Red Wings Screen HT™

HAMPTON RESEARCH
Solutions for Crystal Growth

User Guide **HR2-073 (pg 1)**

Features

Red Wings Screen HT™ is a high throughput reagent kit designed to provide a rapid screening method for the crystallization of biological macromolecules. The kit is straightforward, effective, and practical for the determination of preliminary crystallization conditions. The kit is also effective in determining the solubility of a macromolecule in a wide range of reagents and pH.

Red Wings Screen HT is supplied in a sterile, polypropylene Deep Well block, each reservoir containing 1 ml of sterile filtered reagent. The block is heat sealed using a special polypropylene backed film.

Red Wings Screen offer a sparse matrix of trial crystallization reagent conditions based upon the original Red Wings Screen of Alexei Savchenko's group.[3] The primary screen variables are salt, pH, and precipitant (salts, polymers, volatile organics, and non-volatile organics).

General Description

Red Wings Screen HT is supplied in a sterile, polypropylene 96 Deep Well block, each reservoir containing 1 ml of sterile filtered reagent. The block is heat sealed using a special polypropylene backed film.

Each Red Wings Screen HT kit is supplied with an adhesive sealing film which can be used to seal the block after removing the heat seal. Additional adhesive sealing films can be obtained from Hampton Research or laboratory supply companies which offer high throughput plates and seals.

Sample Preparation

The macromolecular sample should be homogenous, as pure as is practically possible (>95%) and free of amorphous and particulate material. Remove amorphous material by centrifugation or microfiltration prior to use. [1,2,4]

The recommended sample concentration is 5 to 25 mg/ml in sterile filtered, deionized water or dilute (25 mM or less) buffer. For initial screens, the sample should be free of unnecessary additives in order to observe the effect of the Red Wings Screen HT variables. However, agents that promote and preserve sample stability and homogeneity can and should be included in the sample. For additional sample preparation recommendation see Crystal Growth 101 - Preliminary Sample Preparation bulletin from Hampton Research.

Preparing the Deep Well Block for Use

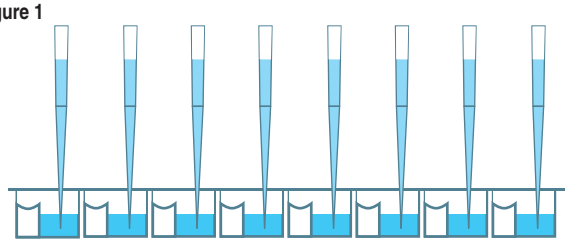
Allow the block to equilibrate to room temperature. To remove stray reagent from the sealing film, centrifuge the block at 500 rpm for 5 minutes. To remove film, grasp a corner of the film and gently peel film from the block. Alternatively, the film can be pierced to access reagents.

Performing the Screen

Manual Method - Sitting Drop Vapor Diffusion

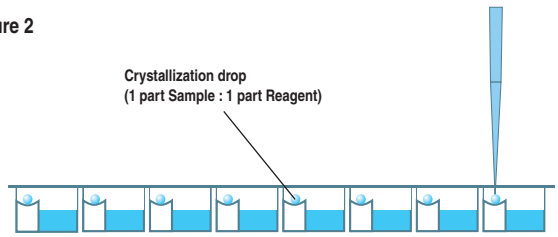
1. Using a 96 well sitting drop vapor diffusion plate, pipet the recommended volume (typically 100 microliters) of crystallization reagent from the Deep Well block into the reservoirs of the crystallization plate. The Deep Well block is compatible with 8 and 12 channel pipets as well as many automated liquid handling systems. Use clean pipet tips for each reagent set transfer and change pipet tips when changing reagents. For an 8 channel pipet, transfer reagents A1-H1 to reservoirs A1-H1 of the crystallization plate. Repeat this procedure for reagent columns 2 through 12. Change pipet tips when moving between reagent columns. For a 12 channel pipet, transfer reagents A1-A12 to reservoirs A1-A12 of the crystallization plate. Repeat this procedure for reagent rows B through H. See Figure 1. Time and pipet tips can be conserved by batch pipetting multiple plates with the same (row or column) of reagent before changing reagent and pipet tips.

Figure 1



2. Using clean pipet tips, pipet 0.05 to 2 microliters of crystallization reagent from the crystallization plate reservoir to the sitting drop well. Some 96 well crystallization plates allow this procedure to be performed using a multichannel pipet where other plates require the use of a single channel pipet. Change the pipet tip between reagents. See Figure 2.

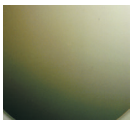
Figure 2

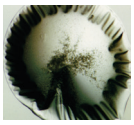


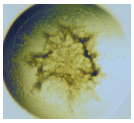
3. Using a clean pipet tip, pipet 0.05 to 2 microliters of sample to the reagent drop in the sitting drop well. One may choose to simply dispense the sample with no mixing or dispense with mixing by gently aspirating and dispensing the sample several times, keeping the tip in the drop during mixing to avoid foaming. Work carefully but quickly to minimize evaporation from the crystallization plate. See Figure 2.

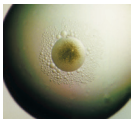
4. Seal the crystallization plate as per the manufacturer's recommendation. Most 96 well crystallization plates are sealed using a clear sealing tape, film, or cap mat. View and score the experiment as desired. See Hampton

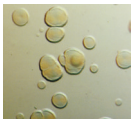
Figure 3
Typical observations in a crystallization experiment

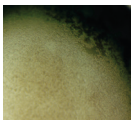

Clear Drop


Skin / Precipitate

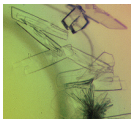

Precipitate


Precipitate / Phase

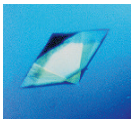

Quasi Crystals


Microcrystals


Needle Cluster


Plates


Rod Cluster


Single Crystal

Research technical bulletin Crystal Growth 101 - Viewing Crystallization Experiments for additional information on viewing drops.

5. Seal the remaining reagent in the Deep Well block using sealing film.

Red Wings Screen HT Deep Well Block and Automated Liquid Handling Systems

The polypropylene Deep Well block is designed to be compatible with the SBS standard 96 microwell format and is therefore compatible with numerous automated liquid handling systems that accept 8 x 12 96 well assay blocks. Follow the manufacturer's recommendation for handling deep well microplates.

Examine the Drop

Carefully examine the drops under a stereo microscope (10 to 100x magnification) immediately after setting up the screen. Record all observations and be particularly careful to scan the focal plane for small crystals. Observe the drops once each day for the first week, then once a week there after. Records should indicate whether the drop is clear, contains precipitate, and or crystals. It is helpful to describe the drop contents using descriptive terms. Adding magnitude is also helpful. Example: 4+ yellow/brown fine precipitate, 2+ small bipyramid crystals, clear drop, 3+ needle shaped crystals in 1+ white precipitate. One may also employ a standard numerical scoring scheme (Clear = 0, Precipitate = 1, Crystal = 10, etc). Figure 3, on the left side of page 2 shows typical examples of what one might observe in a crystallization experiment.

Interpreting Red Wings Screen HT

Clear drops indicate that either the relative supersaturation of the sample and reagent is too low or the drop has not yet completed equilibration. If the drop remains clear after 3 to 4 weeks consider repeating the screen condition and doubling the sample concentration. If more than 70 of the 96 screen drops are clear consider doubling the sample concentration and repeating the entire screen.

Drops containing precipitate indicate either the relative supersaturation of the sample and reagent is too high, the sample has denatured, or the sample is heterogeneous. To reduce the relative supersaturation, dilute the sample twofold and repeat the screen condition. If more than 70 of the 96 screen drops contain precipitate and no crystals are present, consider diluting the sample concentration in half and repeating the entire screen. If sample denaturation is suspect, take measures to stabilize the sample (add reducing agent, ligands, glycerol, salt, or other stabilizing agents). If the sample is impure, aggregated, or heterogeneous take measures to pursue homogeneity. It is possible to obtain crystals from precipitate so do not discard nor ignore a drop containing precipitate. If possible, examine drops containing precipitate under polarizing optics to differentiate precipitate from microcrystalline material.

If the drop contains a macromolecular crystal the relative supersaturation of the sample and reagent is appropriate for crystal nucleation and growth. The next step is to optimize the preliminary conditions (pH, salt type, salt concentration, precipitant type, precipitant concentration, sample concentration, temperature, additives, and other crystallization variables) which produced the crystal in order to improve crystal size and quality.

Compare the observations between the 4°C and room temperature incubation to determine the effect of temperature on sample solubility. Different results in the same drops at different temperatures indicate that sample solubility is temperature dependent and that one should include temperature as a variable in subsequent screens and optimization experiments.

Retain and observe plates until the drops are dried out. Crystal growth can occur within 15 minutes or one year.

HT Formulation

Crystallization reagents are formulated using the highest purity chemicals, ultrapure water (18.2 Megohm-cm, 5 ppb TOC) and are sterile filtered using 0.22 micron filters into sterile Deep Well blocks (no preservatives added).

Crystallization reagents are readily reproduced using Hampton Research Optimize™ and StockOptions™ stock solutions of salts, polymers and buffers. Optimize and StockOptions stock reagents make reproducing crystallization screen reagents accurate, precise, fast, convenient and easy. Dilutions can be performed directly into the crystallization plate using Optimize and StockOptions stock reagents.

Crystallization reagents containing buffers are formulated by creating a 1.0 M stock buffer, titrated to the desired pH using Hydrochloric acid or Sodium hydroxide. The buffer is then

diluted with the other reagent components and water. No further pH adjustment is required.

Crystallization reagents are stable at room temperature and are best if used within 12 months of receipt. To enhance reagent stability the crystallization reagents can be stored at 4°C or -20°C. Avoid ultraviolet light to preserve reagent stability.

If the sample contains phosphate, borate, or carbonate buffers it is possible to obtain inorganic crystals (false positives) when using crystallization reagents containing divalent cations such as magnesium, calcium, or zinc. To avoid false positives use phosphate, borate, or carbonate buffers at concentrations of 10 mM or less or exchange the phosphate, borate, or carbonate buffer with a more soluble buffer that does not complex with divalent cations.

References and Readings

1. Crystallization of nucleic acids and proteins, Edited by A. Ducruix and R. Giege, The Practical Approach Series, Oxford Univ. Press, 1992.

2. Current approaches to macromolecular crystallization. McPherson, A. Eur. J. Biochem. 189, 1-23, 1990.

3. Salvage or recovery of failed targets by in situ proteolysis. Yufeng Tong, Aiping Dong, Xiaohui Xu, Amy Wernimont. Methods Mol Biol. 2014;1140:179-88. doi: 10.1007/978-1-4939-0354-2_14.

4. Protein and Nucleic Acid Crystallization. Methods, A Companion to Methods in Enzymology, Academic Press, Volume 1, Number 1, August 1990.

Technical Support

Inquiries regarding Red Wings Screen HT reagent formulation, interpretation of screen results, optimization strategies and general inquiries regarding crystallization are welcome. Please e-mail, fax, or telephone your request to Hampton Research. Fax and e-mail Technical Support are available 24 hours a day. Telephone technical support is available 8:00 a.m. to 4:00 p.m. USA Pacific Standard Time.

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Well #	Salt	Well #	Buffer ◊	Well #	Precipitant
1. (A1)	None	1. (A1)	0.1 M Sodium acetate trihydrate pH 4.6	1. (A1)	1.5 M Ammonium sulfate
2. (A2)	0.1 M Ammonium sulfate	2. (A2)	0.1 M BIS-TRIS pH 5.5	2. (A2)	25% w/v Polyethylene glycol 3,350
3. (A3)	0.2 M Ammonium acetate	3. (A3)	0.1 M HEPES pH 7.5	3. (A3)	25% w/v Polyethylene glycol 3,350
4. (A4)	None	4. (A4)	None	4. (A4)	0.2 M Magnesium formate dihydrate
5. (A5)	0.2 M Lithium sulfate monohydrate	5. (A5)	0.1 M HEPES pH 7.5	5. (A5)	25% w/v Polyethylene glycol 3,350
6. (A6)	None	6. (A6)	0.1 M Sodium acetate trihydrate pH 4.6	6. (A6)	2.0 M Sodium formate
7. (A7)	0.2 M Potassium acetate	7. (A7)	None	7. (A7)	20% w/v Polyethylene glycol 3,350
8. (A8)	0.2 M Magnesium nitrate hexahydrate	8. (A8)	None	8. (A8)	20% w/v Polyethylene glycol 3,350
9. (A9)	0.2 M Ammonium acetate	9. (A9)	0.1 M Sodium citrate tribasic dihydrate pH 5.6	9. (A9)	15% w/v Polyethylene glycol 4,000
10. (A10)	0.2 M Sodium acetate trihydrate	10. (A10)	0.1 M TRIS hydrochloride pH 8.5	10. (A10)	30% w/v Polyethylene glycol 4,000
11. (A11)	None	11. (A11)	0.1 M TRIS hydrochloride pH 8.5	11. (A11)	8% w/v Polyethylene glycol 8,000
12. (A12)	None	12. (A12)	0.1 M HEPES pH 7.0	12. (A12)	30% v/v Jeffamine® ED-2001 pH 7.0
13. (B1)	None	13. (B1)	0.1 M BIS-TRIS propane pH 7.0	13. (B1)	1.5 M Ammonium sulfate
14. (B2)	0.1 M Ammonium sulfate	14. (B2)	0.1 M BIS-TRIS pH 6.5	14. (B2)	25% w/v Polyethylene glycol 3,350
15. (B3)	0.2 M Ammonium acetate	15. (B3)	0.1 M Tris pH 8.5	15. (B3)	25% w/v Polyethylene glycol 3,350
16. (B4)	None	16. (B4)	0.1 M BIS-TRIS pH 6.5	16. (B4)	0.5 M Magnesium formate dihydrate
17. (B5)	0.2 M Magnesium acetate tetrahydrate	17. (B5)	None	17. (B5)	20% w/v Polyethylene glycol 3,350
18. (B6)	None	18. (B6)	0.1 M BIS-TRIS propane pH 7.0	18. (B6)	2.0 M Sodium formate
19. (B7)	0.2 M Potassium chloride	19. (B7)	None	19. (B7)	20% w/v Polyethylene glycol 3,350
20. (B8)	0.2 M Sodium acetate trihydrate	20. (B8)	None	20. (B8)	20% w/v Polyethylene glycol 3,350
21. (B9)	0.2 M Ammonium sulfate	21. (B9)	0.1 M Sodium acetate trihydrate pH 4.6	21. (B9)	25% w/v Polyethylene glycol 4,000
22. (B10)	None	22. (B10)	0.1 M Sodium acetate trihydrate pH 4.6	22. (B10)	8% w/v Polyethylene glycol 4,000
23. (B11)	None	23. (B11)	0.1 M HEPES pH 7.5	23. (B11)	20% w/v Polyethylene glycol 10,000
24. (B12)	None	24. (B12)	0.1 M HEPES pH 7.0	24. (B12)	30% v/v Jeffamine® ED-2001 pH 7.0
25. (C1)	None	25. (C1)	0.1 M Tris pH 8.5	25. (C1)	1.5 M Ammonium sulfate
26. (C2)	0.1 M Ammonium sulfate	26. (C2)	0.1 M HEPES pH 7.5	26. (C2)	25% w/v Polyethylene glycol 3,350
27. (C3)	0.2 M Ammonium chloride	27. (C3)	None	27. (C3)	20% w/v Polyethylene glycol 3,350
28. (C4)	None	28. (C4)	0.1 M HEPES pH 7.5	28. (C4)	0.5 M Magnesium formate dihydrate
29. (C5)	0.2 M Sodium phosphate monobasic monohydrate	29. (C5)	None	29. (C5)	20% w/v Polyethylene glycol 3,350
30. (C6)	None	30. (C6)	0.1 M Tris pH 8.5	30. (C6)	2.0 M Sodium formate
31. (C7)	0.2 M Potassium phosphate monobasic	31. (C7)	None	31. (C7)	20% w/v Polyethylene glycol 3,350
32. (C8)	0.2 M Lithium citrate tribasic tetrahydrate	32. (C8)	None	32. (C8)	20% w/v Polyethylene glycol 3,350
33. (C9)	0.2 M Lithium sulfate monohydrate	33. (C9)	0.1 M TRIS hydrochloride pH 8.5	33. (C9)	30% w/v Polyethylene glycol 4,000
34. (C10)	0.2 M Sodium acetate trihydrate	34. (C10)	0.1 M Sodium cacodylate trihydrate pH 6.5	34. (C10)	30% w/v Polyethylene glycol 8,000
35. (C11)	None	35. (C11)	None	35. (C11)	30% w/v Polyethylene glycol 1,500
36. (C12)	0.02 M Calcium chloride dihydrate	36. (C12)	0.1 M Sodium acetate trihydrate pH 4.6	36. (C12)	30% v/v (+/-)-2-Methyl-2,4-pentanediol
37. (D1)	None	37. (D1)	0.1 M Sodium acetate trihydrate pH 4.6	37. (D1)	2.5 M Ammonium sulfate
38. (D2)	0.1 M Ammonium sulfate	38. (D2)	0.1 M Tris pH 8.5	38. (D2)	25% w/v Polyethylene glycol 3,350
39. (D3)	0.2 M Ammonium phosphate dibasic	39. (D3)	None	39. (D3)	20% w/v Polyethylene glycol 3,350
40. (D4)	0.2 M Calcium acetate hydrate	40. (D4)	None	40. (D4)	20% w/v Polyethylene glycol 3,350
41. (D5)	0.2 M Ammonium citrate dibasic	41. (D5)	None	41. (D5)	20% w/v Polyethylene glycol 3,350
42. (D6)	None	42. (D6)	0.1 M Sodium acetate trihydrate pH 4.6	42. (D6)	3.5 M Sodium formate
43. (D7)	0.2 M Potassium fluoride	43. (D7)	None	43. (D7)	20% w/v Polyethylene glycol 3,350
44. (D8)	0.2 M Sodium iodide	44. (D8)	None	44. (D8)	20% w/v Polyethylene glycol 3,350
45. (D9)	0.2 M Magnesium chloride hexahydrate	45. (D9)	0.1 M TRIS hydrochloride pH 8.5	45. (D9)	30% w/v Polyethylene glycol 4,000
46. (D10)	0.2 M Sodium chloride	46. (D10)	0.1 M BIS-TRIS pH 6.5	46. (D10)	25% w/v Polyethylene glycol 3,350
47. (D11)	None	47. (D11)	0.1 M MES monohydrate pH 6.5	47. (D11)	12% w/v Polyethylene glycol 20,000
48. (D12)	0.2 M Calcium chloride dihydrate	48. (D12)	0.1 M HEPES sodium pH 7.5	48. (D12)	28% v/v Polyethylene glycol 400

◊ Buffer pH is that of a 1.0 M stock prior to dilution with other reagent components: pH with HCl or NaOH.

Red Wings Screen HT™ (Deep Well Block) contains 96 unique reagents beginning at position A1.
To determine the formulation of each reagent, simply read across the page.

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Well #	Salt	Buffer ◊	Precipitant
49. (E1)	None	0.1 M BIS-TRIS propane pH 7.0	2.5 M Ammonium sulfate
50. (E2)	None	0.1 M HEPES pH 7.5	2.0 M Ammonium formate
51. (E3)	0.2 M Ammonium formate	None	20% w/v Polyethylene glycol 3,350
52. (E4)	0.2 M Calcium chloride dihydrate	None	20% w/v Polyethylene glycol 3,350
53. (E5)	None	0.1 M Sodium acetate trihydrate pH 4.6	3.2 M Sodium chloride
54. (E6)	None	0.1 M BIS-TRIS propane pH 7.0	3.5 M Sodium formate
55. (E7)	0.2 M Potassium sodium tartrate tetrahydrate	None	20% w/v Polyethylene glycol 3,350
56. (E8)	0.15 M Potassium bromide	None	30% w/v Polyethylene glycol monomethyl ether 2,000
57. (E9)	0.2 M Ammonium sulfate	0.1 M Sodium cacodylate trihydrate pH 6.5	30% w/v Polyethylene glycol 8,000
58. (E10)	0.2 M Sodium chloride	0.1 M HEPES pH 7.5	25% w/v Polyethylene glycol 3,350
59. (E11)	0.05 M Magnesium chloride hexahydrate	0.1 M HEPES pH 7.5	30% v/v Polyethylene glycol monomethyl ether 550
60. (E12)	2.0 M Ammonium sulfate	None	5% v/v 2-Propanol
61. (F1)	None	0.1 M Tris pH 8.5	2.5 M Ammonium sulfate
62. (F2)	None	0.1 M TRIS hydrochloride pH 8.5	2.0 M Ammonium phosphate monobasic
63. (F3)	0.2 M Ammonium iodide	None	20% w/v Polyethylene glycol 3,350
64. (F4)	0.2 M Potassium sulfate	None	20% w/v Polyethylene glycol 3,350
65. (F5)	None	0.1 M BIS-TRIS propane pH 7.0	3.2 M Sodium chloride
66. (F6)	None	0.1 M Tris pH 8.5	3.5 M Sodium formate
67. (F7)	0.2 M Magnesium chloride hexahydrate	0.1 M HEPES pH 7.5	25% w/v Polyethylene glycol 3,350
68. (F8)	0.1 M Potassium thiocyanate	None	30% w/v Polyethylene glycol monomethyl ether 2,000
69. (F9)	0.2 M Calcium acetate hydrate	0.1 M Sodium cacodylate trihydrate pH 6.5	18% w/v Polyethylene glycol 8,000
70. (F10)	None	0.1 M Citric acid pH 3.5	25% w/v Polyethylene glycol 3,350
71. (F11)	0.2 M Ammonium sulfate	0.1 M MES monohydrate pH 6.5	30% w/v Polyethylene glycol monomethyl ether 5,000
72. (F12)	0.2 M Sodium citrate tribasic dihydrate	0.1 M Sodium cacodylate trihydrate pH 6.5	15% v/v 2-Propanol
73. (G1)	0.2 M Potassium sodium tartrate tetrahydrate	0.1 M Sodium citrate tribasic dihydrate pH 5.6	2.0 M Ammonium sulfate
74. (G2)	0.2 M Ammonium acetate	0.1 M BIS-TRIS pH 5.5	25% w/v Polyethylene glycol 3,350
75. (G3)	0.2 M Ammonium nitrate	None	20% w/v Polyethylene glycol 3,350
76. (G4)	0.2 M Potassium thiocyanate	None	20% w/v Polyethylene glycol 3,350
77. (G5)	None	0.1 M Tris pH 8.5	3.2 M Sodium chloride
78. (G6)	None	None	1.0 M Sodium phosphate monobasic monohydrate, Potassium phosphate dibasic / pH 6.9
79. (G7)	0.2 M Magnesium chloride hexahydrate	0.1 M Tris pH 8.5	25% w/v Polyethylene glycol 3,350
80. (G8)	0.2 M Trimethylamine N-oxide dihydrate	0.1 M Tris pH 8.5	20% w/v Polyethylene glycol monomethyl ether 2,000
81. (G9)	0.2 M Magnesium acetate tetrahydrate	0.1 M Sodium cacodylate trihydrate pH 6.5	20% w/v Polyethylene glycol 8,000
82. (G10)	None	0.1 M Sodium acetate trihydrate pH 4.5	25% w/v Polyethylene glycol 3,350
83. (G11)	None	0.1 M BIS-TRIS pH 6.5	20% w/v Polyethylene glycol monomethyl ether 5,000
84. (G12)	None	0.1 M HEPES sodium pH 7.5	20% w/v Polyethylene glycol 4,000 10% v/v 2-Propanol
85. (H1)	2.0 M Ammonium sulfate	0.1 M HEPES sodium pH 7.5	2% v/v Polyethylene glycol 400
86. (H2)	0.2 M Ammonium acetate	0.1 M BIS-TRIS pH 6.5	25% w/v Polyethylene glycol 3,350
87. (H3)	0.2 M Ammonium tartrate dibasic	None	20% w/v Polyethylene glycol 3,350
88. (H4)	0.2 M Lithium sulfate monohydrate	0.1 M BIS-TRIS pH 6.5	25% w/v Polyethylene glycol 3,350
89. (H5)	None	0.1 M HEPES sodium pH 7.5	1.4 M Sodium citrate tribasic dihydrate
90. (H6)	0.2 M Sodium tartrate dibasic dihydrate	None	20% w/v Polyethylene glycol 3,350
91. (H7)	0.2 M Magnesium formate dihydrate	None	20% w/v Polyethylene glycol 3,350
92. (H8)	0.2 M Ammonium acetate	0.1 M Sodium citrate tribasic dihydrate pH 5.6	30% w/v Polyethylene glycol 4,000
93. (H9)	0.1 M Succinic Acid pH 7.0	None	15% w/v Polyethylene glycol 3,350
94. (H10)	None	0.1 M BIS-TRIS pH 6.5	28% w/v Polyethylene glycol monomethyl ether 2,000
95. (H11)	None	0.1 M Sodium citrate tribasic dihydrate pH 5.6	2.5 M 1,6-Hexanediol
96. (H12)	None	0.1 M Sodium citrate tribasic dihydrate pH 5.6	20% w/v Polyethylene glycol 4,000 20% v/v 2-Propanol

◊ Buffer pH is that of a 1.0 M stock prior to dilution with other reagent components: pH with HCl or NaOH.

Red Wings Screen HT™ (Deep Well Block) contains 96 unique reagents beginning at position A1.
To determine the formulation of each reagent, simply read across the page.

34 Journey
Aliso Viejo, CA 92656-3317 U.S.A.
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HAMPTON RESEARCH
Solutions for Crystal Growth

Sample: _____ Sample Concentration: _____

Sample Buffer: _____ Date: _____

Reservoir Volume: _____ Temperature: _____

Drop Volume: Total _____ µl Sample _____ µl Reservoir _____ µl Additive _____ µl

1 Clear Drop
2 Phase Separation
3 Regular Granular Precipitate
4 Birefringent Precipitate or Microcrystals
5 Posettes or Spherulites
6 Needles (1D Growth)
7 Plates (2D Growth)
8 Single Crystals (3D Growth < 0.2 mm)
9 Single Crystals (3D Growth > 0.2 mm)

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Red Wings Screen HT™ - HR2-073 Scoring Sheet		Date:	Date:	Date:
1. (A1)	1.5 M Ammonium sulfate, 0.1 M Sodium acetate trihydrate pH 4.6			
2. (A2)	25% w/v Polyethylene glycol 3,350, 0.1 M Ammonium sulfate, 0.1 M BIS-TRIS pH 5.5			
3. (A3)	25% w/v Polyethylene glycol 3,350, 0.2 M Ammonium acetate, 0.1 M HEPES pH 7.5			
4. (A4)	0.2 M Magnesium formate dihydrate			
5. (A5)	25% w/v Polyethylene glycol 3,350, 0.2 M Lithium sulfate monohydrate, 0.1 M HEPES pH 7.5			
6. (A6)	2.0 M Sodium formate, 0.1 M Sodium acetate trihydrate pH 4.6			
7. (A7)	20% w/v Polyethylene glycol 3,350, 0.2 M Potassium acetate			
8. (A8)	20% w/v Polyethylene glycol 3,350, 0.2 M Magnesium nitrate hexahydrate			
9. (A9)	15% w/v Polyethylene glycol 4,000, 0.2 M Ammonium acetate, 0.1 M Sodium citrate tribasic dihydrate pH 5.6			
10. (A10)	30% w/v Polyethylene glycol 4,000, 0.2 M Sodium acetate trihydrate, 0.1 M TRIS hydrochloride pH 8.5			
11. (A11)	8% w/v Polyethylene glycol 8,000, 0.1 M TRIS hydrochloride pH 8.5			
12. (A12)	30% v/v Jeffamine ED-2001 pH 7.0, 0.1 M HEPES pH 7.0			
13. (B1)	1.5 M Ammonium sulfate, 0.1 M BIS-TRIS propane pH 7.0			
14. (B2)	25% w/v Polyethylene glycol 3,350, 0.1 M Ammonium sulfate, 0.1 M BIS-TRIS pH 6.5			
15. (B3)	25% w/v Polyethylene glycol 3,350, 0.2 M Ammonium acetate, 0.1 M Tris pH 8.5			
16. (B4)	0.5 M Magnesium formate dihydrate, 0.1 M BIS-TRIS pH 6.5			
17. (B5)	20% w/v Polyethylene glycol 3,350, 0.2 M Magnesium acetate tetrahydrate			
18. (B6)	2.0 M Sodium formate, 0.1 M BIS-TRIS propane pH 7.0			
19. (B7)	20% w/v Polyethylene glycol 3,350, 0.2 M Potassium chloride			
20. (B8)	20% w/v Polyethylene glycol 3,350, 0.2 M Sodium acetate trihydrate			
21. (B9)	25% w/v Polyethylene glycol 4,000, 0.2 M Ammonium sulfate, 0.1 M Sodium acetate trihydrate pH 4.6			
22. (B10)	8% w/v Polyethylene glycol 4,000, 0.1 M Sodium acetate trihydrate pH 4.6			
23. (B11)	20% w/v Polyethylene glycol 10,000, 0.1 M HEPES pH 7.5			
24. (B12)	30% v/v Jeffamine M-600 pH 7.0, 0.1 M HEPES pH 7.0			
25. (C1)	1.5 M Ammonium sulfate, 0.1 M Tris pH 8.5			
26. (C2)	25% w/v Polyethylene glycol 3,350, 0.1 M Ammonium sulfate, 0.1 M HEPES pH 7.5			
27. (C3)	20% w/v Polyethylene glycol 3,350, 0.2 M Ammonium chloride			
28. (C4)	0.5 M Magnesium formate dihydrate, 0.1 M HEPES pH 7.5			
29. (C5)	20% w/v Polyethylene glycol 3,350, 0.2 M Sodium phosphate monobasic monohydrate			
30. (C6)	2.0 M Sodium formate, 0.1 M Tris pH 8.5			
31. (C7)	20% w/v Polyethylene glycol 3,350, 0.2 M Potassium phosphate monobasic			
32. (C8)	20% w/v Polyethylene glycol 3,350, 0.2 M Lithium citrate tribasic tetrahydrate			
33. (C9)	30% w/v Polyethylene glycol 4,000, 0.2 M Lithium sulfate monohydrate, 0.1 M TRIS hydrochloride pH 8.5			
34. (C10)	30% w/v Polyethylene glycol 8,000, 0.2 M Sodium acetate trihydrate, 0.1 M Sodium cacodylate trihydrate pH 6.5			
35. (C11)	30% w/v Polyethylene glycol 1,500			
36. (C12)	30% v/v (+/-)-2-Methyl-2,4-pentanediol, 0.02 M Calcium chloride dihydrate, 0.1 M Sodium acetate trihydrate pH 4.6			
37. (D1)	2.5 M Ammonium sulfate, 0.1 M Sodium acetate trihydrate pH 4.6			
38. (D2)	25% w/v Polyethylene glycol 3,350, 0.1 M Ammonium sulfate, 0.1 M Tris pH 8.5			
39. (D3)	20% w/v Polyethylene glycol 3,350, 0.2 M Ammonium phosphate dibasic			
40. (D4)	20% w/v Polyethylene glycol 3,350, 0.2 M Calcium acetate hydrate			
41. (D5)	20% w/v Polyethylene glycol 3,350, 0.2 M Ammonium citrate dibasic			
42. (D6)	3.5 M Sodium formate, 0.1 M Sodium acetate trihydrate pH 4.6			
43. (D7)	20% w/v Polyethylene glycol 3,350, 0.2 M Potassium fluoride			
44. (D8)	20% w/v Polyethylene glycol 3,350, 0.2 M Sodium iodide			
45. (D9)	30% w/v Polyethylene glycol 4,000, 0.2 M Magnesium chloride hexahydrate, 0.1 M TRIS hydrochloride pH 8.5			
46. (D10)	25% w/v Polyethylene glycol 3,350, 0.2 M Sodium chloride, 0.1 M BIS-TRIS pH 6.5			
47. (D11)	12% w/v Polyethylene glycol 20,000, 0.1 M MES monohydrate pH 6.5			
48. (D12)	28% v/v Polyethylene glycol 400, 0.2 M Calcium chloride dihydrate, 0.1 M HEPES sodium pH 7.5			

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Red Wings Screen HT™ - HR2-073 Scoring Sheet	Date:	Date:	Date:
49. (E1) 2.5 M Ammonium sulfate, 0.1 M BIS-TRIS propane pH 7.0			
50. (E2) 2.0 M Ammonium formate, 0.1 M HEPES pH 7.5			
51. (E3) 20% w/v Polyethylene glycol 3,350, 0.2 M Ammonium formate			
52. (E4) 20% w/v Polyethylene glycol 3,350, 0.2 M Calcium chloride dihydrate			
53. (E5) 3.2 M Sodium chloride, 0.1 M Sodium acetate trihydrate pH 4.6			
54. (E6) 3.5 M Sodium formate, 0.1 M BIS-TRIS propane pH 7.0			
55. (E7) 20% w/v Polyethylene glycol 3,350, 0.2 M Potassium sodium tartrate tetrahydrate			
56. (E8) 30% w/v Polyethylene glycol monomethyl ether 2,000, 0.15 M Potassium bromide			
57. (E9) 30% w/v Polyethylene glycol 8,000, 0.2 M Ammonium sulfate, 0.1 M Sodium cacodylate trihydrate pH 6.5			
58. (E10) 25% w/v Polyethylene glycol 3,350, 0.2 M Sodium chloride, 0.1 M HEPES pH 7.5			
59. (E11) 30% v/v Polyethylene glycol monomethyl ether 550, 0.05 M Magnesium chloride hexahydrate, 0.1 M HEPES pH 7.5			
60. (E12) 2.0 M Ammonium sulfate, 5% v/v 2-Propanol			
61. (F1) 2.5 M Ammonium sulfate, 0.1 M Tris pH 8.5			
62. (F2) 2.0 M Ammonium phosphate monobasic, 0.1 M TRIS hydrochloride pH 8.5			
63. (F3) 20% w/v Polyethylene glycol 3,350, 0.2 M Ammonium iodide			
64. (F4) 20% w/v Polyethylene glycol 3,350, 0.2 M Potassium sulfate			
65. (F5) 3.2 M Sodium chloride, 0.1 M BIS-TRIS propane pH 7.0			
66. (F6) 3.5 M Sodium formate, 0.1 M Tris pH 8.5			
67. (F7) 25% w/v Polyethylene glycol 3,350, 0.2 M Magnesium chloride hexahydrate, 0.1 M HEPES pH 7.5			
68. (F8) 30% w/v Polyethylene glycol monomethyl ether 2,000, 0.1 M Potassium thiocyanate			
69. (F9) 18% w/v Polyethylene glycol 8,000, 0.2 M Calcium acetate hydrate, 0.1 M Sodium cacodylate trihydrate pH 6.5			
70. (F10) 25% w/v Polyethylene glycol 3,350, 0.1 M Citric acid pH 3.5			
71. (F11) 30% w/v Polyethylene glycol monomethyl ether 5,000, 0.2 M Ammonium sulfate, 0.1 M MES monohydrate pH 6.5			
72. (F12) 15% v/v 2-Propanol, 0.2 M Sodium citrate tribasic dihydrate, 0.1 M Sodium cacodylate trihydrate pH 6.5			
73. (G1) 2.0 M Ammonium sulfate, 0.2 M Potassium sodium tartrate tetrahydrate, 0.1 M Sodium citrate tribasic dihydrate pH 5.6			
74. (G2) 25% w/v Polyethylene glycol 3,350, 0.2 M Ammonium acetate, 0.1 M BIS-TRIS pH 5.5			
75. (G3) 20% w/v Polyethylene glycol 3,350, 0.2 M Ammonium nitrate			
76. (G4) 20% w/v Polyethylene glycol 3,350, 0.2 M Potassium thiocyanate			
77. (G5) 3.2 M Sodium chloride, 0.1 M Tris pH 8.5			
78. (G6) 1.0 M Sodium phosphate monobasic monohydrate, Potassium phosphate dibasic / pH 6.9			
79. (G7) 25% w/v Polyethylene glycol 3,350, 0.2 M Magnesium chloride hexahydrate, 0.1 M Tris pH 8.5			
80. (G8) 20% w/v Polyethylene glycol monomethyl ether 2,000, 0.2 M Trimethylamine N-oxide dihydrate, 0.1 M Tris pH 8.5			
81. (G9) 20% w/v Polyethylene glycol 8,000, 0.2 M Magnesium acetate tetrahydrate, 0.1 M Sodium cacodylate trihydrate pH 6.5			
82. (G10) 25% w/v Polyethylene glycol 3,350, 0.1 M Sodium acetate trihydrate pH 4.5			
83. (G11) 20% w/v Polyethylene glycol monomethyl ether 5,000, 0.1 M BIS-TRIS pH 6.5			
84. (G12) 10% v/v 2-Propanol, 20% w/v Polyethylene glycol 4,000, 0.1 M HEPES sodium pH 7.5			
85. (H1) 2.0 M Ammonium sulfate, 2% v/v Polyethylene glycol 400, 0.1 M HEPES sodium pH 7.5			
86. (H2) 25% w/v Polyethylene glycol 3,350, 0.2 M Ammonium acetate, 0.1 M BIS-TRIS pH 6.5			
87. (H3) 20% w/v Polyethylene glycol 3,350, 0.2 M Ammonium tartrate dibasic			
88. (H4) 25% w/v Polyethylene glycol 3,350, 0.2 M Lithium sulfate monohydrate, 0.1 M BIS-TRIS pH 6.5			
89. (H5) 1.4 M Sodium citrate tribasic dihydrate, 0.1 M HEPES sodium pH 7.5			
90. (H6) 20% w/v Polyethylene glycol 3,350, 0.2 M Sodium tartrate dibasic dihydrate			
91. (H7) 20% w/v Polyethylene glycol 3,350, 0.2 M Magnesium formate dihydrate			
92. (H8) 30% w/v Polyethylene glycol 4,000, 0.2 M Ammonium acetate, 0.1 M Sodium citrate tribasic dihydrate pH 5.6			
93. (H9) 15% w/v Polyethylene glycol 3,350, 0.1 M Succinic Acid pH 7.0			
94. (H10) 28% w/v Polyethylene glycol monomethyl ether 2,000, 0.1 M BIS-TRIS pH 6.5			
95. (H11) 2.5 M 1,6-Hexanediol, 0.1 M Sodium citrate tribasic dihydrate pH 5.6			
96. (H12) 20% v/v 2-Propanol, 20% w/v Polyethylene glycol 4,000, 0.1 M Sodium citrate tribasic dihydrate pH 5.6			

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