

Molecular
Dimensions



HELIX™ HT-96 MD1-69

Crystallize a diverse range of nucleic acid topologies and molecular weights.

This screen targets all types of DNA/RNA, triplexes, quadruplexes, pseudoknots, i-motifs, and large molecular weight nucleic acids.

Developed by Dr. Gary Parkinson from School of Pharmacy, University College London, UK.

MD1- 69 is presented as a 96 x 1 mL conditions in a deep-well block.

Features of HELIX™:

- Crystallize nucleic acid fragments of all molecular weights.
- Ideal for double stranded DNA and RNA, pseudo knots, G-quadruplexes, i-motifs, triplex, ribozymes.
- Optimized for MAD, SAD, SIRAS data collections- no cacodylate.
- Cryo-cooling optimization.
- Screening of additives.
- Adaptable for HT Screening.

MPD, NaCl, and $(\text{NH}_4)_2\text{SO}_4$ have been used as precipitating agents in HELIX and can be used for any molecular weight target while the size of the PEGs used as crystallization agents tend to follow the molecular weights of the target molecules. Components that have significant impact on crystallization for DNA and RNA are quite restricted and well defined. However, it is still important to explore alternative conditions. HELIX explores the use of PEGs as a precipitating agent.

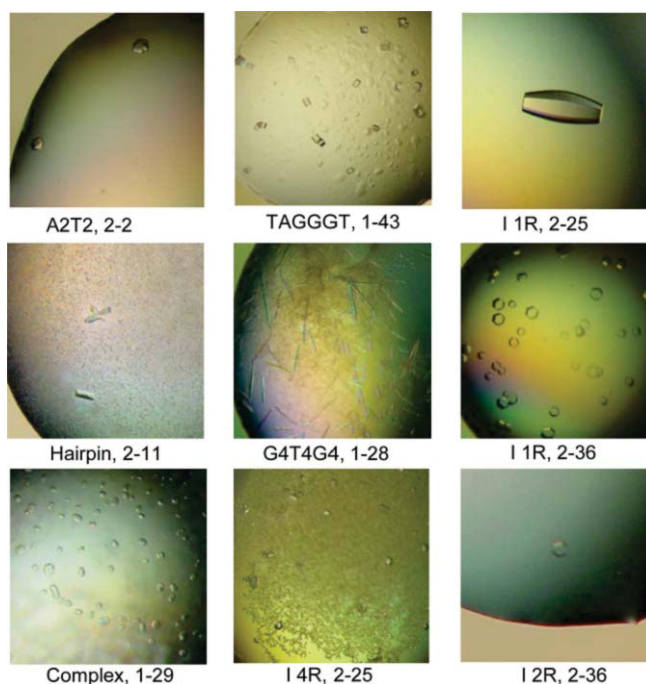
Introduction

HELIX™ is designed to satisfy the requirements for structural biologists to experimentally determine initial nucleation and crystal growth conditions, and subsequently enable successful cryo-cooling along with the unimpeded collection of anomalous data.

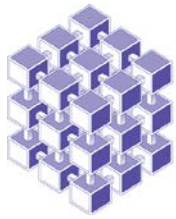
An analysis of the PDB covering the 1324 deposited DNA/RNA crystallization conditions has been crucial in the design of these new 96 conditions.

HELIX Formulation

A buffering range from pH 4.6 to 8.5, combining ammonium acetate, MES, HEPES, and Bis-Tris, ensures appropriate buffering when setting-up crystallizations from 4 °C to 37 °C. The lower pH conditions, for example, enable protonation of cytosine for the formation of i-motifs and promote triplex DNA formation.



Light micrographs showing nine crystals obtained with HELIX for the sequences assayed. Magnifications differ and crystal sizes vary from 20 to 200 nm.



Co-crystallization & Ligand-binding.

Co-crystallization of ligands to nucleic acids needs to ensure that the electrostatic component of ligand binding associations are not reduced significantly by the use of high salt concentrations. It is for this reason HELIX has limited the use of ammonium sulfate as a precipitating agent, and retained the focus on MPD, PEGs.

Reducing the ionic strength in conditions with precipitants retains the electrostatic component thus promotes the formation of ligand-binding.

Cryo-Protection

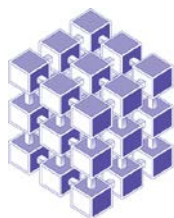
It is now a key requirement to take the initial screening conditions and cryo-protect the crystals for flash-cooling. When flash-cooled the effects of radiation damage on the crystals are limited and allow transportation and long-term storage of the crystal.

HELIX has focused on conditions allowing for an easy transition from the initial well condition towards suitable cryo-conditions. Furthermore, within the kit, 24 conditions have been set aside to enable direct cryo-protection without further modification. For those conditions containing salts known to enhance crystallizations a small percentage of suitable cryo-protectants have been added to aid in subsequent cryo-protection optimization.

Why no cacodylate?

Well apart from the obvious toxicity issue of using cacodylate you might say that nucleic acids would benefit from a chemical that potentially mics a phosphate backbone. Unfortunately the cons outweigh the pros.

The use of cacodylic acid is avoided in HELIX to remove any competing fluorescence signal from the arsenic that will mask the fluorescence emission from selenium, bromine and iodine atoms. The determination of the precise wavelength for the anomalous peak is required to maximize the diffracted anomalous diffraction signal from anomalous scattering elements.



Instructions for Use:

A range of temperature options are available for screening Nucleic Acids, ranging from 4°C – 37 °C.

Common temperatures are 4°C (18%), 16-20°C (34%), 22-25°C (26%), 37°C (5%).

Recommended storage for HELIX is 4°C. Or aliquot into deep-well blocks or microplates and freeze until needed.

Allow 10 mL tubes to equilibrate to room temperature prior to use.

Formulation Notes:

HELIX reagents are formulated using ultrapure water (>18.0 MΩ) and are sterile-filtered using 0.22 μm filters. No preservatives are added.

Final pH may vary from that specified on the datasheet. Molecular Dimensions will be happy to discuss the precise formulation of individual reagents.

Individual reagents and stock solutions for optimization are available from Molecular Dimensions.

Enquiries regarding HELIX formulation, interpretation of results or optimization strategies are welcome. Please e-mail, fax or phone your query to Molecular Dimensions.

Contact and product details can be found at www.moleculardimensions.com

Manufacturer's safety data sheets are available to download from our website.

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HELIX™ HT-96

Wells A1 – D12

MD1-69

Well #	Conc.	Salt 1	Conc.	Salt 2	Conc.	Additive 1	Conc.	Additive 2	Conc.	Buffer	pH	Conc.	Precipitant
A1	0.05 M	Potassium chloride	0.1 M	Lithium chloride	0.012 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	25 % v/v	PEG 400
A2					0.001 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	25 % v/v	PEG 400
A3	0.1 M	Sodium chloride	0.05 M	Lithium chloride	0.01 M	Magnesium chloride hexahydrate			0.05 M	MES	6.5	25 % v/v	PEG 400
A4	0.2 M	Potassium chloride			0.005 M	Hexamine cobalt(III) chloride			0.05 M	MES	6.5	25 % v/v	PEG 400
A5	0.1 M	Lithium chloride			0.01 M	Manganese(II) chloride tetrahydrate			0.05 M	MES	6.5	25 % v/v	PEG 400
A6	0.1 M	Sodium chloride			0.005 M	Strontium chloride hexahydrate			0.05 M	MES	6.5	25 % v/v	PEG 400
A7	0.05 M	Potassium chloride	0.1 M	Lithium chloride	0.012 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	35 % v/v	MPD
A8					0.001 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	35 % v/v	MPD
A9	0.1 M	Sodium chloride	0.1 M	Lithium chloride	0.01 M	Magnesium chloride hexahydrate			0.05 M	MES	6.5	35 % v/v	MPD
A10	0.2 M	Potassium chloride			0.005 M	Hexamine cobalt(III) chloride			0.05 M	MES	6.5	35 % v/v	MPD
A11	0.1 M	Lithium chloride			0.01 M	Manganese(II) chloride tetrahydrate			0.05 M	MES	6.5	35 % v/v	MPD
A12	0.1 M	Sodium chloride			0.005 M	Strontium chloride hexahydrate			0.05 M	MES	6.5	35 % v/v	MPD
B1	0.05 M	Potassium chloride	0.1 M	Lithium chloride	0.012 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	17 % w/v	PEG 4000
B2					0.001 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	17 % w/v	PEG 4000
B3	0.1 M	Sodium chloride	0.05 M	Lithium chloride	0.01 M	Magnesium chloride hexahydrate			0.05 M	MES	6.5	17 % w/v	PEG 4000
B4	0.2 M	Potassium chloride			0.005 M	Hexamine cobalt(III) chloride			0.05 M	MES	6.5	17 % w/v	PEG 4000
B5	0.1 M	Lithium chloride			0.01 M	Manganese(II) chloride tetrahydrate			0.05 M	MES	6.5	17 % w/v	PEG 4000
B6	0.1 M	Sodium chloride			0.005 M	Strontium chloride hexahydrate			0.05 M	MES	6.5	17 % w/v	PEG 4000
B7	0.05 M	Potassium chloride	0.1 M	Lithium chloride	0.012 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	2.6 M	Sodium malonate dibasic monohydrate
B8					0.001 M	Spermine tetrahydrochloride			0.05 M	MES	6.5	2.6 M	Sodium malonate dibasic monohydrate
B9	0.1 M	Sodium chloride	0.05 M	Lithium chloride	0.01 M	Magnesium chloride hexahydrate			0.05 M	MES	6.5	2.6 M	Sodium malonate dibasic monohydrate
B10	0.2 M	Potassium chloride			0.005 M	Hexamine cobalt(III) chloride			0.05 M	MES	6.5	2.6 M	Sodium malonate dibasic monohydrate
B11	0.1 M	Lithium chloride			0.01 M	Manganese(II) chloride tetrahydrate			0.05 M	MES	6.5	2.6 M	Sodium malonate dibasic monohydrate
B12	0.1 M	Sodium chloride			0.002 M	Strontium chloride hexahydrate			0.05 M	MES	6.5	2.6 M	Sodium malonate dibasic monohydrate
C1	0.1 M	Ammonium sulfate							0.05 M	Bis-Tris	7.0	44 % v/v	PEG 200
C2	0.2 M	Potassium chloride			0.01 M	Calcium chloride dihydrate			0.05 M	Bis-Tris	7.0	40 % v/v	PEG 400
C3	0.1 M	Sodium chloride			0.002 M	Spermine tetrahydrochloride			0.05 M	Bis-Tris	7.0	37 % w/v	PEG 1000
C4	0.1 M	Sodium chloride	0.2 M	Potassium chloride	0.02 M	Magnesium chloride hexahydrate			0.05 M	Bis-Tris	7.0	35 % w/v	PEG 2000
C5	0.05 M	Potassium chloride			0.005 M	Spermine tetrahydrochloride			0.05 M	Bis-Tris	7.0	33 % w/v	PEG 3350
C6	0.1 M	Ammonium sulfate	0.1 M	Sodium chloride	1.0 M	Sodium formate			0.05 M	Bis-Tris	7.0	20 % w/v	PEG 8000
C7					1.0 M	Sodium formate			0.05 M	Bis-Tris	7.0	20 % w/v	PEG 20,000
C8	0.05 M	Lithium sulfate							0.05 M	Bis-Tris	7.0	40 % v/v	PEG 500 MME
C9	0.1 M	Sodium citrate tribasic dihydrate							0.05 M	Bis-Tris	7.0	40 % v/v	PEG 500 MME
C10	0.1 M	Ammonium sulfate			0.002 M	Spermine tetrahydrochloride			0.05 M	Bis-Tris	7.0	40 % v/v	MPD
C11									0.05 M	Bis-Tris	7.0	35 % v/v	MPD
C12	0.1 M	Sodium chloride			0.02 M	Magnesium chloride hexahydrate			0.05 M	Bis-Tris	7.0	30 % v/v	MPD
D1	0.1 M	Potassium chloride							0.05 M	Bis-Tris	7.0	27 % v/v	MPD
D2					5 % v/v	PEG 400			0.05 M	Bis-Tris	7.0	24 % v/v	MPD
D3	0.2 M	Lithium sulfate							0.05 M	Bis-Tris	7.0	20 % v/v	MPD
D4					20 % v/v	Glycerol	5 % w/v	PEG 3350	0.05 M	Bis-Tris	7.0	1.0 M	Ammonium sulfate
D5					10 % v/v	Glycerol	5 % v/v	PEG 500 MME	0.05 M	Bis-Tris	7.0	1.2 M	Ammonium sulfate
D6					25 % v/v	Glycerol			0.05 M	Bis-Tris	7.0	1.4 M	Ammonium sulfate
D7	0.15 M	Potassium chloride			25 % v/v	Glycerol			0.05 M	Bis-Tris	7.0	1.6 M	Ammonium sulfate
D8	0.1 M	Sodium chloride			25 % v/v	Glycerol			0.05 M	Bis-Tris	7.0	1.8 M	Ammonium sulfate
D9					5 % v/v	PEG 400			0.05 M	Bis-Tris	7.0	2.0 M	Ammonium sulfate
D10	0.1 M	Potassium chloride							0.05 M	Bis-Tris	7.0	22 % w/v	PEG 2000 MME
D11	0.2 M	Lithium chloride							0.05 M	Bis-Tris	7.0	22 % w/v	PEG 2000 MME
D12	0.1 M	Sodium chloride							0.05 M	Bis-Tris	7.0	22 % w/v	PEG 2000 MME



HELIX™ HT-96

Wells E1 – H12

MD1-69

Well #	Conc. Salt 1	Conc. Salt 2	Conc.	Additive 1	Conc.	Additive 2	Conc.	Buffer	pH	Conc.	Precipitant
E1	0.1 M Potassium chloride						0.05 M	HEPES	6.5	20 % v/v	MPD
E2			0.005 M	Spermine tetrahydrochloride			0.05 M	HEPES	6.5	10 % v/v	MPD
E3	0.1 M Sodium chloride	0.2 M Potassium chloride					0.05 M	HEPES	6.5	10 % v/v	MPD
E4	0.2 M Sodium chloride						0.05 M	HEPES	6.5	25 % v/v	MPD
E5	0.05 M Lithium sulfate						0.05 M	HEPES	6.5	15 % v/v	MPD
E6	0.3 M Sodium chloride						0.05 M	HEPES	6.5	30 % v/v	MPD
E7	0.05 M Sodium chloride		0.01 M	Magnesium chloride hexahydrate			0.05 M	HEPES	6.5	25 % v/v	PEG 400
E8	0.2 M Potassium chloride						0.05 M	Bis-Tris	7.0	10 % v/v	PEG 400
E9	0.05 M Lithium sulfate						0.05 M	HEPES	6.5	18 % w/v	PEG 1000
E10							0.05 M	Bis-Tris	7.0	1.5 M	Lithium sulfate
E11	0.1 M Sodium chloride	0.1 M Potassium chloride					0.05 M	HEPES	6.5	1.9 M	Lithium sulfate
E12			0.02 M	Magnesium chloride hexahydrate			0.05 M	Bis-Tris	7.0	15 % w/v	PEG 2000 MME
F1	0.05 M Lithium sulfate						0.05 M	HEPES	6.5	10 % w/v	PEG 2000 MME
F2							0.05 M	Bis-Tris	7.0	14 % w/v	PEG 2000 MME
F3	0.1 M Lithium chloride						0.05 M	Bis-Tris	7.0	10 % v/v	PEG 500 MME
F4	0.05 M Potassium chloride						0.05 M	Bis-Tris	7.0	1.1 M	Ammonium sulfate
F5	0.05 M Lithium sulfate						0.05 M	HEPES	6.5	1.4 M	Ammonium sulfate
F6	0.1 M Sodium chloride		0.01 M	Lithium chloride			0.05 M	HEPES	6.5	1.8 M	Ammonium sulfate
F7	0.3 M Lithium sulfate						0.05 M	Bis-Tris	7.0	18 % w/v	PEG 1000
F8	0.3 M Sodium chloride						0.05 M	HEPES	6.5	15 % w/v	PEG 1000
F9	0.25 M Potassium chloride						0.05 M	HEPES	6.5	12 % w/v	PEG 1000
F10	0.1 M Lithium sulfate						0.05 M	Bis-Tris	7.0	25 % v/v	PEG 350 MME
F11							0.05 M	HEPES	6.5	20 % v/v	PEG 350 MME
F12	0.05 M Lithium sulfate						0.05 M	HEPES	6.5	15 % v/v	PEG 350 MME
G1			0.005 M	Spermine tetrahydrochloride			0.05 M	Sodium acetate	4.5	18 % v/v	MPD
G2	0.1 M Sodium chloride						0.05 M	Sodium acetate	4.5	24 % v/v	MPD
G3	0.15 M Potassium chloride						0.05 M	Sodium acetate	4.5	32 % v/v	MPD
G4			0.01 M	Magnesium chloride hexahydrate			0.05 M	Sodium acetate	5.0	21 % v/v	MPD
G5	0.1 M Sodium chloride	0.1 M Potassium chloride	0.005 M	Spermine tetrahydrochloride			0.05 M	Sodium acetate	5.0	14 % v/v	MPD
G6			0.05 M	Lithium chloride			0.05 M	Sodium acetate	5.0	15 % v/v	PEG 400
G7							0.05 M	MES	5.5	21 % v/v	MPD
G8			0.02 M	Magnesium chloride hexahydrate			0.05 M	MES	5.5	21 % v/v	MPD
G9							0.05 M	MES	5.5	12 % w/v	PEG 2000
G10			0.02 M	Magnesium sulfate heptahydrate			0.05 M	MES	5.5	21 % v/v	MPD
G11			0.005 M	Spermine tetrahydrochloride			0.05 M	MES	5.5	15 % v/v	PEG 400
G12			10 % v/v	Glycerol			0.05 M	MES	5.5	1.5 M	Ammonium sulfate
H1	0.05 M Sodium chloride	0.1 M Potassium chloride	0.01 M	Calcium chloride dihydrate			0.05 M	Bis-Tris	7.0	25 % w/v	PEG 2000
H2	0.25 M Sodium chloride		0.005 M	Spermine tetrahydrochloride			0.05 M	Bis-Tris	7.0	20 % w/v	PEG 3350
H3	0.1 M Potassium chloride		0.005 M	Barium chloride dihydrate			0.05 M	Bis-Tris	7.0	12 % w/v	PEG 8000
H4	0.3 M Sodium chloride						0.05 M	Bis-Tris	7.0	8 % w/v	PEG 20,000
H5	0.05 M Sodium chloride	0.2 M Lithium chloride	0.01 M	Calcium chloride dihydrate			0.05 M	Bis-Tris	7.0	20 % w/v	PEG 2000
H6	0.1 M Potassium chloride		0.002 M	Spermine tetrahydrochloride			0.05 M	Bis-Tris	7.0	15 % w/v	PEG 2000 MME
H7	0.05 M Potassium chloride	0.1 M Lithium sulfate	0.02 M	Magnesium sulfate heptahydrate			0.05 M	Bis-Tris	7.0	15 % w/v	PEG 2000 MME
H8	0.1 M Sodium chloride		0.002 M	Barium chloride dihydrate			0.05 M	Bis-Tris	7.5	6 % w/v	PEG 20,000
H9	0.1 M Potassium chloride		0.002 M	Spermine tetrahydrochloride			0.05 M	Bis-Tris	7.5	7 % w/v	PEG 8000
H10	0.05 M Lithium sulfate		0.03 M	Magnesium sulfate heptahydrate			0.05 M	Bis-Tris	8.0	15 % w/v	PEG 3350
H11	0.2 M Sodium chloride						0.05 M	Bis-Tris	8.5	9 % w/v	PEG 3350
H12			0.052 M	Magnesium chloride hexahydrate			0.05 M	Bis-Tris	8.5	18 % w/v	PEG 3350



Abbreviations:

MPD: 2-methyl, 2,4-pentanediol, **PEG:** Poly Ethylene Glycol,

Manufacturer's safety data sheets are available from our website or by scanning the QR code here:



Ordering details:

Catalogue Description

Catalogue Code

HELIX™

MD1-68

HELIX™ HT-96

MD1-69

HELIX™ single reagents

MDSR-68-tube number

HELIX™ HT-96 single reagents

MDSR-69-tube number

For HELIX™ stock reagents visit our Optimization page on our website.