 M $AgNO_3$ solutions (observed silver nitrate loading of 0.5 mg/cm^2 after seven prints) were used to prepare the samples reported here. The developer solution was prepared according to a standard recipe for making D-76, as of, 10 g of sodium sulphite was dissolved in 75 mL hot water (50-55°C) and then 0.2 g of metol (monomethyl-p-aminophenol hemisulphate) along with 0.5 g of hydroquinone were added and mixed thoroughly. To this solution, 0.2 g of borax was added. Finally, cold water was added to make the solution volume 100 ml.

Fig.3.1.1: Developer solutions – D-72, D-78, D-76

3.2. Print- Expose- Develop Method

Photographic processing is one such metallization scheme that relies on forming silver halide layers on a substrate, exposing silver halides to light to form a latent image, followed by 'development' in an appropriate chemical bath to amplify the latent image.

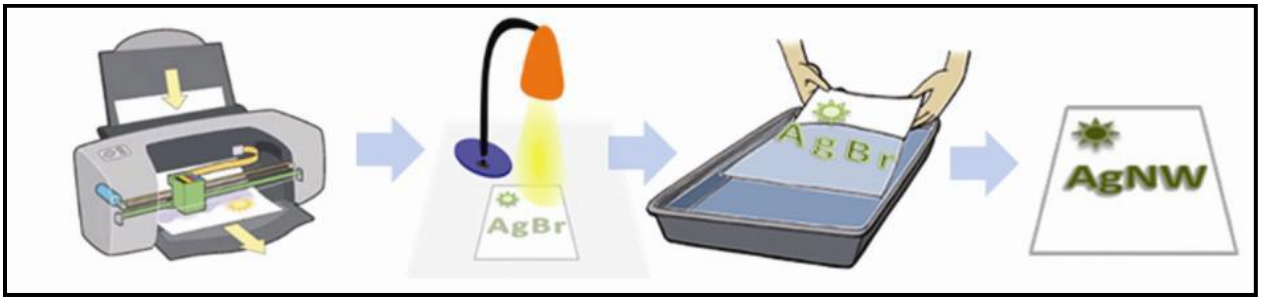


Figure 3.2.1: Schematic representation of the print–expose–develop cycle[2]

Printing

In this step the precursor for the synthesis of silver halide i.e. $AgNO_3$ and KBr-KI (95:5%) is printed on the desired substrate in layer by layer manner using an HP desktop inkjet printer, to form the required patterns. The silver and halide salt solutions react on the substrate as given below: $AgNO_{3\,(aq)} + KX_{(aq)} \rightarrow AgX_{(aq)} + KNO_{3\,(aq)}$

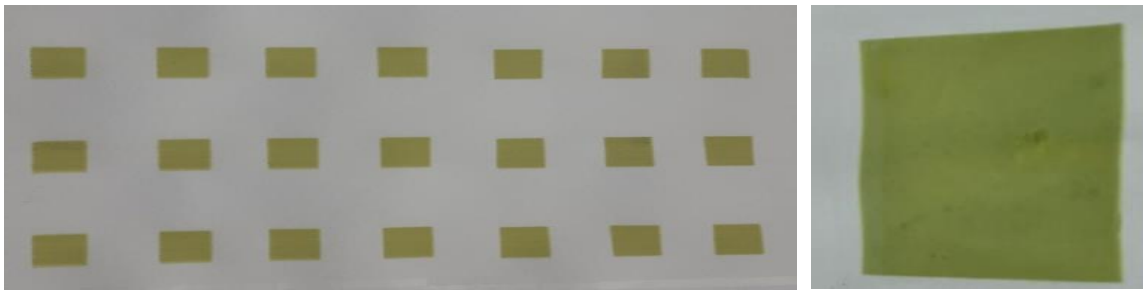


Fig.3.2.2: Silver halide salt after printing

Exposure

The printed substrate is exposed under a halogen lamp for 15 minutes for the formation of the latent images. A latent image is a small cluster of metallic silver atoms formed in or on a silver halide crystal due to reduction of interstitial silver ions by photoelectrons which are generated while exposing under the halogen lamp. The size of a silver cluster in the latent image can be as small as a few silver atoms. However, in order to act as an effective latent image center, at least four silver atoms are necessary[2]. A slight yellowish thin film of the patterns is observed after the exposure leading to the visual confirmation of formation of latent image on the substrate.

Fig.3.2.3: Exposure to halogen lamp for photo-reduction of Silver halide salt

Development

The exposed substrate containing thin film of AgX is then immersed in the developer solution bath. A developer solution converts silver halide crystals to metallic silver grains, but it acts only on those having latent image centers. This conversion is due to electrochemical reduction, wherein the latent image centers act as a catalyst.

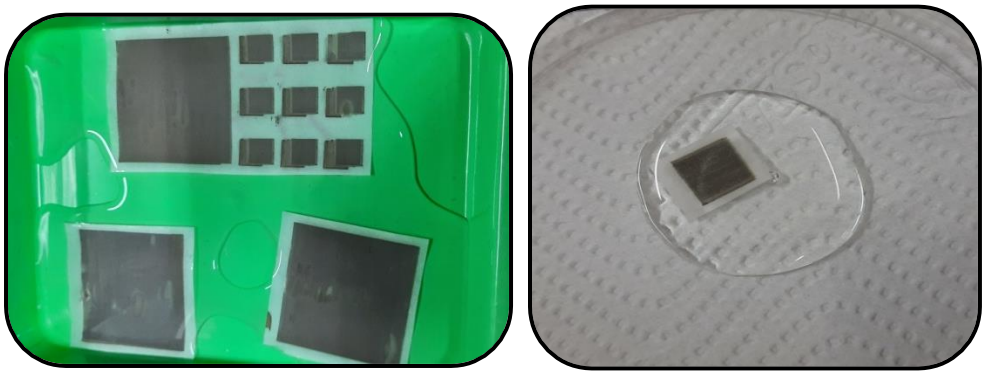


Fig.3.2.4: Development of Silver nanostructures in developer solution to obtain latent image.

Latent Image formation

The nucleation of the atomic silver from the silver halide crystal; which are called as the latent image centers. The stepwise formation and growth of the latent image silver specks as a result of light absorption is assumed as follows[3]:

$$Ag^{+}\,i + e^{-} \rightarrow Ag$$

$$Ag + Ag^{+}\,i + e^{-} \rightarrow Ag2$$

$$Ag + Ag^{+}\,i + e^{-} \rightarrow Ag3$$

$$Ag + Ag^{+}\,i + e^{-} \rightarrow Ag4$$

The smaller sized specks of silver (Ag, Ag2, Ag3) are termed as the pre-latent center image and sub latent center image respectively depending on the number of silver atoms present in the speck. The pattern printed on the substrate consists of photosensitive crystals containing primarily silver bromide with some amount of silver iodide. These silver halide crystals also contain a few free silver ions (interstitial silver ions) in the spaces between the crystalline lattice atoms. These interactions of photons from the halogen lamp with the bromide ions present in the crystal result in the removal of an electron from the bromide ions. By the loss of an electron, a bromide ion is converted into a neutral bromine atom. The free electrons move through the crystal until they reach a sensitivity site, where they become trapped and impart a negative charge to the site. The negatively charged sensitivity site then attracts positively charged free interstitial silver ions. When a silver ion reaches the negatively charged sensitivity site, it is reduced and forms a neutral atom of metallic silver (Ag). The sites containing these neutral silver atoms are now called latent image sites. This process occurs numerous times within a crystal. The overall distribution of latent image sites in a film after exposure constitutes the latent image.

Mechanism of photographic development

Photographic development is a process that amplifies a latent image formed. The development process is based on a redox reaction that involves silver halide grains and developing agents with the aid of the catalyzing effect of latent image centers. The initiation of development of a latent image involves electron transfer from developing agents to latent image centers. Silver clusters grow through capture of an electron and an interstitial silver ion one after the other.

In the case of growth of silver clusters during exposure, electrons are provided by silver halide grains as a result of light absorption of grains. In the case of development, electrons are provided by developers, since a latent image center is such a deep electron trap as to readily accept an electron from a developing agent in a developer solution. In the first step of the development processes, a development center on a silver halide grain receives an electron from a developing agent in a developer which is an electronic process and consequently the center attracts and combines with an interstitial silver ion which is an ionic process. The repetition of the above stated electronic and ionic processes finally reduces all the silver ions in a grain to silver atoms, releasing halide ions into a developer solution.

3.3. Loading Calculation

- Preparation of 2M $AgNO_3$ solution

  Volume to be prepared = 5 mL Mol. Weight = 169.87 g/mol

  Calculated Mass = 1.6987 g of $AgNO_3$

  No. of prints: 7

  Printing pattern: *KKKK AAAAAAA KKK*

  Print setting: Glossy paper – Best quality

Sr. no.	Readings	∆D= D^{o}th – Dn	∑ Readings	Avg = ∆D/n
0th	**23.6212**			
1	**23.4979**	**0.1233**	**0.1233**	
2	**23.4023**	**0.2189**	**0.10945**	
3				
4	**23.6012**	**0.02**	**0.006666667**	
5				
6				
				0.065200333
7	**23.5003**	**0.1209**	**0.030225**	
8				
9				
10				
11	**23.3394**	**0.2818**	**0.05636**	
12				
13				
14				
15				
		SUM	**0.326001667**	

Table 3.3.1: Loading calculations for Ag Nanostructures for 7 prints

Entity to be printed: Square

Dimensions: Length = 1 cm Breadth = 1cm **Area = 1 cm**

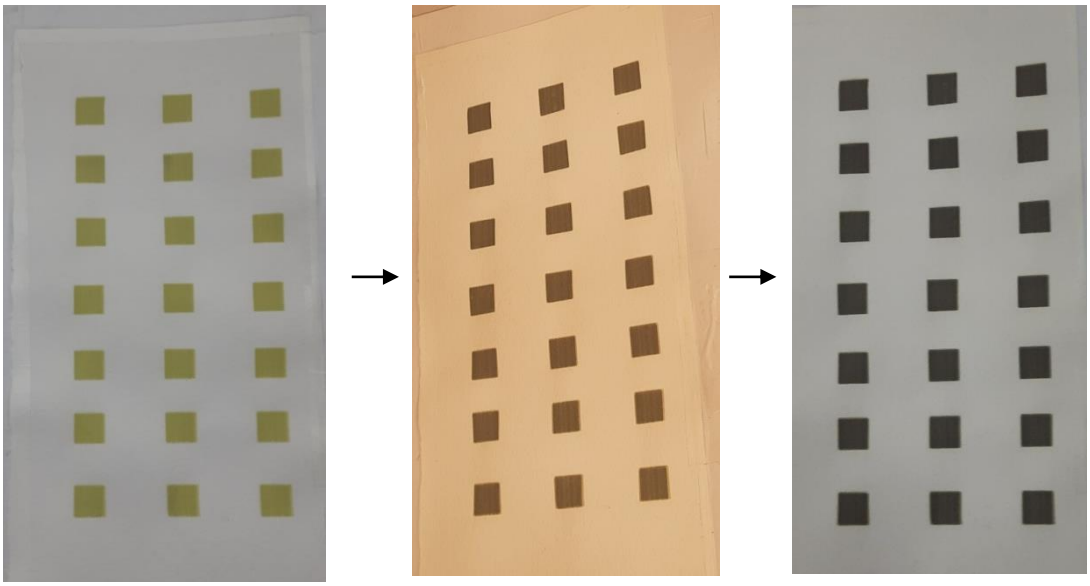
$Volume\ (in\ mL) = Mass/Density of\ H_2O$

= (0.0652003 / 1) * 0.001 = 6.52003 E-05 L

Moles = 0.000130401

Mass of Ag = 0.000130401 * 107.8682 = 0.014066085

Deposition/Area = 0.000562643 gm/cm^2 = 0.5 mg/cm^2



Print Exposure Development

Fig.3.3.1: Represents Ag NS's after photographic processing on 1 cm^2 – Squares – 7 prints

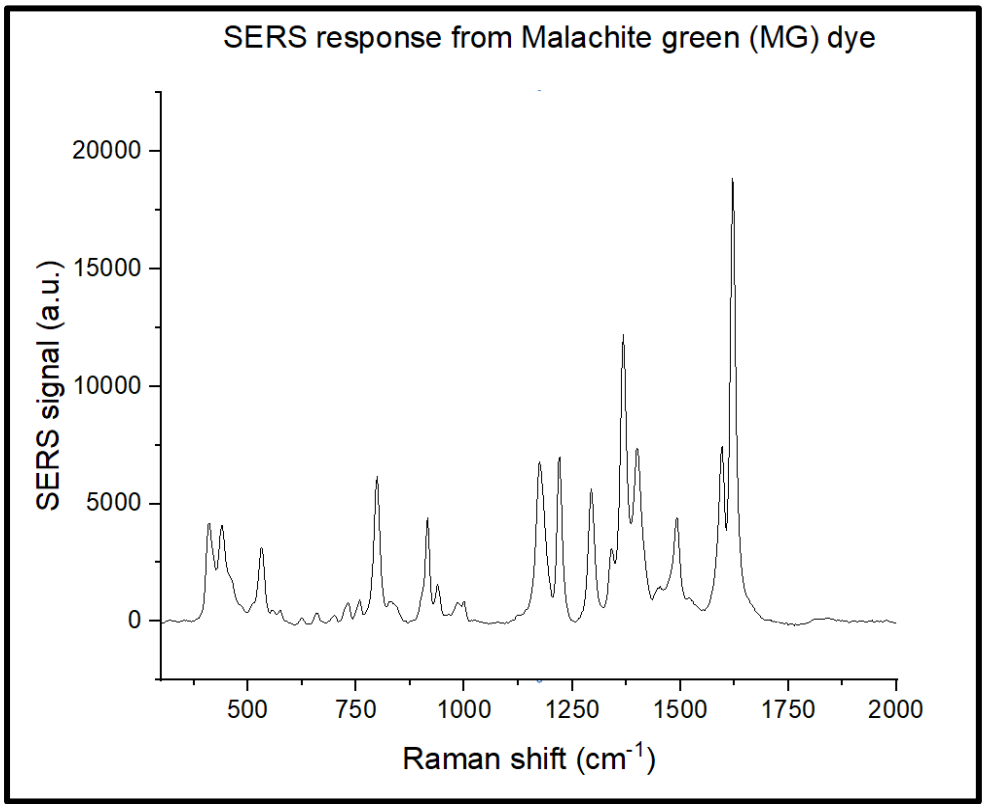


Fig. 3.3.2: SERS response of 1mM Mg dye on Ag (printed).

3.4. Aim: To check whether the Silver Halide Salt is printed throughout the substrate i.e. Chromatography paper and bypasses the cross-section thoroughly by changing precursor solutions having less surface tension or changing viscosity of solutions too.

I. With $AgNO_3$ in H_2O: C_2H_5OH in 30:70 (%v/v) solution; alongwith, KBr.KI in 80:20 (%v/v) H_2O: C_2H_5OH solution.

Printing, Exposure and Development of freshly prepared $AgNO_3$ in H_2O: C_2H_5OH in 30:70 (%v/v) solution. Alongwith, KBr.KI in 80:20 (%v/v) H_2O: C_2H_5OH solution.

		Molarity (M)	Volume (mL) – H2O: EtOH	Calc. Mass(mg)
$AgNO_3$		**2 (1.9047 ± 0.001)**	**2.1 (30:70) [0.7: 1.4]**	**679.5 ± 0.1**
95	**KBr**	**3.8094 ± 0.001**	**2 (80: 20)**	**861.3 ± 0.1**
5	**KI**			**63.2 ± 0.1**

Table 3.4.1: Molarity calculations for Ag Nanostructure's for 7 prints- H_2O: EtOH

Printing pattern: *AAAA KKKK AAA KK*

No. of prints: 7 prints

Print setting: Glossy paper – Best quality

D^oth Value = 23.3506 gm

D^nth value	Difference	Avg (gm)
23.2376	**0.1130**	**0.3614**
23.1128	**0.2378**	
22.9800	**0.3706**	
22.9133	**0.4373**	
22.8958	**0.4548**	
22.8937	**0.4569**	
22.8914	**0.4592**	
	SUM = 2.5296	

Table 3.4.2: Loading calculations for Ag Nanostructure's for 7 prints- H_2O: EtOH

Entity to be printed: Square

Dimensions: Length = 5 cm Breadth = 5 cm **Area = 25 cm^2**

$$Volume\ (in\ mL) = Mass/Density$$

= 0.3614 / [(0.3*1) + (0.7*0.789)] * 0.001 = 4.24 E-05 L

Moles = 0.00008076

Mass of Ag = 0.00008076 * 107.8682 = 0.0087112

Deposition/Area = 0.0003484 gm/cm^2 = 0.3484 mg/cm^2

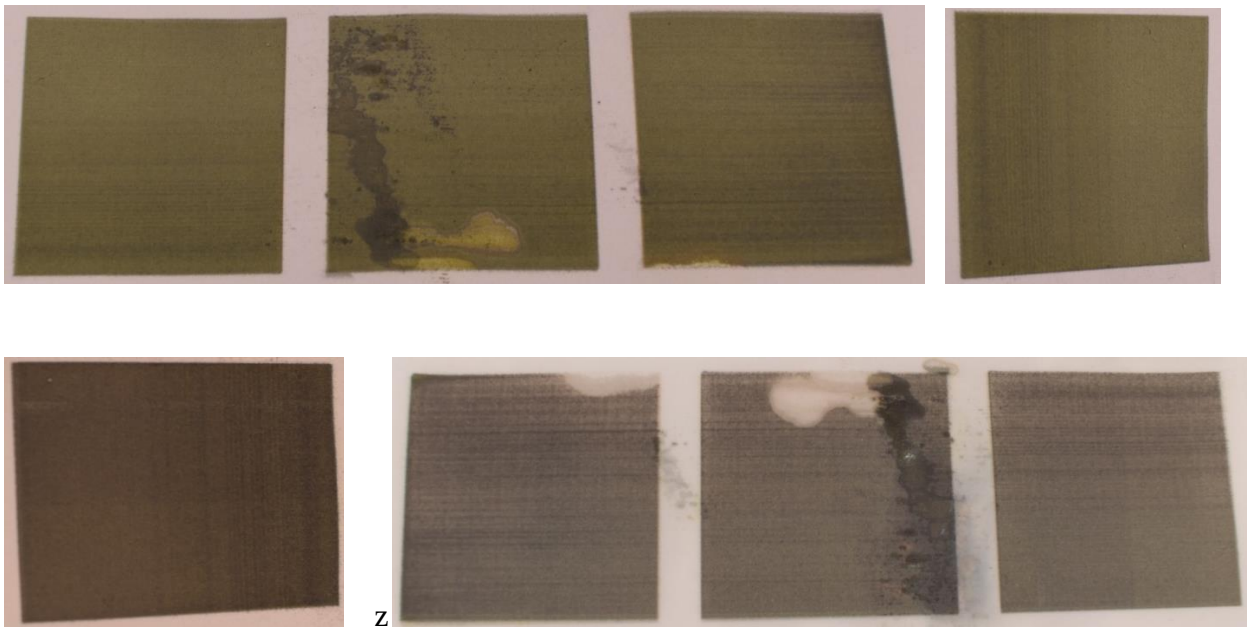


Fig.3.4.1: Ag NS's after printing (slight yellowish film can be seen), exposure and development.

II. With $AgNO_3$ in 5:4:1 (%v/v) H_2O: Glycerol: C_2H_5OH (%v/v) solution; alongwith, KBr.KI in 5:4:1 (%v/v) H_2O: Glycerol: C_2H_5OH solution.

Printing, Exposure and Development of freshly prepared $AgNO_3$ in 5:4:1 (%v/v) H_2O: Glycerol: C_2H_5OH (%v/v) solution. Alongwith, KBr.KI in 5:4:1 (%v/v) H_2O: Glycerol: C_2H_5OH solution.

		Molarity (M)	**Volume (mL) – H2O: Glycerol: EtOH**	**Calc. Mass(mg)**
$AgNO_3$		**2 (1.904 ± 0.001)**	**2.6 (5:4:1) [1.6: 0.8: 0.2]**	**679.5 ± 0.1**
95 : 5	**KBr**	**3.809 ± 0.001**	**2 (5: 4: 1)**	**861.3 ± 0.1**
	KI			**63.2 ± 0.1**

Table 3.4.3: Molarity calculations for Ag Nanostructure's for 7 prints- H2O: Glycerol: EtOH

Printing pattern: *AAAA KKKK AAA KK*

No. of prints: 7 prints.

Print setting: Glossy paper – Best quality

D^0th = 23.5260 gm D^nth = 23.4203 gm*

D^nth value	**Difference**	**Avg (gm)**
23.4182	**0.1078**	**0.2165**
23.3502	**0.1758**	
23.2284	**0.2976**	
23.1613	**0.3647**	
23.3459*	**0.0744**	
23.2135	**0.2068**	
23.1316	**0.2887**	
	SUM = 1 .5158	

Table 3.4.4: Loading calculations for Ag Nanostructure's for 7 prints- H2O: Glycerol: EtOH

Entity to be printed: Square

Dimensions: Length = 5 cm Breadth = 5 cm **Area = 25 cm^2**

Density = Mass of solution in glass vial / Volume taken = 1.6471 / 2.6

= 0.6335 gm / cm^3

$Volume\ (in\ mL) = Mass/Density$

= 0.3614 / [(0.3*1) + (0.7*0.789)] * 0.001 = 4.24 E-05 L

Moles = 0.00006836

Mass of Ag = 0.00006836 * 107.8682 = 0.007374 gm

Deposition/Area = 0.0002949 gm/cm^2 = 0.295 mg/cm^2

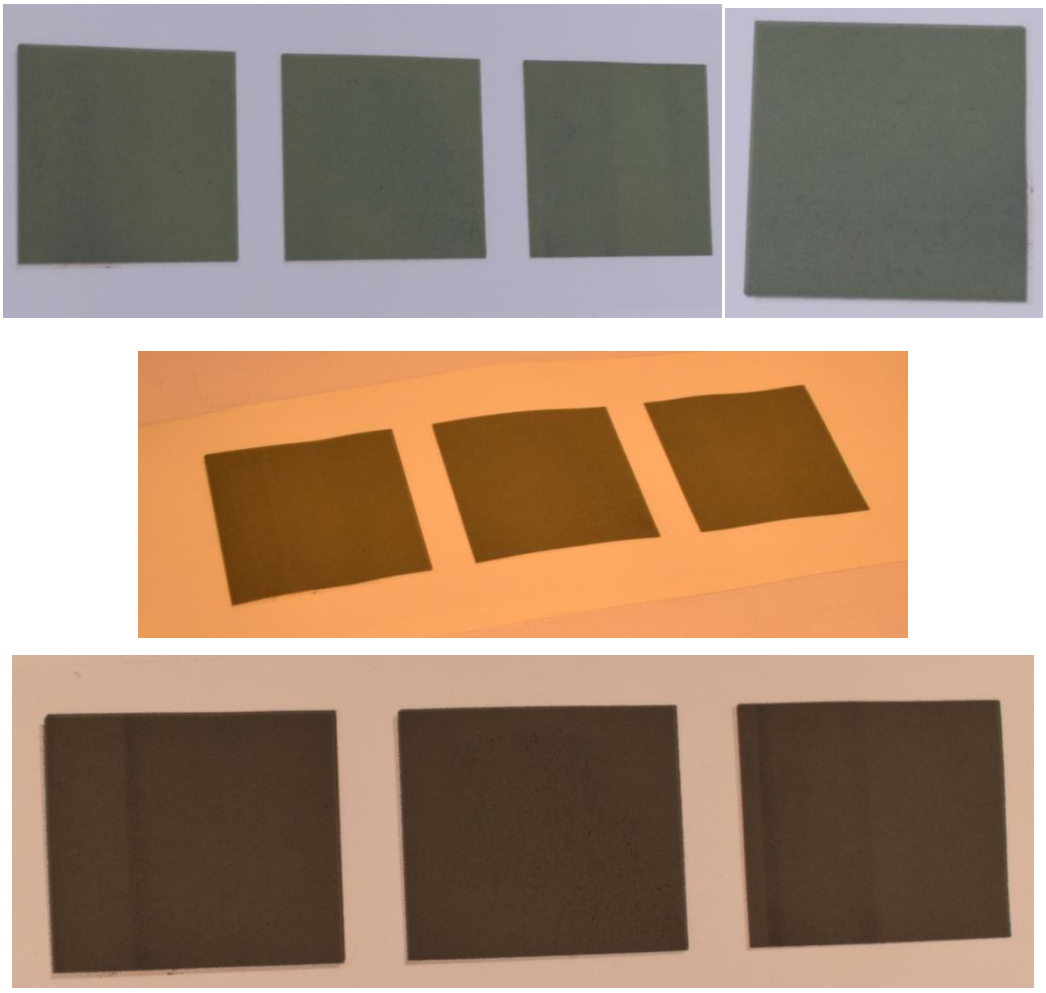


Fig.3.4.2: Print-Exposure-Development of Silver halide salt on Chromatography paper

3.5. Results

Comparison of backside of substrates of $AgNO_3$ printed using different precursor solutions.

1

Image (1).Ag NS's printed with H_2O on $1cm^2$ - squares

2

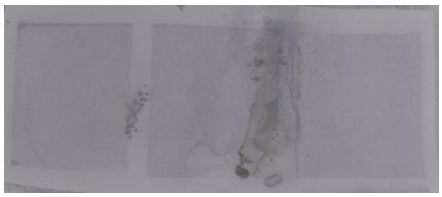


Image (2).Ag NS's printed with H_2O & EtOH on 5 cm^2 - squares.

3

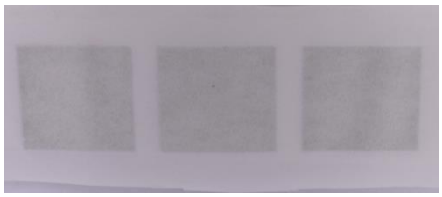


Image (3).Ag NS's printed with H_2O, Glycerol & EtOH on 5 cm^2 - squares.

Fig. 3.5.1: backside of substrates of $AgNO_3$ printed using different precursor solutions.

The experiment was performed to check whether the Silver Halide salt did deposit throughout the Chromatography paper so that enhancement in SERS signal can be checked. Less surface tension solvents were used (the cartridge did not drip after loading it with solution).

No significant difference was observed between the shades of the two printed structures.

CHAPTER 4 FABRICATION OF Au BASED SERS SUBSTRATE

4.1 Preparation of Aqueous Au solution.

Compound / Mol Wt.	Molarity (mM)	Volume (in mL)	Calc. Mass (mg)
$HAuCl_4.3H_2O$ 393.83 g/mol	1	10	3.9383
	5	10	19.6915
	7	10	27.5681
	10	10	39.3830
	20	10	78.7660

Table 4.1.1: Molarity calculations for Ag Nanostructure's for 7 prints for Au solution

Figure 4.1.1: (a). Au 20mM aq. Solution.

Optical properties of Plasmonic nanoparticles can be changed by changing their shape, size, composition, and structure[5]. The effect of nanoparticle concentration on their ability to provide surface enhancement has also been studied. The sometimes intuitive approach of increasing the number of nanoparticles to increase the signal intensity may prevent molecules from adsorbing on plasmonic nanoparticles because of nanoparticle aggregation, limiting the ability for the molecule's signal to be enhanced. It has been shown by researchers that there are ideal concentrations of plasmonic nanoparticles for use in surface-enhanced Raman spectroscopy (SERS) based on nanoparticle surface geometry and surface plasmon resonance. Ideally, the ratio of nanoparticles to analyte should be kept low enough to establish a monolayer of nanoparticles on a surface, with enough analyte to not completely cover a nanoparticle's surface[6, 7].

4.2. Experimental protocol for preparation and analysis of SERS substrate

I. Stepwise procedure of formation of SERS substrate with different time of deposition.

- Cut the Aluminium tape into 3 cm x 1 cm of a number of pieces.
- Dip the tapes into Isopropyl Alcohol (IPA) and sonicate them for 5-6 minutes.
- After sonication, remove the tapes without hampering the tapes and clean them with a kimwipe soaked with IPA.
- Verifying that the surface is smooth and free of dust, points and rough cuts.
- Dropcast 100 uL of three drops on each tape using micropipette and let them deposit for different time-period such as :
- After the specific time period, wash the Sample 1 with Deionized water and dry it. Similarly, follow the same procedure for all the samples.ss

Sr.No.	Time period (in minute)
Sample 1	**2**
Sample 2	**5**
Sample 3	**8**
Sample 4	**10**
Sample 5	**30**
Sample 6	**60**
Sample 7	**120**

Table 4.2.1: Sample number & deposition time

4.3. RAMAN Spectroscopy- Procedure

- Dip the spot from the dropcasted sample into dye (1mM Malachite Green or 100mM Metalin Yellow) for 30 minutes. Follow the same for all the samples.
- Remove the dropcasted spot from dye placed in glass vials and place them on tissue.
- Remove the paper layer (bottom layer) from the dropcasted spot and stick the spot on the testing glass slide.
- Switch ON the machine and connect the device with the software (SNOWY PEAK).
- Check and calibrate the Raman spectroscopy machine with Silicon peak (512).
- Put the slide over the stage such that the laser must point on the substrate.
- From the ‘Acquisition pane’, choose the Laser power and Integration time parameters and then proceed to acquire the plot. Plot consists of Raman Intensity (a.u.) on Y-axis & Counts/ frequency/ wavenumbers on X-axis

4.4. Preparation of 1, 5, 7, 10, 20 mM Au-Al SERS substrate using dropcast method.

- Dropcast 100 uL on each Aluminium tape and let them deposit for different time-period such as:

Sr.No.	Time period (in minute)
Sample 1	**2**
Sample 2	**5**
Sample 3	**8**
Sample 4	**10**
Sample 5	**30**
Sample 6	**60**
Sample 7	**120**

Table 4.4.1: Sample number & deposition time

Dropcasted the above prepared samples with 1mM Malachite Green dye for 30 minutes.
SERS analysis was done to check the deposition of Au nanoparticle on surface of Al strips.

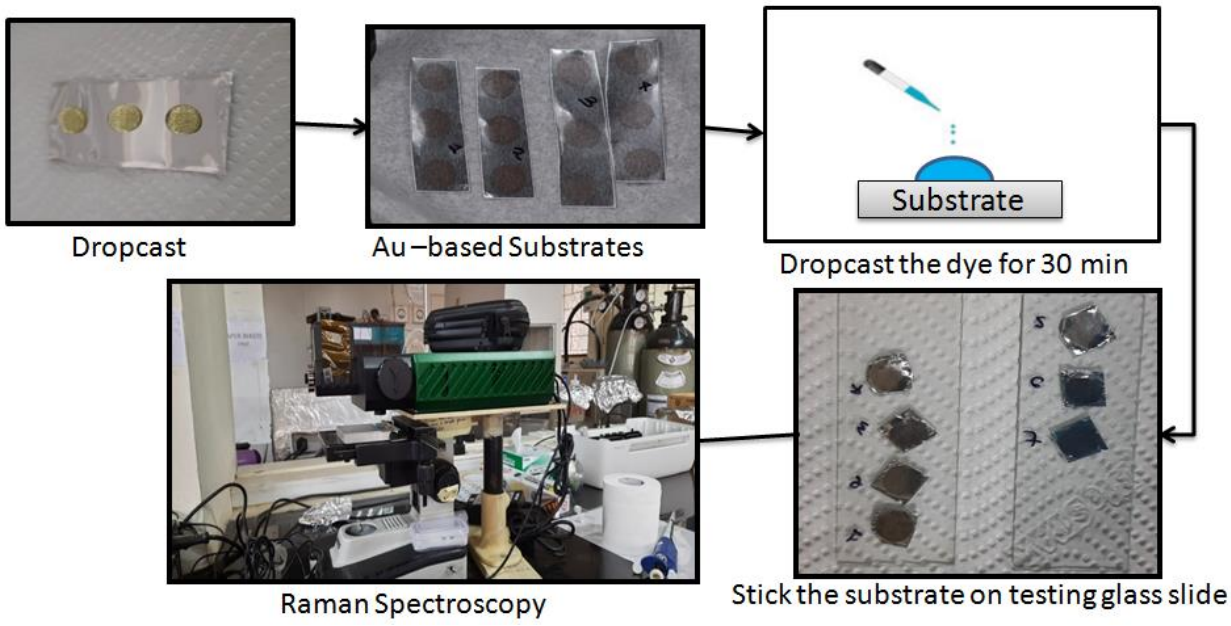


Fig.4.4.1: Schematics for Substrate preparation – SERS analysis

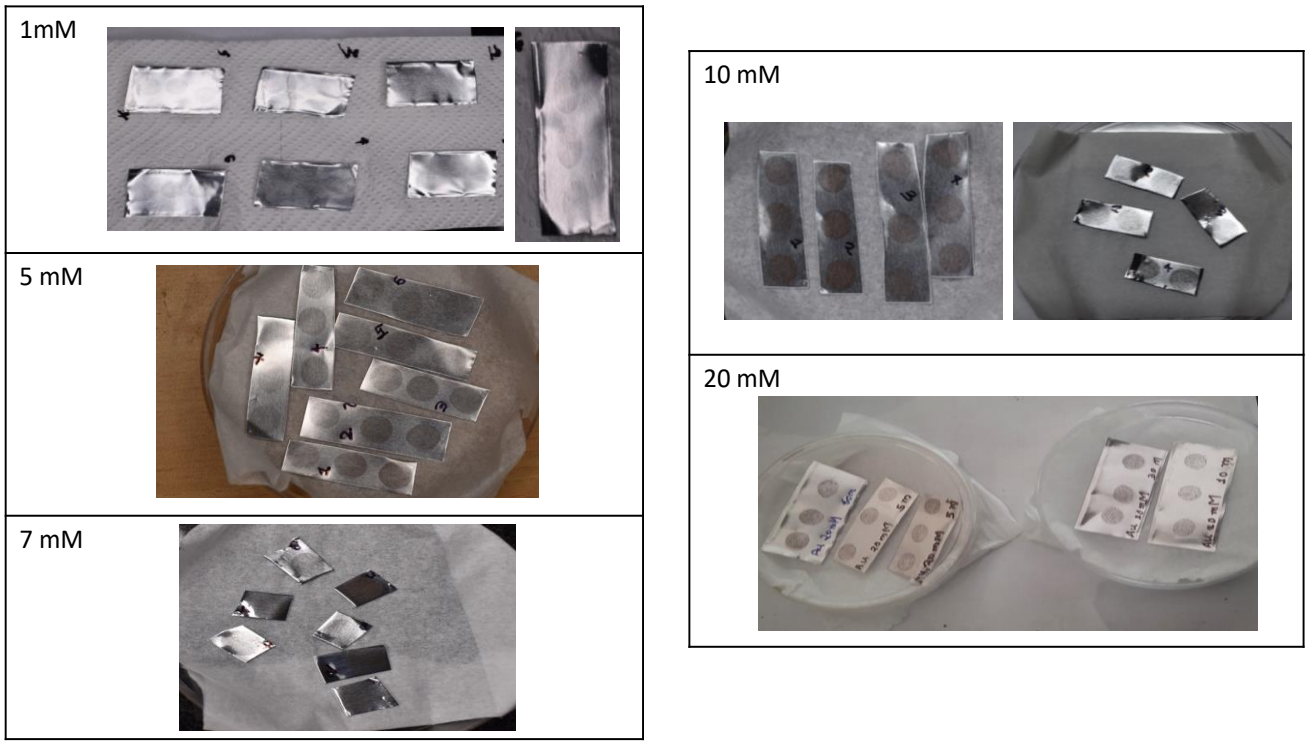


Fig.4.4.2: SERS Substrates -1, 5, 7, 10, 20mM concentration

Raman spectroscopy was performed on the prepared samples by dropcasting 10µL - 1mM MG dye. Initially, the sample was dipped into the dye and washed with DI Water after 30 minutes; but after washing the proper SERS peaks were not obtained; so shift to dropcasting of MG dye was preferred.

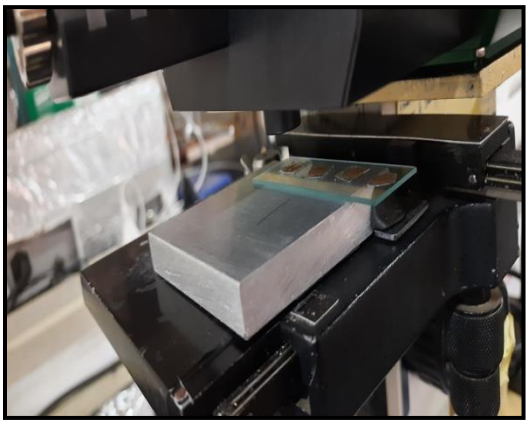


Fig.4.4.3: Testing slide placed on sample stage for Raman spectroscopy

4.5 Results

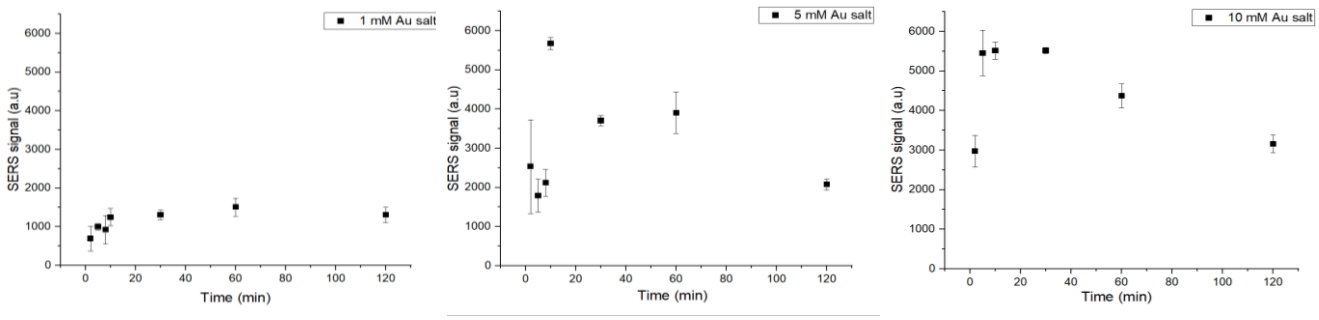


Fig.4.5.1: Representative SERS spectra of 1mM, 5mM, 10mM – vs. different deposition time

Raman spectroscopy results showed increased SERS enhancement with concentration 5mM, 10mM for 10 minutes deposition time as compared to different concentration vs. time samples. Deposition on samples of 1, 7, and 20mM was observed but SERS peak with proper intensity were not observed; as change in concentration and deposition time did not result in increased enhancement in SERS signal.

SEM images showed smoother and finer dendritic structure in 5 mM concentration than 10 mM concentration vs. 10 min deposition time (Fig.1, 3 - on right side); uniformly structures were obtained throughout the sample. (Fig.2, 4 - on right side).

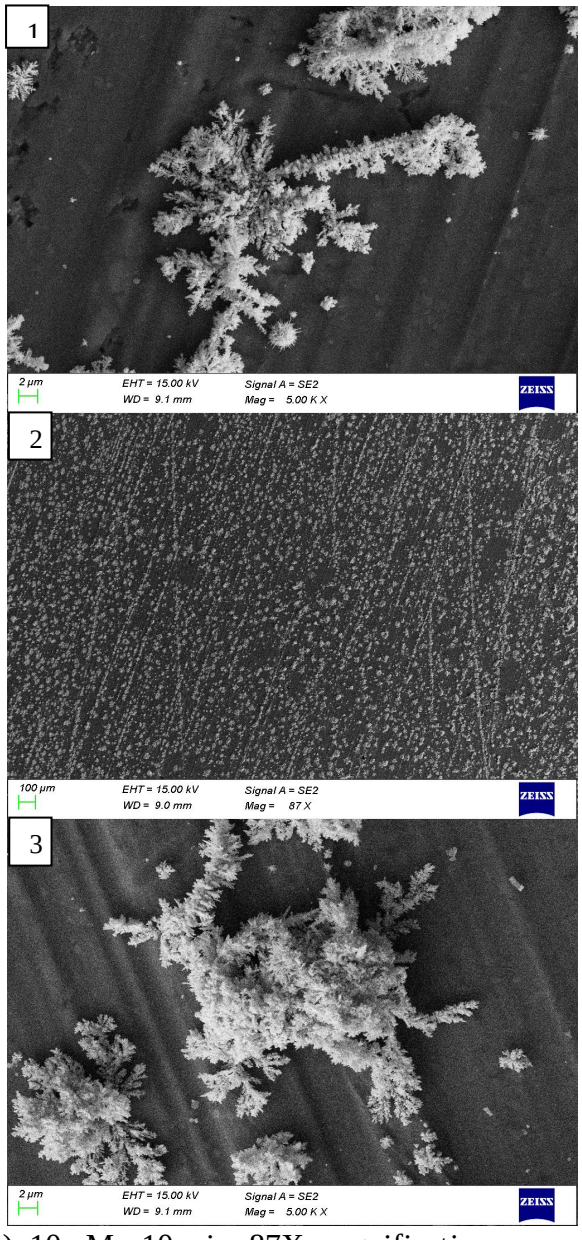


Fig.4.5.2:
SEM Image for
(1). 5mM - 10 min- 5kX magnification.

(2). 10mM - 10 min- 5kX magnification.

(3). 5mM - 10 min- 87X magnification. (4). 10mM - 10 min- 87X magnification

EDS of the samples was obtained as shown below:

Sr.no. / Elements	Sample 1 2 min	Sample 2 5 min	Sample 3 8 min	Sample 4 10 min	Sample 5 30 min	Sample 6 60 min	Sample 7 120 min
O	2.73	3.23	3.55	4.11	2.62	2.76	2.67
Si	0.50	-	0.54	0.49	0.54	0.51	0.57
Fe	0.45	0.43	0.43	-	0.34	0.3	0.41
Al	**90.83 ±1.61**	**90.62 ±1.61**	**90.66 ±1.61**	**91.00 ±1.61**	**94.27 ±1.61**	**93.9 ±1.61**	**92.89 ±1.61**
Au	**6.13 ±1.46**	**5.62 ±1.46**	**5.13 ±1.46**	**4.39 ±1.46**	**2.34 ±1.46**	**2.73 ±1.46**	**3.45 ±1.46**

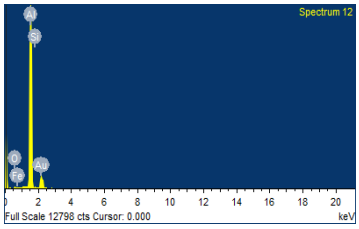


Table 4.5.1: EDS results of 5 mM substrate - representing average Atomic wt % of each sample taken on 3 different spots - alongwith standard deviation values.

Sr.no. / Elements	Sample 1 2 min	Sample 2 5 min	Sample 3 8 min	Sample 4 10 min	Sample 5 30 min	Sample 6 60 min	Sample 7 120 Min
O	2.49	3.19	2.79	2.88	3.01	3.16	3.26
Si	0.45	-	-	0.49	-	0.55	0.59
Fe	0.36	0.44	-	-	0.46	0.49	0.46
Al	**92.63 ±1.17**	**89.5 ±1.17**	**91.4 ±1.17**	**91.63 ±1.17**	**90.71 ±1.17**	**89.34 ±1.17**	**91.12 ±1.17**
Au	**4.33 ±0.64**	**5.69 ±0.64**	**5.77 ±0.64**	**5.04 ±0.64**	**6.13 ±0.64**	**5.20 ±0.64**	**4.7 ±0.64**

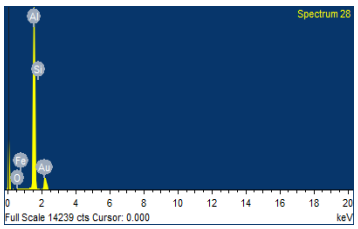


Table 4.5.2: EDS results of 10 mM substrate - representing average Atomic wt % of each sample taken on 3 different spots - alongwith standard deviation values.

CONCLUSION

In this project, Ag nanostructures were printed on chromatography paper changing the precursor solutions. It did not result in formation of nanostructure throughout the cross-section of chromatography paper; the Ag salt printed initially as soon as it comes in contact with halide salt it reacts and forms silver halide salt which does not permit it to reach throughout the chromatography paper as pre-determined. SERS active Au substrates were prepared on Al foil by varying the concentration and deposition time; a maximum SERS peak was obtained with 5mM and 10mM sample with 10 minutes deposition time. EDS results showed deposition of Gold to 6.13 atomic wt %. Malachite Green dye shows SERS response, which further was optimized for different concentration and reaction time.

Future trajectory consists of fabrication and optimization of Au-Ag i.e. bimetallic SERS substrates as Gold NPs are easier to synthesize, have better biocompatibility and long-term stability, also, silver NPs have a more intense SPR, which is of great advantage for SERS and sensing applications.

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ANNEXURE – V

SUPERVISOR EVALUATION OF INTERN

Student Name: OM BALASAHEB TAMBE Date: 30 July 22

Work Supervisor: Dr. VENUGOPAL SANTHANAM Title: ASSOCIATE PROFESSOR

Institute/Industry: INDIAN INSTITUTE OF SCIENCE, BANGALORE

Dates of Internship: From 01 JUNE 2022 To 30 JULY 2022

Please evaluate your intern on following factors: (Tick √)

Parameters	Needs improvement	Satisfactory	Good	Excellent
Behaviors				✓
Cooperates with co-workers and supervisors				✓
Shows interest in work				✓
Learns quickly			✓	
Produces high quality work			✓	
Accepts responsibility				✓
Uses technical knowledge and expertise			✓	
Demonstrates creativity/originality			✓	
Analyzes problems effectively			✓	
Communicates well				✓
Has a professional attitude				✓
Is punctual				✓
Uses time effectively				✓

Overall performance of student intern (circle one):

(Needs improvement / Satisfactory / Good / Excellent)

Signature of Institute / Industry supervisor

प्रोफ़ेसर / Professor
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