Supplementary data

SERS-based quantification of albuminuria in the normal-to-mildly increased range

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Supplementary Table 1. The individual albumin concentration and albumin to creatinine ratio (ACR) values of each sample.

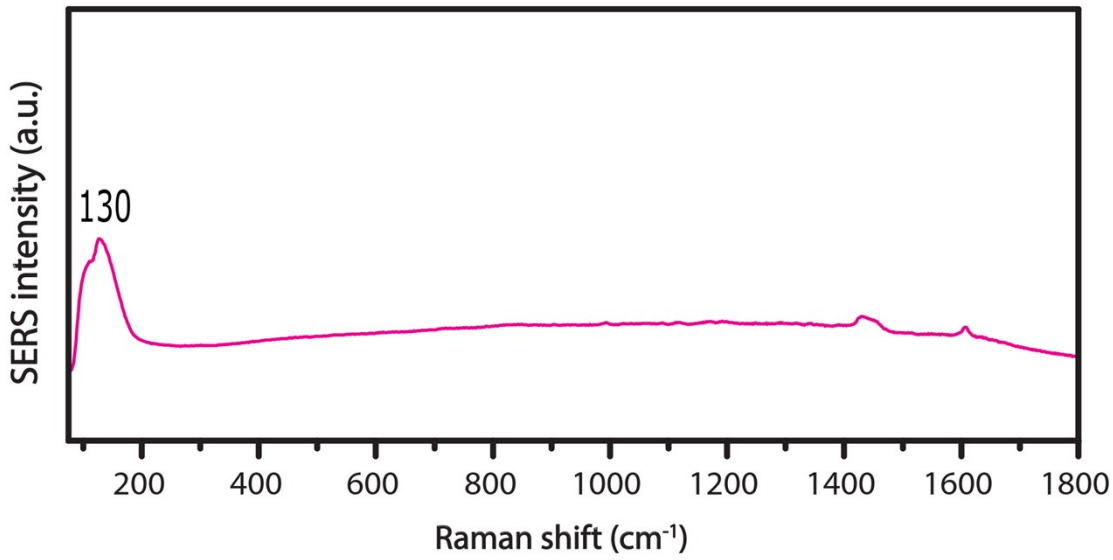
Sample nr.	Albumin concentration (mg/L)	ACR
C1	7.1	0.04
C2	3.6	0.04
C3	1.3*	0.03
C4	0.4*	0.01
C5	11.8	0.14
C6	0*	0
C7	0.1*	0.11
C8	1.2*	1.18
C9	0*	0
C10	0.3*	0.01
C11	7.8	0.05
C12	3.5	0.03
C13	0.5*	0.01
C14	0*	0
C15	0.3*	0.81
C16	0*	0
C17	3.9	0.02
C18	5.6	0.03
S1	119.7	78.1
S2	93.5	69.6
S3	30.1	31.5

S4	18.1	46.2
S5	27.5	91.1
S6	4.7	10.5
S7	10.2	29.1
S8	25.6	23
S9	5.7	15.2

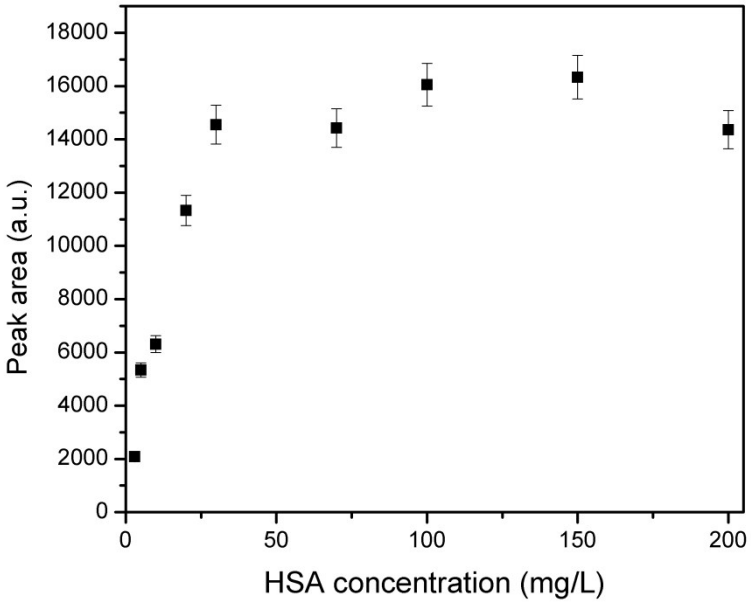
*The clinical laboratory provides accurate results for albumin concentrations above 3 μg/mL.

Supplementary Table 2. The demographic data of the six groups defined by the 3, 6 and 10 μg/mL cut-off values along with the median albumin concentration.

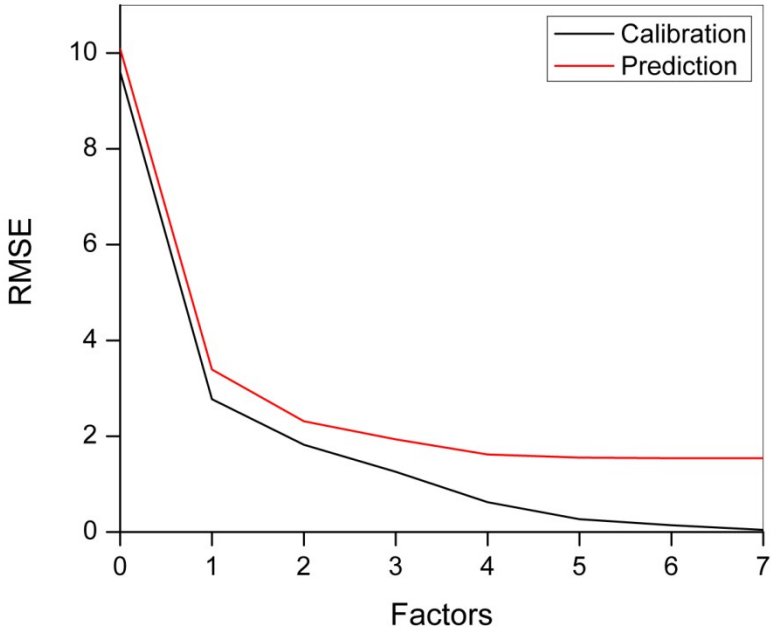
Concentration range	Nr. of subjects	Median albumin concentration(μg/ml) (range)
< 3 μg/ml	11	0.3 (0-2.9)
> 3 μg/ml	16	9.01 (3-119.7)
< 6 μg/ml	17	0.5 (0-5.68)
> 6 μg/ml	10	21.87 (7.1-119.7)
< 10 μg/ml	19	1.18 (0-7.1)
> 10 μg/ml	8	26.55 (10.22-119.7)



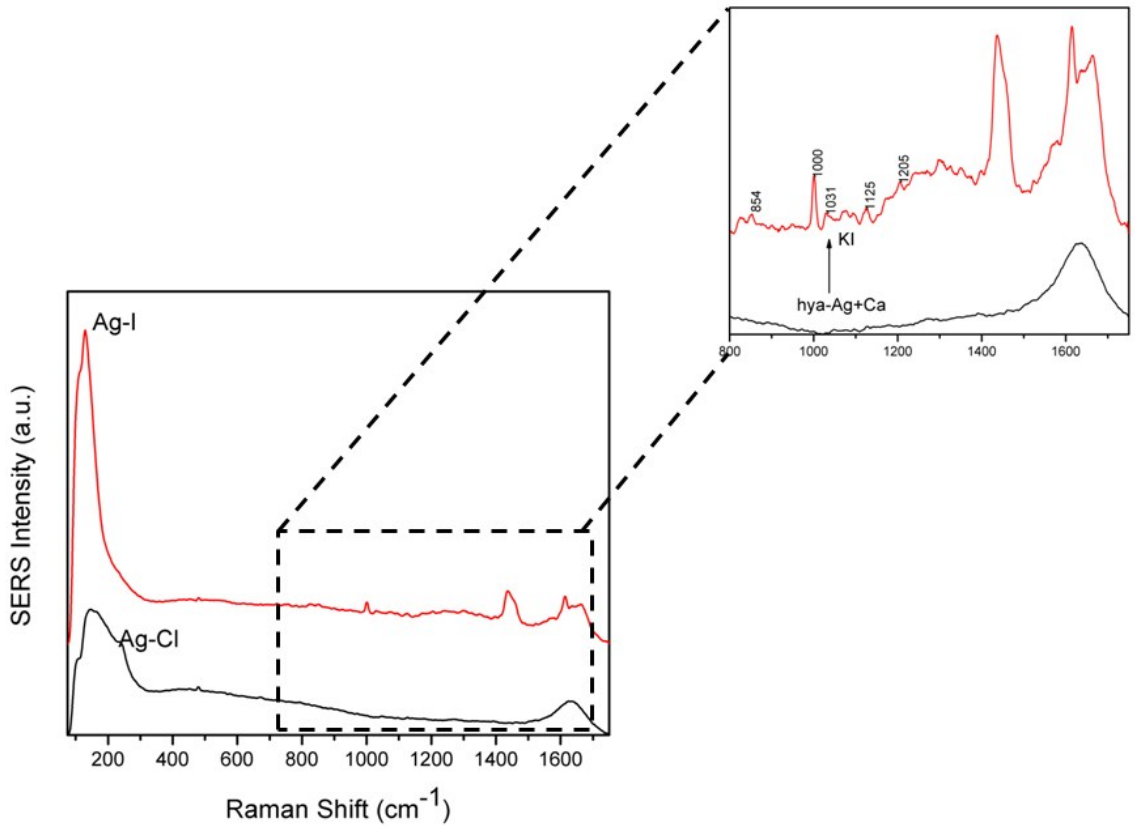
Supplementary Figure 1. The SERS spectra of IMNPs. The presence of iodide ions on the surface of the NPs is proved by Ag-I band at 130 cm^{-1}. The artifact peaks are also visible at approximately 1600 cm^{-1} and a broad band around 1400 cm^{-1}.



Supplementary Figure 2. Area of 1002 cm^{-1} peak plotted against HSA concentration in 3-200 µg/mL interval.

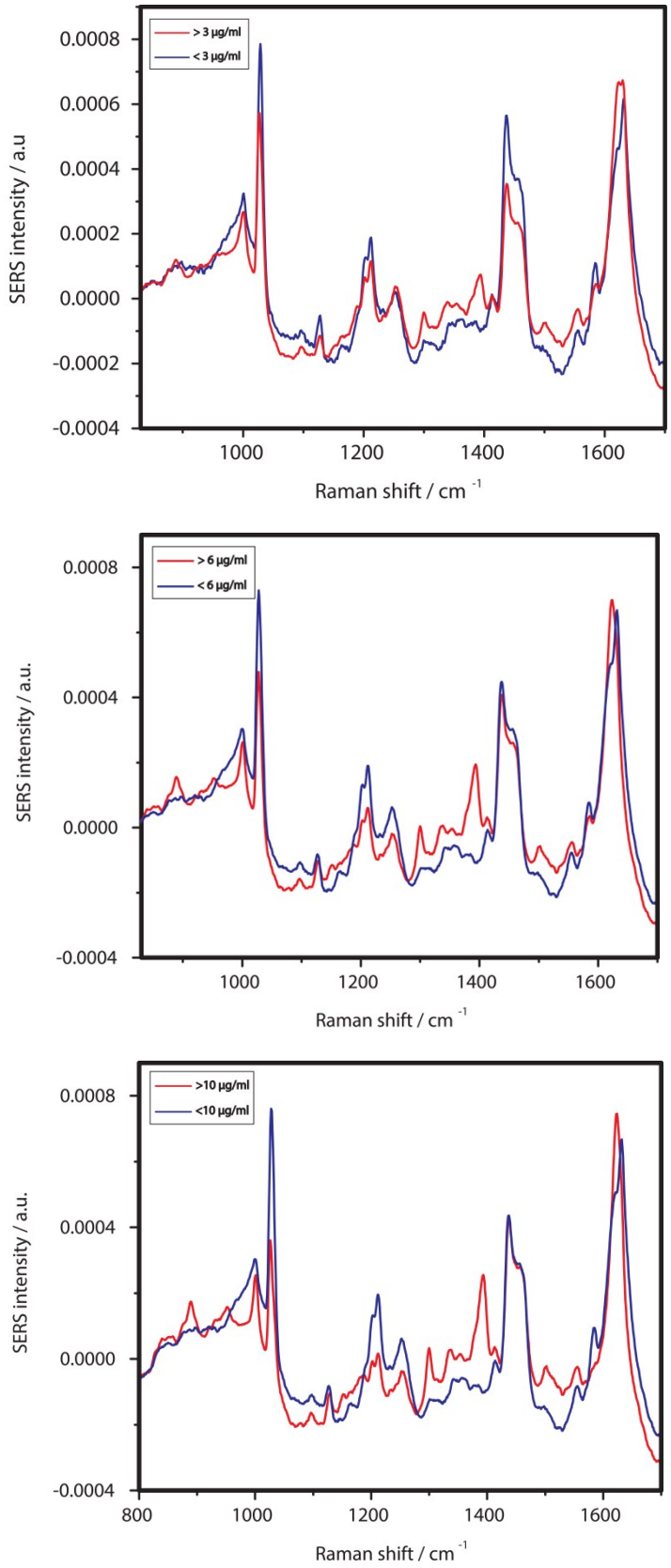


Supplementary Figure 3. Plot of RMSE against the number of factors in PLS model.

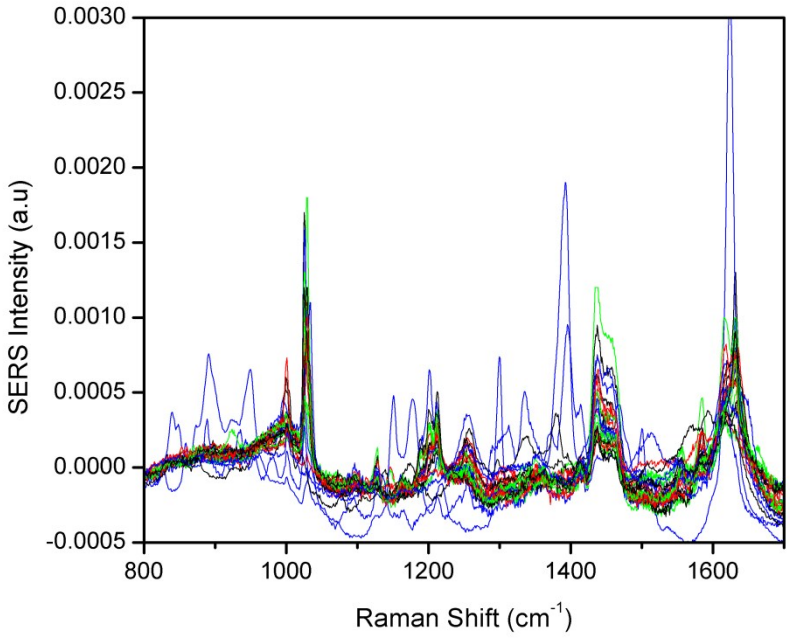


Supplementary Figure 4. SERS spectrum of HSA using iodide-modified AgNPs (red spectrum) and as synthesisezed AgNPS as substrate (black spectrum). Three measurements were averaged for each spectrum.

In order to prove the crucial role played by iodide in mediating the acquisition of SERS signal from albumin, we performed the following experiment: in 90 µl AgNPs colloidal solution obtained by reduction with hydroxylamine hydrochloride, we added 10 µl HSA (1 mg/mL) and 1 µl of $MgSO_4$ (10^{-2} M). No SERS signal could be observed from HSA because Mg^{2+} adions only lead to the adsorption of Cl^- ions, as evidenced by the weak 240 cm^{-1} SERS band (Supplementary Figure 4, black spectrum). In the same mixture, we added 1 µl of KI (0.1 M) and the characteristic SERS bands of HSA appeared at 1000, 1030 and 850 cm^{-1} along with the SERS band of Ag-I at 130 cm^{-1} (Supplementary Figure 4, red spectrum). The presence of the Ag-I SERS band at 130 cm^{-1} and lack of the Ag-Cl band at 240 cm^{-1}, indicates that chloride is replaced by iodide on the AgNPs surface. Moreover, iodide adions mediate the chemisorption of HSA to the AgNPs surface and turn on of the SERS spectrum of HSA.[1,2]



Supplementary Figure 5. The SERS spectra of urine samples, from the six groups defined by 3, 6 and 10 µg/mL cut-off values, as shown in the figure.



Supplementary Figure 6. SERS spectra from all 27 urine samples, after de-trending and mean normalization.

References

1. N. Leopold, A. Stefancu, K. Herman, I. S. Tódor, S. D. Iancu, V. Moisoiu and L. F. Leopold, *Beilstein J. Nanotechnol.*, 2018, **9**, 2236-2247.
2. A. Stefancu, S. D. Iancu, V. Moisoiu and N. Leopold, Specific and selective SERS active sites generation on silver nanoparticles by cationic and anionic adatoms, Rom. Rep. Phys., 2018, **70**, in press.