

HELIX: a new modular nucleic acid crystallization screen.**Supporting information**Julia Viladoms^a and Gary N. Parkinson^{a*}^a Department of Pharmaceutical and Biological Chemistry, UCL School of Pharmacy, 29-39 Brunswick Square, London, WC1N 1AX, United Kingdom

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Table S1. Formulation of the HELIX nucleic acid crystallization screen.

Tube	Buffer system	Precipitants	Salts	Additives	Subset
1-1	0.05 M MES pH 6.5	25% v/v PEG 400	0.05 M KCl, 0.10 M LiCl	0.012 M spermine	variX
1-2	0.05 M MES pH 6.5	25% v/v PEG 400		0.001 M spermine	variX
1-3	0.05 M MES pH 6.5	25% v/v PEG 400	0.10 M NaCl, 0.05 M LiCl	0.010 M MgCl ₂	variX
1-4	0.05 M MES pH 6.5	25% v/v PEG 400	0.2 M KCl	0.005 M [Co(NH ₃) ₆]Cl ₃	variX
1-5	0.05 M MES pH 6.5	25% v/v PEG 400	0.1 M LiCl	0.010 M MnCl ₂	variX
1-6	0.05 M MES pH 6.5	25% v/v PEG 400	0.1 M NaCl	0.005 M SrCl ₂	variX
1-7	0.05 M MES pH 6.5	35% v/v MPD	0.05 M KCl, 0.10 M LiCl	0.012 M spermine	variX
1-8	0.05 M MES pH 6.5	35% v/v MPD		0.001 M spermine	variX
1-9	0.05 M MES pH 6.5	35% v/v MPD	0.10 M NaCl, 0.05 M LiCl	0.010 M MgCl ₂	variX
1-10	0.05 M MES pH 6.5	35% v/v MPD	0.2 M KCl	0.005 M [Co(NH ₃) ₆]Cl ₃	variX
1-11	0.05 M MES pH 6.5	35% v/v MPD	0.1 M LiCl	0.010 M MnCl ₂	variX
1-12	0.05 M MES pH 6.5	35% v/v MPD	0.1 M NaCl	0.005 M SrCl ₂	variX
1-13	0.05 M MES pH 6.5	17% w/v PEG 4000	0.05 M KCl, 0.10 M LiCl	0.012 M spermine	variX
1-14	0.05 M MES pH 6.5	17% w/v PEG 4000		0.001 M spermine	variX
1-15	0.05 M MES pH 6.5	17% w/v PEG 4000	0.10 M NaCl, 0.05 M LiCl	0.010 M MgCl ₂	variX
1-16	0.05 M MES pH 6.5	17% w/v PEG 4000	0.2 M KCl	0.005 M [Co(NH ₃) ₆]Cl ₃	variX
1-17	0.05 M MES pH 6.5	17% w/v PEG 4000	0.1 M LiCl	0.010 M MnCl ₂	variX
1-18	0.05 M MES pH 6.5	17% w/v PEG 4000	0.1 M NaCl	0.005 M SrCl ₂	variX
1-19	0.05 M MES pH 6.5	2.6 M Na ₂ malonate	0.05 M KCl, 0.10 M LiCl	0.012 M spermine	variX

1-20	0.05 M MES pH 6.5	2.6 M Na ₂ malonate		0.001 M spermine	variX
1-21	0.05 M MES pH 6.5	2.6 M Na ₂ malonate	0.10 M NaCl, 0.05 M LiCl	0.010 M MgCl ₂	variX
1-22	0.05 M MES pH 6.5	2.6 M Na ₂ malonate	0.2 M KCl	0.005 M [Co(NH ₃) ₆]Cl ₃	variX
1-23	0.05 M MES pH 6.5	2.6 M Na ₂ malonate	0.1 M LiCl	0.010 M MnCl ₂	variX
1-24	0.05 M MES pH 6.5	2.6 M Na ₂ malonate	0.1 M NaCl	0.005 M SrCl ₂	variX
1-25	0.05 M BIS-TRIS pH 7.0	44% v/v PEG 200	0.1 M (NH ₄) ₂ SO ₄		Cryo
1-26	0.05 M BIS-TRIS pH 7.0	40% v/v PEG 400	0.2 M KCl	0.010 M CaCl ₂	Cryo
1-27	0.05 M BIS-TRIS pH 7.0	37% w/v PEG 1000	0.1 M NaCl	0.002 M spermine	Cryo
1-28	0.05 M BIS-TRIS pH 7.0	35% w/v PEG 2000	0.1 M NaCl, 0.2 M KCl	0.020 M MgCl ₂	Cryo
1-29	0.05 M BIS-TRIS pH 7.0	33% w/v PEG 3350	0.05 KCl	0.005 M spermine	Cryo
1-30	0.05 M BIS-TRIS pH 7.0	20% w/v PEG 8000, 1 M Naformate	0.1 M (NH ₄) ₂ SO ₄ , 0.1 M NaCl		Cryo
1-31	0.05 M BIS-TRIS pH 7.0	20% w/v PEG 20000, 1 M Naformate		0.010 M CaCl ₂	Cryo
1-32	0.05 M BIS-TRIS pH 7.0	40% v/v PEG-MME 550	0.05 M LiCl		Cryo
1-33	0.05 M BIS-TRIS pH 7.0	40% v/v PEG-MME 550	0.1 M Na citrate		Cryo
1-34	0.05 M BIS-TRIS pH 7.0	40% v/v MPD	0.1 M (NH ₄) ₂ SO ₄	0.002 M spermine	Cryo
1-35	0.05 M BIS-TRIS pH 7.0	35% v/v MPD			Cryo
1-36	0.05 M BIS-TRIS pH 7.0	30% v/v MPD	0.10 M NaCl	0.020 M MgCl ₂	Cryo
1-37	0.05 M BIS-TRIS pH 7.0	27% v/v MPD	0.10 M KCl		Cryo
1-38	0.05 M BIS-TRIS pH 7.0	24% v/v MPD		5% v/v PEG 400	Cryo
1-39	0.05 M BIS-TRIS pH 7.0	20% v/v MPD	0.20 M LiCl		Cryo
1-40	0.05 M BIS-TRIS pH 7.0	1.0 M (NH ₄) ₂ SO ₄	0.15 M KCl	20% v/v glycerol, 5% w/v PEG 3350	Cryo
1-41	0.05 M BIS-TRIS pH 7.0	1.2 M (NH ₄) ₂ SO ₄	0.1 M NaCl	10% v/v glycerol, 5% v/v PEG mme-550	Cryo
1-42	0.05 M BIS-TRIS pH 7.0	1.4 M (NH ₄) ₂ SO ₄		25% v/v glycerol	Cryo
1-43	0.05 M BIS-TRIS pH 7.0	1.6 M (NH ₄) ₂ SO ₄		25% v/v glycerol	Cryo
1-44	0.05 M BIS-TRIS pH 7.0	1.8 M (NH ₄) ₂ SO ₄		25% v/v glycerol	Cryo
1-45	0.05 M BIS-TRIS pH 7.0	2.0 M (NH ₄) ₂ SO ₄		5% v/v PEG 400	Cryo
1-46	0.05 M BIS-TRIS pH 7.0	22% w/v PEG-MME 2000	0.1 M KCl		Cryo
1-47	0.05 M BIS-TRIS pH 7.0	22% w/v PEG-MME 2000	0.2 M LiCl		Cryo
1-48	0.05 M BIS-TRIS pH 7.0	22% w/v PEG-MME 2000	0.1 M NaCl		Cryo
2-1	0.05 M HEPES pH 6.5	20% v/v MPD	0.1 M KCl		Quad
2-2	0.05 M HEPES pH 6.5	10% v/v MPD		0.005 M spermine	Quad
2-3	0.05 M HEPES pH 6.5	10% v/v MPD	0.1 M NaCl, 0.2 M KCl		Quad
2-4	0.05 M HEPES pH 6.5	25% v/v MPD	0.2 M NaCl		Quad

2-5	0.05 M HEPES pH 6.5	15% v/v MPD	0.05 M LiCl		Quad
2-6	0.05 M HEPES pH 6.5	30% v/v MPD	0.3 M NaCl		Quad
2-7	0.05 M HEPES pH 6.5	25% v/v PEG 400	0.05 M NaCl	0.01 M MgCl ₂	Quad
2-8	0.05 M BIS-TRIS pH 7.0	10% v/v PEG 400	0.2 M KCl		Quad
2-9	0.05 M HEPES pH 6.5	18% v/v PEG 1000	0.05 M LiCl		Quad
2-10	0.05 M BIS-TRIS pH 7.0	1.5 M Li ₂ SO ₄			Quad
2-11	0.05 M HEPES pH 6.5	1.9 M Li ₂ SO ₄	0.1 M NaCl, 0.1 M KCl		Quad
2-12	0.05 M BIS-TRIS pH 7.0	15% w/v PEG-MME 2000		0.02 M MgCl ₂	Quad
2-13	0.05 M HEPES pH 6.5	10% w/v PEG-MME 2000	0.05 M LiCl		Quad
2-14	0.05 M BIS-TRIS pH 7.0	14% w/v PEG-MME 2000			Quad
2-15	0.05 M BIS-TRIS pH 7.0	10% v/v PEG-MME 550	0.1 M LiCl		Quad
2-16	0.05 M BIS-TRIS pH 7.0	1.1 M (NH ₄) ₂ SO ₄	0.05 M KCl		Quad
2-17	0.05 M HEPES pH 6.5	1.4 M (NH ₄) ₂ SO ₄	0.05 M LiCl		Quad
2-18	0.05 M HEPES pH 6.5	1.8 M (NH ₄) ₂ SO ₄	0.10 M NaCl	0.01 M LiCl	Quad
2-19	0.05 M BIS-TRIS pH 7.0	18% w/v PEG 1000	0.30 M LiCl		Quad
2-20	0.05 M HEPES pH 6.5	15% w/v PEG 1000	0.3 M NaCl		Quad
2-21	0.05 M HEPES pH 6.5	12% w/v PEG 1000	0.25 M KCl		Quad
2-22	0.05 M BIS-TRIS pH 7.0	25% v/v PEG-MME 350	0.10 M LiCl		Quad
2-23	0.05 M HEPES pH 6.5	20% v/v PEG-MME 350			Quad
2-24	0.05 M HEPES pH 6.5	15% v/v PEG-MME 350	0.05 M LiCl		Quad
2-25	0.05 M Acetate pH 4.5	18% v/v MPD		0.005 M spermine	C-prot
2-26	0.05 M Acetate pH 4.5	24% v/v MPD	0.1 M NaCl		C-prot
2-27	0.05 M Acetate pH 4.5	32% v/v MPD	0.15 M KCl		C-prot
2-28	0.05 M Acetate pH 5.0	21% v/v MPD		0.01 M MgCl ₂	C-prot
2-29	0.05 M Acetate pH 5.0	14% v/v MPD	0.1 M NaCl, 0.1 M KCl	0.005 M spermine	C-prot
2-30	0.05 M Acetate pH 5.0	15% v/v PEG 400	0.05 M LiCl		C-prot
2-31	0.05 M MES pH 5.5	21% v/v MPD			C-prot
2-32	0.05 M MES pH 5.5	21% v/v MPD		0.02 M MgCl ₂	C-prot
2-33	0.05 M MES pH 5.5	12% v/v PEG 2000			C-prot
2-34	0.05 M MES pH 5.5	21% v/v MPD		0.020 M MgSO ₄	C-prot
2-35	0.05 M MES pH 5.5	15% v/v PEG 400		0.005 M spermine	C-prot
2-36	0.05 M MES pH 5.5	1.5 M (NH ₄) ₂ SO ₄		10% v/v glycerol	C-prot
2-37	0.05 M BIS-TRIS pH 7.0	25% w/v PEG 2000	0.05 M NaCl, 0.1 M KCl	0.01 M CaCl ₂	Macro
2-38	0.05 M BIS-TRIS pH 7.0	20% w/v PEG 3350	0.25 M NaCl	0.005 M spermine	Macro
2-39	0.05 M BIS-TRIS pH 7.0	12% w/v PEG 8000	0.1 M KCl	0.005 M BaCl ₂	Macro
2-40	0.05 M BIS-TRIS pH 7.0	8% w/v PEG 20000	0.30 M NaCl		Macro
2-41	0.05 M BIS-TRIS pH 7.0	20% w/v PEG 2000	0.05 M NaCl, 0.1 M LiCl	0.01 M CaCl ₂	Macro
2-42	0.05 M BIS-TRIS pH 7.0	15% w/v PEG MME- 2000	0.1 M KCl	0.002 M spermine	Macro
2-43	0.05 M BIS-TRIS pH 7.0	15% w/v PEG MME- 2000	0.05 M KCl, 0.1 M LiCl	0.020 M MgSO ₄	Macro

2-44	0.05 M BIS-TRIS pH 7.5	6% w/v PEG 20000	0.10 M NaCl	0.002 M BaCl ₂	Macro
2-45	0.05 M BIS-TRIS pH 7.5	7% w/v PEG 8000	0.1 M KCl	0.002 M spermine	Macro
2-46	0.05 M BIS-TRIS pH 8.0	15% w/v PEG 3350	0.05 M LiCl	0.030 M MgSO ₄	Macro
2-47	0.05 M BIS-TRIS pH 8.5	9% w/v PEG 3350	0.20 M NaCl		Macro
2-48	0.05 M BIS-TRIS pH 8.5	18% w/v PEG 3350		0.052 M MgCl ₂	Macro

BIS-TRIS: Bis(2-hydroxyethyl)amino-tris(hydroxymethyl)methane; HEPES: 2-[4-(2-hydroxyethyl)piperazin-1-yl]ethanesulfonic acid; MES: 2-(N-morpholino)ethanesulfonic acid; MPD: 2-Methyl-2,4-pentanediol; PEG: Polyethylene glycol; PEG-MME: polyethylene glycol monomethyl ether.

Table S2. Summary crystallographic data derived from the crystals obtained using the HELIX screen.

Name	Sequence	Laue group	Unit Cell a, b, c (Å)	Angles α, β, γ (°)	Resolution Max (Å)	Data Collected	Sequence Published PDBID
auto complementary duplex	d(CGAATTAATTCG)	ND	ND		-	-	3UXW
hairpin	d(TATAAGAAAATCTTATA)	-3m	50.97(1), 50.97(1), 97.68(5)	90,90, 120	3.4	Yes	-
complex	d(ACCCTAACCTA) + d(AGGGTTAGGGTT)	ND	ND		-	-	-
tetramolecular quadruplex	d(TAGGGT)	4/m	40.9(2), 40.85(7), 26.95(3)	90, 90, 90	-	-	-
bimolecular quadruplex	d(GGGGTTTTGGGG)	-3m	25.1(2), 25.8(2), 145(1)	90, 90, 120	5	ND	1JPQ
unimolecular quadruplex	d(AGGGAGGGCGCTGGGAGGAGGG)	ND	ND		ND	ND	3QXR
unimolecular i-motif	d(TAACCTAA)	mmm	31.09(1), 31.09(1), 81.88(2)	90, 90, 120	2.90	Yes	-
bimolecular i-motif	d(ACCCTAACCT)	mmm	27.24(1), 27.24(1), 152.80(6)	90, 90, 120	2.71	Yes	-
tetramolecular i-motif	d(CCCTAACCTAACCTAACCT)	ND	ND		-	-	-

Crystals were flash-cooled at 100 K in supercooled N₂ gas (Oxford Cryosystems) and maintained at 100 K during the data collections. Cell dimensions were determined using iMosflm (Battye et al., 2011) for the hairpin, unimolecular and bimolecular i-motifs, and Crysalis^{Pro} software for the tetramolecular quadruplex and bimolecular quadruplex.