

MIDAS™ HT-96 MD1–60

MIDAS™: a Modern Intelligent Dynamic Alternative Screen -

A revolutionary 96 condition crystallization screen based on alternative polymeric precipitants¹, developed and tested in the Laboratory of Dr. Clemens Grimm at University of Würzburg, Germany.

MD1-60 is presented as a 96 x 1.1 mL condition deep-well block.

Features of MIDAS™

- Ideal for both protein, protein/protein complexes, protein-nucleic acid complexes and sensitive macromolecular complexes.
- Narrow range of pH and salt concentrations centered on physiological values.
- Every condition contains at least one alternative polymeric precipitant.
- Designed to complement PEG and salt-based screens.
- Compatible with liquid-handling robots.

MIDAS:

To address this issue, recent work by Clemens Grimm *et al* have devised a protein crystallization screen (MIDAS) that systematically searches for crystallization conditions with alternative polymeric precipitants. To expand the precipitant diversity even more, they also scanned the current water-soluble polymer market for chemical variants or alternatives to the precipitants mentioned above. Owing to their particular properties as surface-active substances, ion exchangers and/or viscosity modifiers, many polymer variants have recently been developed.

MIDAS entails a relatively narrow range of pH and salt concentrations centered on physiological values to increase its suitability for sensitive macromolecular complexes, while every condition contains at least one alternative polymeric precipitant.

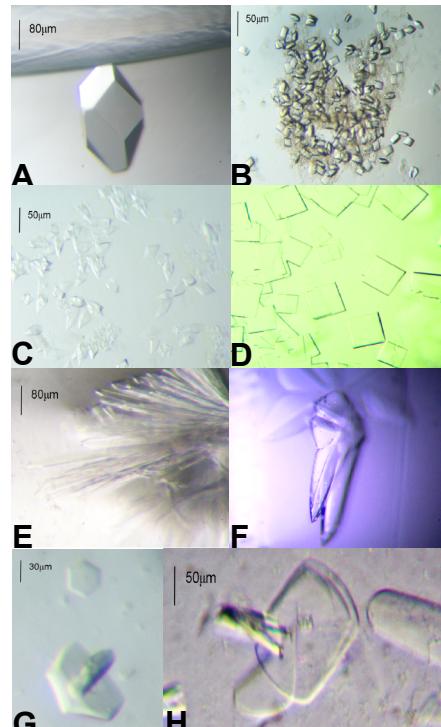


Figure 1. Examples of protein crystals grown using MIDAS.
(A) Lysozyme crystals obtained in 35% Sokalan HP 56, (B) Xylanase crystals obtained in 20% Jeffamine M2070, (C) Crystals of the cytokine receptor-ligand complex obtained in 45% pentaerythritol propoxylate (5/4 PO/OH).
(D) Crystals of streptavidin core obtained in 5% polyacrylate 2100, sodium salt, (E) Histone tail recognizing MBT repeats in 35% polyacrylate 2100, sodium salt, (F) Lysozyme crystals in 30% Sokalan CP 42,(G) spliceosomal assembly complex (SAC) 7 obtained in 6% polyvinyl pyrrolidone K15,

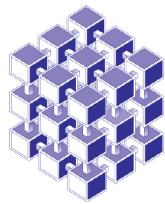
Introduction:

MIDAS is a 96 condition crystallization screen based on alternative polymeric precipitants. Devised and tested (Figure 1) in the Laboratory of Dr. Clemens Grimm *et al* of Würzburg University in Germany, MIDAS is a revolutionary crystallization screen that has moved away from the reliance on polyethylene glycols (PEGs) as the main precipitant (only 3 conditions in MIDAS contain a PEG).

For decades PEGs or their monomethyl ethers (PEG MMEs), have dominated crystallization screens. Out of 8289 entries scanned in the PDB, almost half of the crystallization conditions contained a PEG component and most commercial screens available today contain PEGs. However, the success rate of PEGs might be influenced due to their widespread dominance in crystallization screens.

PEG Alternatives:

There are many alternatives to PEGs and a variety have recently been described as being useful for macromolecular crystallogenesis. Alternative polymers (Figure 2) such as the Jeffamine polyether-amines, pentaerythritol propoxylate and pentareythritol, polyvinyl pyrrolidone, polypropylene glycol, polyvinyl alcohol and polyacrylate have so far only sporadically been introduced into standard crystallization screens.

**Formulation Notes:**

MIDAS reagents are formulated using ultrapure water ($>18.0\text{ }\Omega\text{-cm}$) and are sterile-filtered using $0.22\text{ }\mu\text{m}$ filters. No preservatives are added.

50% Stock solutions of Jeffamine are adjusted to pH 7.0 using HCl prior to inclusion in the reagents.

Sokalan ® CP12 S was adjusted to pH 7 prior to using.

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 MIDAS 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.

References :

1. Grimm, C., Chari, A., Reuter, K. & Fischer, U. (2010). *Acta Cryst. D66*, 685-697.

Ordering details:

Catalogue Description	Catalogue Code
MIDAS™ 10 mL screen	MD1-59
MIDAS™ HT-96 screen	MD1-60
MIDAS™ 10 mL screen single reagents	MDSR-59-tube number
MIDAS™ HT-96 screen single reagents	MDSR-60-well number

For MIDAS™ stock reagents see our website under Optimization.

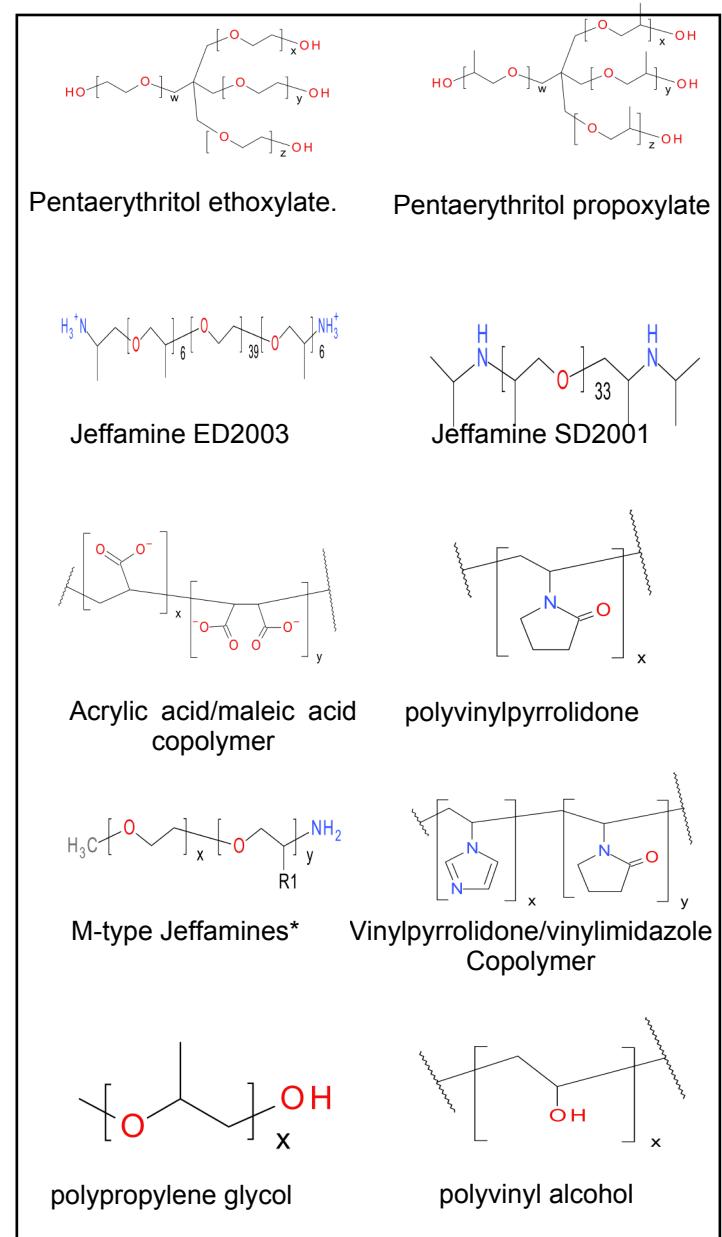
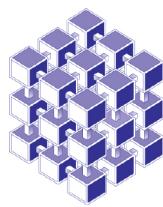


Figure 2. Examples of alternative precipitants used in MIDAS™

*R1 = —H for EO or —CH₃ for PO. The PO/EO molar ratio is 29/6 for Jeffamine M2005, 10/31 for Jeffamine M2070 and 9/1 for Jeffamine M600.

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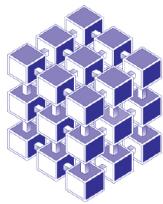
MIDAS™

(Wells A1- D12)

MD1-60

Well No.	% conc	Precipitant	% conc	Salt/Additive	pH	% conc	Buffer
A1	50 v/v	polypropylene glycol 400	5 %	dimethyl sulfoxide	6	0.1 M	HEPES-NaOH
A2	12 w/v	polyvinyl pyrrolidone K15	-	-	5.5	0.1 M	MES- NaOH
A3	45 w/v	polyacrylate 2100, sodium salt	-	-	6.5	0.1 M	HEPES-NaOH
A4	14 v/v	acrylic acid/maleic acid copolymer (50:50), sodium salt	-	-	-	-	-
A5	12.5 w/v	polyacrylate 2100, sodium salt	0.5 M	ammonium phosphate	-	-	-
A6	19 v/v	acrylic acid/maleic acid copolymer (50:50), sodium salt	-	-	8.5	0.1 M	Tris-HCl
A7	10 v/v	polypropylene glycol 400	-	-	-	-	-
A8	5 w/v	polyacrylate 2100, sodium salt	-	-	-	-	-
A9	25 v/v	pentaerythritol propoxylate (5/4 PO/OH)	-	-	6	0.1 M	MES- NaOH
A10	24 % w/v	polyvinyl pyrrolidone K15	0.1 M	sodium sulfate	-	-	-
A11	35 % v/v	pentaerythritol ethoxylate (15/4 EO/OH)	0.2 M	calcium chloride	6.5	0.1 M	HEPES-NaOH
A12	35 % v/v	polypropylene glycol 400	-	-	7	0.1 M	K/Na Phosphate
B1	20% v/v	Jeffamine D2000	0.2 M	sodium chloride	5.5	0.1 M	MES- NaOH
	10 % v/v	Jeffamine M2005	-	-	-	-	-
B2	15 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	0.2 M	sodium thiocyanate	7	0.1 M	HEPES-NaOH
B3	5 % w/v	polyvinyl alcohol type II	0.2 M	potassium acetate	7	0.1 M	HEPES-NaOH
	10 % v/v	Jeffamine T403	-	-	-	-	-
B4	45 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	0.2 M	sodium chloride	6	0.1 M	MES- NaOH
B5	8 % w/v	polyvinyl alcohol type II	10 % v/v	1- propanol	7	0.1 M	HEPES-NaOH
B6	30 % w/v	polyvinyl pyrrolidone K15	0.1 M	lithium sulfate	7	0.1 M	HEPES-NaOH
B7	40 % v/v	polypropylene glycol 400	0.2 M	imidazole	7	-	-
B8	8 % w/v	acrylic acid/maleic acid copolymer (50:50), sodium salt	0.06 M	lithium sulfate	7.5	0.1 M	HEPES-NaOH
	3 % v/v	pentaerythritol ethoxylate (3/4 EO/OH)	-	-	-	-	-
B9	35 % v/v	Jeffamine SD2001	0.1 M	sodium chloride	8	0.1 M	Tris-HCl
B10	30 % v/v	Jeffamine M600	10 % v/v	dimethyl sulfoxide	-	-	-
B11	20 % v/v	polypropylene glycol 400	10 % v/v	1-propanol	-	-	-
B12	28 % w/v	acrylic acid/maleic acid copolymer (50:50), sodium salt	-	-	6.5	0.1 M	HEPES-NaOH
C1	15 % w/v	Jeffamine ED2003	10 % v/v	ethanol	-	-	-
C2	30 % w/v	Jeffamine ED2003	0.2 M	sodium chloride	6	0.1 M	MES- NaOH
C3	25 % v/v	Jeffamine SD2001	0.1 M	sodium malonate	5.5	0.1 M	MES- NaOH
C4	15 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	0.2 M	sodium chloride	6	0.1 M	MES- NaOH
C5	35 % v/v	pentaerythritol ethoxylate (3/4 EO/OH)	0.2 M	Magnesium chloride	-	-	-
C6	40 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	15 % v/v	ethanol	-	-	-
C7	50 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	-	-	8	0.1 M	Tris-HCl
C8	12.5 % w/v	polyvinyl pyrrolidone K15	0.2 M	sodium chloride	8	0.1 M	Tris-HCl
	10 % w/v	PEG 4000	-	-	-	-	-
C9	25 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	0.1 M	sodium chloride	-	-	-
	10 % v/v	dimethyl sulfoxide,	-	-	-	-	-
C10	35 % w/v	polyacrylate 2100, sodium salt	0.2 M	ammonium sulfate	7.5	0.1 M	HEPES-NaOH
C11	30 % v/v	pentaerythritol ethoxylate (15/4 EO/OH)	0.1 M	magnesium formate	8.5	0.1 M	Tris-HCl
C12	20 % v/v	Glascol W13	0.2 M	sodium sulfate	7.5	0.1 M	HEPES-NaOH
D1	60 % v/v	polypropylene glycol 400	-	-	8	0.1 M	Tris-HCl
D2	30 % v/v	pentaerythritol ethoxylate (15/4 EO/OH)	-	-	7.5	0.1 M	HEPES-NaOH
	6 % w/v	polyvinyl pyrrolidone K15	-	-	-	-	-
D3	45 % v/v	polypropylene glycol 400	10 % v/v	ethanol	-	-	-
D4	10 % v/v	pentaerythritol ethoxylate (3/4 EO/OH)	10 % v/v	1-butanol	-	-	-
D5	12.5 % w/v	polyacrylate 2100, sodium salt	-	-	7	0.1 M	HEPES-NaOH
	6 % v/v	Jeffamine SD2001	-	-	-	-	-
D6	6 % w/v	polyvinyl pyrrolidone K15	-	-	6.5	0.1 M	HEPES-NaOH
D7	20 % w/v	Jeffamine ED2003	-	-	6.5	0.1 M	HEPES-NaOH
D8	20 % v/v	glycerol ethoxylate	10 % v/v	tetrahydrofuran	8.0	0.1 M	Tris-HCl
D9	25 % v/v	Jeffamine D2000	0.2 M	imidazole	7	-	-
D10	30 % v/v	Jeffamine SD2001	0.2 M	potassium chloride	6.5	0.1 M	HEPES-NaOH
D11	30 % v/v	polypropylene glycol 400	0.1 M	sodium chloride	-	-	-
D12	20 % v/v	Jeffamine SD2001	15 % v/v	1-propanol	-	-	-

Recommended storage of kits is at room temperature (18°C) or 4°C. If stored at 4°C, turbidity of some solutions may occur. The effect is fully reversible; bringing the tubes back to room temperature and shaking results in clear solutions after 10 min and does not affect the quality of the solution.



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MIDAS™

(Wells E1- H12)

MD1-60

Well No.	% conc	Precipitant	% conc	Salt/Additive	pH	% conc	Buffer
E1	25 % v/v	Jeffamine T403	0.2 M	lithium sulfate	8	0.1 M	Tris-HCl
E2	35 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	0.2 M	potassium acetate	-	-	-
E3	20 % v/v	pentaerythritol ethoxylate (15/4 EO/OH)	0.2 M	potassium chloride	9.5	0.1 M	Glycine
E4	40 % v/v	pentaerythritol propoxylate (5/4 PO/OH)	0.2 M	sodium thiocyanate	7	0.1 M	HEPES-NaOH
E5	15 % v/v	Jeffamine T403	0.2 M	potassium chloride	6.5	0.1 M	HEPES-NaOH
	15 % v/v	Jeffamine ED2003	-	-	-	-	-
E6	15 % v/v	pentaerythritol ethoxylate (15/4 EO/OH), 3 % v/v	0.2 M	potassium acetate	6	0.1 M	MES- NaOH
E7	30 % w/v	polyacrylate 2100, sodium salt	0.1 M	sodium malonate	7	0.1 M	HEPES-NaOH
E8	10 % v/v	Jeffamine D2000	10 % v/v	ethanol	-	-	-
	10 % v/v	Jeffamine M2005	-	-	-	-	-
E9	25 % w/v	Jeffamine ED2003	0.1 M	lithium sulfate	8	0.1 M	Tris-HCl
E10	10 % v/v	Jeffamine T403	-	-	8	0.1 M	Tris-HCl
	10 % w/v	Jeffamine ED2003	-	-	-	-	-
E11	25 % w/v	polyacrylate 2100, sodium salt	0.1 M	lithium sulfate	6.5	0.1 M	HEPES-NaOH
E12	15 % w/v	polyacrylate 2100, sodium salt	0.2 M	magnesium chloride	7.5	0.1 M	HEPES-NaOH
F1	40 % v/v	Jeffamine D2000	-	-	6.5	0.1 M	HEPES-NaOH
F2	10 % w/v	polyacrylate 2100, sodium salt	0.5 M	sodium chloride	8	0.1 M	Tris-HCl
F3	14 % v/v	Jeffamine ED900	-	-	7	0.1 M	K/Na Phosphate
	11 % v/v	Jeffamine SD2001	-	-	-	-	-
F4	20 % w/v	polyacrylate 2100, sodium salt	0.2 M	sodium chloride	9	0.1 M	Bicine
F5	20 % v/v	Jeffamine D2000	0.2 M	sodium malonate	5.5	0.1 M	MES- NaOH
F6	30 % v/v	Jeffamine M2070	0.2 M	potassium chloride	8	0.1 M	Tris-HCl
F7	20 % v/v	Jeffamine M2070	20 % w/v	dimethyl sulfoxide	-	-	-
F8	40 % w/v	pentaerythritol propoxylate (17/8 PO/OH)	0.2 M	magnesium chloride	5.5	0.1 M	MES- NaOH
F9	20 % w/v	polyacrylate 5100, sodium salt	-	-	8	0.1 M	Tris-HCl
F10	28 % v/v	poly(ethylene imine) branched	-	-	7	0.1 M	HEPES-NaOH
F11	20 % v/v	Sokalan® CP 7	0.1 M	ammonium formate	7	0.1 M	HEPES-NaOH
F12	20 % w/v	Sokalan® HP 56	0.2 M	sodium sulfate	8	0.1 M	Tris-HCl
G1	25 % v/v	Sokalan® CP 7	0.1 M	potassium chloride	7	0.1 M	HEPES-NaOH
G2	20 % v/v	Sokalan® CP 5	0.3 M	ammonium formate	7	0.1 M	HEPES-NaOH
G3	40 % v/v	glycerol ethoxylate	-	-	-	-	-
G4	30 % v/v	glycerol ethoxylate	-	-	8.5	0.1 M	Tris-HCl
G5	15 % v/v	Sokalan® HP 66 K	-	-	7	0.1 M	HEPES-NaOH
	3 % v/v	poly(ethylene imine)	-	-	-	-	-
G6	35% v/v	glycerol ethoxylate	0.2 M	lithium citrate	-	-	-
G7	30 % v/v	glycerol ethoxylate	0.2 M	ammonium acetate	6.5	0.1 M	MES- NaOH
G8	20 % v/v	Sokalan® CP 42	5% v/v	methanol	8	0.1 M	Tris-HCl
G9	25 % v/v	Sokalan® CP 42	10 % v/v	tetrahydrofuran	7	0.1 M	Tris-HCl
G10	20 % v/v	Sokalan® CP 42	0.1 M	lithium acetate	6	0.1 M	Bis-Tris- NaOH
G11	15 % v/v	Sokalan® CP 12 S	0.1 M	lithium citrate	5.5	0.1 M	Bis-Tris- NaOH
G12	15 % v/v	Sokalan® CP 5	-	-	6	0.1 M	Bis-Tris- NaOH
H1	25 % v/v	Sokalan® CP 42	-	-	6	0.1 M	Bis-Tris- NaOH
H2	25 % v/v	Sokalan® HP 66 K	0.2 M	ammonium acetate	7	0.1 M	HEPES-NaOH
H3	20 % v/v	glycerol ethoxylate	-	-	8.5	0.1 M	Tris-HCl
	3 % v/v	poly(ethylene imine)	-	-	-	-	-
H4	25 % v/v	glycerol ethoxylate	0.2 M	ammonium chloride	7.5	0.1 M	HEPES-NaOH
H5	40 % v/v	Glascol® W13	0.2 M	potassium citrate	-	-	-
H6	30 % w/v	polyacrylate 5100, sodium salt	10 % v/v	ethanol	6	0.1 M	MES- NaOH
H7	15 % v/v	Sokalan® CP 42	0.2 M	potassium citrate	-	-	-
H8	30 % v/v	Sokalan® CP 42	-	-	8.5	0.1 M	Tris-HCl
H9	25 % w/v	Sokalan® HP 56	0.2 M	ammonium acetate	7	0.1 M	HEPES-NaOH
H10	25 % v/v	Sokalan® CP 5	-	-	8.5	0.1 M	Tris-HCl
H11	10 % w/v	poly(vinyl pyrrolidone) K15	0.2 M	ammonium formate	-	-	-
	20 % w/v	PEG 4000	-	-	-	-	-
H12	15 % w/v	poly(vinyl pyrrolidone) K15	-	-	8	0.1 M	Tris-HCl
	25 % w/v	PEG MME 5000	-	-	-	-	-

Sokalan® are water-soluble polymers based on acrylic acid, maleic acid, vinylpyrrolidone, vinylimidazole and/or hydrophobic monomers.