Supplementary Information for Molecular mechanics of DNA bricks: *In situ* structure, mechanical properties and ionic conductivity.

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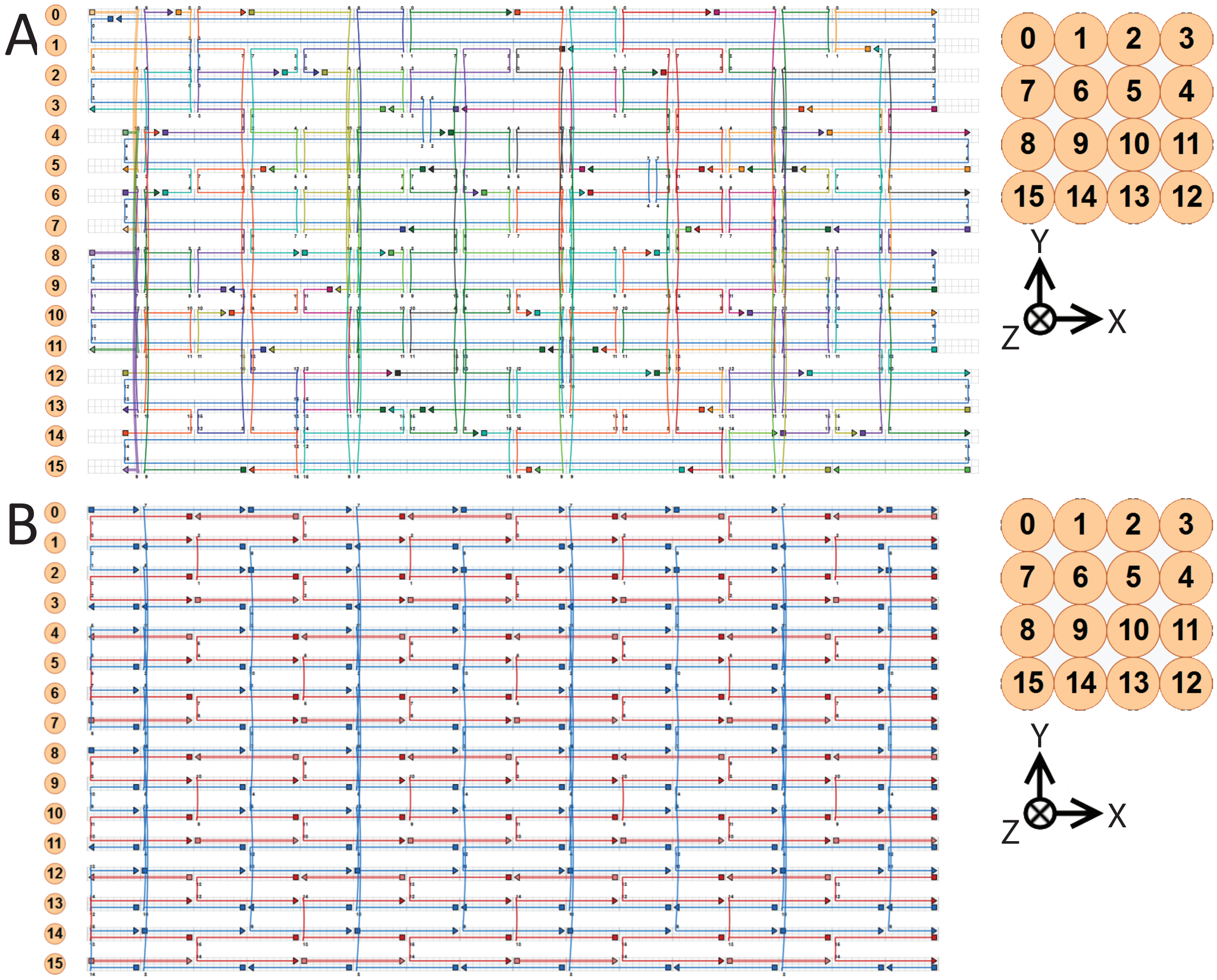

Figure S1. caDNAno schematics of the 4×4 DNA rod structures. (A) The 4×4 DNA rod structure realized via the DNA origami method. (Left) caDNAno schematic of the design. The blue line indicates the scaffold strand; all other colors indicate the staple strands. (Right) Physical location of the helices numbered in the schematics. (B) The 4×4 DNA rod structure realized via the DNA brick method. (Left) caDNAno schematic of the design. Colors correspond to alternating strands. Same color strands are not covalently connected to each other. (Right) Physical location of the helices numbered in the schematics.

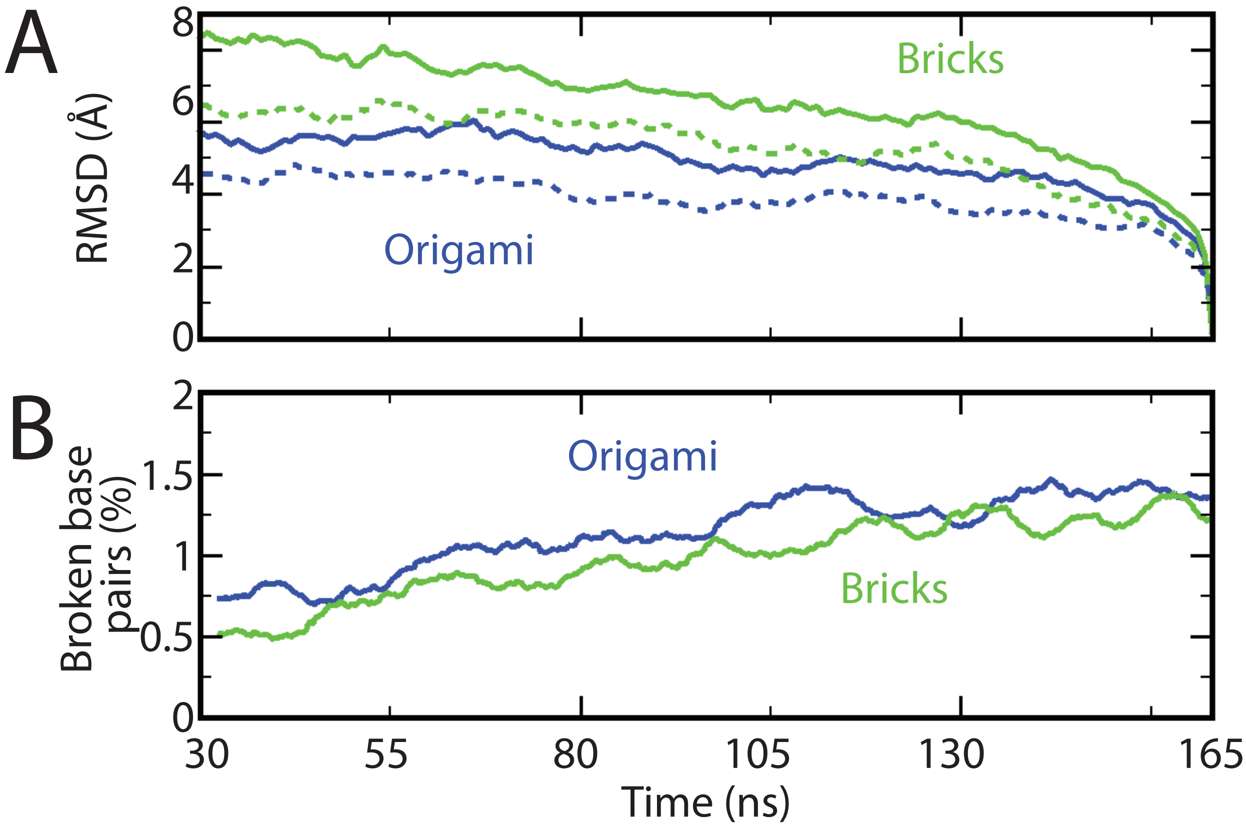

Figure S2. Structural analysis of MD simulations of DNA origami and DNA brick rod objects. (A) RMSD of the DNA origami (blue) and DNA brick (green) objects during the respective MD simulations with respect to the conformation they attain at the end of the simulations. Solid lines indicate data computed for the entire DNA structures, dashed lines indicate the calculations done having the terminal array cells (1, 2, 15, and 16) excluded. (B) The number of broken base pairs versus simulation time for DNA origami (blue) and DNA brick (green) objects. Array cells 1 and 16 were not included in the calculations.

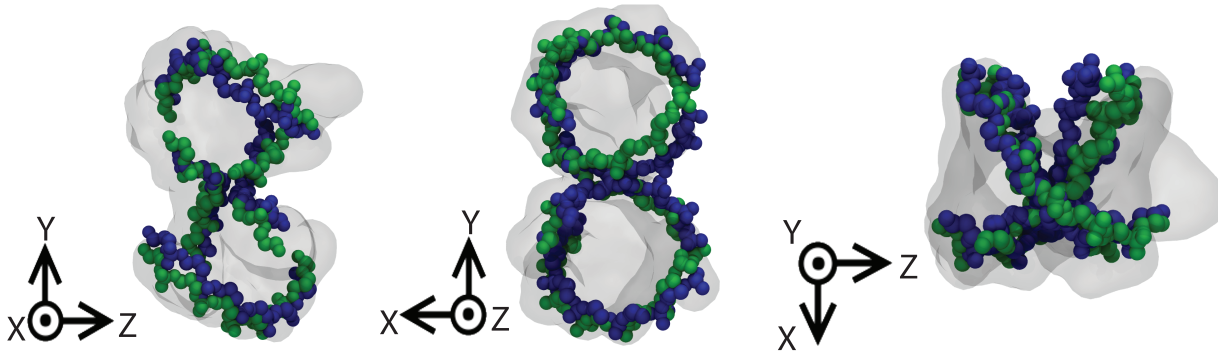

Figure S3. Superposition of the DNA origami and DNA brick junctions. The backbone of DNA origami and DNA brick junctions are shown as blue and green spheres, respectively; the semitransparent surface indicates the overall shape of DNA brick junction. The structures of the junctions were obtained by averaging over the respective unrestrained equilibration trajectories (sampled at 1 ns) and over 30 representative junctions within each structure. The backbone RMSD between the average structures is 4.4 Å.

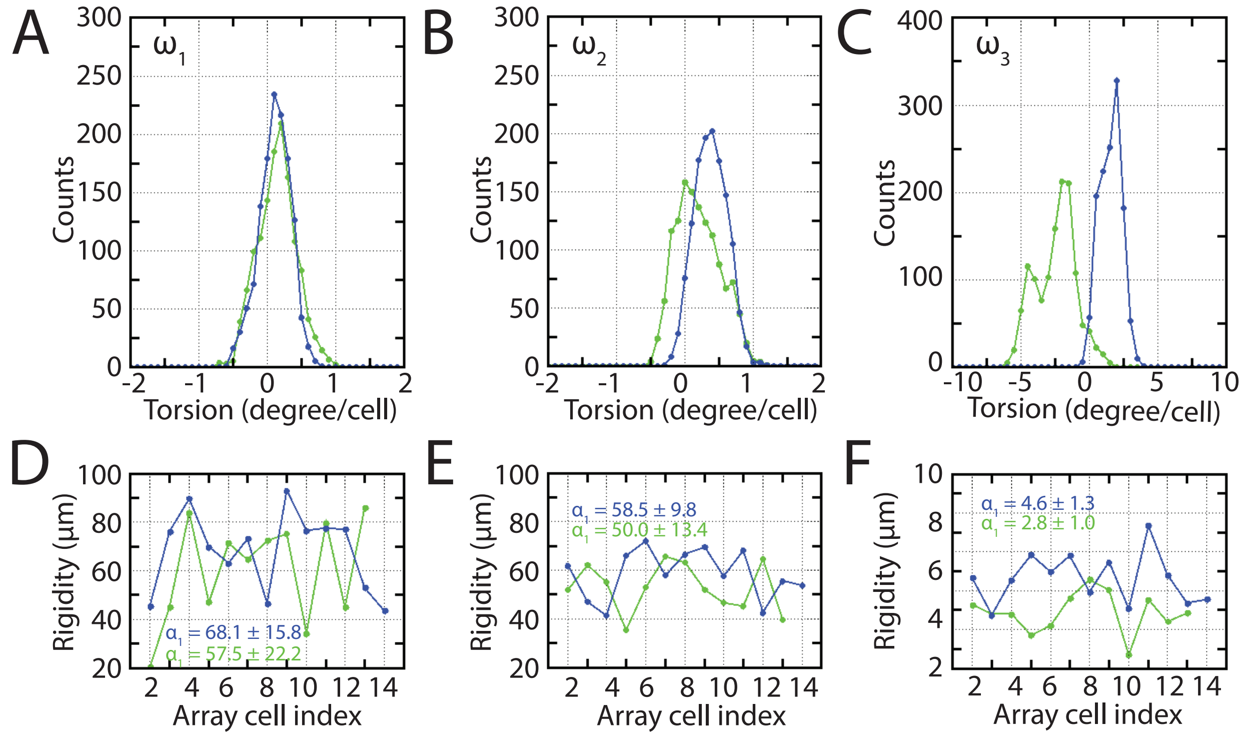

Figure S4. (A-C) Representative distributions of torsions ω_1, ω_2, and ω_3. Histograms of torsions ω_1 (A), ω_2 (B), and ω_3 (C) between array cell 5 and 6 for origami (blue) and brick (green). (D-F) Bending ($\alpha_{1,2}$) and twist (α_3) moduli for the DNA origami (blue) and DNA brick (green) realizations of the 4×4 DNA rod structure.

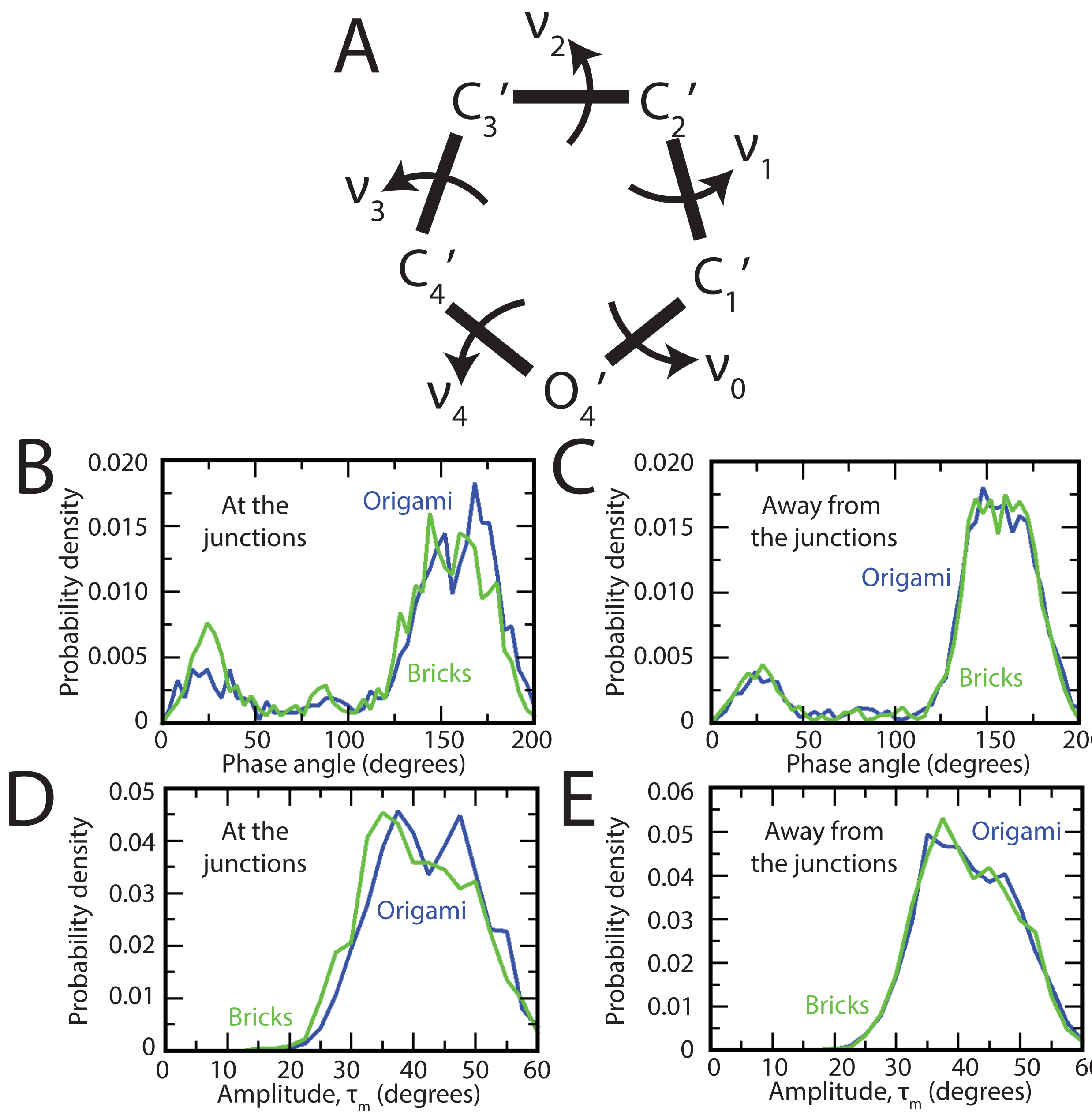

Figure S5. Distributions of the rotation pseudoparameters defining the amount of sugar group puckering in the DNA backbones of DNA origami (blue) and DNA brick (green) rod objects. (A) Schematic representation of a DNA sugar group. The five dihedral angles, v_{1-5}, can be represented using the phase angle, P, and amplitude, τ_m, parameters defined as $\tan(P) = ((v_4 + v_1) - (v_3 + v_0))/(2v_2(P_c))$, where $P_c = \sin(\pi/5) + \sin(\pi/2.5)$, and $\tau_m = (v_2)/\cos(P)$ [1]. (B–E) The distribution of the phase angle, P (middle row), and the amplitude, τ_m (bottom row), in the simulations of the DNA origami (blue) and DNA brick (green) objects. The left column (panels B and D) show the distributions for the sugar groups located within 1 base pair of the junctions. The right columns (panels C and E) show the distributions for the sugar groups located more than 1 base pair away from the junctions.

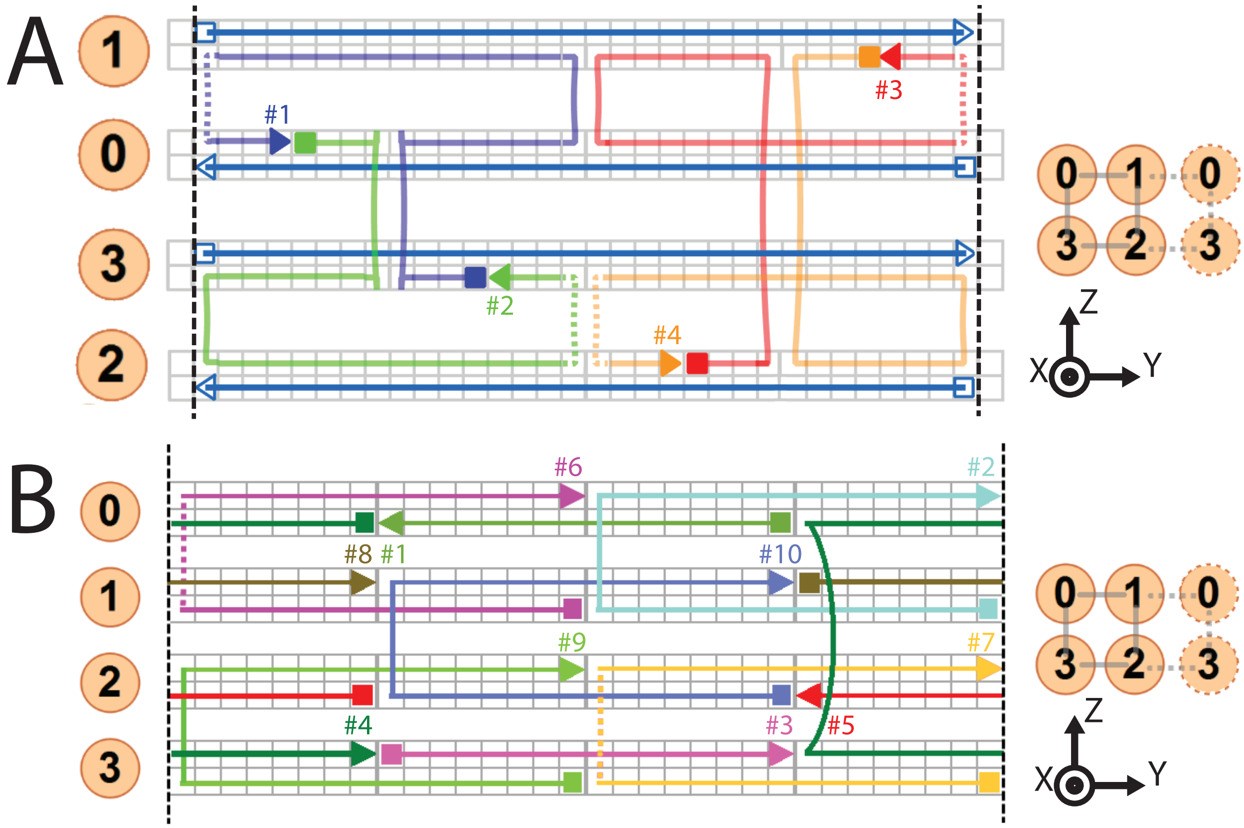

Figure S6. caDNAno schematics of DNA origami and DNA brick plates. (A) Unit cell of the DNA origami plate [3]. Parts of the scaffold strand are shown in blue, all other colors represent staples. Due to the periodic nature of the unit cell, some crossovers occur over the unit cell boundaries. Under the periodic boundary conditions, the boundary at the left hand side of the structure is equivalent to the boundary at the right hand side of the structure so that the dashed black lines at both sides of the structure correspond to the same region of the design. Dotted crossovers, such as in strand #1, occur over the system boundary along the Y-axis. The schematic representation of the structure (right column) illustrates those connections using dotted lines (helix 1 connects to the periodic image of helix 0 whereas helix 2 connects to the periodic image of helix 3). The structure does not repeat itself in the Z direction, giving the appearance of an infinite two-duplex-thick membrane. (B) Unit cell of the DNA brick plate. All DNA strands are shown in different colors. Strands that cross over the dashed boundaries connect across the unit cell boundary along the Y-axis.

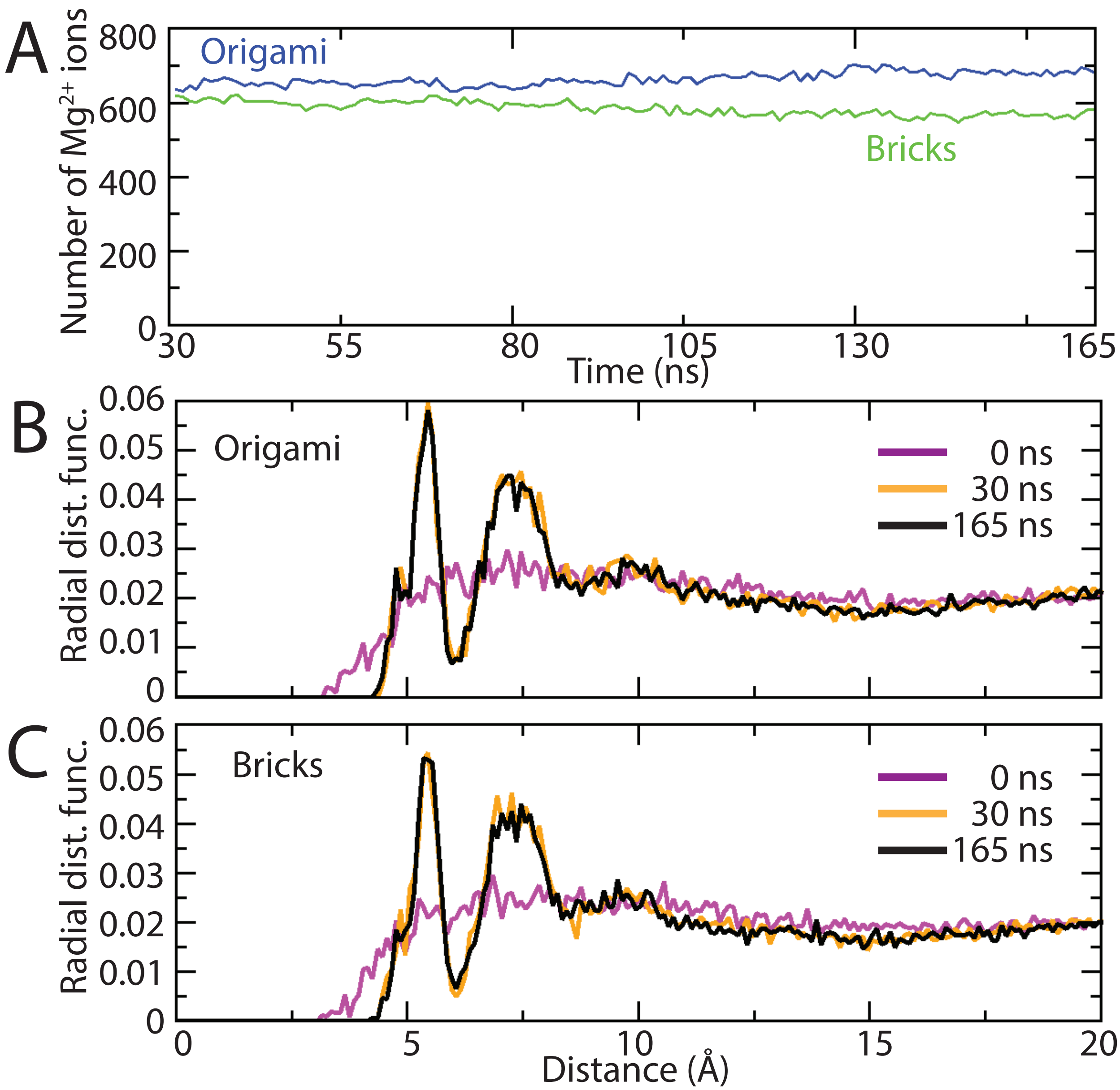

Figure S7. The distributions of Mg^{2+} ions during MD simulations of DNA origami and DNA brick objects. (A) The number of Mg^{2+} ions within a $4 \times 4 \times 35$ nm^3 internal volume of the DNA rod objects. (B) The radial distribution function of Mg^{2+} ions with respect to the phosphorous atoms of the DNA backbone of the DNA origami structure at 0, 30, and 165 ns of the respective MD trajectory. (B) The radial distribution function of Mg^{2+} ions with respect to the phosphorous atoms of the DNA backbone of the DNA brick structure at 0, 30, and 165 ns of the respective MD trajectory.

Table S1: Summary of production simulations

System type	Dimensions (length × # helices)	# nucleotides	# atoms	Simulation time (ns)				
				Equilibration	Applied bias simulation			
					0.1	0.25	0.5(V)	Cycle
origami rod	128 bp×(4h×4h)	4096	1,285,191	∼165	—	—	—	—
brick rod	128 bp×(4h×4h)	4096	1,286,032	∼165	—	—	—	—
brick unit cell	32 bp×(2h×2h)	256	50,066	∼410	48	48	48	∼288[a]
origami unit cell	32 bp×(2h×2h)	256	50,050	∼490	48	48	48	∼230[b]

[a] The electric field was applied for 48 ns and then removed for 96 ns and then repeated one more time.

[b] The electric field was applied for 57.6 ns and then removed for 57.6 ns and then repeated one more time. [3]

Table S2: The nucleotide sequence of strands used to build the 4×4 DNA origami rod

Number	Sequence
Scaffold	M13mp18 sequence provided by CaDNAno program [2].
1	ATCAATAGGCG
2	AATGGGCGAAAAACCGTCTGGACTCCAGTT
3	ATCGTCATAAATATTCCGTGCCAGCAGGGTGGAGG
4	AACAGTTCCTCACTGCGCAACAGCAACGACGGCGC
5	TCAAAAATCATGAGTGAGATTAGCAAACGCCAGGGCT
6	TATCGCGTTTTTGCGGAGAGAGTTACATTTCGACG
7	GCGGATCCCTGACTATTATAGT
8	AGTAAGCAAACTTGGGCGCCTGCATTACTGTGTGA
9	TTTAAGCCCCAGCCAAAAGAACAGCCAGCTTTCC
10	AATTGCTGAAAGAGGAAGGGCAAAGACTAAC
11	GAGTGTTGTTCCAGTTTGGAATTATAAATAGGCG
12	TTAGTAGGTTTGATAAGAGGTCATTTTAATTCTGA
13	GGAAGTTTCATTCCATATAATGTTTAGCAAT
14	GCAACTAATGAAAAGGTGGCA
15	CCGGAAACCAG
16	TCGGCAAAATCCCCAAGAGTCGTCAGGATTAGAG
17	ACCATTAGATGCAGCAAGGGCCTCTTCCAGTGCCATC
18	GGCGCGAGCAGTACGGTGTCT
19	AAACTGCGCAATCTAGAGGGGATTCTCTAGCCAGC
20	CACTTCACCAGGAGCTTCACCCCTCAAATGCTTTA
21	CTGATGGCTTATCCCAATTCTGCGAACGAGTAGAT
22	CATACGGGGATGTGTTTTCCCCCCCAAAATAAATC
23	ATATATATTTTCAATGCCTGAGTAATGCGGAGACAGAG
24	AACTGTAGCTCAACATGTTTTAAATATCAGAAGCACTCAGAGCATAAA
25	TCAATTCTACTAAAAAATTTT
26	GGGCTCGAATTGCAAAGCGCGT
27	GGCGGTGATGGTGACGTCAAACACTATTAAAGAACGT
28	GACGGCCGCTTTCCACAACATACGAGCCGTAGGA
29	TTGCCCTTCACAACCCGAAAGAGAATGACCATAAA
30	AAACCTGTATTGAATCAAGCGAACCAGACCGGACC

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Table S2: The nucleotide sequence of strands used to build the 4×4 DNA origami rod

Number	Sequence
31	TCAAGCCTGGGGTGCCTAAATTTTTGTACAGGAAG
32	ACATTATGGGTCTTTATGCATCAAAAAGATTATATAATGCATCCA
33	GCTAAATCGGTTGGCGGGAGA
34	CGTAATCATGT
35	AATTGTTACCTTCCTGGGTAATCGTAAAACTAAAG
36	TCAATGCCTGCTTTTTCTTGCTGGTTTTTGCTCCTGTT
37	TAACATTAATTGCGTTGCGAGAAAACGACTTCAAA
38	CCCGGGCTGTTTCATGAATCGGCCAACGCCGTCC
39	TCGACCTGTTGGGCGATGAACGGGCGCATCGTAAC
40	AGCCTTTATTTAAATTGTAAA
41	AGGATCCTGTTTTTGCGTATCCAACAGACTGCGGA
42	TATTAATTGCCTGAGGGGACGAAAGGGTGAGAAA
43	GCAAGATTTTTGAGTCAAATCACCA
44	GGCGTGTAGATCGTGGGAACAAACGGCAACCCGTCATC
45	ACAGTATCGGCCTCCAGTTTGAGAGTCTGGAGCA
46	ATTCACAAATGGTCAATAACCCAGTTGATGAGCTT
47	TAGAACCCTCAACCGTTCTAG
48	AATGGGATAGGTCACGTTGACCGCTTCCATTC
49	CGTGCATCTGCAGGAAGATCGGTGCGCGGTC
50	GGCTGTAGGTAGGCGAAAGAGGCCGCCTGGCC
51	TCAATATGATATTCATATATCAAGGATATAGTAGTAAATAAAGCAA
52	AGAATAAGGGCGATCGCACTCTAGCCCGAGATTTG
53	AACGCATGTCAAAGCTTGCCAATTCCACAGTCGGG
54	ATCTACAAAGGCTCAGAAAAGAGTCACGAAAGTG
55	CTGATAAATTAATGCCGGAGAAGCA
56	GAGCGAGTAACGGATTGACCCATTCGCTGGTGCCGAAA
57	ATATGCCATCAAAAATAATTCGCGTCTGGTCCGC
58	ATTGTATAGGGTAGCTGCGATTAATTTAAATGCATTTGG
59	AATATTTCAACGATACTTTTTACCAAAA
60	TTTCATCAACATTAAATGGTCATAGTACCGAGAGA
61	ACGTACCCCGGTTGATAATATCAGGTCCGCCAGCTAAG

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Table S2: The nucleotide sequence of strands used to build the 4×4 DNA origami rod

Number	Sequence
62	AGCTCATTTTTTAACCAAGAAGCATACGTTGTAATGA
63	CGCATTAAACCCTGTAGTTGGGTAAATTAAGCGCATT
64	CGTTAATATTTTGTTAAAATT

Table S3: The nucleotide sequence of strands used to build the 4×4 DNA brick rod

Number	Sequence
1	ATCAATAG
2	ATCGTCATAAATATTC
3	ATGCTTTAAACAGTTC
4	CATAAATCAAAAATCA
5	ATTATAGT
6	AACCGTCTGGACTCCA
7	TCCAACAGGGGCGAAA
8	CAGACCGGAAGCAAACTTGGGCGCCAGGGTGG
9	GAGCTTCAAAGCGAAC
10	TATCGCGTTTTAATTCTGAGACGGGCAACAGC
11	CCCGAAAGACTTCAAA
12	AAAGATTAAGAGGAAGGGCAAAGAATTAGCAA
13	AAGCGGATTGCATCAA
14	CAGAAGCACTCAGAGC
15	TAGAGAGTACCTTTAA
16	GAGGTCATTTTTGCGG
17	TTGCTGAATATAATGC
18	AAGAACGT
19	CAAGAGTGCACTATTA
20	TTGTTCCAGTTTGGAATTATAAATCAAAAGAA
21	TTAGTAGGGTTGAGTG
22	CTGCGAACGAGTAGATCATTAGATACATTTCG
23	CAGTTGATTCCCAATT
24	GTTTCATTCCATATAATGTTTAGCTATATTTT
25	AGTACGGTGTCTGGAA
26	GCAACTAATGAAAAGG
27	GTTTTTTTTTTTTCCG
28	ATCCTGTTTGATGGTGACGTCAAAGTCAGGAT
29	GCCCCAGGAGGCGAAACTGCGCAACTGTTGGG
30	CGGTCCACGCTGGTTTTTGCTCCTTTTGATAA
31	AGAGAGTTGCAGCAAGGGCCTCTTCGCTATTA

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Table S3: The nucleotide sequence of strands used to build the 4×4 DNA brick rod

Number	Sequence
32	CCGCCTGGTGGCCCTGATGGCTTAGAGCTTAA
33	ATCCAATAAATCATACGGGGATGTGCTGCAAG
34	TAGTAGTAGCATTAACTGTAGCTCAACATGTT
35	TCTACTAAAAAATTTT
36	GCGTTTTTTTTTGGG
37	ATGAATCGGCCAACGCCGTCCAATACTGCGGA
38	CGTGCCAGCTGCATTACTGTGTGAAATTGTTA
39	CAGTCGGGAAACCTGTATTGAATCGCCCTCAA
40	CTCACTGCCCGCTTTCCACAACATACGAGCCG
41	CATTAATTGCGTTGCGAGAAAACGAGAATGAC
42	TGAGTGAGCTAACTCAAGCCTGGGGTGCCTAA
43	TACCAAAAACATTATGCGTCTTTACCCTGACT
44	ATCGGTTGGCGGGAGA
45	CGTAATCA
46	CTCGAATTGCAAAGCG
47	ATCCCCGGGTACCGAGAGAGGCGGTTTGCGTA
48	AGGTCGACTCTAGAGGGGATTCTCCGTAATCG
49	AAGCTTGCATGCCTGCTTTTTCTTTTCACCAG
50	AACGACGGCCAGTGCCATCATATGTACCCCGG
51	AGTCACGACGTTGTAATGATTGCCCTTCACAA
52	ACGCCAGGGTTTTCCCGCCCAAAAACAGGAAG
53	TTTCAACGGTTGGGTAAATTAAGCAATAAAGC
54	AGCCTTTATTTAAATT
55	GAAACCAG
56	ACCGCTTCTGGTGCCGAAATCGGCAAAATCCC
57	CAGCCAGCTTTCCGGCGTGTAGATGGGCGCAT
58	CAGGAAGATCGCACTCTAGCCCGAGATTTGAC
59	ACGACAGTATCGGCCTCCAGTTTGAGGGGACG
60	TGTAGGTAAAGATTCACAAATGGTCAATAACC
61	CAATGCCTGAGTAATGCGGAGACAGTCAAATC
62	TCATATATTTTAAATGCATTTGGGGCGCGAGC

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Table S3: The nucleotide sequence of strands used to build the 4 × 4 DNA brick rod

Number	Sequence
63	TAGAACCCTCAACCGT
64	GGGATAGGTCACGTTG
65	CGTAACCGTGCATCTG
66	AAAGGGTGAGAAAGGC
67	ACCATCAATATGATAT
68	CGTTTTTTTTTTTAAT
69	CAAACGGCGGATTGACCCATTCGCGATTCAGG
70	CGATGAACCGTGGGAA
71	GAGCAAACAAGAGAATAAGGGCGATCGGTGCG
72	ATTGCCTGAGAGTCTG
73	CAAAGGCTATCAGGTCCGCCAGCTGGCGAAAG
74	ATTTTTGAGAGATCTA
75	GCCGGAGAGGGTAGCTGCGATTAACAAGGATA
76	AAATTAAT
77	CGAGTAACAACCCGTC
78	TAAAACTAGCATGTCA
79	TTGATAATCAGAAAAG
80	ATTGTATAAGCAAATA
81	TGTTTTTTTTTTTGAG
82	TTTCATCAACATTAAATGGTCATAGCTGTTTC
83	CCTTCCTGTAGCCAGC
84	AATAATTCGCGTCTGGTCCGCTCACAATTCCA
85	TAGGAACGCCATCAAA
86	CTCATTTTTTAACCAAGAAGCATAAAGTGTAA
87	ATTTTTGTTAAATCAG
88	TTAAAATTCGCATTAAACCCTGTAATACTTTT
89	ATATTTTG
90	ATTGGACGCTATTGATAGACGGTTTTTCGCCC
91	ATGACGATTCCGCAGT
92	GATTCAATGAATATTTCCGGTCTGGTTCGCTT
93	TAAAGCATTTGAGGGC

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Table S3: The nucleotide sequence of strands used to build the 4×4 DNA brick rod

Number	Sequence
94	CGTTTTCTGAACTGTTACGCGATATTTGAAGT
95	GATTTATGGTCATTCT
96	TAAAGACGTGATTTTTTAATCTTTTTGATGCA
97	ACTATAATAGTCAGGG
98	TTTGACGTTGGAGTCCACGTTCTTTAATAGTG
99	ACTCTCTAATCCTGACCTGTTGGAGTTTGCTT
100	AGGAGCAATTAAAGGTTGGAACAACACTCAAC
101	ATGACCTCTTATCAAATGAAGCTCGAATTAAA
102	TAAGCCATCCGCAAAAGTTCGCAGAATTGGGA
103	TTCAGCAATTAAGCTCCTTTCGGGCTTCCTCT
104	GAGCTACAGCATTATAAATGAAACTTCCAGAC
105	ATATTTAAAACATGTTATCCGCTTTGCTTCTG
106	CACTCTTGTTCCAAAC
107	CCTACTAAATCTACTC
108	ATCAACTGTTATATGG
109	ACCGTACTTTAGTTGC
110	GCCGATTTCGGAAAAA
111	ATTTATAAGGGATTTTAACAGGATTTTCGCCT
112	TCGGGCTATTCTTTTG
113	ATCTAATGGTCAAATCGTGGACCGCTTGCTGC
114	ACCATTTGCGAAATGT
115	GCTAAACAGGTTATTGCCAGGCGGGTATGATT
116	CCCAAATGAAAATATA
117	CCTTTTCAGCTCGCGCTACTACTATTAGTAGA
118	CCGCCTCTCCCAAAAAAAAAAAACCACCATCA
119	GCGCCCAATACGCAAACGATTCATTAATGCAG
120	AAGAAAAACCACCCTGCCTGGGGCAAACCAGC
121	CCGTCTCACTGGTGAACCCGACTGGAAAGCGG
122	GGCAATCAGCTGTTGCAACTCTCTCAGGGCCA
123	TCTTTGCCTTGTGAAGAATTAATGTGAGTTAG
124	GCTTAATTTTGCTAATTATTGGATGTTAATGC

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Table S3: The nucleotide sequence of strands used to build the 4×4 DNA brick rod

Number	Sequence
125	GCTCTGAGGCTTTATTTTTGGTACAACCGAT
126	AAAAACGCGCGTTGGC
127	CTGGCACGACAGGTTT
128	GCAGTGAGCGCAACGC
129	CTCACTCACATAATGT
130	TATGACCATGATTACGAATTCGAGCTCGGTAC
131	TCACACAGGAAACAGC
132	TGAGCGGATAACAATTGTCGACCTGCAGGCAT
133	ATGTTGTGTGGAATTG
134	TATGCTTCCGGCTCGTCCGTCGTTTTACAACG
135	CCCAGGCTTTACACTT
136	TACAGGGTTTAGGCACCCTGGCGTTACCCAAC
137	TCTCCCGCAAAAGTAT
138	GCGAATGGCGCTTTGCCTGGTTTCCGGCACCA
139	TTGCGCAGCCTGAATCCCGGGGATCCTCTAGA
140	TCGCCCTTCCCAACAGGCTGGCTGGAGTGCGA
141	AAGAGGCCCGCACCGAGCAAGCTTGGCACTGG
142	AGCTGGCGTAATAGCGACTGTCGTTGAATCTT
143	ACATCCCCCTTTCGCCTCGTGACTGGGAAAAC
144	TTAATCGCCTTGCAGCAGGCATTGCATTTAAA
145	AAAATTTTTATCCTTGCGTTGAAATAAAGGCT
146	GAAGCGGTGCCGGAAA
147	TCTTCCTGAGGCCGAT
148	TACCTACACATTACTC
149	ATATATGAGGGTTCTA
150	CCTATCCCATTAAAAA
151	ATCTACACCAACGTGAGCCGTTTGTTCCCACG
152	CGGTTACGATGCGCCC
153	CAAACTGGCAGATGCAGTTTGCTCCAGACTCT
154	CACCCTTTCGTCCCCT
155	TGTCTCCGGCCTTTCTAGCCTTTGTAGATCTC

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Table S3: The nucleotide sequence of strands used to build the 4×4 DNA brick rod

Number	Sequence
156	TTGATGGTGATTTGAC
157	ACGGTTGAATATCATATCTCCGGCATTAATTT
158	GTTACTCGCTCAAAAAAAAAAACGGTCAATCC
159	GAGAATCCGACGGGTTTGATGAAAGCTGGCTA
160	TAGTTTTACGATTACGGTTCATCGATTCTCTT
161	CATATGATTGACATGCGAATTATTTTTGATGG
162	ATTATCAACCGGGGTACAGGCAATGACCTGAT
163	TTTTGGGCCTTTTCTGAAAATGAGCTGATTTA
164	TATACAATCTTCCTGTTCAAAAATAGCTACCC
165	AATTTAAATATTTGCTAATTTTAACAAAATAT
166	AAAAAACATTTAATGT
167	CAGGAAGGCCAGACGC
168	CGTTCCTATTGGTTAA
169	ACAAAAATTTAATGCG
170	TTAAATAT

Table S4: The nucleotide sequence of strands used to build the DNA origami plate. This design is reproduced from our previous study [3]

Number[a]	Sequence
Scaffold 0	ATGTTGTGTGGAATTGTGAGCGGATAACAATT
Scaffold 1	CCCGACTGGAAAGCGGGCAGTGAGCGCAACGC
Scaffold 2	GCTGGGGCAAACCAGCGTGGACCGCTTGCTGC
Scaffold 3	AACACTCAACCCTATCTCGGGCTATTCTTTTG
Staple 1	GGGTTCCGCTCACCGCTTTCCAGTCGGGAATT
Staple 2	GTTATGAGTGTTGCAGCAAGCGGTCCACGATA
Staple 3	GTTTCTCACTGCCAATTCCACACAACATGCGT
Staple 4	TGCGGCCCCAGCCAAAAGAATAGCCCGAGCTG

[a] The strands are numbered as in Fig. S3A schematic.

Table S5: The nucleotide sequence of strands used to build the DNA brick plate.

Number[a]	Sequence
1	GCCAAGGGGCGGTGAG
2	GTTATGAGTGTTGCAGTTTCCAGTCGGGAATT
3	CGATAAGAAAACCGAC
4	CGGAACCCAATTCCCGGGGCCGCACAGCTCGG
5	TGAGAAACACGCATGT
6	CAAGCGGTCCACGATAGGGTTCCGCTCACCGC
7	TGCGGCCCCAGCCAAATCCACACAACATGCGT
8	CTCATAACTATCGTGG
9	AGAATAGCCCGAGCTGGTTTCTCACTGCCAAT
10	TGTGTGGAATTGGCAGACCGCTTGCTGCAACA

[a] The strands are numbered as in Fig. S3B schematic.

[1] C. Altona and M. Sundaralingam. Conformational analysis of the sugar ring in nucleosides and nucleotides. a new description using the concept of pseudorotation. *J. Am. Chem. Soc.*, 94:8205–8212, 1972.

[2] S. M. Douglas, A. H. Marblestone, S. Teerapittayanon, A. Vazquez, G. M. Church, and W. M. Shih. Rapid prototyping of 3D DNA-origami shapes with caDNAno. *Nucleic Acids Res.*, 37(15):5001–6, Aug. 2009.

[3] C.-Y. Li, E. A. Hemmig, J. Kong, J. Yoo, S. Hernández-Ainsa, U. F. Keyser, and A. Aksimentiev. Ionic conductivity, structural deformation and programmable anistropy of DNA origami in electric field. *ACS Nano*, 9(2):1420–1433, 2015.