Molecular fingerprinting of biological nanoparticles with a label-free optofluidic platform

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Figs. S1 to S6
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Other Supplementary Materials for this manuscript include the following:

Movies S1 to S2

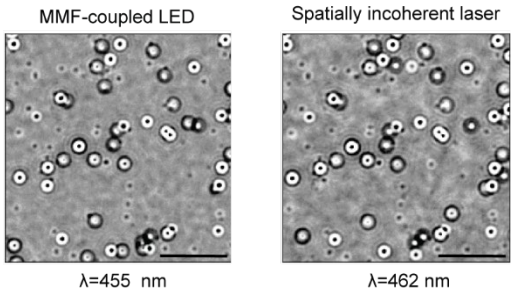

Fig. S1. Equivalence between spatially incoherent illumination schemes. Representative zoom-in images of the same sample comparing two different light sources used for producing spatially incoherent illumination. Both illumination schemes use the same MMF fibre. The spatial coherence of the laser source is drastically reduced upon passing through a rotating ground glass diffuser prior to coupling to the MMF fibre. Scale bars: 5 μm.

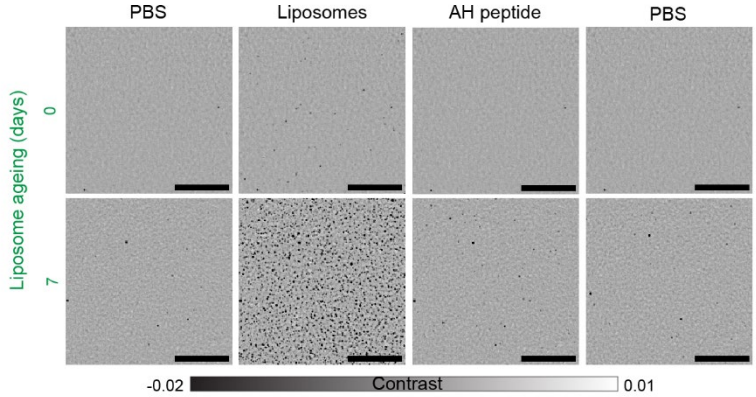

Fig. S2. Effect of liposome ageing on SLB formation. Liposome ageing affects the probability of spontaneous vesicle rupture but not the overall SLB quality upon treatment with the peptide. Representative zoom-in images for each preparation method at the different stages of the peptide-mediated supported lipid bilayer formation process. Scale bars: 5 μm.

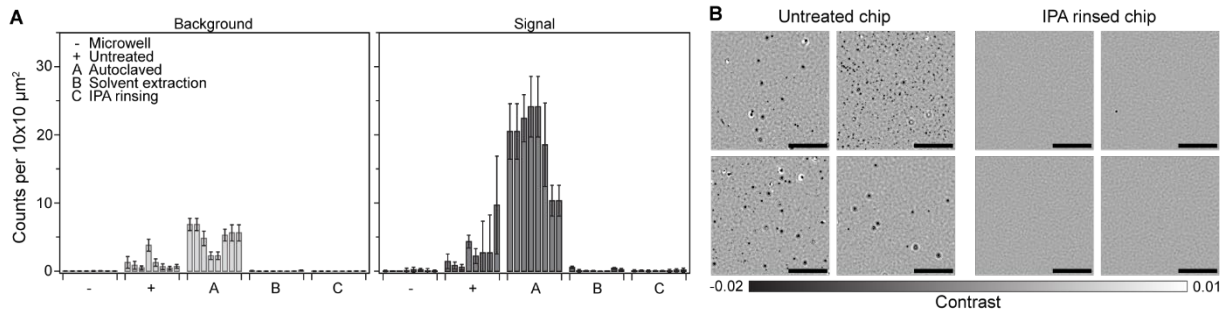

Fig. S3. Effect of different PDMS cleaning strategies to remove uncured oligomers. (A) Mean substrate defect densities as a function of different PDMS cleaning strategies. Microwells acted as negative control due to the lower PDMS surface area and thus lower likelihood for uncured oligomers to leach towards the substrate surface. Untreated PDMS chips were considered as a positive control. Bars represent the median defect density recorded within a single FOV (0.01 µm^2) over a sensing area of 0.2 mm^2, with error bars indicating the standard deviation. **(B)** representative zoom-in images of the substrate upon addition of PBS for an untreated and IPA rinsed PDMS chip, respectively. Scale bars: 10 µm.

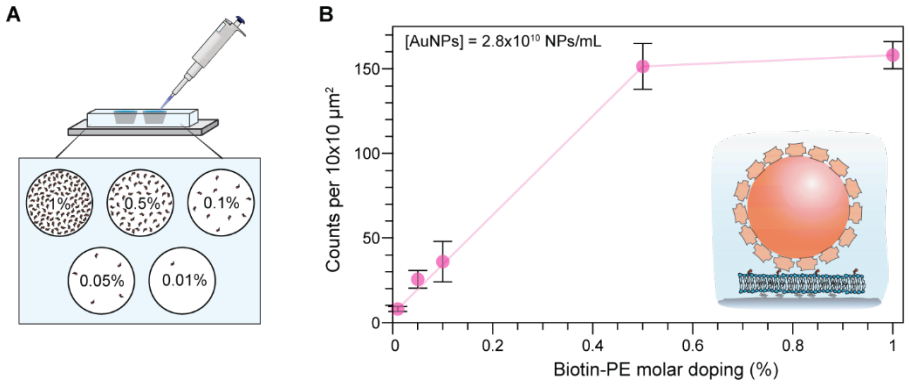

Fig. S4. Lipid composition of the SLB determines the capture site density. **(A)** Cartoon depicting the control over the capture sites by tuning the percentage of biotin-PE lipid included in the SLB formation. **(B)** Biotin-DOPE doping response curve for a fixed AuNP-SAv concentration. Inset depicts the pulldown assay consisting of a biotinylated SLB and AuNP-SAv.

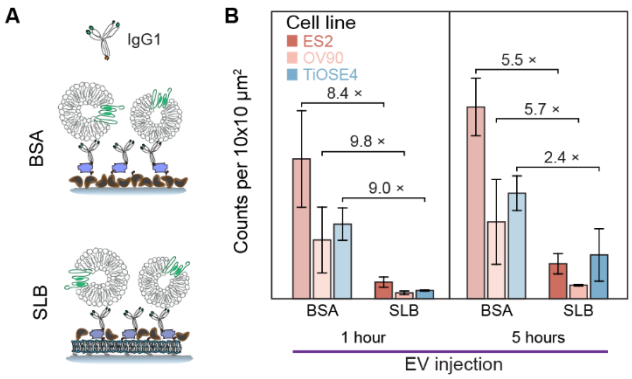

Fig. S5. Non-specific binding comparison between BSA and SLB. (A) Cartoon depicting the two surface functionalisation strategies compared using IgG1 isotype control antibody as the capture probe. **(B)** Number of non-specifically bound EVs for the two functionalisation schemes across three different ovarian cell lines. Numbers above the bars indicate the fold increase of non-specific binding to BSA functionalised chips compared to SLB ones. Errors bars indicate the standard deviation over the mean (N = 2).

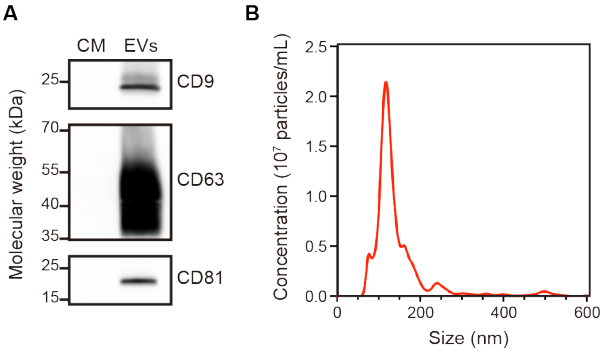

Fig. S6. ES2 extracellular vesicle characterisation. (A) Western blot analysis against the characteristic EV tetraspanin markers: CD9, CD63, CD81. CM: cultured medium. **(B)** Size and concentration determination via nanoparticle tracking analysis.

Movie S1.

Following the on-chip surface functionalisation in real time: peptide assisted bilayer formation. The video shows a zoomed area corresponding to 31.2 μm x 31.2 μm of the bilayer formation process using 30 nm extruded liposomes. The total imaged area covers a total area of 0.2 mm^2. The data is recorded at 1 Hz and is represented with a contrast range of -0.04 to 0.03. Scale bar: 10 μm.

Movie S2.

Following the on-chip surface functionalisation in real time: liposome fusion bilayer formation. The video shows a zoomed area corresponding to 31.2 μm x 31.2 μm of the bilayer formation process using sonicated liposomes ranging in size from 100-3000 nm. The total imaged area covers a total area of 0.2 mm^2 The data were recorded at 1 Hz and are represented with a contrast range of -0.04 to 0.03. Scale bar: 10 μm.