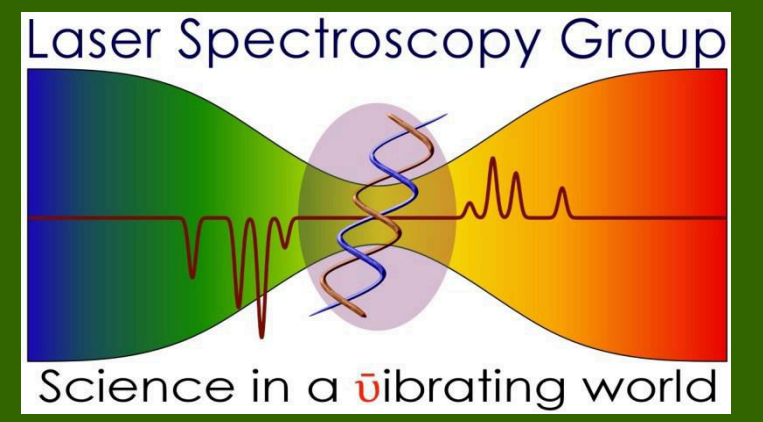




# Raman Spectroscopic Study of Serum Components

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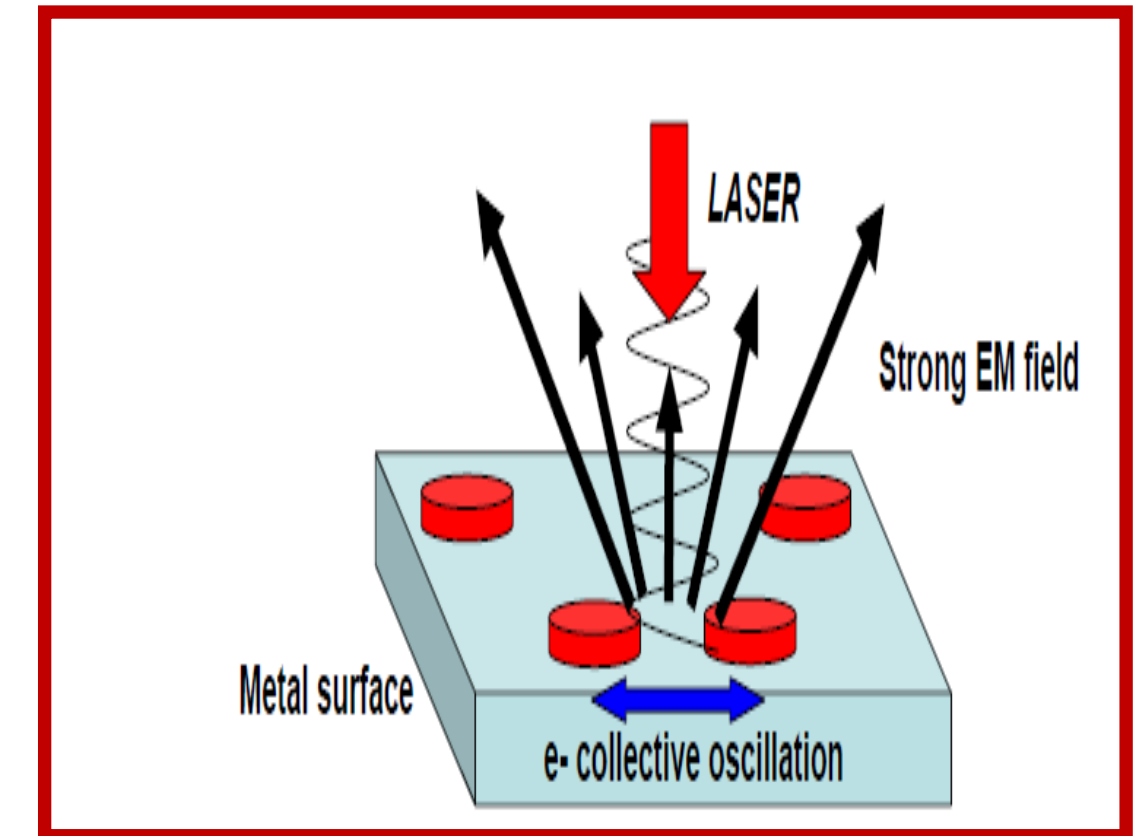
## Introduction

- Early diagnosis of disease is important as treatment is often simpler & more likely to be effective
- To make this possible, technological developments which enable detection of biochemical changes in blood, to avoid biopsy, are needed as the blood contains thousands of bio-markers present in the form by-products within the diseased part of the organism
- The challenge is to detect very low concentrations of the analytes need to be detected at the early stage of disease
- In this study, Surface Enhanced Raman Spectroscopy (SERS) is used for the detection of the bio-molecules (tryptophan & serum albumin) present in serum.
- The samples are presented in colloid dispersion both in static drop and also in flow system Lab-on-a-chip (LOC)

## SERS Enhancements

Electromagnetic Enhancement

Chemical Enhancement



## Materials and Methods

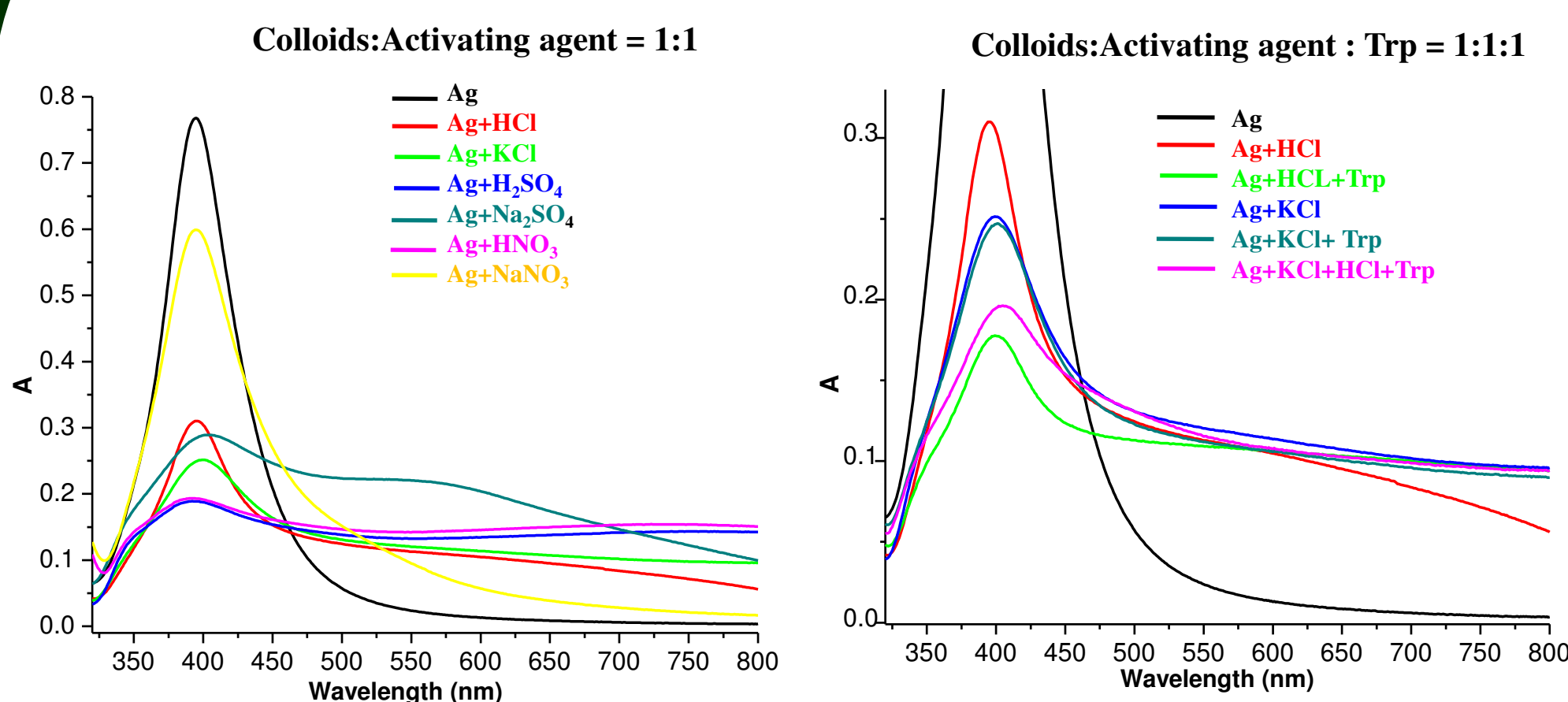
Silver (Ag) colloids: [1] NaBH<sub>4</sub> reduced [2] Sodium citrate reduced capped with PVP

Activating agents: KCl, HCl, NaNO<sub>3</sub>, HNO<sub>3</sub>, Na<sub>2</sub>SO<sub>4</sub>, H<sub>2</sub>SO<sub>4</sub>.

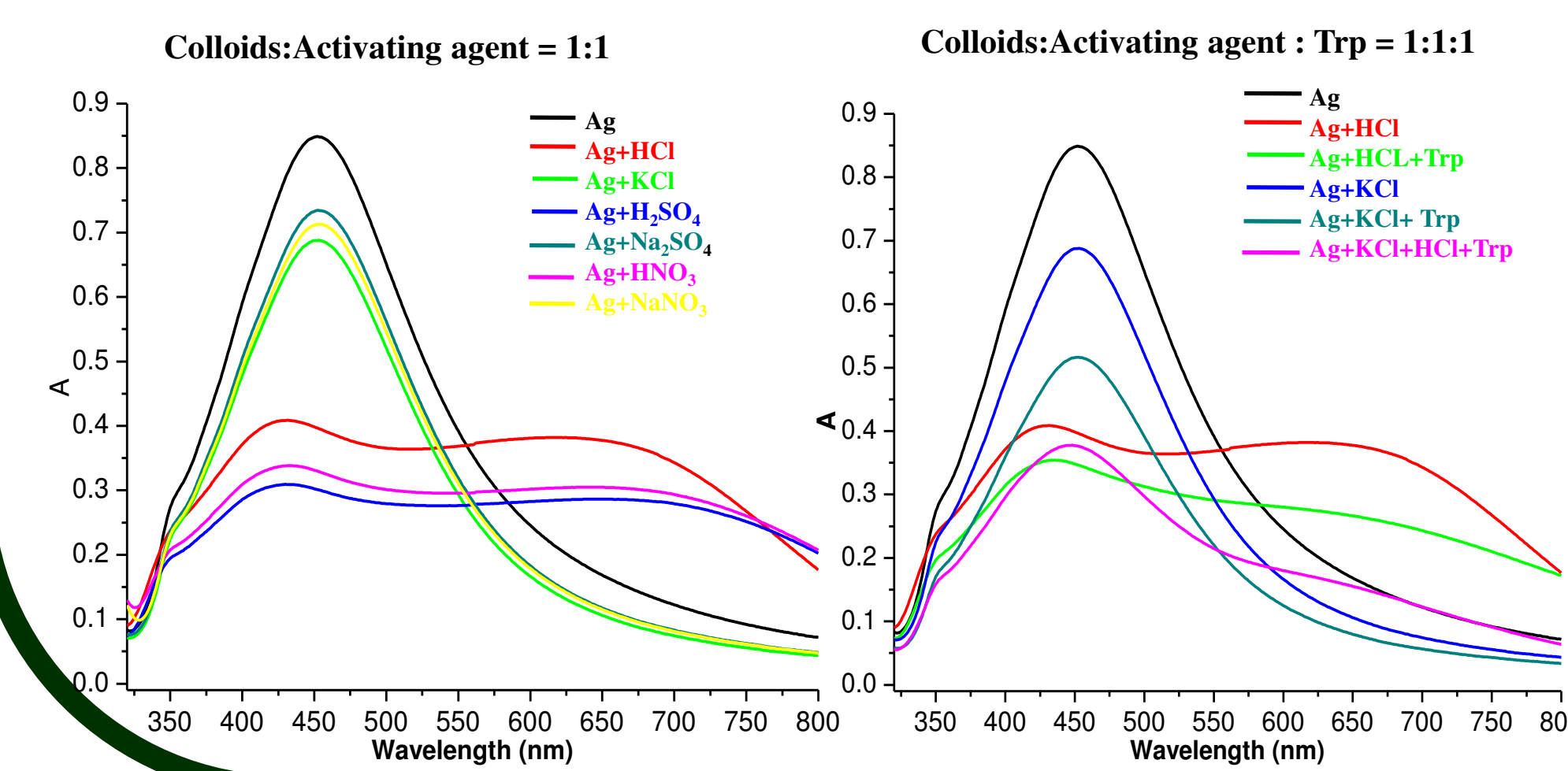
Analytes: Tryptophan (Trp) and Bovine Serum Albumin (BSA)

## UV-VIS Spectra

### NaBH<sub>4</sub> reduced Ag colloids



### Sodium citrate reduced, capped with PVP Ag colloids

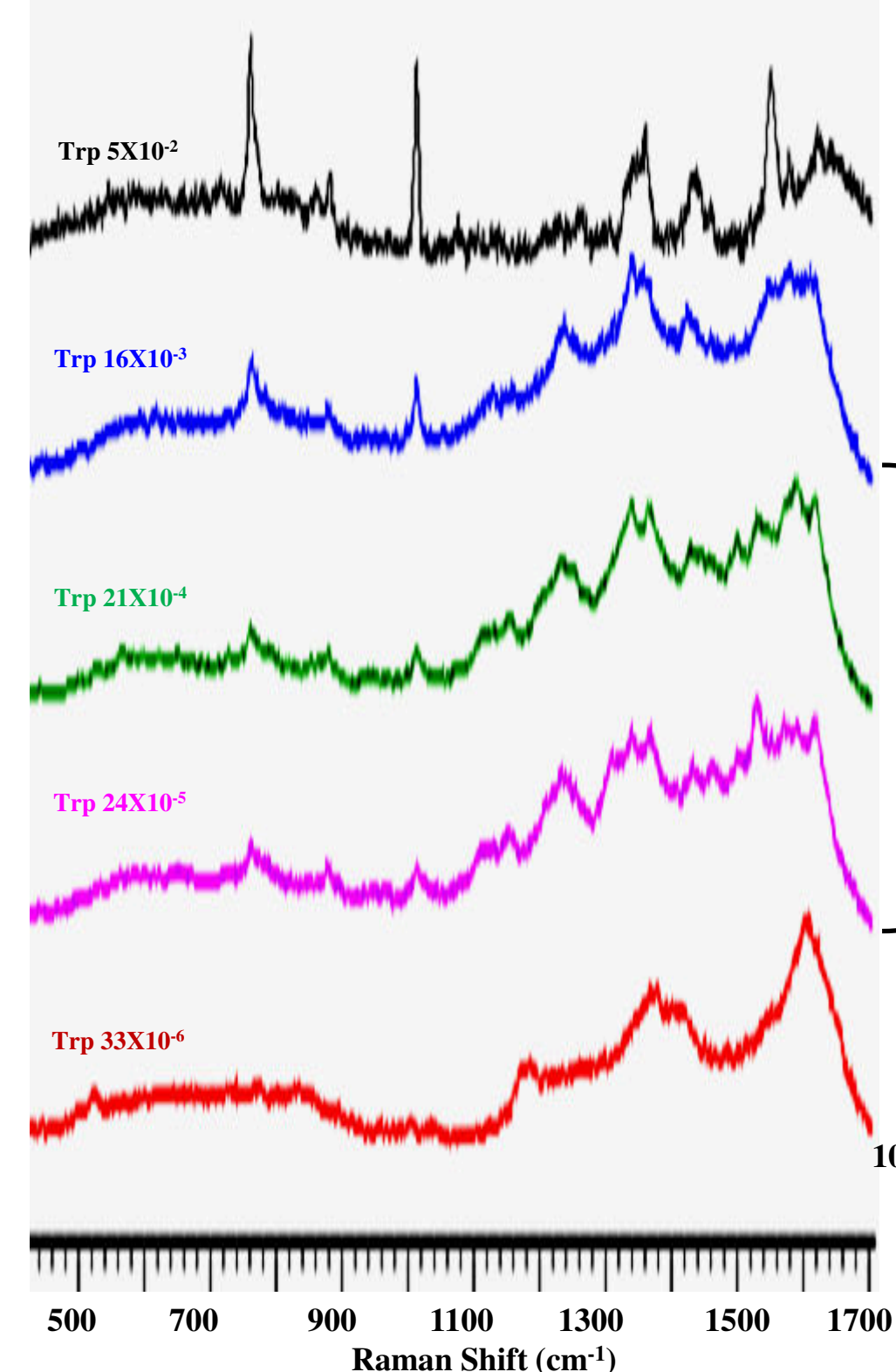


## SERS of Tryptophan

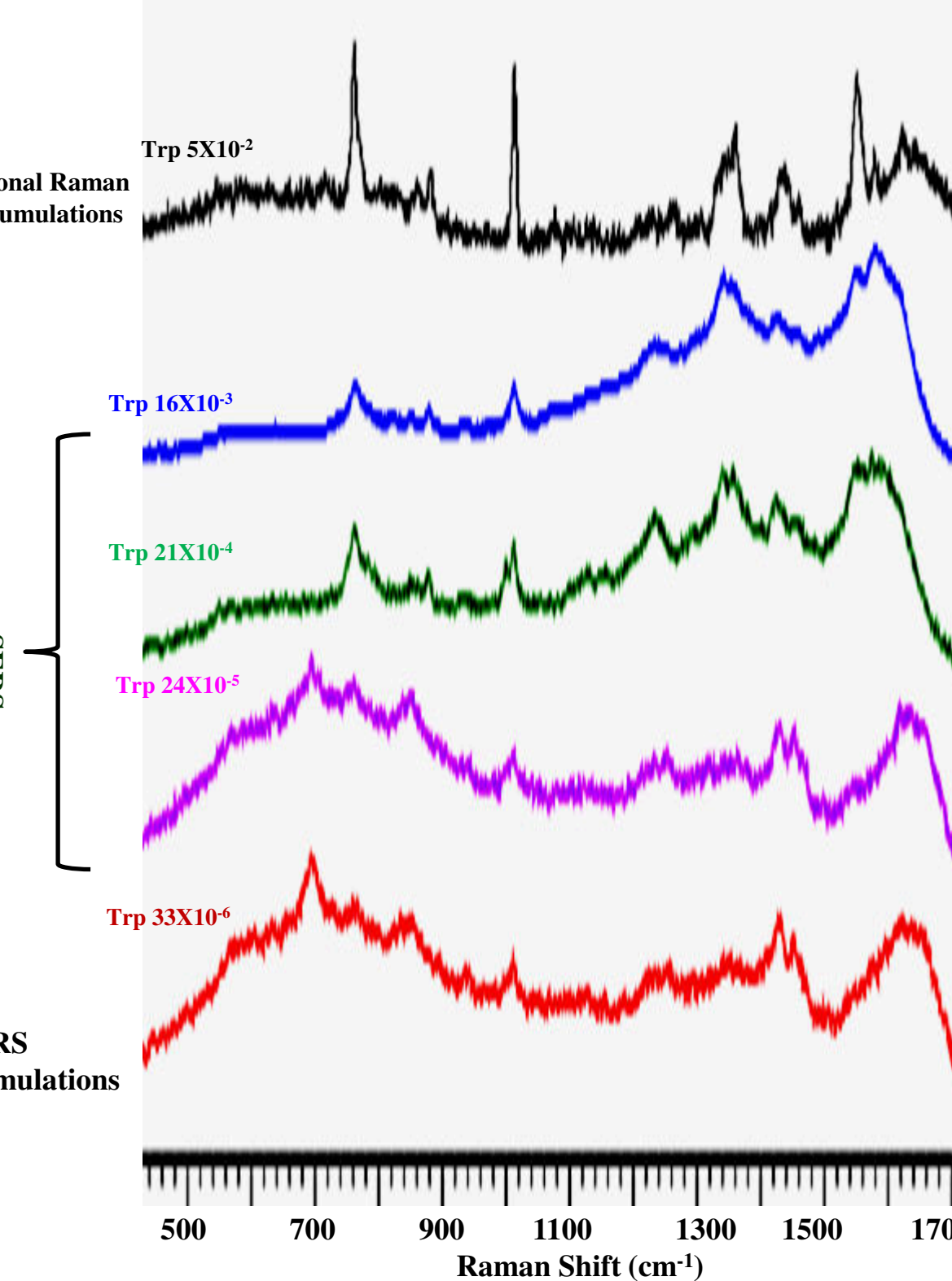
In solution Phase

Colloids were activated with HCl solution, before adding tryptophan to it. Mixture was excited at 514nm (~5mW) using 50X long working objective

### NaBH<sub>4</sub> reduced Ag colloids



### Sodium citrate reduced, capped with PVP Ag colloids

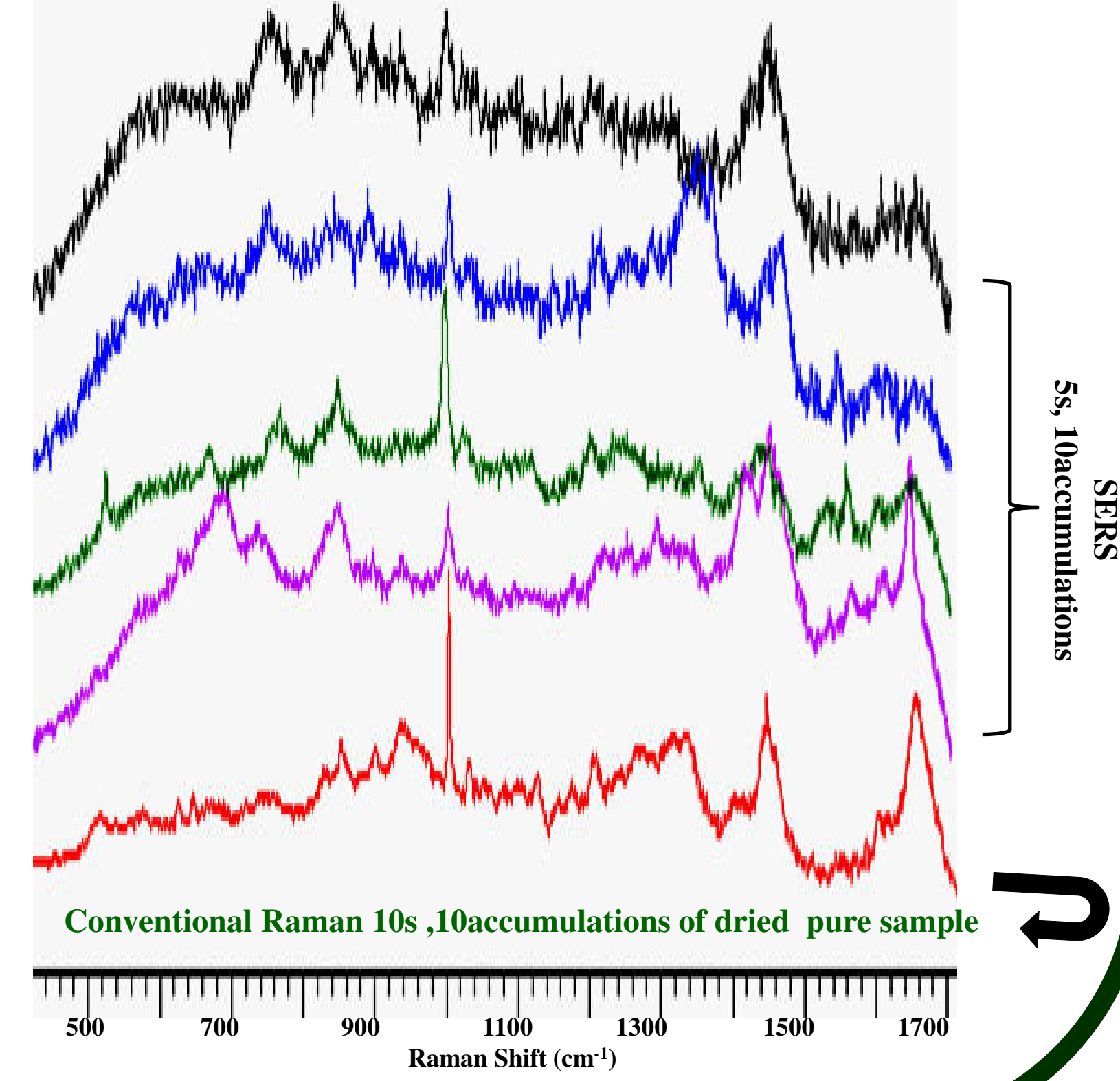


## SERS of BSA

In pellet form

BSA was added to activated colloids (HCl) and then incubated for 1 hour at room temp. and the mixture was centrifuged to make a pellet. Dry pellet was excited at 514nm (~0.1mW)

### Sodium citrate reduced, capped with PVP Ag colloids



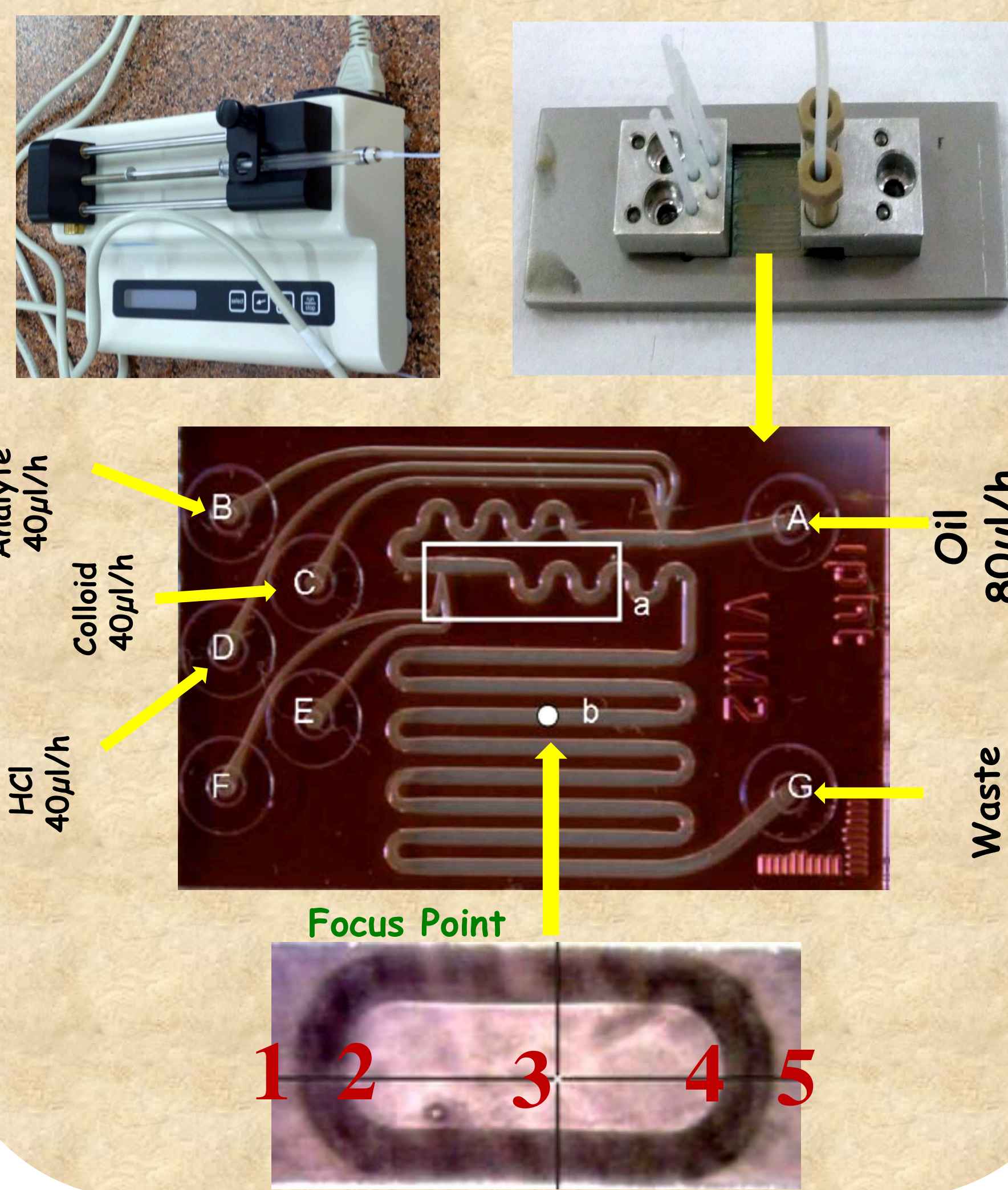
## Limitations of SERS

- ❑ Reproducibility of SERS spectra is a key concern, under similar experimental conditions
- ❑ This limitation can be overcome by performing SERS spectroscopic measurements in flow cells like LOC

## LOC has various advantages

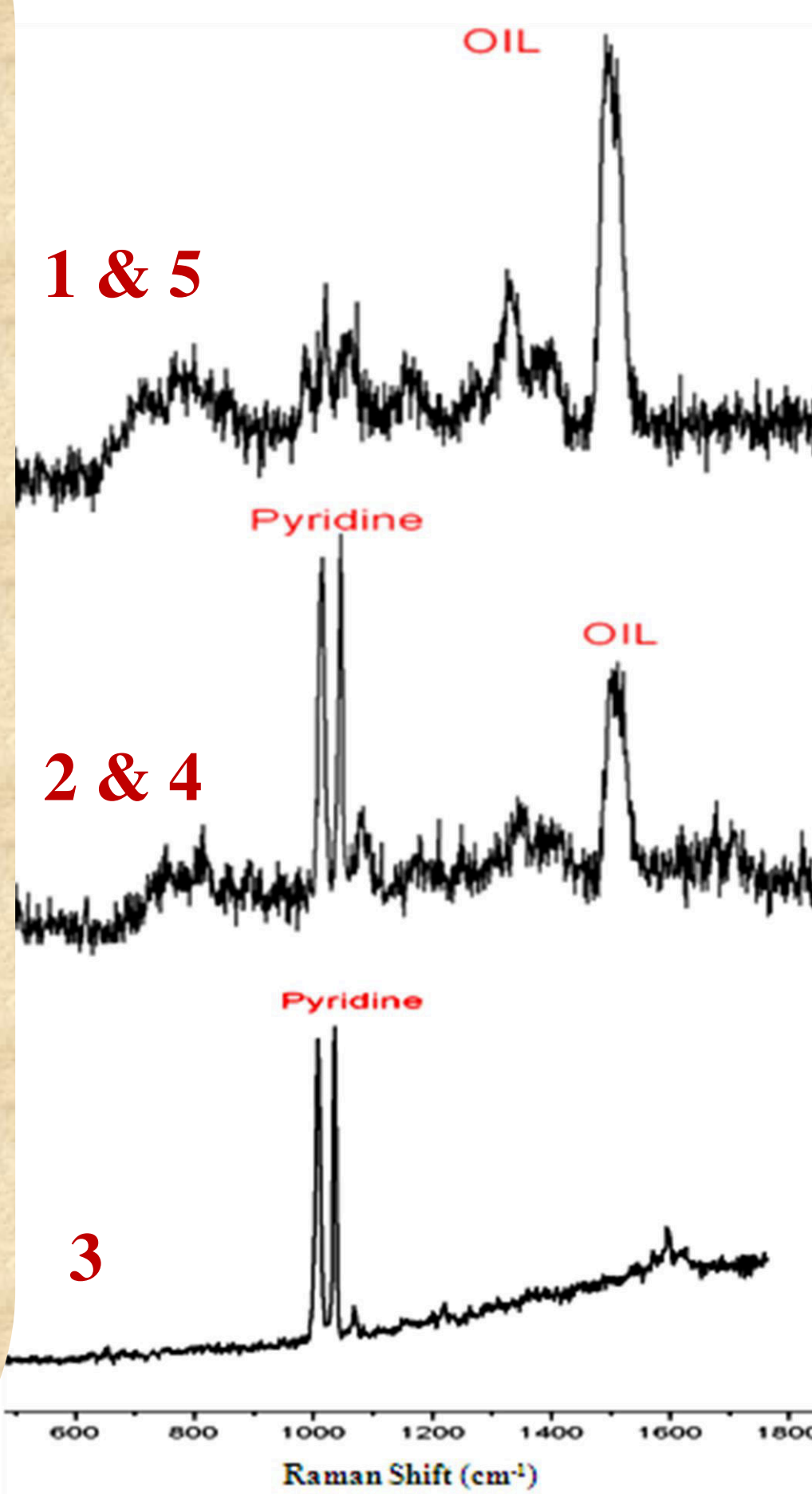
- Higher analysis throughput
- Cost effective
- Decreasing analysis time for diffusion controlled reaction
- Automated and human Error Free
- Reasonably reproducibility

## LOC Setup



## SERS of Pyridine in flow system

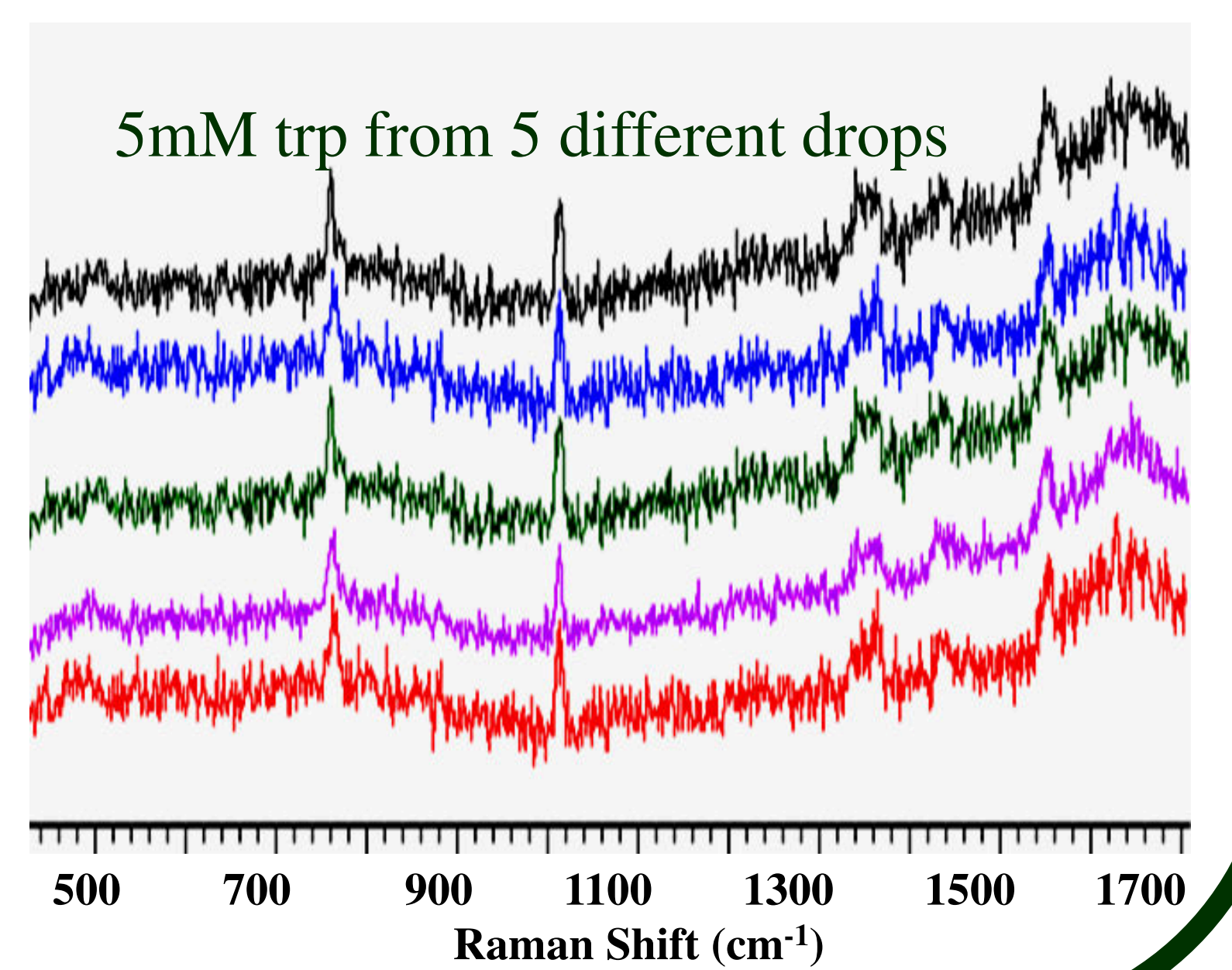
Exposure time -1s; No. of Accumulation -1.



## SERS of Tryptophan in flow system

Sodium citrate reduced Ag colloids capped with (1%) PVP and HCl as activating agent were used

Excited at 514 nm; exposure time -2s



## Conclusions

- ❖ 1% PVP capped Ag colloids gives a consistent weak background
- ❖ Both NaBH<sub>4</sub> and citrate reduced nanoparticles require acidic environment for better aggregation
- ❖ SERS is more reproducible in automated flow system (LOC)

## References

- ❖ Z. Zhang, *et. al* Cancer Res. **64**, 5882 (2004).
- ❖ F. Gentile *et al* Microelectronic Engineering, **87**, 798 (2010).
- ❖ P.C. Lee *et al* J. Phys. Chem. **86**, 3391-3395 (1982)
- ❖ K. R. Strehle *et al* Anal. Chem. **79**, 1542 (2007)

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