Supporting Information

Selective Placement of Functionalised DNA Origami via Thermal Scanning Probe Lithography Patterning

Tingting Zheng [a], Caoimhe O'Neill [b], John F. Marshall [c], Thomas Iskratsch [b, *], Matteo Palma[a,*]

[a] Department of Chemistry, Queen Mary University of London, Mile End Road, London E1 4NS, United Kingdom.

[b] School of Engineering and Materials Science, Queen Mary University of London, Mile End Road, London E1 4NS, United Kingdom.

[c] Barts Cancer Institute, Cancer Research UK Centre of Excellence, Queen Mary University of London, Charterhouse Square, London EC1M 6BQ, United Kingdom

* t.iskratsch@qmul.ac.uk
* m.palma@qmul.ac.uk

DNA sequences tables:

1. List of unmodified staple strands

A01	CGGGGTTTCCTCAAGAGAAGGATTTTGAATTA
A02	AGCGTCATGTCTCTGAATTTACCGACTACCTT
A03	TTCATAATCCCCTTATTAGCGTTTTTCTTACC
A04	ATGGTTTATGTCACAATCAATAGATATTAAAC
A05	TTTGATGATTAAGAGGCTGAGACTTGCTCAGTACCAGGCG
A06	CCGGAACCCAGAATGGAAAGCGCAACATGGCT
A07	AAAGACAACATTTTCGGTCATAGCCAAAATCA
A08	GACGGGAGAATTAACTCGGAATAAGTTTATTTCCAGCGCC
A09	GATAAGTGCCGTCGAGCTGAAACATGAAAGTATACAGGAG
A10	TGTACTGGAAATCCTCATTAAAGCAGAGCCAC
A11	CACCGGAAAGCGCGTTTTCATCGGAAGGGCGA
A12	CATTCAACAAACGCAAAGACACCAGAACACCCTGAACAAA
A13	TTTAACGGTTCGGAACCTATTATTAGGGTTGATATAAGTA
A14	CTCAGAGCATATTCACAAACAAATTAATAAGT
A15	GGAGGGAATTTAGCGTCAGACTGTCCGCCTCC
A16	GTCAGAGGGTAATTGATGGCAACATATAAAAGCGATTGAG
A17	TAGCCCGGAATAGGTGAATGCCCCCTGCCTATGGTCAGTG
A18	CCTTGAGTCAGACGATTGGCCTTGCGCCACCC
A19	TCAGAACCCAGAATCAAGTTTGCCGGTAAATA
A20	TTGACGGAAATACATACATAAAGGGCGCTAATATCAGAGA
A21	CAGAGCCAGGAGGTTGAGGCAGGTAACAGTGCCCG
A22	ATTAAAGGCCGTAATCAGTAGCGAGCCACCCT
A23	GATAACCCACAAGAATGTTAGCAAACGTAGAAAATTATTC
A24	GCCGCCAGCATTGACACCACCCTC
A25	AGAGCCGCACCATCGATAGCAGCATGAATTAT
A26	CACCGTCACCTTATTACGCAGTATTGAGTTAAGCCCAATA
A27	AGCCATTTAAACGTCACCAATGAACACCAGAACCA
A28	ATAAGAGCAAGAAACATGGCATGATTAAGACTCCGACTTG
A29	CCATTAGCAAGGCCGGGGGAATTA
A30	GAGCCAGCGAATACCCAAAAGAACATGAAATAGCAATAGC
A31	TATCTTACCGAAGCCCAAACGCAATAATAACGAAAATCACCAG
A32	CAGAAGGAAACCGAGGTTTTTAAGAAAAGTAAGCAGATAGCCG
A33	CCTTTTTTCATTTAACAATTTCATAGGATTAG
A34	TTTAACCTATCATAGGTCTGAGAGTTCCAGTA
A35	AGTATAAAATATGCGTTATACAAAGCCATCTT
A36	CAAGTACCTCATTCCAAGAACGGGAAATTCAT
A37	AGAGAATAACATAAAAACAGGGAAGCGCATTA
A38	AAAACAAAATTAATTAAATGGAAACAGTACATTAGTGAAT
A39	TTATCAAACCGGCTTAGGTTGGGTAAGCCTGT
A40	TTAGTATCGCCAACGCTCAACAGTCGGCTGTC
A41	TTTCCTTAGCACTCATCGAGAACAATAGCAGCCTTTACAG
A42	AGAGTCAAAAATCAATATATGTGATGAAACAAACATCAAG
A43	ACTAGAAATATATAACTATATGTACGCTGAGA
A44	TCAATAATAGGGCTTAATTGAGAATCATAATT

A45	AACGTCAAAAATGAAAAGCAAGCCGTTTTTATGAAACCAA
A46	GAGCAAAAGAAGATGAGTGAATAACCTTGCTTATAGCTTA
A47	GATTAAGAAATGCTGATGCAAATCAGAATAAA
A48	CACCGGAATCGCCATATTTAACAAAATTTACG
A49	AGCATGTATTTCATCGTAGGAATCAAACGATTTTTTGTTT
A50	ACATAGCGCTGTAAATCGTCGCTATTCATTTCAATTACCT
A51	GTTAAATACAATCGCAAGACAAAGCCTTGAAA
A52	CCCATCCTCGCCAACATGTAATTTAATAAGGC
A53	TCCCAATCCAAATAAGATTACCGCGCCCAATAAATAATAT
A54	TCCCTTAGAATAACGCGAGAAAACTTTTACCGACC
A55	GTGTGATAAGGCAGAGGCATTTTCAGTCCTGA
A56	ACAAGAAAGCAAGCAAATCAGATAACAGCCATATTATTTA
A57	GTTTGAAATTCAAATATATTTTAG
A58	AATAGATAGAGCCAGTAATAAGAGATTTAATG
A59	GCCAGTTACAAAATAATAGAAGGCTTATCCGGTTATCAAC
A60	TTCTGACCTAAAATATAAAGTACCGACTGCAGAAC
A61	GCGCCTGTTATTCTAAGAACGCGATTCCAGAGCCTAATTT
A62	TCAGCTAAAAAAGGTAAAGTAATT
A63	ACGCTAACGAGCGTCTGGCGTTTTAGCGAACCCAACATGT
A64	ACGACAATAAATCCCGACTTGCGGGAGATCCTGAATCTTACCA
A65	TGCTATTTTGCACCCAGCTACAATTTTGTTTTGAAGCCTTAAA
B01	TCATATGTGTAATCGTAAAACTAGTCATTTTC
B02	GTGAGAAAATGTGTAGGTAAAGATACAACTTT
B03	GGCATCAAATTTGGGGCGCGAGCTAGTTAAAG
B04	TTCGAGCTAAGACTTCAAATATCGGGAACGAG
B05	ACAGTCAAAGAGAATCGATGAACGACCCCGGTTGATAATC
B06	ATAGTAGTATGCAATGCCTGAGTAGGCCGGAG
B07	AACCAGACGTTTAGCTATATTTTCTTCTACTA
B08	GAATACCACATTCAACTTAAGAGGAAGCCCGATCAAAGCG
B09	AGAAAAGCCCCAAAAAGAGTCTGGAGCAAACAATCACCAT
B10	CAATATGACCCTCATATATTTTAAAGCATTAA
B11	CATCCAATAAATGGTCAATAACCTCGGAAGCA
B12	AACTCCAAGATTGCATCAAAAAGATAATGCAGATACATAA
B13	CGTTCTAGTCAGGTCATTGCCTGACAGGAAGATTGTATAA
B14	CAGGCAAGATAAAAATTTTTAGAATATTCAAC
B15	GATTAGAGATTAGATACATTTCGCAAATCATA
B16	CGCCAAAAGGAATTACAGTCAGAAGCAAAGCGCAGGTCAG
B17	GCAAATATTTAAATTGAGATCTACAAAGGCTACTGATAAA
B18	TTAATGCCTTATTTCAACGCAAGGGCAAAGAA
B19	TTAGCAAATAGATTTAGTTTGACCAGTACCTT
B20	TAATTGCTTTACCCTGACTATTATGAGGCATAGTAAGAGC
B21	ATAAAGCCTTTGCGGGAGAAGCCTGGAGAGGGTAG
B22	TAAGAGGTCAATTCTGCGAACGAGATTAAGCA
B23	AACACTATCATAACCCATCAAAAATCAGGTCTCCTTTTGA
B24	ATGACCCTGTAATACTTCAGAGCA
B25	TAAAGCTATATAACAGTTGATTCCCATTTTTG
B26	CGGATGGCACGAGAATGACCATAATCGTTTACCAGACGAC
B27	TAATTGCTTGGAAGTTTCATTCCAAATCGGTTGTA

B28	GATAAAAACCAAAATATTAAACAGTTCAGAAATTAGAGCT
B29	ACTAAAGTACGGTGTCGAATATAA
B30	TGCTGTAGATCCCCCTCAAATGCTGCGAGAGGCTTTTGCA
B31	AAAGAAGTTTTGCCAGCATAAATATTCATTGACTCAACATGTT
B32	AATACTGCGGAATCGTAGGGGGTAATAGTAAAATGTTTAGACT
B33	AGGGATAGCTCAGAGCCACCACCCCATGTCAA
B34	CAACAGTTTATGGGATTTTGCTAATCAAAAGG
B35	GCCGCTTTGCTGAGGCTTGCAGGGGAAAAGGT
B36	GCGCAGACTCCATGTTACTTAGCCCGTTTTAA
B37	ACAGGTAGAAAGATTCATCAGTTGAGATTTAG
B38	CCTCAGAACCGCCACCCAAGCCCAATAGGAACGTAAATGA
B39	ATTTTCTGTCAGCGGAGTGAGAATACCGATAT
B40	ATTCGGTCTGCGGGATCGTCACCCGAAATCCG
B41	CGACCTGCGGTCAATCATAAGGGAACGGAACAACATTATT
B42	AGACGTTACCATGTACCGTAACACCCCTCAGAACCGCCAC
B43	CACGCATAAGAAAGGAACAACTAAGTCTTTCC
B44	ATTGTGTCTCAGCAGCGAAAGACACCATCGCC
B45	TTAATAAAACGAACTAACCGAACTGACCAACTCCTGATAA
B46	AGGTTTAGTACCGCCATGAGTTTCGTCACCAGGATCTAAA
B47	GTTTTGTCAGGAATTGCGAATAATCCGACAAT
B48	GACAACAAGCATCGGAACGAGGGTGAGATTTG
B49	TATCATCGTTGAAAGAGGACAGATGGAAGAAAAATCTACG
B50	AGCGTAACTACAAACTACAACGCCTATCACCGTACTCAGG
B51	TAGTTGCGAATTTTTTCACGTTGATCATAGTT
B52	GTACAACGAGCAACGGCTACAGAGGATACCGA
B53	ACCAGTCAGGACGTTGGAACGGTGTACAGACCGAAACAAA
B54	ACAGACAGCCCAAATCTCCAAAAAAAAATTTCTTA
B55	AACAGCTTGCTTTGAGGACTAAAGCGATTATA
B56	CCAAGCGCAGGCGCATAGGCTGGCAGAACTGGCTCATTAT
B57	CGAGGTGAGGCTCCAAAAGGAGCC
B58	ACCCCCAGACTTTTTCATGAGGAACTTGCTTT
B59	ACCTTATGCGATTTTATGACCTTCATCAAGAGCATCTTTG
B60	CGGTTTATCAGGTTTCCATTAAACGGGAATACACT
B61	AAAACACTTAATCTTGACAAGAACTTAATCATTGTGAATT
B62	GGCAAAAGTAAAATACGTAATGCC
B63	TGGTTTAATTTCAACTCGGATATTCATTACCCACGAAAGA
B64	ACCAACCTAAAAAATCAACGTAACAAATAAATTGGGCTTGAGA
B65	CCTGACGAGAAACACCAGAACGAGTAGGCTGCTCATTCAGTGA
C01	TCGGGAGATATACAGTAACAGTACAAATAATT
C02	CCTGATTAAAGGAGCGGAATTATCTCGGCCTC
C03	GCAAATCACCTCAATCAATATCTGCAGGTCGA
C04	CGACCAGTACATTGGCAGATTCACCTGATTGC
C05	TGGCAATTTTTAACGTCAGATGAAAACAATAACGGATTCG
C06	AAGGAATTACAAAGAAACCACCAGTCAGATGA
C07	GGACATTCACCTCAAATATCAAACACAGTTGA
C08	TTGACGAGCACGTATACTGAAATGGATTATTTAATAAAAG
C09	CCTGATTGCTTTGAATTGCGTAGATTTTCAGGCATCAATA
C10	TAATCCTGATTATCATTTTGCGGAGAGGAAGG

C11	TTATCTAAAGCATCACCTTGCTGATGGCCAAC
C12	AGAGATAGTTTGACGCTCAATCGTACGTGCTTTCCTCGTT
C13	GATTATACACAGAAATAAAGAAATACCAAGTTACAAAATC
C14	TAGGAGCATAAAAGTTTGAGTAACATTGTTTG
C15	TGACCTGACAAATGAAAAATCTAAAATATCTT
C16	AGAATCAGAGCGGGAGATGGAAATACCTACATAACCCTTC
C17	GCGCAGAGGCGAATTAATTATTTGCACGTAAATTCTGAAT
C18	AATGGAAGCGAACGTTATTAATTTCTAACAAC
C19	TAATAGATCGCTGAGAGCCAGCAGAAGCGTAA
C20	GAATACGTAACAGGAAAAACGCTCCTAAACAGGAGGCCGA
C21	TCAATAGATATTAAATCCTTTGCCGGTTAGAACCT
C22	CAATATTTGCCTGCAACAGTGCCATAGAGCCG
C23	TTAAAGGGATTTAGATACCGCCAGCCATTGCGGCACAGA
C24	ACAATTCGACAACTCGTAATACAT
C25	TTGAGGATGGTCAGTATTAACACCTTGAATGG
C26	CTATTAGTATATCCAGAACAATATCAGGAACGGTACGCCA
C27	CGCGAACTAAAACAGAGGTGAGGCTTAGAAGTATT
C28	GAATCCTGAGAAGTGTATCGGCCTTGCTGGTACTTTAATG
C29	ACCACCAGCAGAAGATGATAGCCC
C30	TAAAACATTAGAAGAACTCAAACTTTTTATAATCAGTGAG
C31	GCCACCGAGTAAAAGAACATCACTTGCCTGAGCGCCATTAAAA
C32	TCTTTGATTAGTAATAGTCTGTCCATCACGCAAATTAACCGTT
C33	CGCGTCTGATAGGAACGCCATCAACTTTTACA
C34	AGGAAGATGGGGACGACGACAGTAATCATATT
C35	CTCTAGAGCAAGCTTGCATGCCTGGTCAGTTG
C36	CCTTCACCGTGAGACGGGCAACAGCAGTCACA
C37	CGAGAAAGGAAGGGAAGCGTACTATGGTTGCT
C38	GCTCATTTTTTAACCAGCCTTCCTGTAGCCAGGCATCTGC
C39	CAGTTTGACGCACTCCAGCCAGCTAAACGACG
C40	GCCAGTGCGATCCCCGGGTACCGAGTTTTTCT
C41	TTTCACCAGCCTGGCCCTGAGAGAAAGCCGGCGAACGTGG
C42	GTAACCGTCTTTCATCAACATTAAAATTTTTGTTAAATCA
C43	ACGTTGTATTCCGGCACCGCTTCTGGCGCATC
C44	CCAGGGTGGCTCGAATTCGTAATCCAGTCACG
C45	TAGAGCTTGACGGGGAGTTGCAGCAAGCGGTCATTGGGCG
C46	GTTAAAATTCGCATTAATGTGAGCGAGTAACACACGTTGG
C47	TGTAGATGGGTGCCGGAAACCAGGAACGCCAG
C48	GGTTTTCCATGGTCATAGCTGTTTGAGAGGCG
C49	GTTTGCGTCACGCTGGTTTGCCCCAAGGGAGCCCCCGATT
C50	GGATAGGTACCCGTCGGATTCTCCTAAACGTTAATATTTT
C51	AGTTGGGTCAAAGCGCCATTCGCCCCGTAATG
C52	CGCGCGGGCCTGTGTGAAATTGTTGGCGATTA
C53	CTAAATCGGAACCCTAAGCAGGCGAAAATCCTTCGGCCAA
C54	CGGCGGATTGAATTCAGGCTGCGCAACGGGGGATG
C55	TGCTGCAAATCCGCTCACAATTCCCAGCTGCA
C56	TTAATGAAGTTTGATGGTGGTTCCGAGGTGCCGTAAAGCA
C57	TGGCGAAATGTTGGGAAGGGCGAT
C58	TGTCGTGCACACAACATACGAGCCACGCCAGC

C59	CAAGTTTTTGGGGTCGAAATCGGCAAAATCCGGGAAACC
C60	TCTTCGCTATTGGAAGCATAAAGTGTATGCCCGCT
C61	TTCCAGTCCTTATAAATCAAAAGAGAACCATCACCCAAAT
C62	GCGCTCACAAGCCTGGGGTGCCTA
C63	CGATGGCCCACTACGTATAGCCCGAGATAGGGATTGCGTT
C64	AACTCACATTATTGAGTGTTGTTCCAGAAACCGTCTATCAGGG
C65	ACGTGGACTCCAACGTCAAAGGGCGAATTTGGAACAAGAGTCC
Link-A1C	TTAATTAATTTTTACCATATCAAA
Link-A2C	TTAATTTCATCTTAGACTTTACAA
Link-A3C	CTGTCCAGACGTATACCGAACGA
Link-A4C	TCAAGATTAGTGTAGCAATACT
Link-B1A	TGTAGCATTCCTTTTATAAACAGTT
Link-B2A	TTTAATTGTATTTCCACCAGAGCC
Link-B3A	ACTACGAAGGCTTAGCACCATTA
Link-B4A	ATAAGGCTTGCAACAAAGTTAC
Link-C1B	GTGGGAACAAATTTCTATTTTTGAG
Link-C2B	CGGTGCGGGCCTTCCAAAAACATT
Link-C3B	ATGAGTGAGCTTTTAAATATGCA
Link-C4B	ACTATTAAAGAGGATAGCGTCC
Loop	GCGCTTAATGCGCCGCTACAGGGC

2. Sequences for AuNPs complementary

A48-sticky end	AAAAAAAAAAAAAAAAAAAAAAAATTTTCACCGGAATCGCCAT A TTTAACAAAATTTACG
A49-sticky end	AAAAAAAAAAAAAAAAAAAAAAAATTTTAGCATGTATTTCATC GT AGGAATCAAACGATTTTTTGTTT
A56-sticky end	AAAAAAAAAAAAAAAAAAAAAAAATTTTACAAGAAAGCAAGC A AATCAGATAACAGCCATATTATTTA
Thiol ssDNA	TTTTTTTTTTTTTTTTTTTTTTTTTTTT-SH

3. Sequences for QDs streptavidin conjunction

B32-sticky end	AATACTGCGGAATCGTAGGGGGTAATAGTAAAATGTTTAGACTA GTTGTGGATCCTACT
Biotin ssDNA	Biotin-AGTAGGATCCACAACT

4. Sequences for amino anchors

A5-Amino	TTTGATGATTAAGAGGCTGAGACTTGCTCAGTACCAGGCGCCGA TTCAG-NH_2
B5-Amino	ACAGTCAAAGAGAATCGATGAACGACCCCGGTTGATAATCCCG ATTCAG-NH_2
C5-Amino	TGGCAATTTTTAACGTCAGATGAAAACAATAACGGATTCGCCGA TTCAG-NH_2

A13-Amino	TTTAACGGTTCGGAACCTATTATTAGGGTTGATATAAGTACCGATTCAG-NH_2
B13-Amino	CGTTCTAGTCAGGTCATTGCCTGACAGGAAGATTGTATAACCGATTCAG-NH_2
C13-Amino	GATTATACACAGAAATAAAGAAATACCAAGTTACAAAATCCCGATTCAG-NH_2
A33-Amino	CGCGTCTGATAGGAACGCCATCAACTTTTACACCGATTCAG-NH_2
B33-Amino	AGGGATAGCTCAGAGCCACCACCCCATGTCAACCGATTCAG-NH_2
C33-Amino	GATTATACACAGAAATAAAGAAATACCAAGTTCCGATTCAG-NH_2
A42-Amino	AGAGTCAAAAATCAATATATGTGATGAAACAAACATCAAGCCGATTCAG-NH_2
B42-Amino	AGACGTTACCATGTACCGTAACACCCCTCAGAACCGCCACCCGATTCAG-NH_2
C42-Amino	GTAACCGTCTTTCATCAACATTAAAATTTTTGTTAAATCACCGATTCAG-NH_2
A50-Amino	ACATAGCGCTGTAAATCGTCGCTATTCATTTCAATTACCTCCGATTCAG-NH_2
B50-Amino	AGCGTAACTACAAACTACAACGCCTATCACCGTACTCAGGCCGATTCAG-NH_2
C50-Amino	GGATAGGTACCCGTCGGATTCTCCTAAACGTTAATATTTTCCGATTCAG-NH_2

Supplementary figures

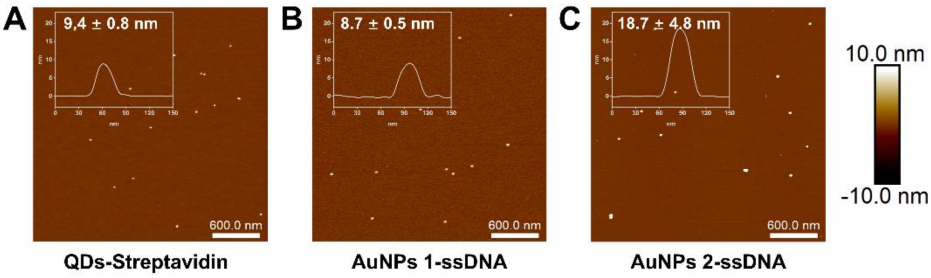

Figure S1. AFM images of different functionsed nanoparticles. A) The size of streptavidin modified QDs are around 9.4 ± 0.8 nm; B) the size of ssDNA conjugated AuNPs 1 are around 8.7 ± 0.5 nm; C) the size of ssDNA conjugated AuNPs 2 are around 18.7 ± 4.8 nm.

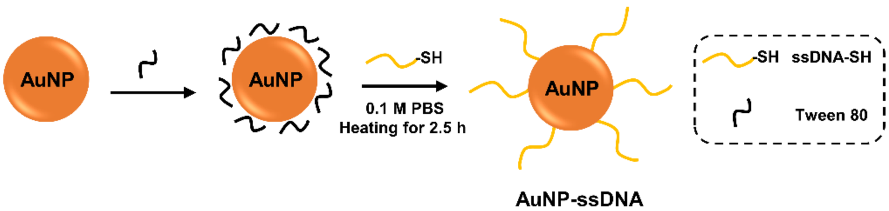

Figure S2. Schematic of Tween 80 mediated thoil-ssDNA (ssDNA-SH) and AuNPs conjugation.

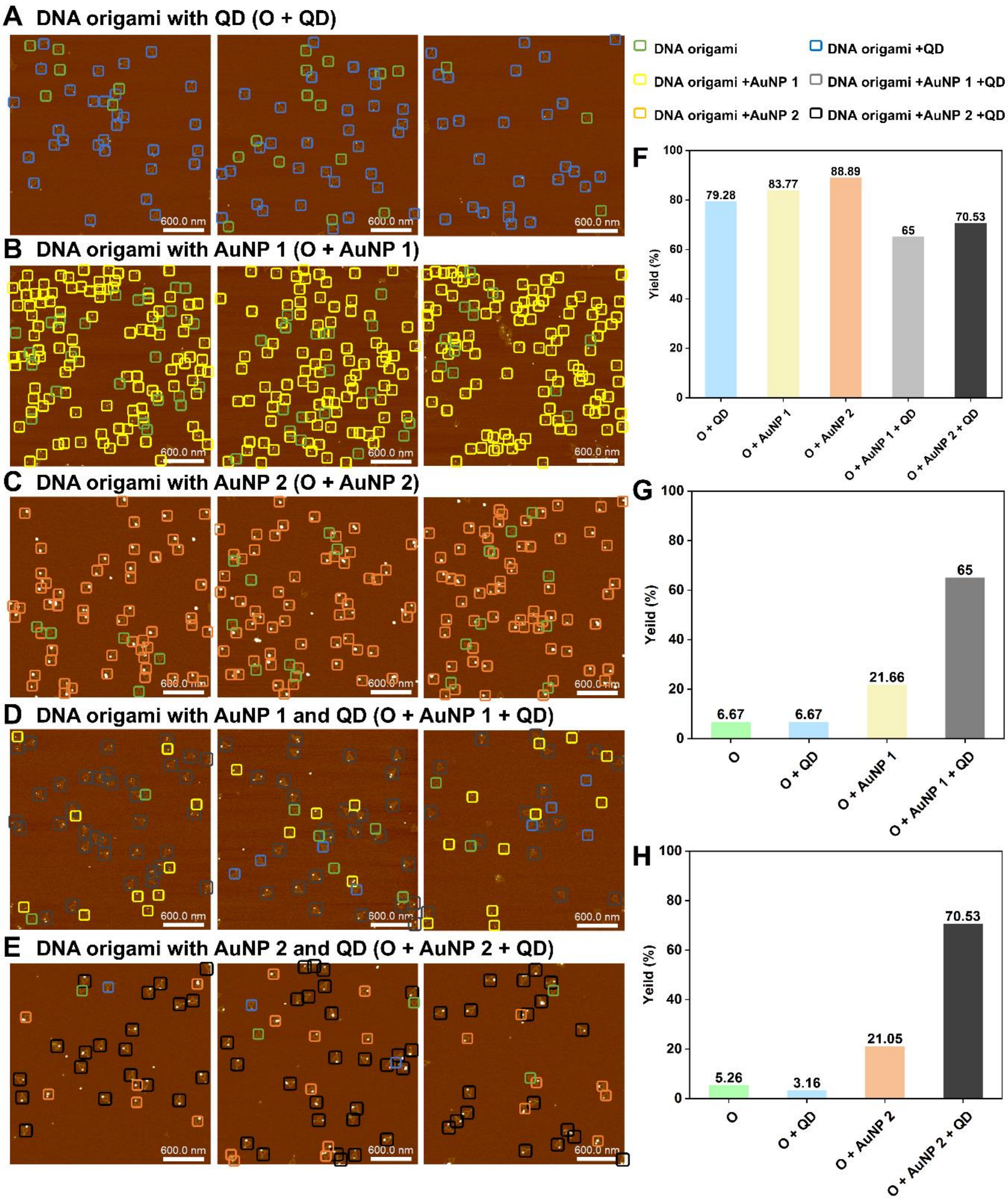

Figure S3. Analysis of the yields of DNA origami functionalization with multiple nanoparticles. A)-E) Select three different area via AFM and counting numbers of each functionalisation DNA origami; F) analysis of the yields of each functionalisation DNA origami; G) analysis of the yield of DNA orgami with AuNP 1 and QD; H) analysis of the yield of DNA orgami with AuNP 2 and QD.

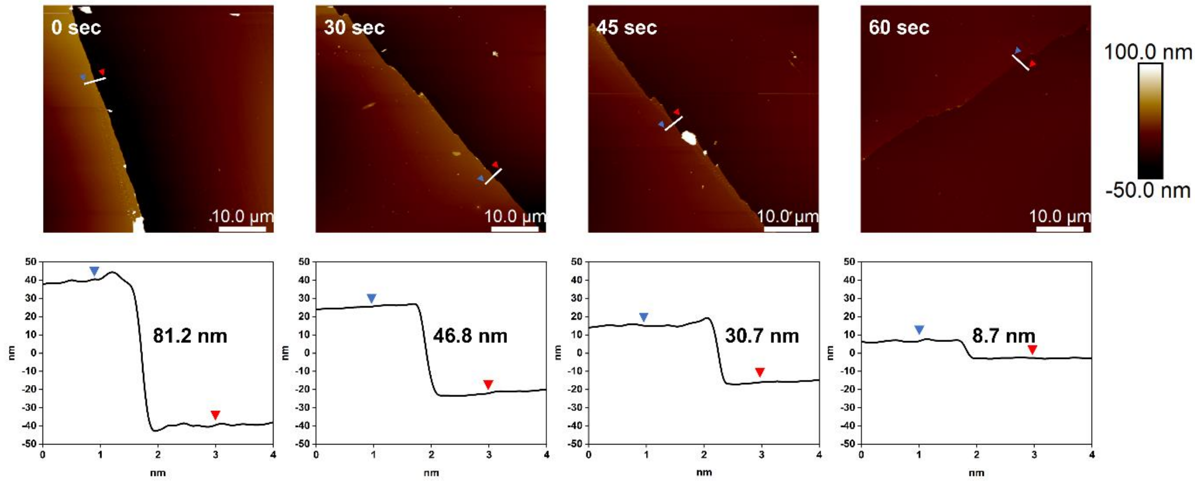

Figure S4. AFM images of the correlation between PPA thickness and the increase of oxygen plasma time. The thickness of PPA decreases by about 1.2 nm per seconds.

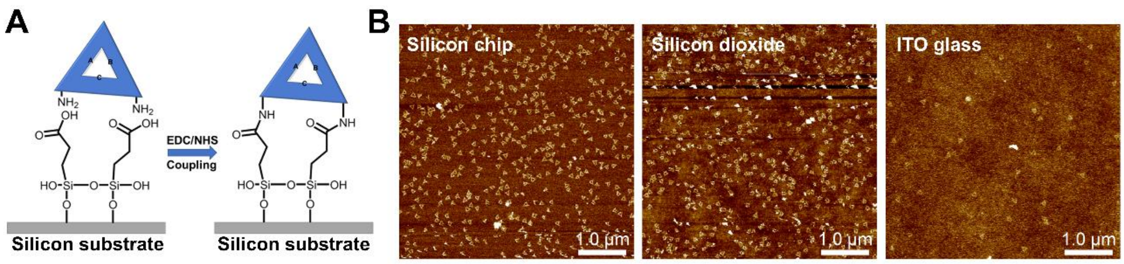

Figure S5. Schematic and validation of amino-functionalised DNA origami. A) Reactions of carboxylic silanisation on the substrate for coupling with amino-anchors of DNA origami; B) the DNA origami placement process can work in different substrates (silicon, silicon dioxide and ITO glass).

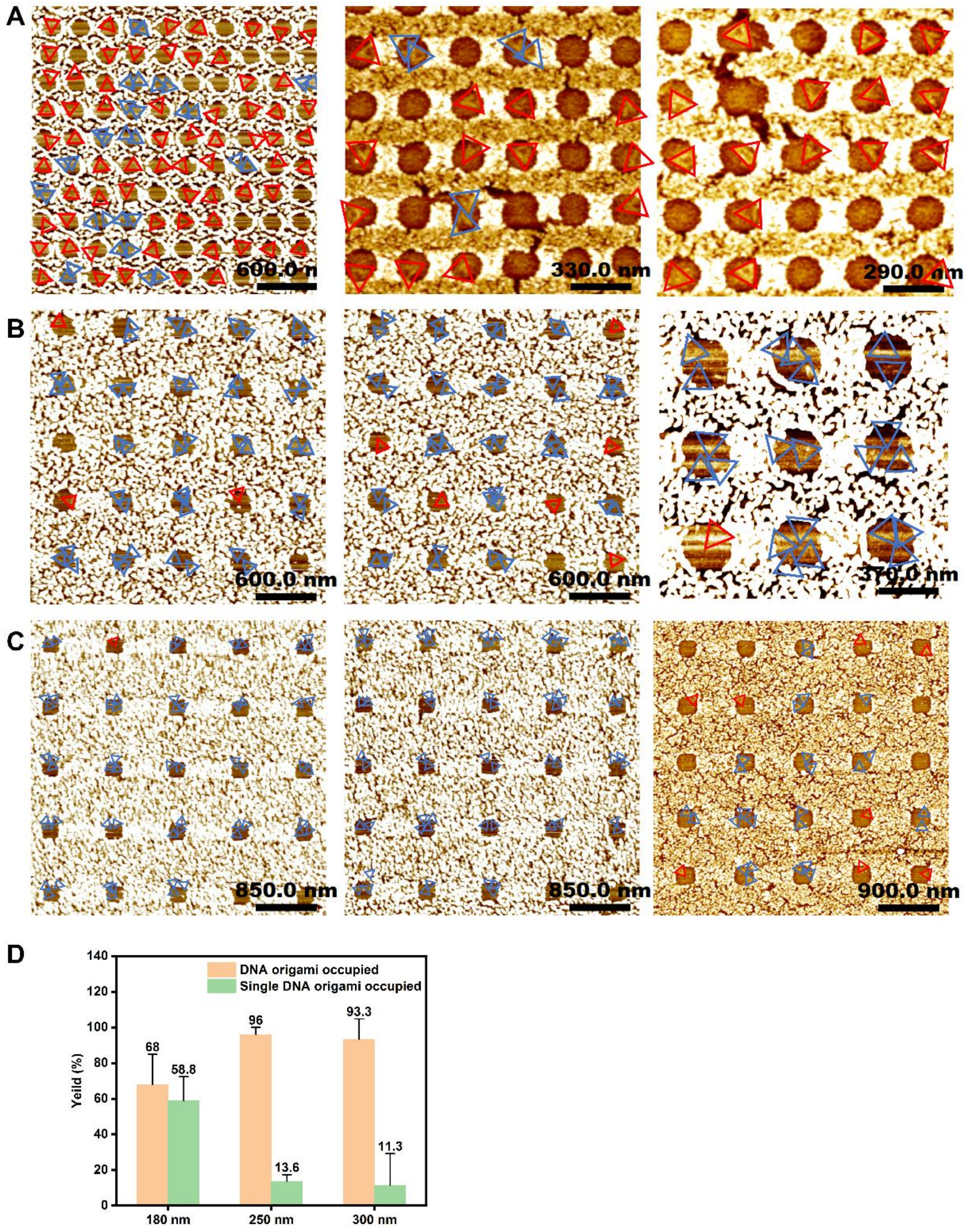

Figure S6. Topographical AFM image of DNA origami immobilised in A) 180 nm nanoapertures, B) 250 nm nanoapertures, and C) 300 nm nanoapertures. Red triangle frames show single DNA origami occupied; blue triangle frames indicate multiple DNA origami occupied. D) analysis of DNA origami immobilization yields shows 68% ± 17% (58.8% ± 13.6% single occupancy), 96% ± 4% (13.6% ± 3.8% single occupancy), and 93.3% ± 11.5% (11.3% ± 17.9% single occupancy) for the 180 nm, 250 nm and 300 nm nanoapertures, respectively.

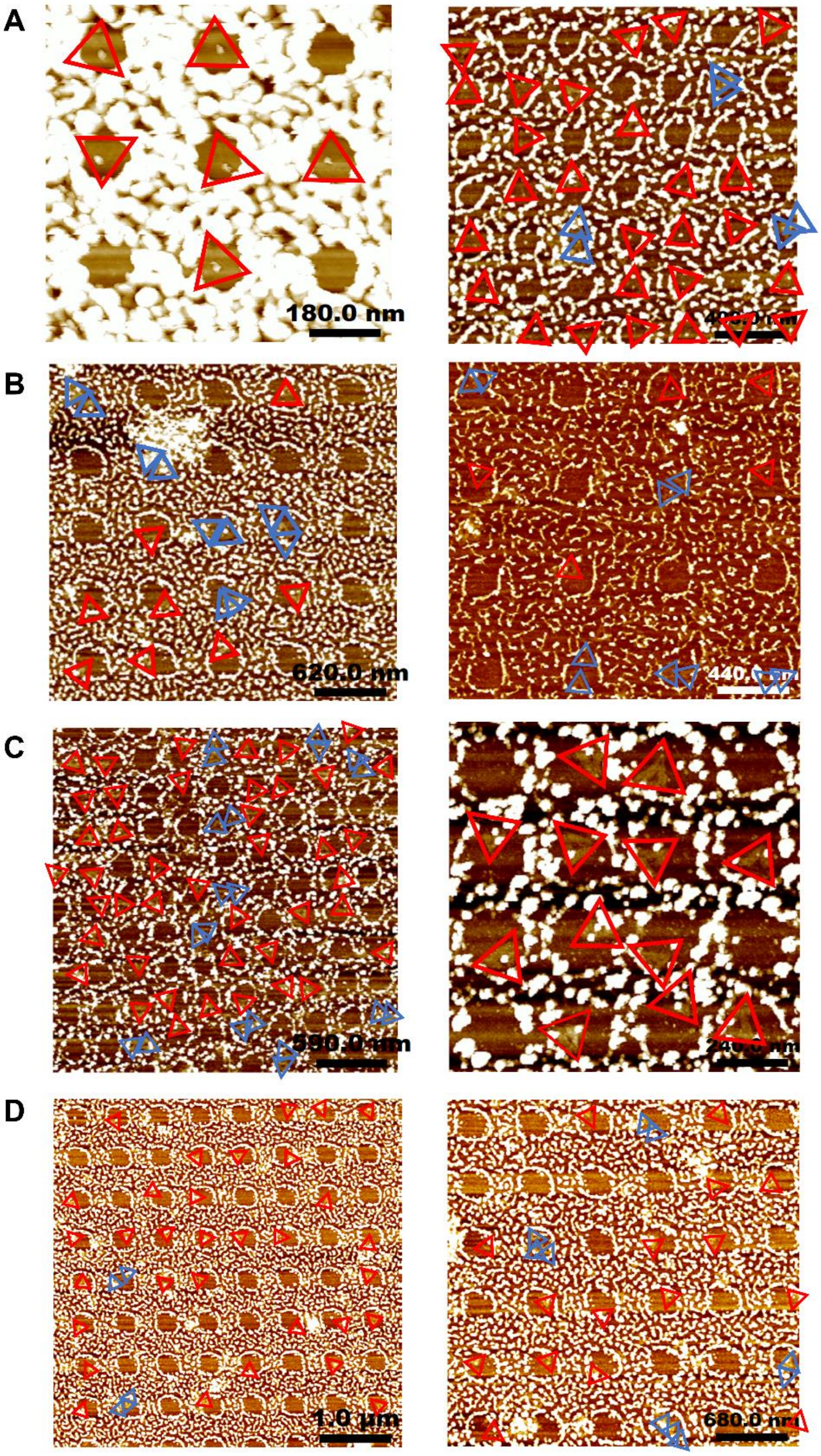

Figure S7. AFM topographical images of multiple nanoparticle-functionalised DNA origami. DNA origami with AuNP 1 in A) 180 nm nanoapertures and B) 250 nm nanoapertures; DNA origami with AuNP 1 and QD immobilised in C) 180 nm nanoapertures and D) 250 nm nanoapertures. Red triangle frames show single DNA origami occupied; blue triangle frames indicate multiple DNA origami occupied.