# A preliminary investigation on human tears by means of surface enhanced Raman spectroscopy

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Tears are exceptionally reach sources of information on the health status of the eyes, as well as of the whole body.

Surface Enhanced Raman Spectroscopy (**SERS**) provides an unique methods for analyzing low concentration organic fluids as tears.*

In this work, home-made SERS substrates were prepared by using gold nanoparticles (GNP's), enhabling spectroscopy of fluid samples by using HeNe laser ($\lambda$= 633 nm) .

* P. Hu et al, J Raman Spectr 45 (2014) 565; W. S. Kim et al, Sensors and Actuators B: Chem 222 (2016), 1112; S. Choi et al, Anal. Chemistry 86 (2014) 11093.

# SERS

Surface Enhanced Raman Spectroscopy

how SERS works?

LIGHT ⟷ em interaction ⟷ NANOSCALE METALS

↓

collective oscillations of conduction electrons :
LOCALIZED SURFACE PLASMON RESONANCES

↓

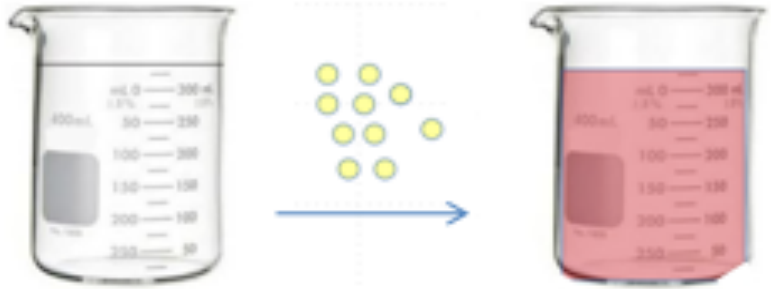
high local enhancements of the electromagnetic energy
(Large improvement of Raman spectral response)

# Gold nanoparticles colloid : Fabrication process

0.01% $HAuCl_4$ solution

+

1% sodium citrate

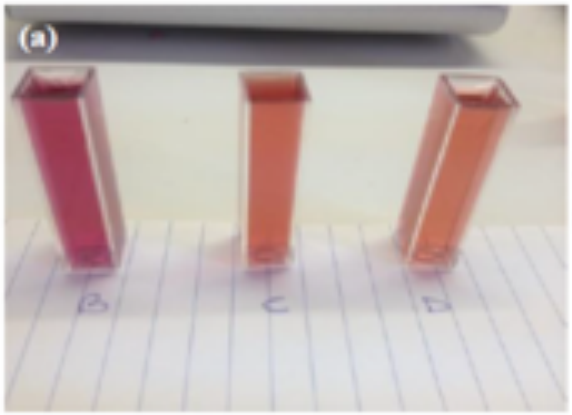


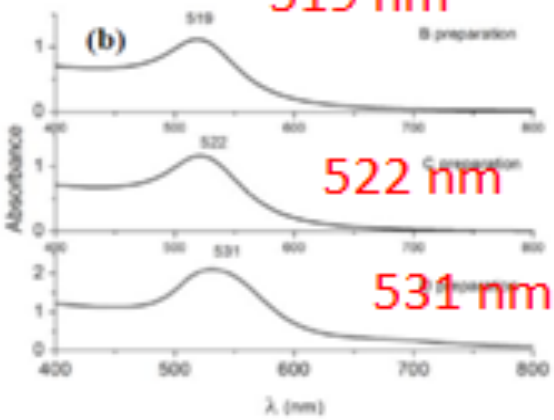
Lee and Meisel method: the reducing agent (sodium citrate) acts as stabilizing agent by adsorbing onto the metal surface and avoiding nanoparticle aggregation through electrostatic repulsions .

C. Camerlingo, M. Portaccio, R. Tatè, M. Lepore, I. Delfino, Fructose and pectin detection in fruit-based products by surface-enhanced Raman spectroscopy. *Sensors* **2017**, 17, 839.

# Gold nanoparticles colloid : characterization

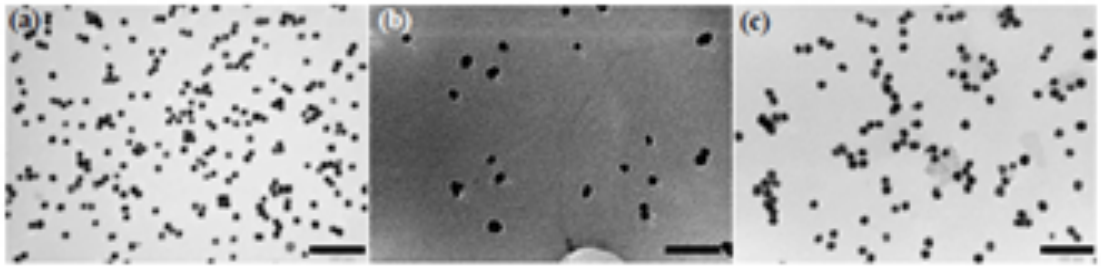
## Absorption spectra




TEM

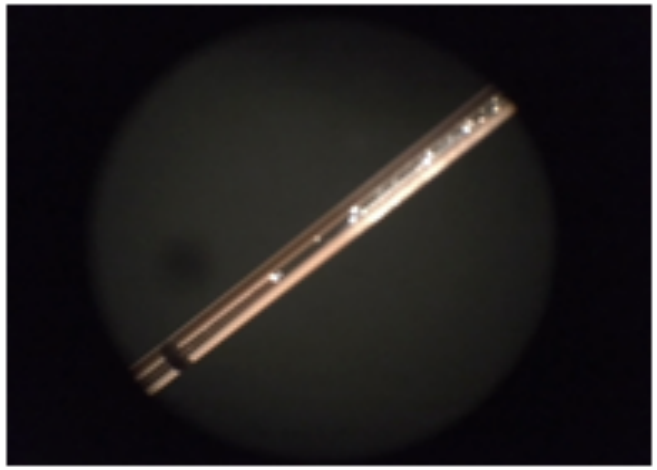
**50±5 nm** **20±2 nm** **27±3 nm**



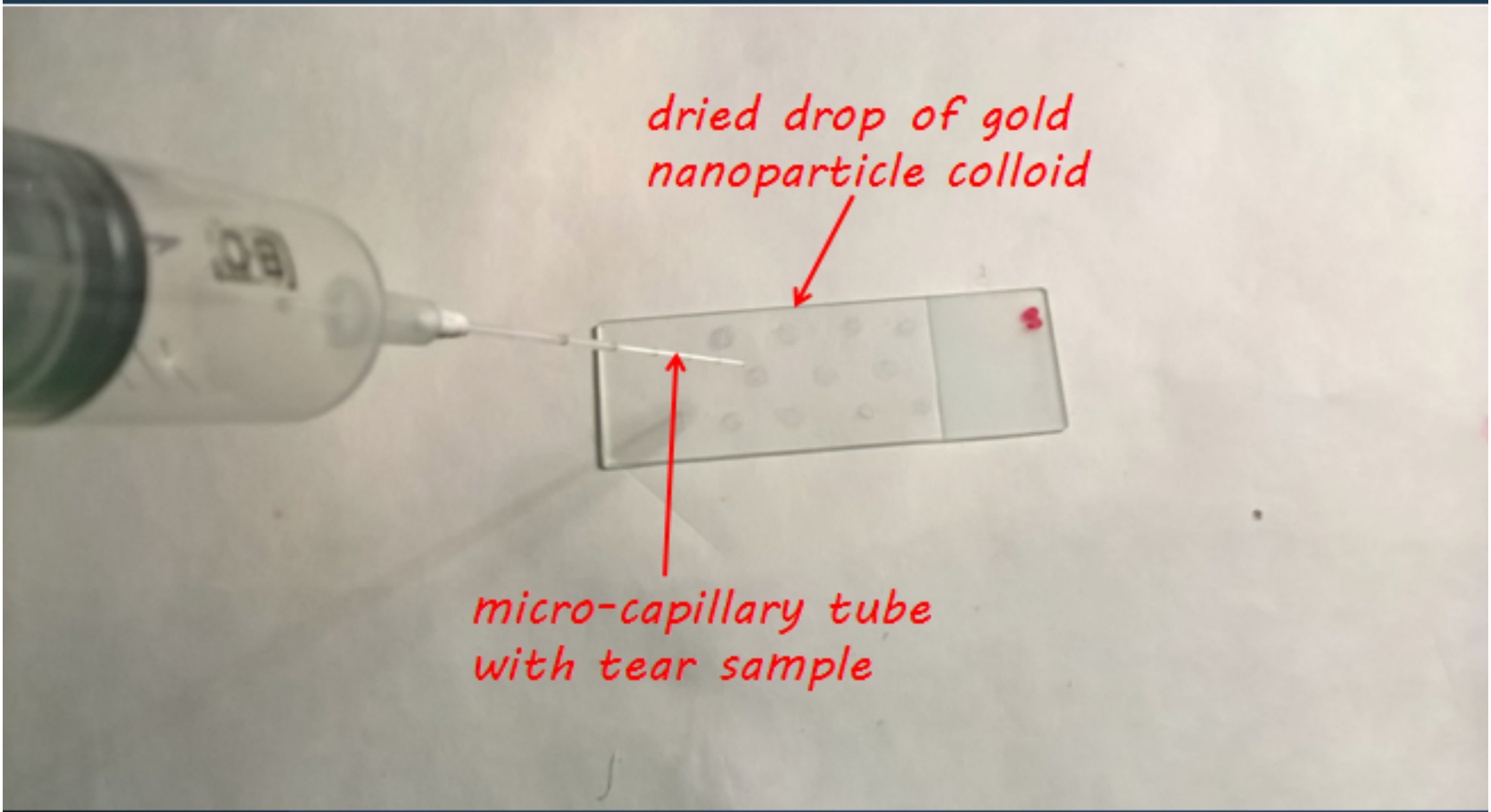

100 nm

C. Camerlingo et al, *Proceedings* **1** (2017), 25

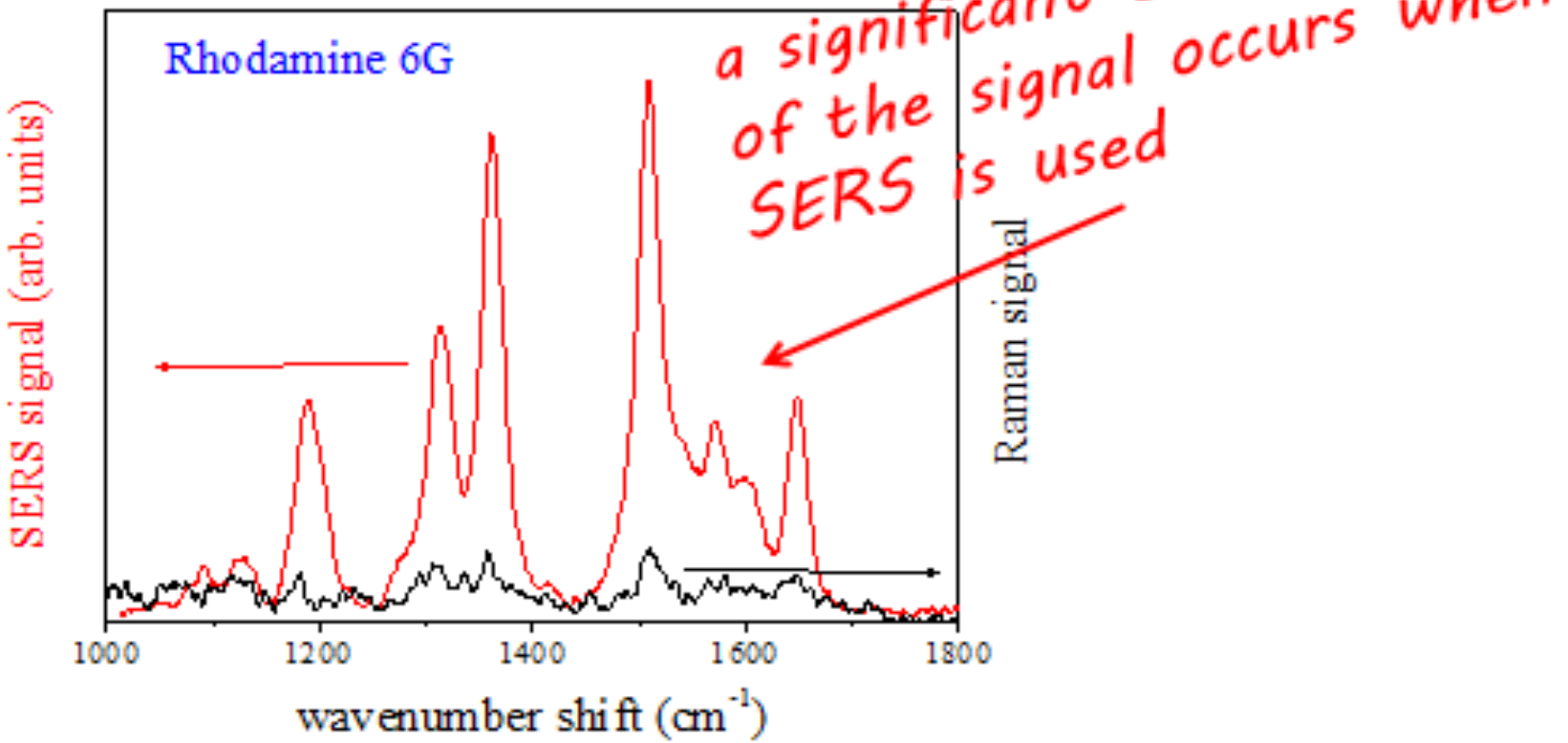
Tears were extracted from informed healthy patients by means of a not-invasive method based on a direct collection of the tear fluid by means of a suitable micro-capillary tube.



# A preliminary investigation on human tears by means of SERS

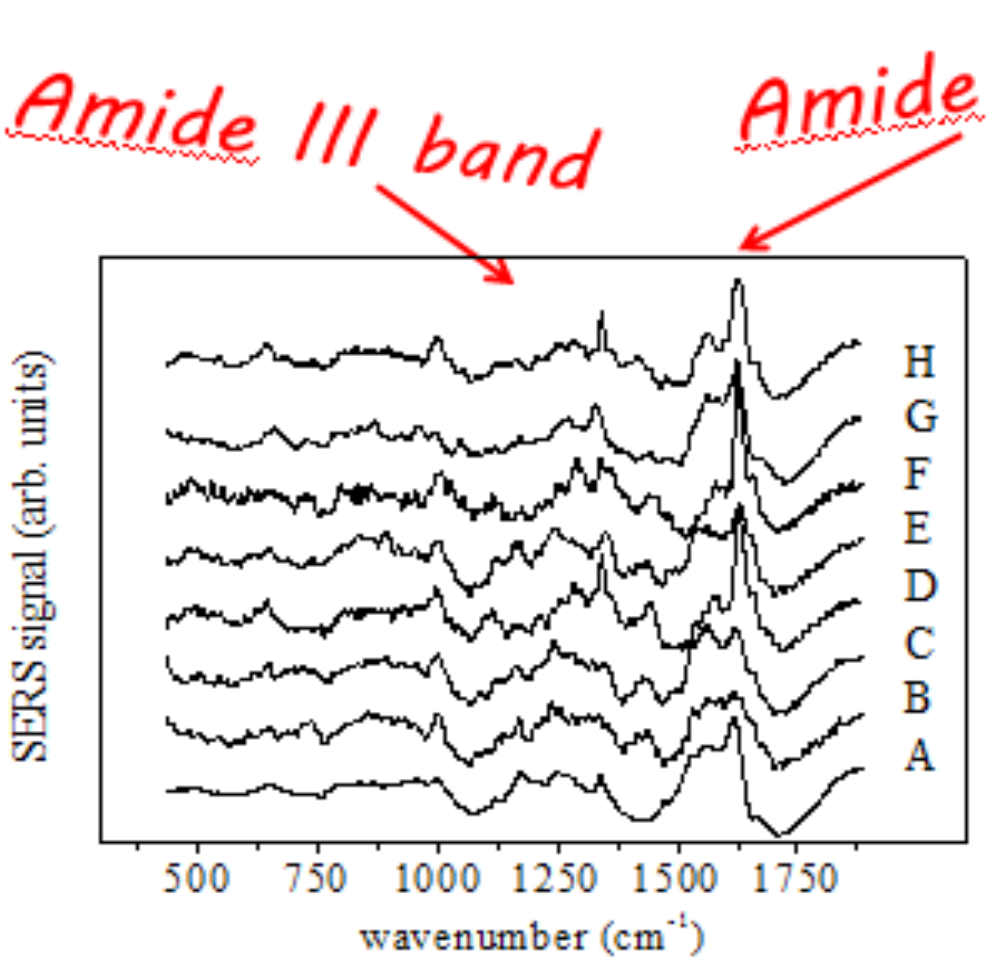


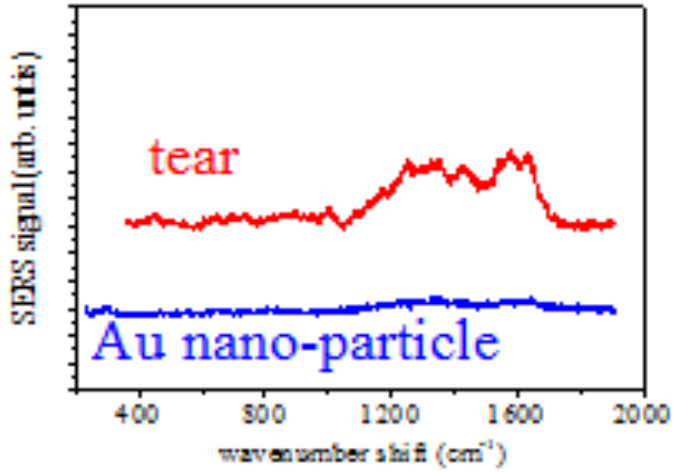
The SERS was preliminary tested on Rhodamine 6G aqueous solutions at different concentrations.



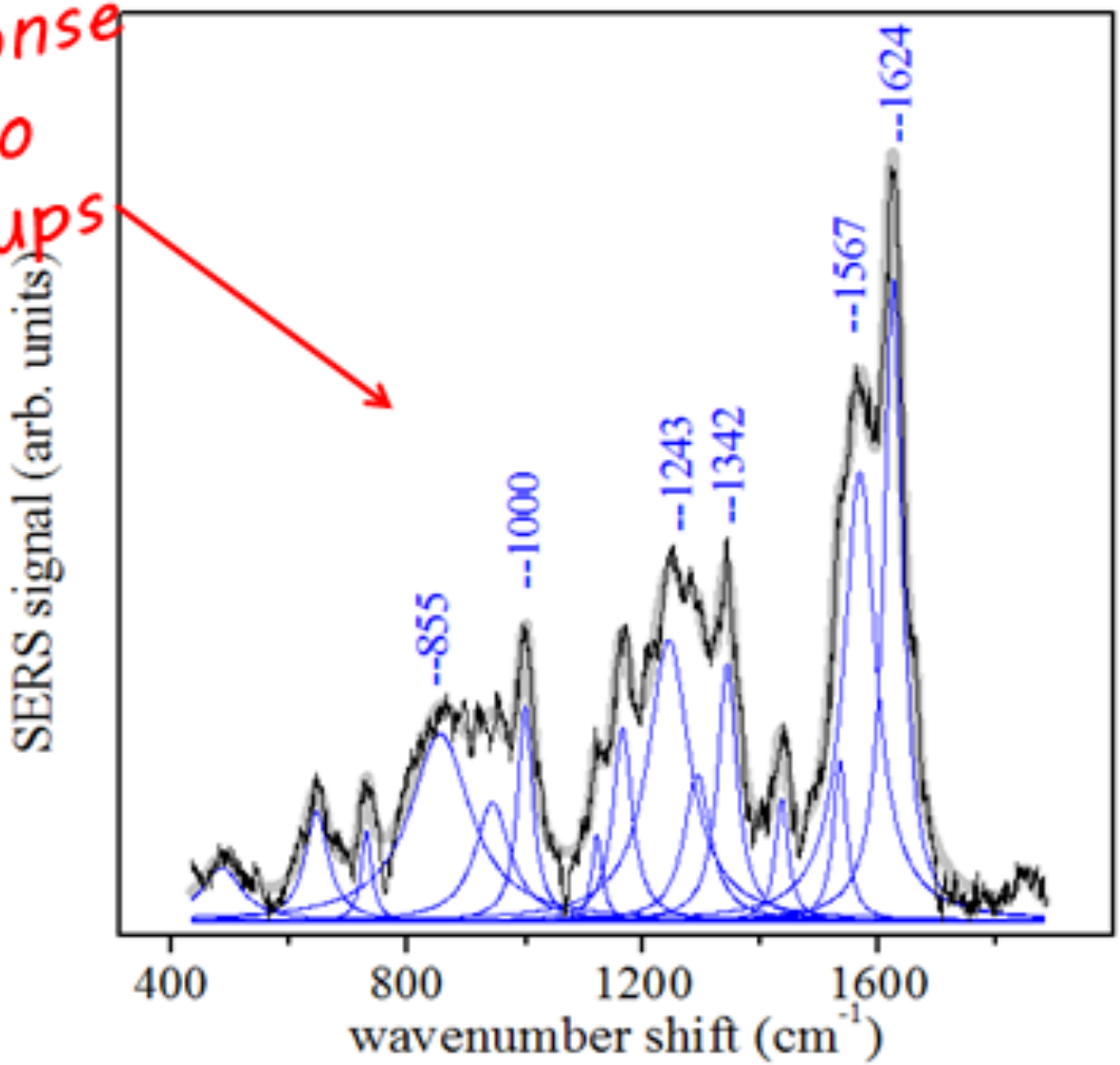
SERS responses of tear samples obtained from 8 healthy patients.

Protein content contributions are largely present in SERS spectra.



The SERS response is mainly due to amino acid groups




Average spectrum obtained by the sum of all the SERS spectra . The signal have been deconvoluted in terms of Lorentzians (blue lines) and the main Raman mode components are indicated.

Changes in amino acid levels can be correlated to tear composition variations

main organic components of human tear

| component | mol. weight (KDa) | *c* (mg/ml) [*] | *c* ($\mu$M) | description |
|---|---|---|---|---|
| Lactoferrin | 80 | 1.8 - 2.7 | 23 -34 | iron binding glycoprotein |
| Lysozyme | 14 | 1.6 - 2.5 | 111 -172 | single chain polypeptide |
| Lipocaline | 20 | 1.2 - 2.95 | 62 -145 | low mol. weight protein |
| Immunoglobulins (IgA) | 162 | 0.2 - 0.3 | 1.5 - 1.9 | glycoproteins (antibodies) |
| Albumine | 66 | 1.3 | 20 | single peptide chain |

[*] Fullard R. J., Sbyder C., Protein levels in nonstimulated and stimulated tears of normal human subjects. Ophthalmology & Visual Science **1990, 31(6), 1119-1126.**

# *Conclusions*

A significatively SERS response from TEAR samples can be obtained on conventional microscope glasses by using home-made gold nanoparticles (GNP's).

The method allows to sense the main organic components of the TEARS with promising perspectives in diagnostic applications.

*This activity is a joint research between CNR* and the Universities "Federico II" of Neaples* and "Luigi Vanvitelli" of Campania (Neaples)*.*

* *CNR-SPIN, Ist. superconduttori, materiali innov. e dispositivi (CC; MLi)*

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