Electronic Supplementary Material (ESI) for Nanoscale.

Electronic Supplementary Information

Raman tweezers microspectroscopy of *circa* 100-nm extracellular vesicles

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Content of ESI file

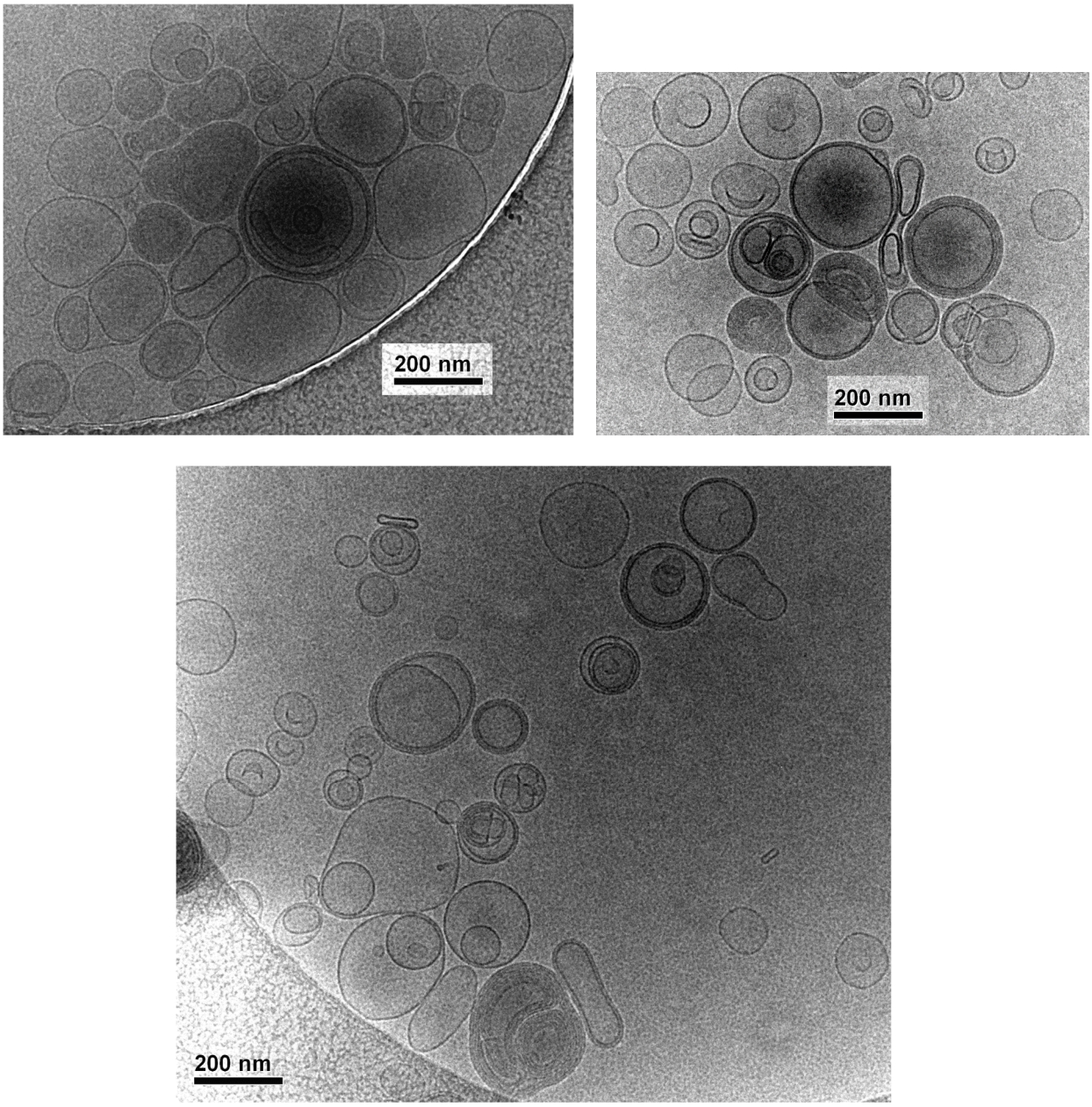

Figure S1. Cryo-TEM images of liposomes DOPC “200 nm”.

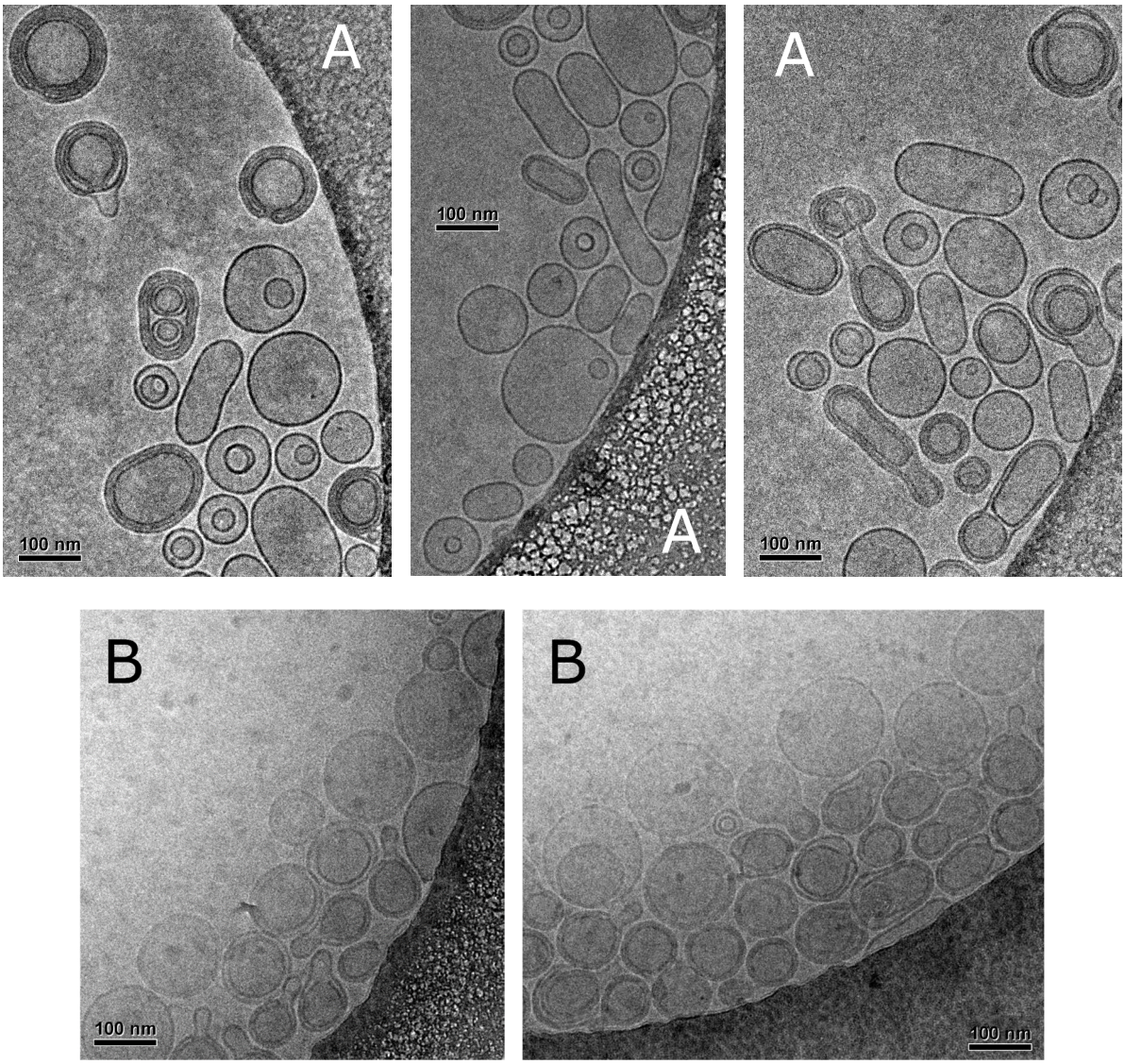

Figure S2. Cryo-TEM images of liposomes DOPC “100 nm”, from two different preparations (A and B).

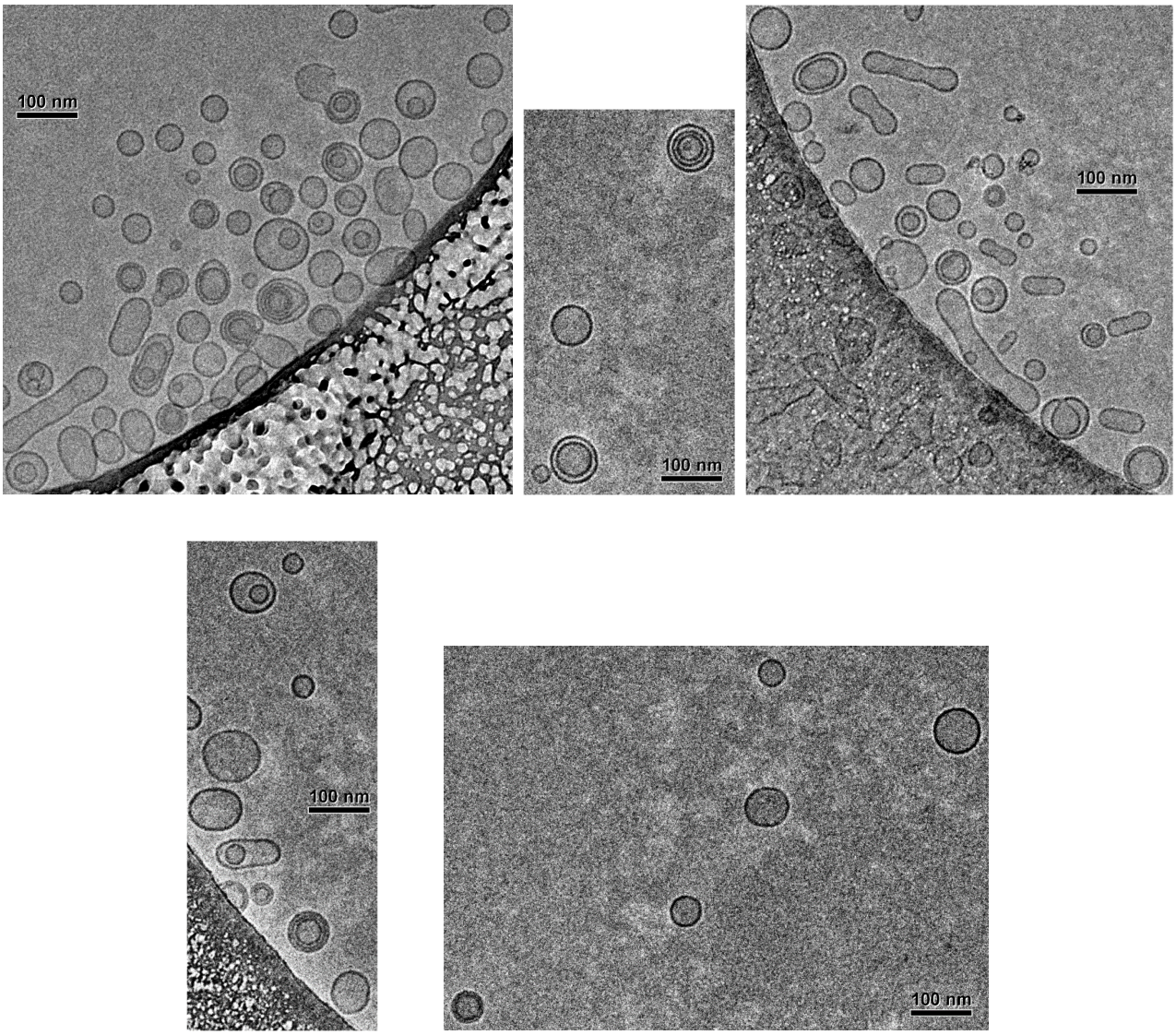

Figure S3. Cryo-TEM images of liposomes DOPC “50 nm”.

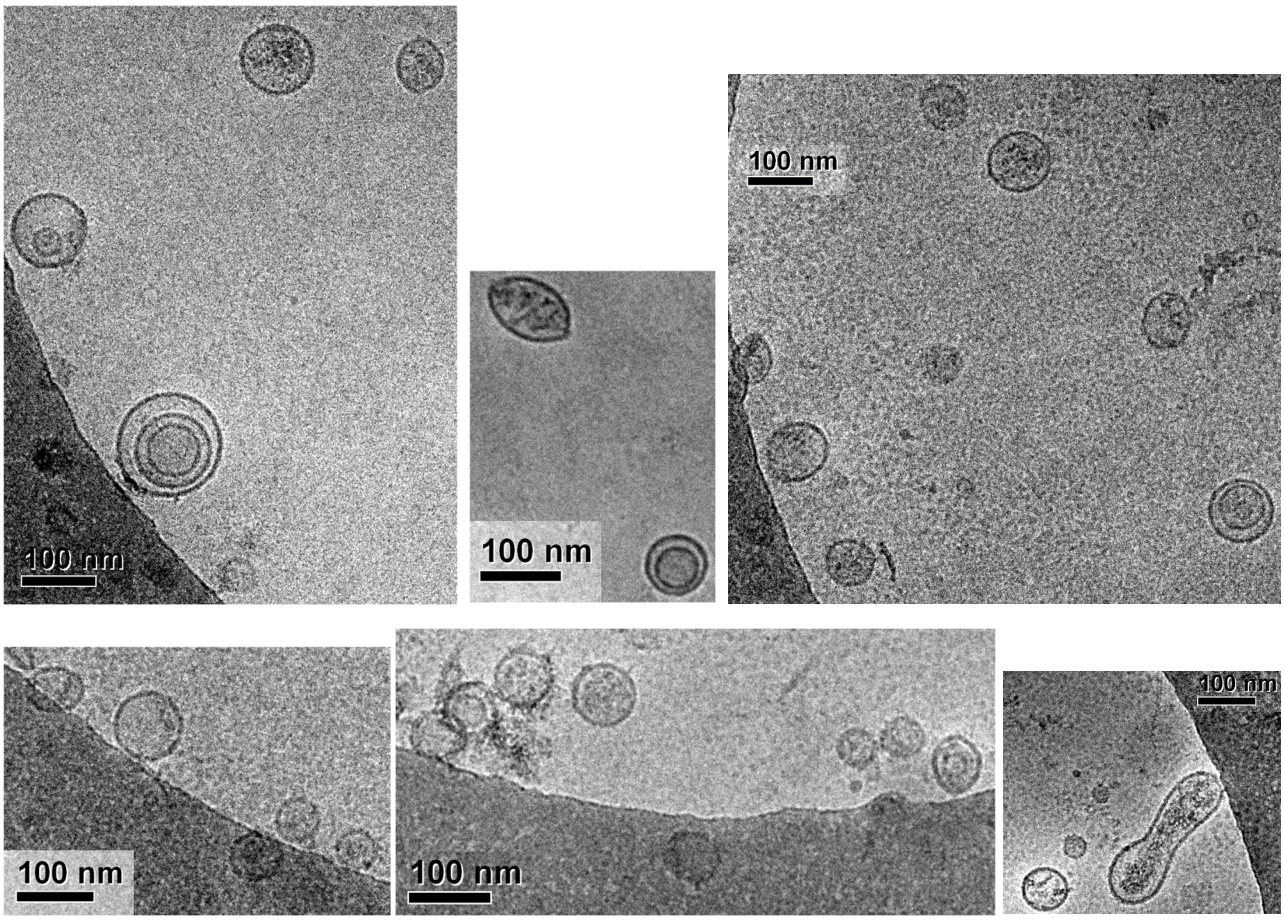

Figure S4. Cryo-TEM images of exosomes from human urine (sample HU-1).

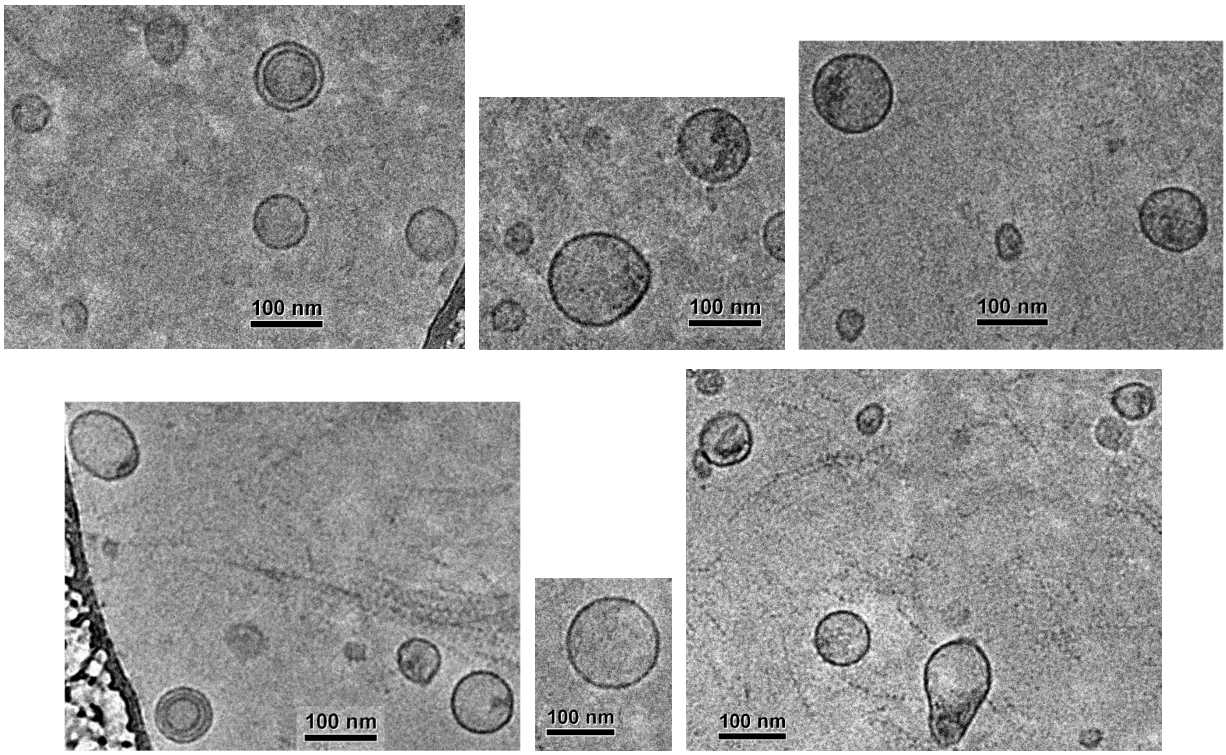

Figure S5. Cryo-TEM images of exosomes from human urine (sample HU-2).

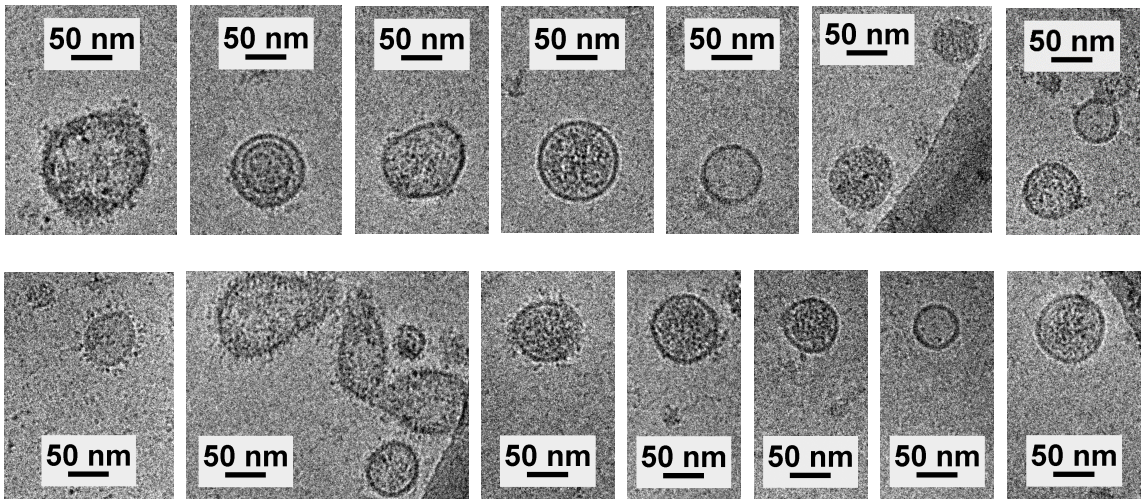

Figure S6. Cryo-TEM images of exosomes from rat hepatocytes (sample RH-1).

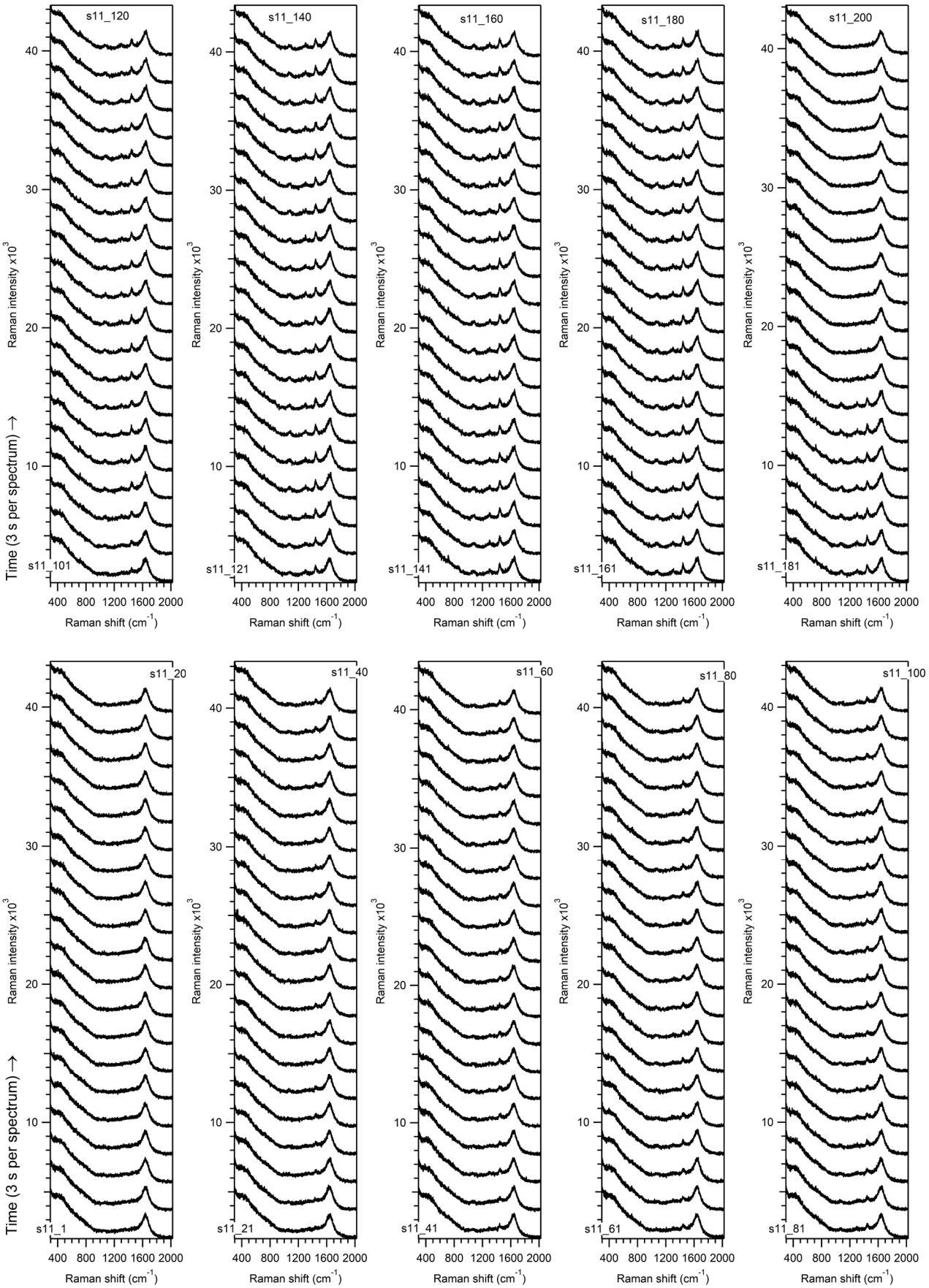

Figure S7. Time sequence (from left bottom to right top) of 200 raw Raman spectra of liposomes DOPC "100 nm" (C_p = 1.5 x 10^{11} ml^{-1}), corresponding to the data of Fig. 3(A).

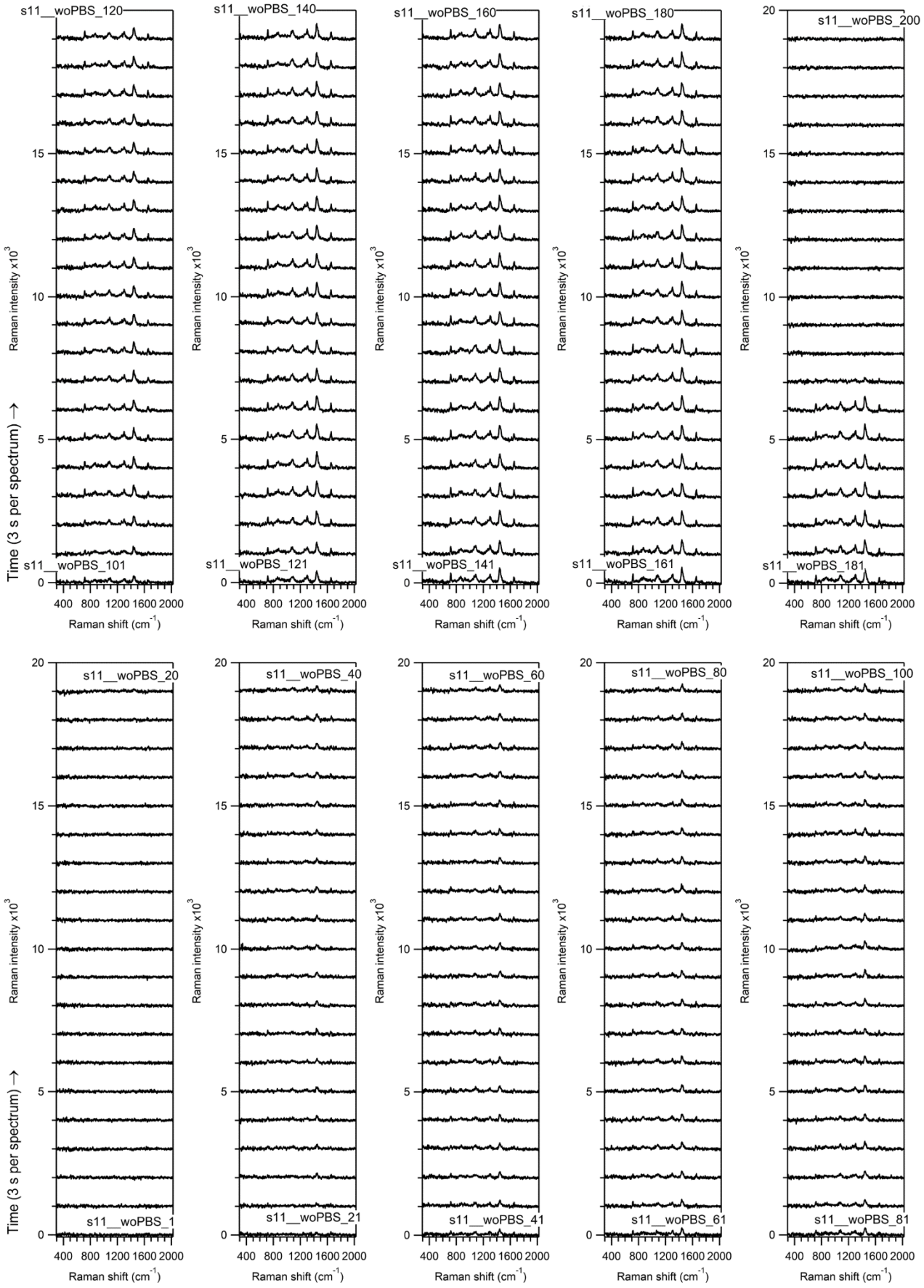

Figure S8. Time sequence (from left bottom to right top) of 200 normalized Raman spectra after PBS subtraction of liposomes DOPC "100 nm" (C_p = 1.5 x 10^{11} ml^{-1}), corresponding to the data of Fig. 3 (B-D), S7.

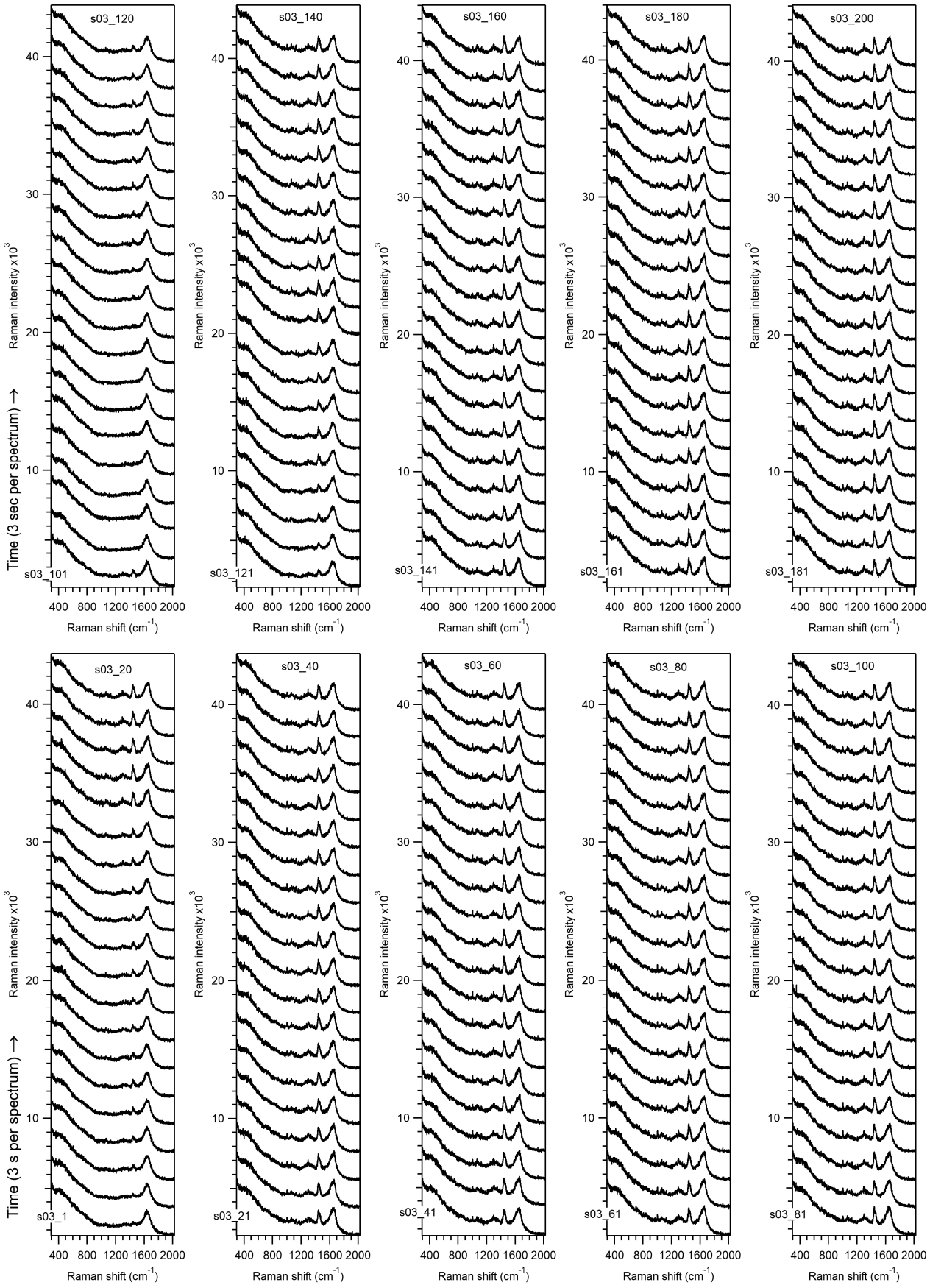

Figure S9. Time sequence (from left bottom to right top) of 200 raw Raman spectra of exosomes from human urine (sample HU-1, C_p = 1.6 x 10^{10} ml^{-1}), corresponding to the data of Fig. 4 (A, B).

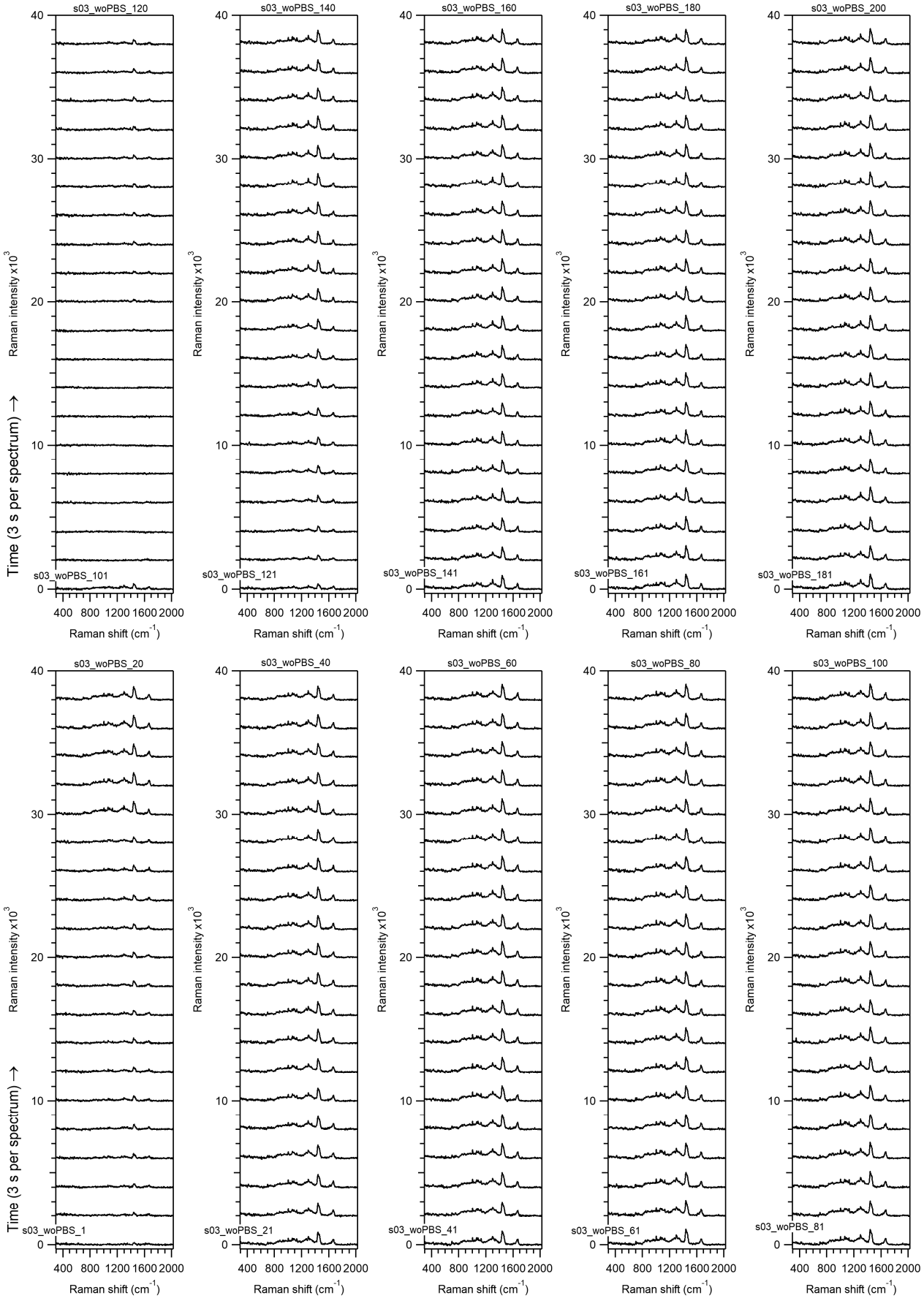

Figure S10. Time sequence (from left bottom to right upper corner) of 200 normalized Raman spectra after PBS subtraction for exosomes from human urine (sample HU-1, C_p = 1.6 x 10^{10} ml^{-1}), corresponding to the data of Fig. 4 (A, B), S9.

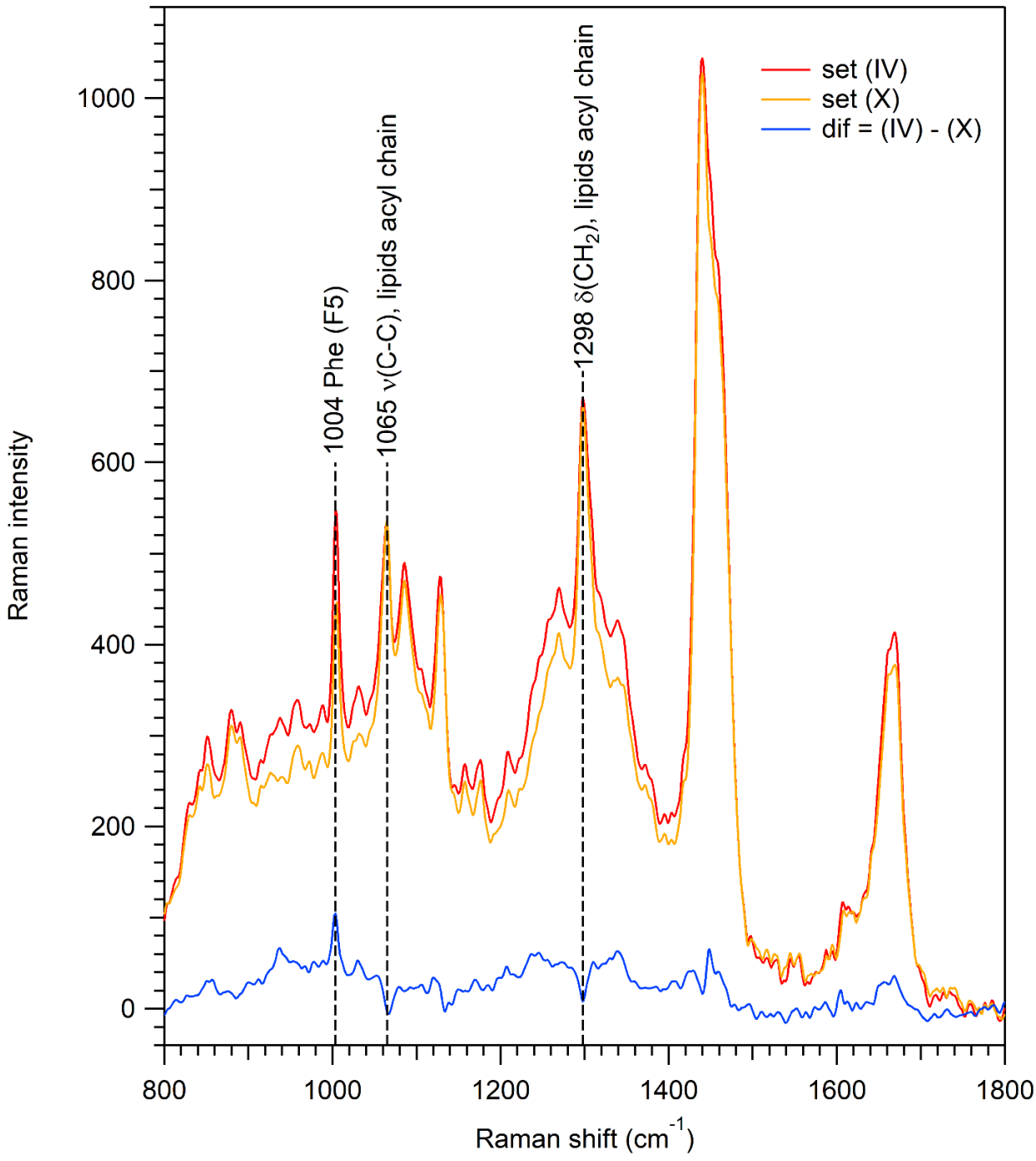

Figure S11. Raman spectra corresponding to the sets (IV, red curve) and (X, yellow curve) in Figure 4B, and their (1:1) difference spectrum (blue curve). The difference spectrum exhibits one pronounced positive peak at 1004 cm^{-1} due to protein's Phe band and two pronounced negative peaks, at 1065 and 1298 cm^{-1} due to lipids acyl chain vibrations. These peaks are marked by dashed vertical lines.

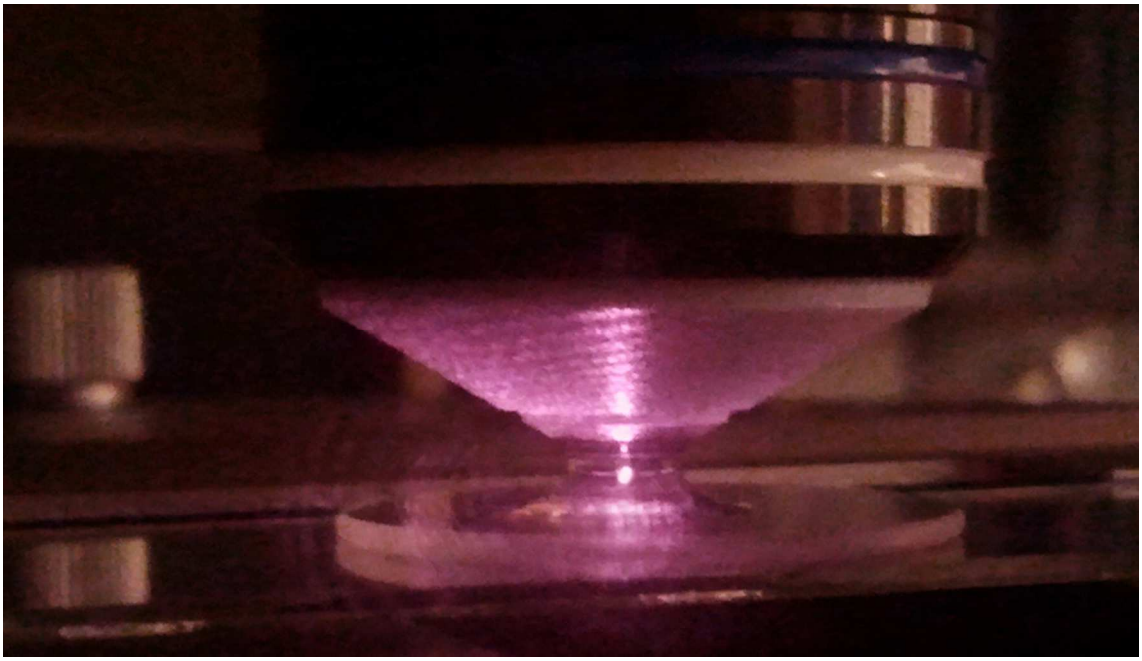

Figure S12. Photo: in the event of multiple-vesicles trapping, a bright diffusive spot appears within the optical trap, in the objective's focal plane.

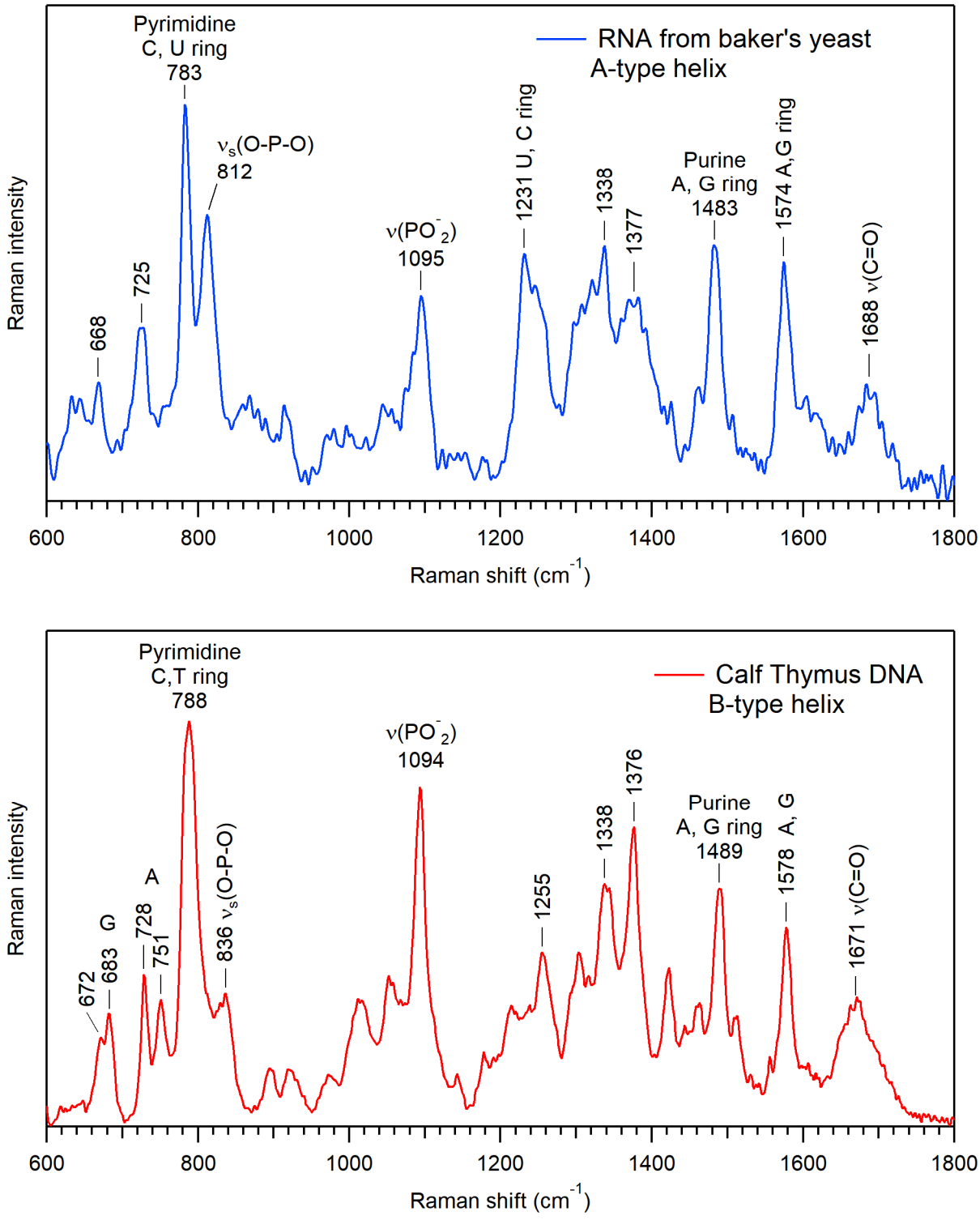

Figure S13. Characteristic Raman spectra of two nucleic acids in PBS solution (pH 7.4) recorded using our Raman setup with excitation at 780 nm. Concentration of nucleic acid is 7 mg/ml in both cases. PBS contribution is subtracted.

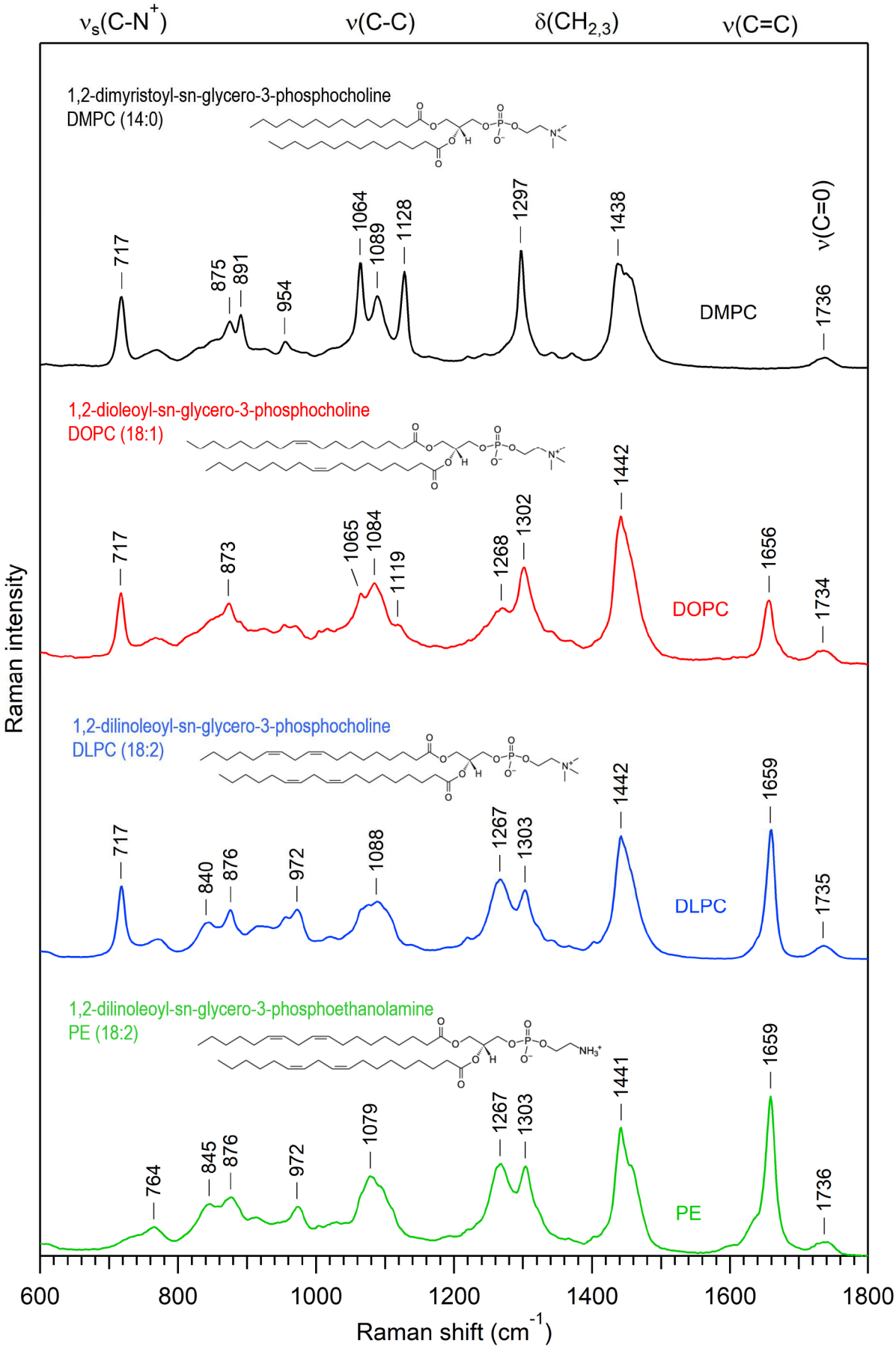

Figure S14. Characteristic Raman spectra of several membrane lipids in liposomes "200 nm" in HEPES buffer (pH 8) recorded using our Raman setup with excitation at 780 nm. Lipids concentration is 100 µg/ml. Water buffer contribution is subtracted.

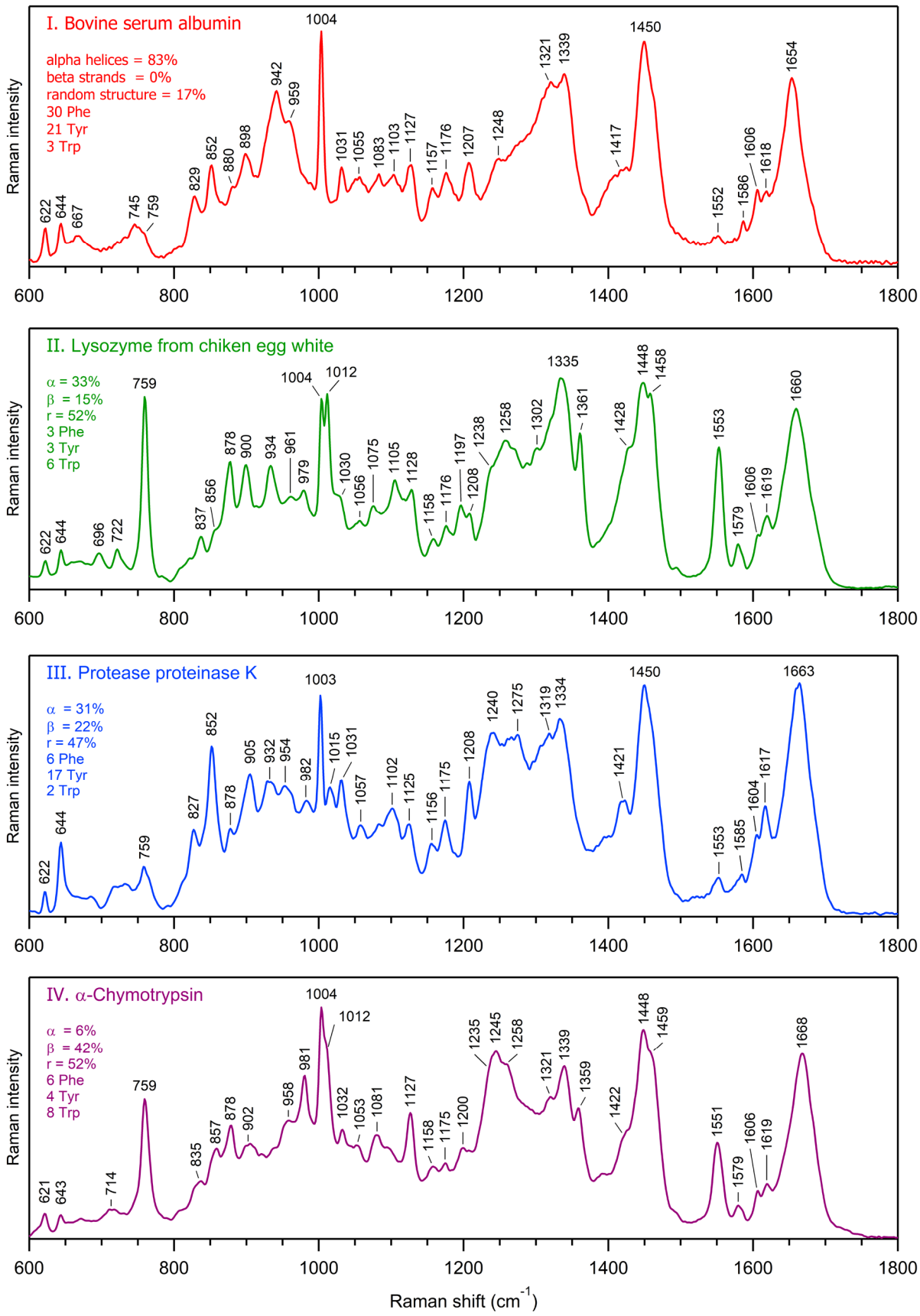

Figure S15. Characteristic Raman spectra of several proteins in HEPES buffer (I, IV) (10 mM, pH 7.4) and PBS (II, III) (pH 7.4) recorded using our Raman setup with excitation at 780 nm. Water buffer contribution is subtracted.

Table S1. Major marker Raman bands for biomolecular components analysis

Biomolecule	Frequency range (cm^{-1})
Nucleic acids	782 – 788
Nucleic acid: A-type helix	810 – 812
Nucleic acid: B-type helix	830 – 840 (shoulder)
Carotenoids	1515 – 1540
Lipids	1050 – 1130, 1720 – 1750
Lipids: Cholesterol	700 – 704
Lipids: $\nu(C\text{-}N^{+})$ of polar head	717
Lipids: Ergosterol	1602 – 1604
Proteins: Phenylalanine	1003 – 1004
Proteins: Tyrosine	643 – 645
Proteins: Tryptophan	758 – 759, 1012