Supplementary Information

SERS reveals the specific interaction of silver and gold nanoparticles with hemoglobin and red blood cell components

*Daniela Drescher [1,2]**, Tina Büchner [1,2]**, Don McNaughton[3], Janina Kneipp [1,2]**

[1]Humboldt-Universität zu Berlin, Department of Chemistry, Brook-Taylor-Str. 2, 12489 Berlin, Germany, [2] BAM Federal Institute for Materials Research and Testing, Richard-Willstätter-Str. 11, 12489 Berlin, Germany, [3]Centre for Biospectrosopy, School of Chemistry, Monash University, 3800 Clayton, Victoria, Australia

Email: janina.kneipp@chemie.hu-berlin.de

* To whom correspondence should be addressed: Prof. Dr. Janina Kneipp, Humboldt-Universität zu Berlin, Department of Chemistry, Brook-Taylor-Str. 2, 12489 Berlin

** These authors contributed equally to the paper

S1. Additional SERS spectra of hemoglobin on AgC nanoparticles at different excitation wavelengths

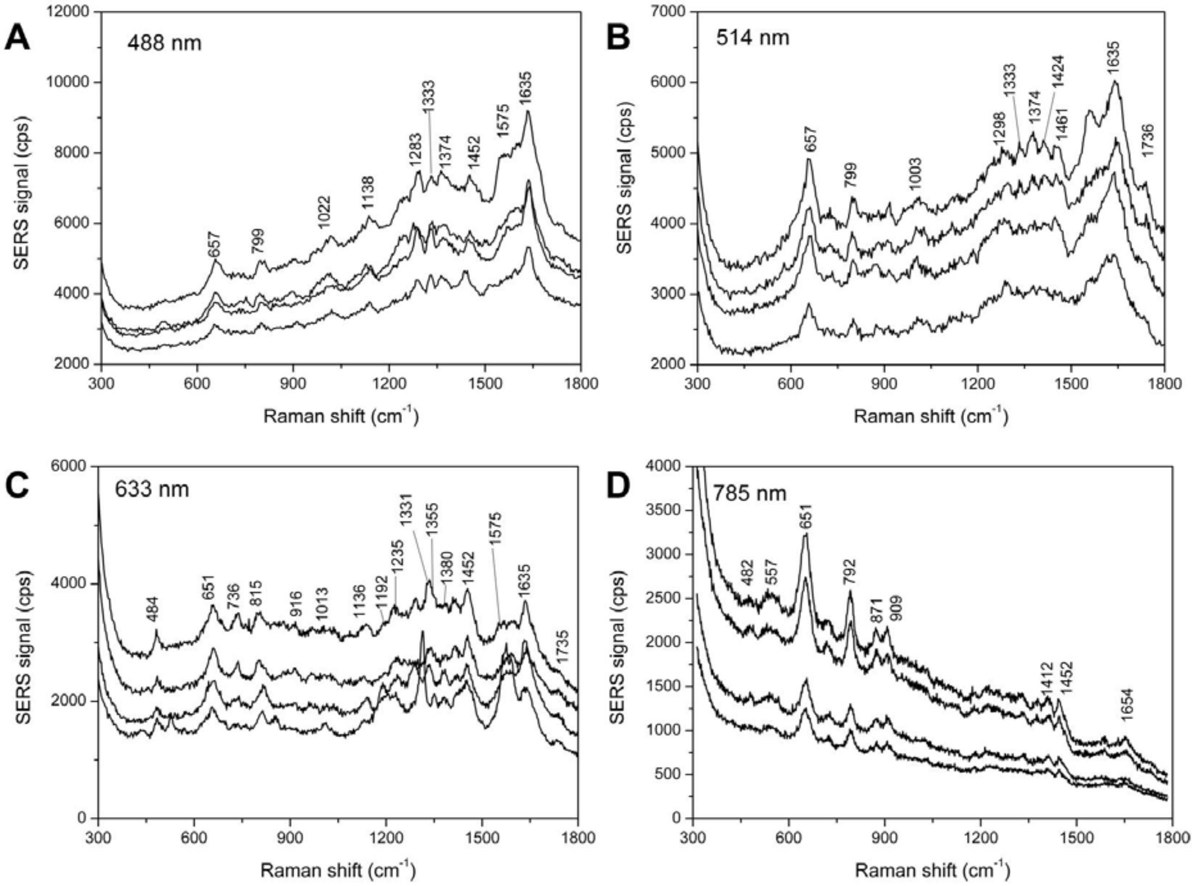

Figure S1. Four SERS spectra of hemoglobin (10 µM) out of 90 spectra (**A**) and 30 spectra (**B, C, D**), respectively obtained with AgC nanoaggregates at different excitation wavelengths (compare with Figure 1). Excitation intensities: A, B) $4{\cdot}10^5$ W/cm^2, **C**) $3{\cdot}10^5$ W/cm^2, D) $2{\cdot}10^6$ W/cm^2, acquisition time: 1 s.

S2. Characterization of red blood cells by normal Raman scattering

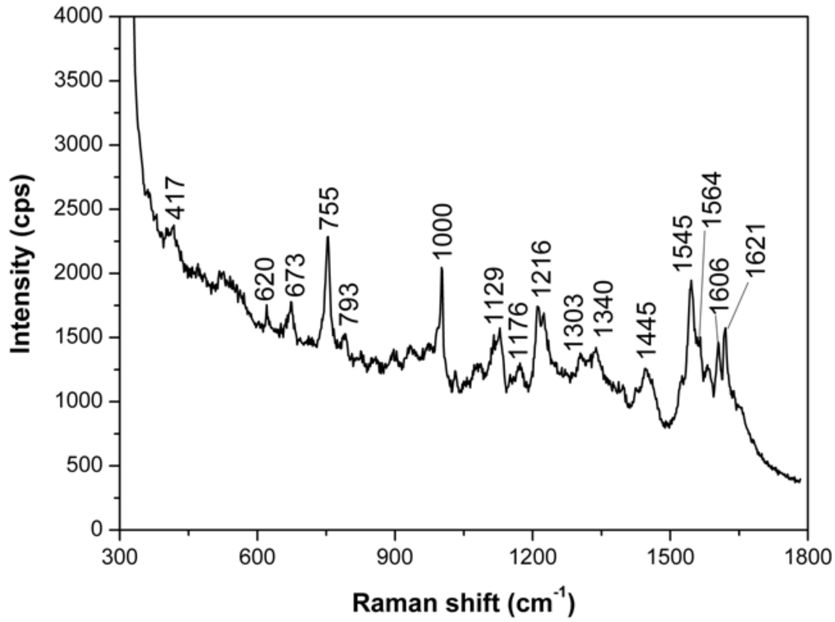

Figure S2. Raman spectrum of red blood cell recorded at an excitation wavelength of 785 nm. Excitation intensity: $2{\cdot}10^6$ W/cm^2, acquisition time: 10 s.

Table S1. Assignments of Raman signals obtained from red blood cells (see Figure S1) based on refs.[1-3] Excitation wavelength: 785 nm.

Raman shift (cm^{-1})	Assignment to globin	Assignment to porphyrin
417		δ(Fe-O-O)
673		δ(pyr deform)$_{sym}$, ν_7
755		ν(pyr breathing), ν_{15}
793		ν(pyr breathing), ν_6
1000	δ(CH$_{ring\ in\ plane}$)$_{sym}$ (Phe)	
1129		ν(C$_\beta$-Methyl), ν_5
1176		ν(pyr half-ring)$_{asym}$, ν_{30}
1216		δ(C$_m$H), ν_{13} oder ν_{42}
1303		δ(C$_m$H), ν_{21}
1340		ν(pyr half-ring)$_{sym}$, ν_{41}
1445		δ(CH_2/CH_3)
1545		ν(C$_\beta$C$_\beta$), ν_{11}
1564		ν(C$_\beta$C$_\beta$), ν_{19}
1606		ν(C$_a$=C$_b$), ν(C=C)$_{vinyl}$
1621		ν(C$_a$=C$_b$), ν(C=C)$_{vinyl}$

S3. UV-vis absorbance spectra of nanoparticles with hemoglobin

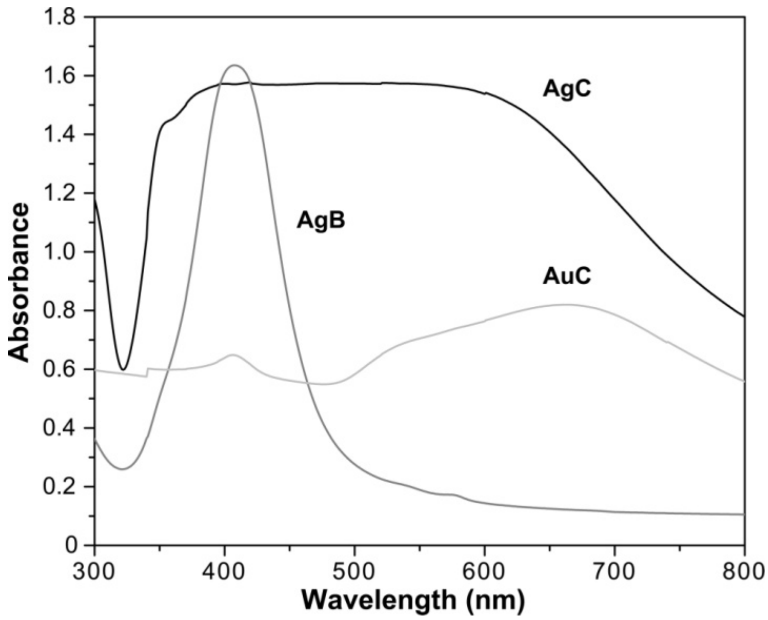

Figure S3. UV-vis absorbance spectra of AgC, AgB and AuC nanoparticles after addition of hemoglobin (10 μM).

S4. UV-vis absorption spectra of hemoglobin at different temperatures

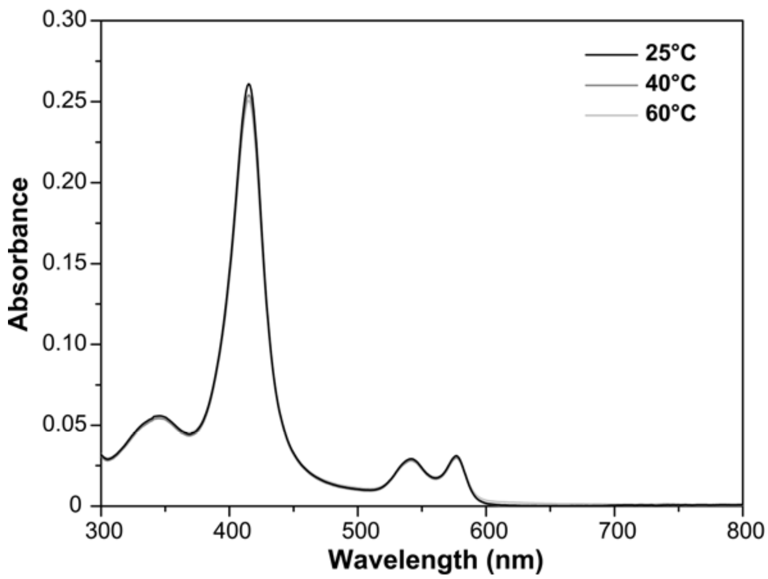

Figure S4. UV-vis absorption spectra of hemoglobin after incubation at different temperatures for 20 min.

S5. Additional SERS spectra of hemoglobin on different nanoparticles after incubation at 40°C and 60°C

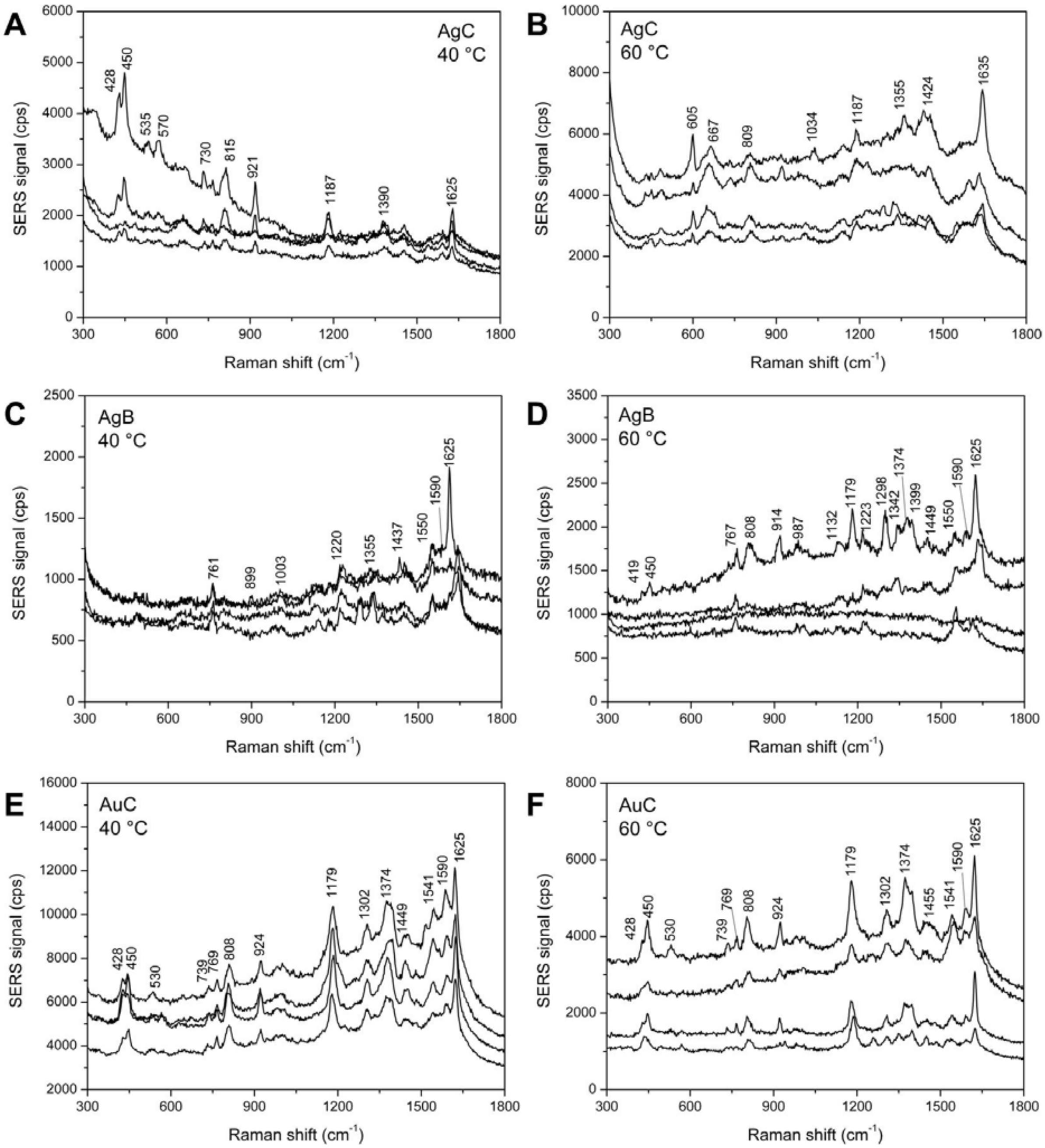

Figure S5. Additional SERS spectra of hemoglobin (10 μM) incubated at 40°C **(A, C, E)** and 60 °C **(B, D, F**), obtained with AgC (**A**, **B**), AgB (**C**, **D**), and AuC (**E**, **F**) nanoaggregates (compare with Figure 3). Excitation wavelength: 633 nm, excitation intensity: $7.9 \cdot 10^3$ W/cm^2, acquisition time: 1 s.

S6/7. Additional SERS spectra of ghost cells and erythrocytes on AgC nanoparticles

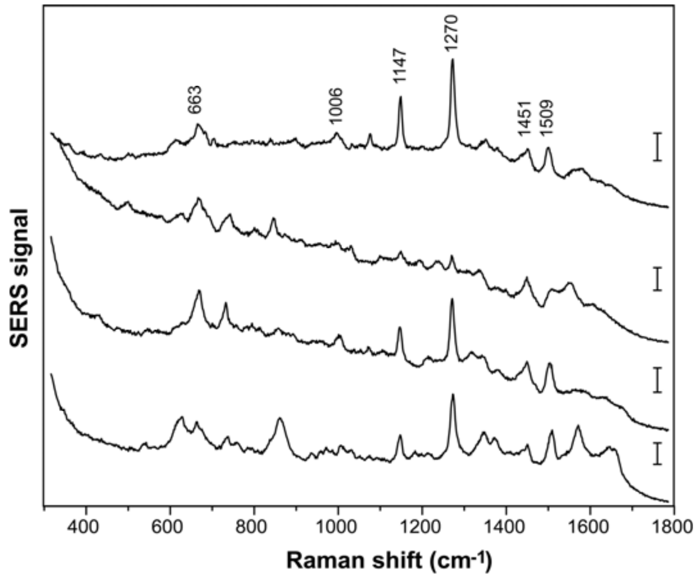

Figure S6. SERS spectra of four ghost cells after addition of AgC nanoparticles. Bands at 663, 1006, 1147, 1270, 1371, 1451 and 1509 cm^{-1} are visible in all ghost cell spectra. Excitation wavelength: 785 nm, excitation intensity: $2{\cdot}10^5$ W/cm^2, acquisition time 1 s, scale bars 1500 cps.

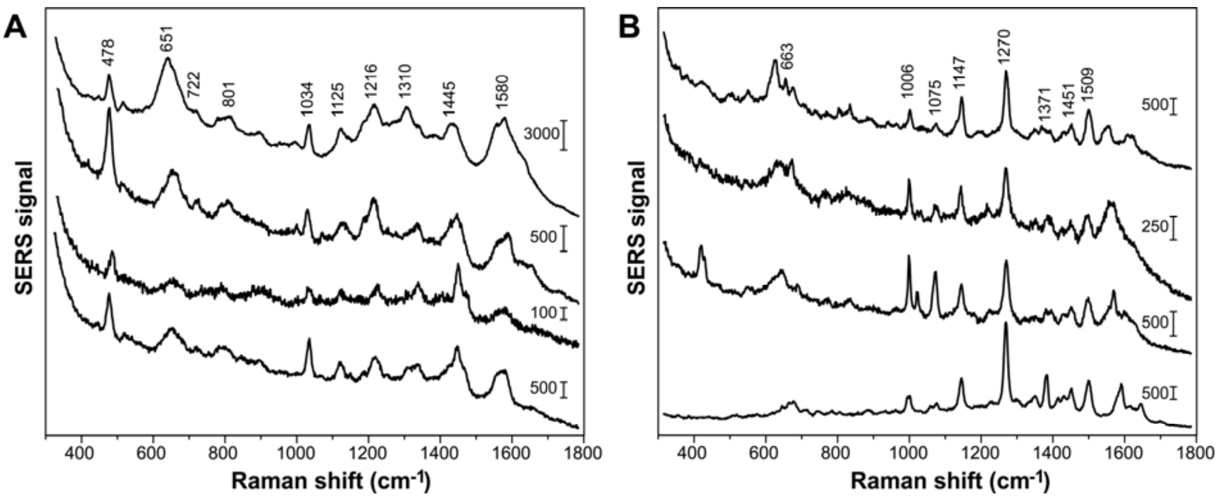

Figure S7. SERS spectra of four different RBCs after incubation with AgC nanoparticles for 15 minutes (**A**) and 18 hours (**B**). The marked bands are found in all cell spectra. Excitation wavelength: 785 nm, excitation intensity: $2{\cdot}10^5$ W/cm^2 (RBCs 15 min), $6{\cdot}10^5$ W/cm^2 (RBCs 18 h), acquisition time 1 s.

S8. Estimation of the maximum fraction of hemoglobin molecules in contact with the nanoparticles

For the estimation of the number of hemoglobin molecules (Hb) adsorbed on nanoparticles, all nanoparticles, and the Hb molecules were considered to be spherical as an approximation. The nanoparticle surface covered by one Hb molecule, was assumed to be that of a circular area defined by the diameter of the Hb spheres (Hb diameter 5.5 nm [4]). The number of Hb per nanoparticle was calculated from the nanoparticle surface assuming a total coverage of the nanoparticles with Hb. For the calculations of the nanoparticles surfaces we used the nanoparticle diameters obtained from TEM measurements (see. Fig. 2 and Table 2) of AgC: 55 nm, AgB: 14 nm, and AuC: 32 nm). The Hb concentration in the experiments was about 10 µM as determined from the UV-vis absorption spectra. The nanoparticle concentrations were calculated from the amounts of $AgNO_3$ or $HAuCl_4$ used for synthesis (both can be assumed to be fully used in the reduction due to excess sodium citrate and borohydride) and from the average particle sizes as determined by TEM. From the Hb and nanoparticle concentration and from the number of Hb molecules per nanoparticle, we estimated the maximum fraction of Hb in contact with the nanoparticles.

References

1 S. Hu, K. M. Smith and T. G. Spiro, *J. Am. Chem. Soc.*, 1996, **118**, 12638-12646.
2 T. G. Spiro, R. S. Czernuszewicz and X.-Y. Li, *Coord. Chem. Rev.*, 1990, **100**, 541-571.
3 B. R. Wood, P. Caspers, G. J. Puppels, S. Pandiancherri and D. McNaughton, *Anal. Bioanal. Chem.*, 2007, **387**, 1691-1703.
4 M. F. Perutz, H. Muirhead, J. M. Cox, L. C. G. Goaman, F. S. Mathews, E. L. McGandy and L. E. Webb, *Nature*, 1968, **219**, 29-32.