

Applications

Crystallization screen for biological macromolecules.

Features

- Reagent formulation developed at Hampton Research.
- Screens an array of polymers of varying molecular weight in a low ionic strength environment versus a wide range of pH.
- Polymer molecular weight range 200 - 20,000.
- pH range 3.5 - 9.0 utilizing 10 unique buffers.
- An array of polymers of varying molecular weight in a medium ionic strength environment in the presence of additives, salts, volatile organics, and polyols versus a wide range of pH.
- Primary screen variables are polymer type, polymer molecular weight, pH and secondary reagents which include additives, salts, volatile organics, and polyols.

General Description

PEGRx HT™ is a crystallization reagent kit designed to provide a rapid screening method for the crystallization of biological macromolecules. PEGRx HT is designed as a 96 reagent crystallization screen that combines the strategies of PEGRx™ 1 and PEGRx™ 2 into a highly effective and efficient format. This kit allows one to evaluate a large variety of potential crystallization conditions with the 96 unique reagents.

PEGRx HT is supplied in a sterile, polypropylene 96 Deep Well block, each reservoir containing 1 ml of sterile filtered reagent. The block is compatible with robotic and multi-channel pipet liquid handling systems and is heat sealed using a special polypropylene backed film. Each PEGRx HT kit is supplied with an adhesive sealing film which can be used to seal the block after removing the heat seal.

Within the 96 Deep Well block, rows A through D feature the 48 reagents of PEGRx 1. PEGRx 1 is a crystallization reagent kit designed to evaluate an array of polymers of varying molecular weight in a low ionic strength environment versus a wide range of pH. Polymer reagents include Polyethylene glycols, Polyethylene glycol monomethyl ethers, and Jeffamines. The molecular weight range between 200 and 20,000 is evaluated in a low ionic strength formulation. Ten different buffers are used to span the range of pH between 3.5 and 9.0. The primary screen variables are polymer type, polymer molecular weight, pH and low ionic strength.

Rows E through H feature the 48 reagents of PEGRx 2. PEGRx 2 is a crystallization reagent kit designed to evaluate an array of polymers of varying molecular weight in a medium ionic strength environment in the presence of additives, salts, volatile organics, and polyols versus a wide range of pH. Polymer reagents include Polyethylene glycols and Polyethylene glycol monomethyl ethers. The polymer molecular weight range between 200 and

20,000 is evaluated in a medium ionic strength formulation. Ten different buffers are used to span the range of pH between 3.5 and 9.0. The primary screen variables are polymer type, polymer molecular weight, pH and secondary reagents which include additives, salts, volatile organics, and polyols.

Refer to the enclosed PEGRx HT reagent formulation for additional information on all 96 reagents.

Sample Preparation

The macromolecular sample should be homogenous, as pure as practically possible (>95%) and free of amorphous and particulate material. Remove amorphous material by centrifugation or micro-filtration prior to use (1, 2, 3).

The recommended sample concentration is 5 to 25 mg/ml in sample buffer. Initially, the sample should be free of any unnecessary additives in order to observe the effect of the PEGRx HT variables. Ideally, the initial screen should be performed with a sample which has been dialyzed against a dilute buffer although ligands, ions, reducing agents, or other additives may be present as required by the sample for solubility, stability, or activity.

Preparing the Deep Well Block for Use

It is recommended the Deep Well block be centrifuged and at 25 degrees Celsius before removing the sealing film. Centrifugation at 500 rpm for five minutes will remove stray reagent from the sealing film. Removing the reagent from the film prevents stray reagent droplets from falling into neighboring wells during film removal. After centrifugation the film can be removed by grasping a corner of the film and gently peeling the film from the plate. Alternatively, the film can be left intact and the pierced for reagent access.

Performing The Screen

Since it is the most frequently reported method of crystallization, the following procedure describes the use of the PEGRx HT with the Sitting Drop Vapor Diffusion method. PEGRx HT is also very compatible with the Hanging Drop, Sandwich Drop, Microbatch, and Microdialysis methods. A complete description of the Hanging, Sitting, Sandwich Drop, Dialysis and other crystallization methods are available from the Hampton Research Crystal Growth 101 Library.

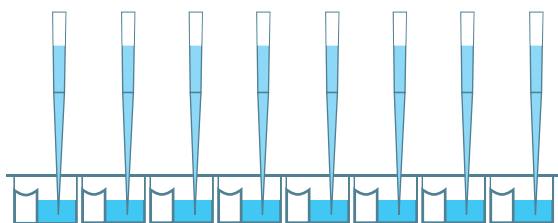
Manual Method - Sitting Drop Vapor Diffusion

1. Using a 96 well sitting drop vapor diffusion plate, pipet the recommended volume (typically 50 to 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 trans-

fer 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 1 through 12. Change pipets when moving between reagent columns.

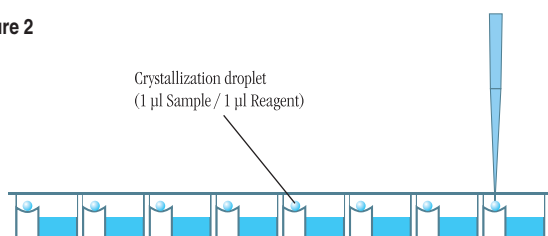
For a 12 channel pipet, transfer reagents A1-A12 to reservoirs A1-A12 of the crystallization plate. Repeat this procedure for reagent rows A through H. See Figure 1 below. Time and pipet tips can be conserved by batch pipetting multiple plates with the same (row or column) of reagent before changing 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 multi-channel pipet where other plates require the use of a single channel pipet. Change the pipet tip between reagents. See Figure 2 below.

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 manufacturers recommendation. Most 96 well crystallization plates are sealed using a clear sealing tape or film. View and score the experiment as desired. See Hampton Research technical bulletin Crystal Growth 101 - Viewing Crystallization Experiments for additional information on viewing drops.

PEGRx 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 8x12 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 thereafter. 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 page 3) shows typical examples of what one might observe in a crystallization experiment.

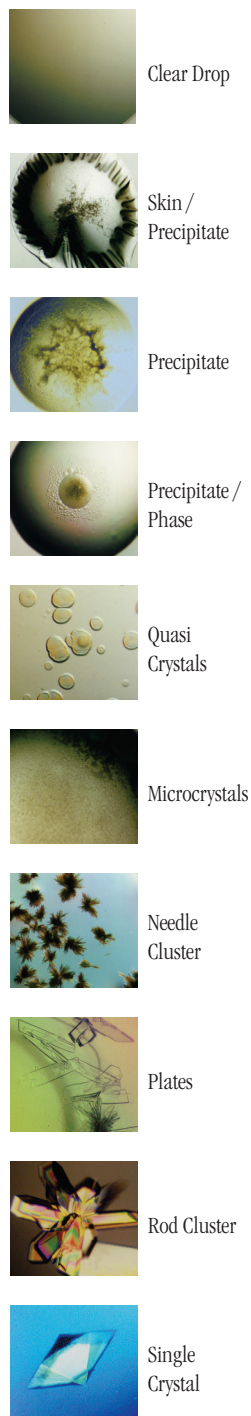
Interpreting PEGRx HT

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.

Figure 3
Typical observations in a crystallization experiment



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.

PEGRx 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).

PEGRx HT reagents are readily reproduced using Hampton Research Optimize™ and StockOptions™ stock solutions of salts, polymers and buffers. Refer to PEGRx 1 and PEGRx 2 Fundamentals for further information regarding reagent formulation. 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.

PEGRx HT reagents are stable at room temperature and are best if used within 12 months of receipt. To enhance reagent stability it is strongly recommended that PEGRx HT reagents 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. Protein and Nucleic Acid Crystallization. Methods, A Companion to Methods in Enzymology, Academic Press, Volume 1, Number 1, August 1990.

Technical Support

Inquiries regarding PEGRx 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 4:30 p.m. USA Pacific Standard Time.

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How to Reproduce PEGRx HT Reagents

PEGRx HT™ reagents and optimization conditions based on PEGRx HT hits can be formulated using volumetric methods and carefully prepared reagent stocks (Table 1). Note the examples below.

Example 1. To prepare 1.0 milliliter of PEGRx™ 1 reagent 14 (Well B2).

Solution Composition: 0.1 M MES monohydrate pH 6.0
20% v/v Jeffamine® M-600® pH 7.0

- 500 µl water³
- 100 µl 1.0 M MES monohydrate pH 6.0
(CAS # 145224-94-8, Catalog # HR2-943-09)
- 400 µl 50% v/v Jeffamine® M-600® pH 7.0
(CAS # 77110-54-4, Catalog # HR2-501)

Make no pH adjustments. Mix well.

Example 2. To prepare 1.0 milliliter of PEGRx™ 2 reagent 15 (Well F3).

Solution Composition: 0.1 M Sodium malonate pH 8.0
0.1 M Tris pH 8.0
30% w/v Polyethylene glycol 1,000

- 570.6 µl water³
- 29.4 µl 3.4 M Sodium malonate pH 8.0
(CAS # 141-82-2, Catalog # HR2-807)
- 100 µl 1.0 M Tris pH 8.0
(CAS # 77-86-1, Catalog # HR2-900-11)
- 300 µl 100% w/v Polyethylene glycol 1,000
(CAS # 25322-68-3, Catalog # HR2-523)

Make no pH adjustments. Mix well.

Example 3. To prepare 1 milliliter of PEGRx 2 reagent 48 (Well H12).

Solution Composition: 3% w/v Dextran sulfate sodium salt
0.1 M BICINE pH 8.5
15% w/v Polyethylene glycol 20,000

- 300 µl water³
- 100 µl 1.0 M BICINE pH 8.5
(CAS # 150-25-4, Catalog # HR2-999-10)
- 100 µl 30% w/v Dextran sulfate sodium salt
(CAS # 9011-18-1, Catalog # HR2-428-51)
- 500 µl 30% w/v Polyethylene glycol 20,000
(CAS # 25322-68-3, Catalog # HR2-609)

Make no pH adjustments. Mix well.

³ ASTM Type I water.

Formulation Notes for PEGRx HT Reagents

1. No additional pH adjustment is made to any reagent after formulation. Use the salts and buffers in Table 1 to reproduce a PEGRx HT reagent.
2. All Optimize solutions and screen reagents are sterile filtered using 0.22 µm filters into sterile containers.
3. Add water first as this will help maintain the solubility of subsequently added reagents.
4. When formulating reagents using a pipet, add the largest volume last (except water). Use this larger volume setting to aspirate and dispense the reagent until the solution is mixed.
5. When formulating reagents using a pipet, use a clean, sterile pipet tip for each reagent added to the solution.
6. Use Optimize™, Custom Shop™, StockOptions™ pH, and StockOptions™ pH buffer kits from Hampton Research to systematically vary the pH as a crystallization variable.
7. The measured final pH of all PEGRx HT reagents is available at www.hamptonresearch.com. Search using catalog number HR2-086 and follow the link to the 'PEGRx HT pH and Conductivity' document.

pH as a Crystallization Variable

The buffers listed in Table 2 can be used to vary the pH as a crystallization variable and are recommended when optimizing a crystal grown from a PEGRx HT kit.

Optimize buffer stocks are supplied as a 100 milliliters sterile filtered solution.

StockOptions buffer kits contain 10 milliliters each of ready to pipet buffers, titrated in 0.1 pH increments over the indicated pH range. The number of reagents offered in a StockOptions buffer kit depends upon the pH range of the buffer. The broader the pH range, the more buffers in the kit.

Custom Shop ready to pipet buffers are made to order, pH titrated buffer stocks from Hampton Research.

Online Information

Visit www.hamptonresearch.com and enter one of the following:

- Reagent Catalog Number
- Kit Catalog Number
- CAS Number
- Reagent Name

To obtain reagent specifications, pH titration tables, user guides, certificates of analysis, material safety data sheets (MSDS), and any other additional information.

MakeTray™

MakeTray is a free, web based program at www.hamptonresearch.com which generates both a pipetting worksheet and a reagent formulation document for crystallization set ups. MakeTray allows one to enter general information about the sample and experiment, which is then printed on the pipet worksheet and the reagent formulation document. The plate size can be customized for any number of wells, so MakeTray works for: 24, 48, and 96 well plates. MakeTray is especially useful for the design and formulation of crystal optimization experiments.

Table 1. Recommended reagents for the formulation of PEGRx HT and optimization reagents.

Each of these reagents are available as an Optimize crystallization grade reagent from Hampton Research. Table 1 provides the common chemical name, the Hampton Research catalog number, supplied stock concentration, the supplied volume, and the CAS number for each reagent. For more information on a specific Optimize reagent, go to

www.hamptonresearch.com. Using Search, enter either the catalog number, CAS number, or chemical name to obtain additional information for the Optimize reagent, including a Certificate of Analysis and MSDS (where applicable).

Additive	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
Dextran sulfate sodium salt	HR2-428-51	30% w/v	1.0 ml	9011-18-1
n-Octyl-β-D-glucoside	HR2-428-71	5% w/v	1.0 ml	29836-26-8
Organics (Volatile)	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
1,4-Dioxane	HR2-617	100%	200 ml	123-91-1
2-Propanol	HR2-619	100%	200 ml	67-63-0
Polyol	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
(+/-)-2-Methyl-2,4-pentanediol	HR2-627	100%	200 ml	107-41-5
Ethylene glycol	HR2-621	100%	100 ml	107-21-1
Polyethylene glycol 200	HR2-601	100%	200 ml	25322-68-3
Polyethylene glycol 400	HR2-603	100%	200 ml	25322-68-3
Salt	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
Ammonium acetate	HR2-565	1.0 M	100 ml	631-61-8
	HR2-799	8.0 M	200 ml	631-61-8
(Salt Continued on page 3)				

Table 1 (Continued). Recommended reagents for the formulation of PEGRx HT and optimization reagents.

Salt	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
Ammonium citrate tribasic pH 7.0	HR2-759	2.5 M	200 ml	3458-72-8
Ammonium sulfate	HR2-541	3.5 M	200 ml	7783-20-2
Cadmium chloride hydrate	HR2-715	1.0 M	100 ml	654054-66-7
Calcium chloride dihydrate	HR2-557	2.0 M	100 ml	10035-04-8
L-Proline	HR2-775	1.0 M	100 ml	147-85-3
Lithium sulfate monohydrate	HR2-545	2.0 M	200 ml	10102-25-7
Magnesium chloride hexahydrate	HR2-559	2.0 M	100 ml	7791-18-6
	HR2-803	5.0 M	200 ml	7791-18-6
Magnesium formate dihydrate	HR2-537	1.0 M	200 ml	557-39-1
DL-Malic acid pH 7.0	HR2-761	3.0 M	200 ml	6915-15-7
Nickel(II) chloride hexahydrate	HR2-687	4.0 M	200 ml	7791-20-0
Potassium formate	HR2-667	14.0 M	200 ml	590-29-4
Potassium sodium tartrate tetrahydrate	HR2-539	1.5 M	200 ml	6381-59-5
Succinic acid pH 7.0	HR2-709	1.2 M	200 ml	110-15-6
Sodium chloride	HR2-637	5.0 M	200 ml	7647-14-5
Sodium formate	HR2-547	7.0 M	200 ml	141-53-7
Sodium malonate pH 5.0	HR2-749	3.4 M	200 ml	141-82-2
Sodium malonate pH 6.0	HR2-751	3.4 M	200 ml	141-82-2
Sodium malonate pH 8.0	HR2-807	3.4 M	200 ml	141-82-2
Tacsimate™ pH 4.0	HR2-823	100%	200 ml	N/A
Tacsimate™ pH 6.0	HR2-827	100%	200 ml	N/A
Tacsimate™ pH 7.0	HR2-755	100%	200 ml	N/A
Polymer	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
Jeffamine® ED-2001 pH 7.0	HR2-597	50% w/v	200 ml	65605-36-9
Jeffamine® M-600® pH 7.0	HR2-501	50% v/v	200 ml	77110-54-4
Polyethylene glycol 200	HR2-601	100%	200 ml	25322-68-3
Polyethylene glycol 300	HR2-517	100%	200 ml	25322-68-3
Polyethylene glycol 400	HR2-603	100%	200 ml	25322-68-3
Polyethylene glycol 1,000	HR2-523	50% w/v	200 ml	25322-68-3
Polyethylene glycol 1,500	HR2-525	50% w/v	200 ml	25322-68-3

(Polymer Continued on page 4)

Table 1 (Continued). Recommended reagents for the formulation of PEGRx HT and optimization reagents.

Polymer	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
Polyethylene glycol 3,350	HR2-527	50% w/v	200 ml	25322-68-3
Polyethylene glycol 4,000	HR2-529	50% w/v	200 ml	25322-68-3
Polyethylene glycol 6,000	HR2-533	50% w/v	200 ml	25322-68-3
Polyethylene glycol 8,000	HR2-535	50% w/v	200 ml	25322-68-3
Polyethylene glycol 10,000	HR2-607	50% w/v	200 ml	25322-68-3
Polyethylene glycol 20,000	HR2-609	30% w/v	200 ml	25322-68-3
Polyethylene glycol monomethyl ether 550	HR2-611	100%	200 ml	9004-74-4
Polyethylene glycol monomethyl ether 2,000	HR2-613	50% w/v	200 ml	9004-74-4
Polyethylene glycol monomethyl ether 5,000	HR2-615	50% w/v	200 ml	9004-74-4
Buffer	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #
BICINE pH 8.5 ²	HR2-999-10	1.0 M	185 ml	150-25-4
BIS-TRIS pH 6.5 ¹	HR2-783	1.0 M	100 ml	6976-37-0
BIS-TRIS propane pH 9.0 ¹	HR2-993-28	1.0 M	185 ml	64431-96-5
Citric acid pH 3.5 ²	HR2-757	1.0 M	100 ml	77-92-9
HEPES pH 7.5 ²	HR2-729	1.0 M	100 ml	7365-45-9
Imidazole pH 7.0 ¹	HR2-819	1.0 M	100 ml	288-32-4
MES monohydrate pH 6.0 ²	HR2-943-09	1.0 M	185 ml	145224-94-8
Sodium acetate trihydrate pH 4.0 ¹	HR2-933-05	1.0 M	185 ml	6131-90-4
Sodium acetate trihydrate pH 4.5 ¹	HR2-789	1.0 M	100 ml	6131-90-4
Sodium citrate tribasic dihydrate pH 5.0 ¹	HR2-935-09	1.0 M	185 ml	6132-04-3
Sodium citrate tribasic dihydrate pH 5.5 ¹	HR2-935-14	1.0 M	185 ml	6132-04-3
Tris pH 8.0 ¹	HR2-900-11	1.0 M	185 ml	77-86-1
¹ pH titrated using Hydrochloric acid (HR2-581) CAS # 7647-01-0				
² pH titrated using Sodium hydroxide (HR2-583) CAS # 1310-73-2				

Table 2. Recommended buffers for screening the pH of PEGRx HT and optimization reagents.

Buffer Solution or Kit	Hampton Research Catalog #	Supplied [Stock]	Supplied Volume	CAS #	pH range
StockOptions™ BICINE	HR2-999-**	1.0 M	185 ml	150-25-4	7.6 - 9.0
StockOptions™ Bis-Tris kit ⁴	HR2-106	1.0 M	10 ml each	6976-37-0	5.5 - 7.5
StockOptions™ Bis-Tris propane	HR2-993-**	1.0 M	185 ml	64431-96-5	6.3 - 9.5
StockOptions™ Citric acid ⁴	HR2-104	1.0 M	10 ml each	77-92-9	2.2 - 6.5
StockOptions™ Hepes kit ⁴	HR2-102	1.0 M	10 ml each	7365-45-9	6.8 - 8.2
StockOptions™ Imidazole	HR2-995-**	1.0 M	185 ml	288-32-4	6.2 - 7.8
StockOptions™ MES kit ⁴	HR2-243	1.0 M	10 ml each	145224-94-8	5.2 - 7.1
StockOptions™ Sodium Acetate kit ⁴	HR2-233	1.0 M	10 ml each	6131-90-4	3.6 - 5.6
StockOptions™ Sodium Citrate kit ⁴	HR2-235	1.0 M	10 ml each	6132-04-3	4.2 - 6.5
StockOptions™ Tris ⁴	HR2-100	1.0 M	10 ml each	77-86-1	7.0 - 9.0

⁴ Individual StockOptions buffers titrated to any pH within the kit's pH range are available in 185 ml volumes from the Hampton Research Custom Shop™

** Refers to the reagent number in the kit. For example, reagent number 1 = HR2-993-01 (pH 6.3)

Technical Support

Inquiries regarding PEGRx HT Fundamentals, 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 4:30 p.m. USA Pacific Standard Time.

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Well #	Buffer ◇	Well #	Polymer
1. (A1)	0.1 M Citric acid pH 3.5	1. (A1)	34% v/v Polyethylene glycol 200
2. (A2)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	2. (A2)	38% v/v Polyethylene glycol 200
3. (A3)	0.1 M HEPES pH 7.5	3. (A3)	42% v/v Polyethylene glycol 200
4. (A4)	0.1 M Sodium acetate trihydrate pH 4.5	4. (A4)	30% v/v Polyethylene glycol 300
5. (A5)	0.1 M BIS-TRIS pH 6.5	5. (A5)	25% v/v Polyethylene glycol 300
6. (A6)	0.1 M BICINE pH 8.5	6. (A6)	20% v/v Polyethylene glycol 300
7. (A7)	0.1 M Sodium acetate trihydrate pH 4.0	7. (A7)	15% v/v Polyethylene glycol 400
8. (A8)	0.1 M MES monohydrate pH 6.0	8. (A8)	22% v/v Polyethylene glycol 400
9. (A9)	0.1 M Tris pH 8.0	9. (A9)	30% v/v Polyethylene glycol 400
10. (A10)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	10. (A10)	30% v/v Polyethylene glycol monomethyl ether 550
11. (A11)	0.1 M Imidazole pH 7.0	11. (A11)	25% v/v Polyethylene glycol monomethyl ether 550
12. (A12)	0.1 M BIS-TRIS propane pH 9.0	12. (A12)	20% v/v Polyethylene glycol monomethyl ether 550
13. (B1)	0.1 M Sodium acetate trihydrate pH 4.0	13. (B1)	10% v/v Jeffamine® M-600® pH 7.0
14. (B2)	0.1 M MES monohydrate pH 6.0	14. (B2)	20% v/v Jeffamine® M-600® pH 7.0
15. (B3)	0.1 M Tris pH 8.0	15. (B3)	30% v/v Jeffamine® M-600® pH 7.0
16. (B4)	0.1 M Citric acid pH 3.5	16. (B4)	14% w/v Polyethylene glycol 1,000
17. (B5)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	17. (B5)	22% w/v Polyethylene glycol 1,000
18. (B6)	0.1 M HEPES pH 7.5	18. (B6)	30% w/v Polyethylene glycol 1,000
19. (B7)	0.1 M Sodium acetate trihydrate pH 4.5	19. (B7)	30% w/v Polyethylene glycol 1,500
20. (B8)	0.1 M BIS-TRIS pH 6.5	20. (B8)	20% w/v Polyethylene glycol 1,500
21. (B9)	0.1 M BICINE pH 8.5	21. (B9)	15% w/v Polyethylene glycol 1,500
22. (B10)	0.1 M Sodium acetate trihydrate pH 4.0	22. (B10)	10% w/v Polyethylene glycol monomethyl ether 2,000
23. (B11)	0.1 M MES monohydrate pH 6.0	23. (B11)	20% w/v Polyethylene glycol monomethyl ether 2,000
24. (B12)	0.1 M Tris pH 8.0	24. (B12)	30% w/v Polyethylene glycol monomethyl ether 2,000
25. (C1)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	25. (C1)	30% v/v Jeffamine® ED-2001 pH 7.0
26. (C2)	0.1 M Imidazole pH 7.0	26. (C2)	20% v/v Jeffamine® ED-2001 pH 7.0
27. (C3)	0.1 M BIS-TRIS propane pH 9.0	27. (C3)	10% v/v Jeffamine® ED-2001 pH 7.0
28. (C4)	0.1 M Citric acid pH 3.5	28. (C4)	25% w/v Polyethylene glycol 3,350
29. (C5)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	29. (C5)	18% w/v Polyethylene glycol 3,350
30. (C6)	0.1 M HEPES pH 7.5	30. (C6)	12% w/v Polyethylene glycol 3,350
31. (C7)	0.1 M Sodium acetate trihydrate pH 4.0	31. (C7)	10% w/v Polyethylene glycol 4,000
32. (C8)	0.1 M MES monohydrate pH 6.0	32. (C8)	14% w/v Polyethylene glycol 4,000
33. (C9)	0.1 M Tris pH 8.0	33. (C9)	28% w/v Polyethylene glycol 4,000
34. (C10)	0.1 M Sodium acetate trihydrate pH 4.5	34. (C10)	30% w/v Polyethylene glycol monomethyl ether 5,000
35. (C11)	0.1 M BIS-TRIS pH 6.5	35. (C11)	20% w/v Polyethylene glycol monomethyl ether 5,000
36. (C12)	0.1 M BICINE pH 8.5	36. (C12)	8% w/v Polyethylene glycol monomethyl ether 5,000
37. (D1)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	37. (D1)	10% w/v Polyethylene glycol 6,000
38. (D2)	0.1 M Imidazole pH 7.0	38. (D2)	20% w/v Polyethylene glycol 6,000
39. (D3)	0.1 M BIS-TRIS propane pH 9.0	39. (D3)	30% w/v Polyethylene glycol 6,000
40. (D4)	0.1 M Citric acid pH 3.5	40. (D4)	28% w/v Polyethylene glycol 8,000
41. (D5)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	41. (D5)	16% w/v Polyethylene glycol 8,000
42. (D6)	0.1 M HEPES pH 7.5	42. (D6)	4% w/v Polyethylene glycol 8,000
43. (D7)	0.1 M Sodium acetate trihydrate pH 4.5	43. (D7)	10% w/v Polyethylene glycol 10,000
44. (D8)	0.1 M BIS-TRIS pH 6.5	44. (D8)	16% w/v Polyethylene glycol 10,000
45. (D9)	0.1 M BICINE pH 8.5	45. (D9)	20% w/v Polyethylene glycol 10,000
46. (D10)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	46. (D10)	18% w/v Polyethylene glycol 20,000
47. (D11)	0.1 M Imidazole pH 7.0	47. (D11)	12% w/v Polyethylene glycol 20,000
48. (D12)	0.1 M BIS-TRIS propane pH 9.0	48. (D12)	8% w/v Polyethylene glycol 20,000

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

PEGRx™ 1 (Deep Well Block) contains forty-eight unique reagents beginning at position A1. To determine the formulation of each reagent, simply read across the page.

Well #	Additive / Salt / Volatile Organic / Polyol	Well #	Buffer ◇	Well #	Polymer
49. (E1)	0.8 M Lithium sulfate monohydrate	49. (E1)	0.1 M Sodium acetate trihydrate pH 4.0	49. (E1)	4% v/v Polyethylene glycol 200
50. (E2)	0.2 M Lithium sulfate monohydrate	50. (E2)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	50. (E2)	26% v/v Polyethylene glycol 200
51. (E3)	0.05 M Calcium chloride dihydrate	51. (E3)	0.1 M MES monohydrate pH 6.0	51. (E3)	45% v/v Polyethylene glycol 200
52. (E4)	28% v/v 2-Propanol	52. (E4)	0.1 M BIS-TRIS pH 6.5	52. (E4)	3% v/v Polyethylene glycol 200
53. (E5)	20% v/v Tacsimate pH 7.0	53. (E5)	0.1 M HEPES pH 7.5	53. (E5)	2% v/v Polyethylene glycol 200
54. (E6)	10% v/v 2-Propanol	54. (E6)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	54. (E6)	26% v/v Polyethylene glycol 400
55. (E7)	0.2 M Ammonium acetate	55. (E7)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	55. (E7)	24% v/v Polyethylene glycol 400
56. (E8)	0.2 M Ammonium sulfate	56. (E8)	0.1 M BIS-TRIS pH 6.5	56. (E8)	18% v/v Polyethylene glycol 400
57. (E9)		57. (E9)	0.1 M HEPES pH 7.5	57. (E9)	40% v/v Polyethylene glycol 400
58. (E10)	6% v/v 2-Propanol	58. (E10)	0.1 M Sodium acetate trihydrate pH 4.5	58. (E10)	26% v/v Polyethylene glycol monomethyl ether 550
59. (E11)	1.8 M Ammonium sulfate	59. (E11)	0.1 M BIS-TRIS pH 6.5	59. (E11)	2% v/v Polyethylene glycol monomethyl ether 550
60. (E12)	0.15 M DL-Malic acid pH 7.0	60. (E12)	0.1 M Imidazole pH 7.0	60. (E12)	22% v/v Polyethylene glycol monomethyl ether 550
61. (F1)	0.1 M Succinic acid pH 7.0	61. (F1)	0.1 M BICINE pH 8.5	61. (F1)	30% v/v Polyethylene glycol monomethyl ether 550
62. (F2)	0.1 M Lithium sulfate monohydrate	62. (F2)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	62. (F2)	20% w/v Polyethylene glycol 1,000
63. (F3)	0.1 M Sodium malonate pH 8.0	63. (F3)	0.1 M Tris pH 8.0	63. (F3)	30% w/v Polyethylene glycol 1,000
64. (F4)	4% v/v (+/-)-2-Methyl-2,4-pentanediol	64. (F4)	0.1 M Citric acid pH 3.5	64. (F4)	20% w/v Polyethylene glycol 1,500
65. (F5)	0.2 M L-Proline	65. (F5)	0.1 M HEPES pH 7.5	65. (F5)	24% w/v Polyethylene glycol 1,500
66. (F6)	10% v/v 2-Propanol	66. (F6)	0.1 M BICINE pH 8.5	66. (F6)	30% w/v Polyethylene glycol 1,500
67. (F7)	0.1 M Sodium chloride	67. (F7)	0.1 M BIS-TRIS propane pH 9.0	67. (F7)	25% w/v Polyethylene glycol 1,500
68. (F8)	0.02 M Nickel(II) chloride hexahydrate, 0.02 M Magnesium chloride hexahydrate, 0.02 M Cadmium chloride hydrate	68. (F8)	0.1 M Sodium acetate trihydrate pH 4.5	68. (F8)	24% w/v Polyethylene glycol monomethyl ether 2,000
69. (F9)	20% v/v 2-Propanol	69. (F9)	0.1 M MES monohydrate pH 6.0	69. (F9)	20% w/v Polyethylene glycol monomethyl ether 2,000
70. (F10)	0.2 M Ammonium citrate tribasic pH 7.0	70. (F10)	0.1 M Imidazole pH 7.0	70. (F10)	20% w/v Polyethylene glycol monomethyl ether 2,000
71. (F11)	4.0 M Potassium formate	71. (F11)	0.1 M BIS-TRIS propane pH 9.0	71. (F11)	2% v/v Polyethylene glycol monomethyl ether 2,000
72. (F12)	50% v/v Tacsimate pH 4.0	72. (F12)	0.1 M Sodium acetate trihydrate pH 4.5	72. (F12)	1% v/v Polyethylene glycol 3,350
73. (G1)	0.10% w/v n-Octyl-β-D-glucoside	73. (G1)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	73. (G1)	22% w/v Polyethylene glycol 3,350
74. (G2)	2% v/v Tacsimate pH 7.0, 5% v/v 2-Propanol	74. (G2)	0.1 M Imidazole pH 7.0	74. (G2)	8% w/v Polyethylene glycol 3,350
75. (G3)	2% v/v 1,4-Dioxane	75. (G3)	0.1 M Tris pH 8.0	75. (G3)	15% w/v Polyethylene glycol 3,350
76. (G4)	18% v/v 2-Propanol	76. (G4)	0.1 M Sodium citrate tribasic dihydrate pH 5.5	76. (G4)	20% w/v Polyethylene glycol 4,000
77. (G5)	6% v/v Tacsimate pH 6.0	77. (G5)	0.1 M MES monohydrate pH 6.0	77. (G5)	25% w/v Polyethylene glycol 4,000
78. (G6)	0.2 M Magnesium formate dihydrate	78. (G6)	0.1 M Sodium acetate trihydrate pH 4.0	78. (G6)	18% w/v Polyethylene glycol monomethyl ether 5,000
79. (G7)	2% v/v Polyethylene glycol 400	79. (G7)	0.1 M Imidazole pH 7.0	79. (G7)	24% w/v Polyethylene glycol monomethyl ether 5,000
80. (G8)	0.2 M Sodium formate	80. (G8)	0.1 M BICINE pH 8.5	80. (G8)	20% w/v Polyethylene glycol monomethyl ether 5,000
81. (G9)	4% v/v 2-Propanol	81. (G9)	0.1 M BIS-TRIS propane pH 9.0	81. (G9)	20% w/v Polyethylene glycol monomethyl ether 5,000
82. (G10)	6% v/v Ethylene glycol	82. (G10)	0.1 M Citric acid pH 3.5	82. (G10)	10% w/v Polyethylene glycol 6,000
83. (G11)	0.15 M Lithium sulfate monohydrate	83. (G11)	0.1 M Citric acid pH 3.5	83. (G11)	18% w/v Polyethylene glycol 6,000
84. (G12)	10% v/v 2-Propanol	84. (G12)	0.1 M Sodium acetate trihydrate pH 4.0	84. (G12)	22% w/v Polyethylene glycol 6,000
85. (H1)	0.2 M Sodium chloride	85. (H1)	0.1 M Sodium acetate trihydrate pH 4.0	85. (H1)	22% w/v Polyethylene glycol 8,000
86. (H2)	20% v/v 2-Propanol	86. (H2)	0.1 M Tris pH 8.0	86. (H2)	5% w/v Polyethylene glycol 8,000
87. (H3)	10% v/v Polyethylene glycol 200	87. (H3)	0.1 M BIS-TRIS propane pH 9.0	87. (H3)	18% w/v Polyethylene glycol 8,000
88. (H4)	15% v/v 2-Propanol	88. (H4)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	88. (H4)	10% w/v Polyethylene glycol 10,000
89. (H5)	0.4 M Sodium malonate pH 6.0	89. (H5)	0.1 M MES monohydrate pH 6.0	89. (H5)	0.5% w/v Polyethylene glycol 10,000
90. (H6)	0.2 M Potassium sodium tartrate tetrahydrate	90. (H6)	0.1 M BIS-TRIS pH 6.5	90. (H6)	10% w/v Polyethylene glycol 10,000
91. (H7)	5% v/v (+/-)-2-Methyl-2,4-pentanediol	91. (H7)	0.1 M HEPES pH 7.5	91. (H7)	10% w/v Polyethylene glycol 10,000
92. (H8)	0.2 M Ammonium acetate	92. (H8)	0.1 M Tris pH 8.0	92. (H8)	16% w/v Polyethylene glycol 10,000
93. (H9)	5% v/v 2-Propanol	93. (H9)	0.1 M Citric acid pH 3.5	93. (H9)	6% v/v Polyethylene glycol 20,000
94. (H10)	1.0 M Sodium malonate pH 5.0	94. (H10)	0.1 M Sodium acetate trihydrate pH 4.5	94. (H10)	2% v/v Polyethylene glycol 20,000
95. (H11)	0.2 M Magnesium chloride hexahydrate	95. (H11)	0.1 M Sodium citrate tribasic dihydrate pH 5.0	95. (H11)	10% w/v Polyethylene glycol 20,000
96. (H12)	3% w/v Dextran sulfate sodium salt	96. (H12)	0.1 M BICINE pH 8.5	96. (H12)	15% w/v Polyethylene glycol 20,000

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

PEGRx™ 2 (Deep Well Block) contains forty-eight unique reagents beginning at position E1.
To determine the formulation of each reagent, simply read across the page.

HAMPTON
RESEARCH

Solutions for Crystal Growth

34 Journey

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Sample: _____ **Sample Concentration:** _____
Sample Buffer: _____ **Date:** _____
Reservoir Volume: _____ **Temperature:** _____
Drop Volume: Total _____ μ l **Sample** _____ μ l **Reservoir** _____ μ l **Additive** _____ μ l

- 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)

PEGRx HT™ - HR2-086 Scoring Sheet		Date:	Date:	Date:	Date:
1. (A1)	0.1 M Citric acid pH 3.5, 34% v/v Polyethylene glycol 200				
2. (A2)	0.1 M Sodium citrate tribasic dihydrate pH 5.5, 38% v/v Polyethylene glycol 200				
3. (A3)	0.1 M HEPES pH 7.5, 42% v/v Polyethylene glycol 200				
4. (A4)	0.1 M Sodium acetate trihydrate pH 4.5, 30% v/v Polyethylene glycol 300				
5. (A5)	0.1 M BIS-TRIS pH 6.5, 25% v/v Polyethylene glycol 300				
6. (A6)	0.1 M BICINE pH 8.5, 20% v/v Polyethylene glycol 300				
7. (A7)	0.1 M Sodium acetate trihydrate pH 4.0, 15% v/v Polyethylene glycol 400				
8. (A8)	0.1 M MES monohydrate pH 6.0, 22% v/v Polyethylene glycol 400				
9. (A9)	0.1 M Tris pH 8.0, 30% v/v Polyethylene glycol 400				
10. (A10)	0.1 M Sodium citrate tribasic dihydrate pH 5.0, 30% v/v Polyethylene glycol monomethyl ether 550				
11. (A11)	0.1 M Imidazole pH 7.0, 25% v/v Polyethylene glycol monomethyl ether 550				
12. (A12)	0.1 M BIS-TRIS propane pH 9.0, 20% v/v Polyethylene glycol monomethyl ether 550				
13. (B1)	0.1 M Sodium acetate trihydrate pH 4.0, 10% v/v Jeffamine® M-600® pH 7.0				
14. (B2)	0.1 M MES monohydrate pH 6.0, 20% v/v Jeffamine® M-600® pH 7.0				
15. (B3)	0.1 M Tris pH 8.0, 30% v/v Jeffamine® M-600® pH 7.0				
16. (B4)	0.1 M Citric acid pH 3.5, 14% w/v Polyethylene glycol 1,000				
17. (B5)	0.1 M Sodium citrate tribasic dihydrate pH 5.5, 22% w/v Polyethylene glycol 1,000				
18. (B6)	0.1 M HEPES pH 7.5, 30% w/v Polyethylene glycol 1,000				
19. (B7)	0.1 M Sodium acetate trihydrate pH 4.5, 30% w/v Polyethylene glycol 1,500				
20. (B8)	0.1 M BIS-TRIS pH 6.5, 20% w/v Polyethylene glycol 1,500				
21. (B9)	0.1 M BICINE pH 8.5, 15% w/v Polyethylene glycol 1,500				
22. (B10)	0.1 M Sodium acetate trihydrate pH 4.0, 10% w/v Polyethylene glycol monomethyl ether 2,000				
23. (B11)	0.1 M MES monohydrate pH 6.0, 20% w/v Polyethylene glycol monomethyl ether 2,000				
24. (B12)	0.1 M Tris pH 8.0, 30% w/v Polyethylene glycol monomethyl ether 2,000				
25. (C1)	0.1 M Sodium citrate tribasic dihydrate pH 5.0, 30% v/v Jeffamine® ED-2001 pH 7.0				
26. (C2)	0.1 M Imidazole pH 7.0, 20% v/v Jeffamine® ED-2001 pH 7.0				
27. (C3)	0.1 M BIS-TRIS propane pH 9.0, 10% v/v Jeffamine® ED-2001 pH 7.0				
28. (C4)	0.1 M Citric acid pH 3.5, 25% w/v Polyethylene glycol 3,350				
29. (C5)	0.1 M Sodium citrate tribasic dihydrate pH 5.5, 18% w/v Polyethylene glycol 3,350				
30. (C6)	0.1 M HEPES pH 7.5, 12% w/v Polyethylene glycol 3,350				
31. (C7)	0.1 M Sodium acetate trihydrate pH 4.0, 10% w/v Polyethylene glycol 4,000				
32. (C8)	0.1 M MES monohydrate pH 6.0, 14% w/v Polyethylene glycol 4,000				
33. (C9)	0.1 M Tris pH 8.0, 28% w/v Polyethylene glycol 4,000				
34. (C10)	0.1 M Sodium acetate trihydrate pH 4.5, 30% w/v Polyethylene glycol monomethyl ether 5,000				
35. (C11)	0.1 M BIS-TRIS pH 6.5, 20% w/v Polyethylene glycol monomethyl ether 5,000				
36. (C12)	0.1 M BICINE pH 8.5, 8% w/v Polyethylene glycol monomethyl ether 5,000				
37. (D1)	0.1 M Sodium citrate tribasic dihydrate pH 5.0, 10% w/v Polyethylene glycol 6,000				
38. (D2)	0.1 M Imidazole pH 7.0, 20% w/v Polyethylene glycol 6,000				
39. (D3)	0.1 M BIS-TRIS propane pH 9.0, 30% w/v Polyethylene glycol 6,000				
40. (D4)	0.1 M Citric acid pH 3.5, 28% w/v Polyethylene glycol 8,000				
41. (D5)	0.1 M Sodium citrate tribasic dihydrate pH 5.5, 16% w/v Polyethylene glycol 8,000				
42. (D6)	0.1 M HEPES pH 7.5, 4% w/v Polyethylene glycol 8,000				
43. (D7)	0.1 M Sodium acetate trihydrate pH 4.5, 10% w/v Polyethylene glycol 10,000				
44. (D8)	0.1 M BIS-TRIS pH 6.5, 16% w/v Polyethylene glycol 10,000				
45. (D9)	0.1 M BICINE pH 8.5, 20% w/v Polyethylene glycol 10,000				
46. (D10)	0.1 M Sodium citrate tribasic dihydrate pH 5.0, 18% w/v Polyethylene glycol 20,000				
47. (D11)	0.1 M Imidazole pH 7.0, 12% w/v Polyethylene glycol 20,000				
48. (D12)	0.1 M BIS-TRIS propane pH 9.0, 8% w/v Polyethylene glycol 20,000				



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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)

PEGRx HT™ - HR2-086 Scoring Sheet

Date: _____ Date: _____ Date: _____

49. (E1)	0.8 M Lithium sulfate monohydrate, 0.1 M Sodium acetate trihydrate pH 4.0, 4% v/v Polyethylene glycol 200			
50. (E2)	0.2 M Lithium sulfate monohydrate, 0.1 M Sodium citrate tribasic dihydrate pH 5.0, 26% v/v Polyethylene glycol 200			
51. (E3)	0.05 M Calcium chloride dihydrate, 0.1 M MES monohydrate pH 6.0, 45% v/v Polyethylene glycol 200			
52. (E4)	28% v/v 2-Propanol, 0.1 M BIS-TRIS pH 6.5, 3% v/v Polyethylene glycol 200			
53. (E5)	20% v/v Tacsimate pH 7.0, 0.1 M HEPES pH 7.5, 2% v/v Polyethylene glycol 200			
54. (E6)	10% v/v 2-Propanol, 0.1 M Sodium citrate tribasic dihydrate pH 5.0, 26% v/v Polyethylene glycol 400			
55. (E7)	0.2 M Ammonium acetate, 0.1 M Sodium citrate tribasic dihydrate pH 5.5, 24% v/v Polyethylene glycol 400			
56. (E8)	0.2 M Ammonium sulfate, 0.1 M BIS-TRIS pH 6.5, 18% v/v Polyethylene glycol 400			
57. (E9)	0.1 M HEPES pH 7.5, 40% v/v Polyethylene glycol 400			
58. (E10)	6% v/v 2-Propanol, 0.1 M Sodium acetate trihydrate pH 4.5, 26% v/v Polyethylene glycol monomethyl ether 550			
59. (E11)	1.8 M Ammonium sulfate, 0.1 M BIS-TRIS pH 6.5, 2% v/v Polyethylene glycol monomethyl ether 550			
60. (E12)	0.15 M DL-Malic acid pH 7.0, 0.1 M Imidazole pH 7.0, 22% v/v Polyethylene glycol monomethyl ether 550			
61. (F1)	0.1 M Succinic acid pH 7.0, 0.1 M BICINE pH 8.5, 30% w/v Polyethylene glycol monomethyl ether 550			
62. (F2)	0.1 M Lithium sulfate monohydrate, 0.1 M Sodium citrate tribasic dihydrate pH 5.5, 20% w/v Polyethylene glycol 1,000			
63. (F3)	0.1 M Sodium malonate pH 8.0, 0.1 M Tris pH 8.0, 30% w/v Polyethylene glycol 1,000			
64. (F4)	4% v/v (+/-)-2-Methyl-2,4-pentanediol, 0.1 M Citric acid pH 3.5, 20% w/v Polyethylene glycol 1,500			
65. (F5)	0.2 M L-Proline, 0.1 M HEPES pH 7.5, 24% w/v Polyethylene glycol 1,500			
66. (F6)	10% v/v 2-Propanol, 0.1 M BICINE pH 8.5, 30% w/v Polyethylene glycol 1,500			
67. (F7)	0.1 M Sodium chloride, 0.1 M BIS-TRIS propane pH 9.0, 25% w/v Polyethylene glycol 1,500			
68. (F8)	0.02 M Nickel(II) chloride hexahydrate, 0.02 M Magnesium chloride hexahydrate, 0.02 M Cadmium chloride hydrate, 0.1 M Sodium acetate trihydrate pH 4.5, 24% w/v Polyethylene glycol monomethyl ether 2,000			
69. (F9)	20% v/v 2-Propanol, 0.1 M MES monohydrate pH 6.0, 20% w/v Polyethylene glycol monomethyl ether 2,000			
70. (F10)	0.2 M Ammonium citrate tribasic pH 7.0, 0.1 M Imidazole pH 7.0, 20% w/v Polyethylene glycol monomethyl ether 2,000			
71. (F11)	4.0 M Potassium formate, 0.1 M BIS-TRIS propane pH 9.0, 2% w/v Polyethylene glycol monomethyl ether 2,000			
72. (F12)	50% v/v Tacsimate pH 4.0, 0.1 M Sodium acetate trihydrate pH 4.5, 1% w/v Polyethylene glycol 3,350			
73. (G1)	0.10% w/v n-Octyl-β-D-glucoside, 0.1 M Sodium citrate tribasic dihydrate pH 5.5, 22% w/v Polyethylene glycol 3,350			
74. (G2)	2% v/v Tacsimate pH 7.0, 5% v/v 2-Propanol, 0.1 M Imidazole pH 7.0, 8% w/v Polyethylene glycol 3,350			
75. (G3)	2% v/v 1,4-Dioxane, 0.1 M Tris pH 8.0, 15% w/v Polyethylene glycol 3,350			
76. (G4)	18% v/v 2-Propanol, 0.1 M Sodium citrate tribasic dihydrate pH 5.5, 20% w/v Polyethylene glycol 4,000			
77. (G5)	6% v/v Tacsimate pH 6.0, 0.1 M MES monohydrate pH 6.0, 25% w/v Polyethylene glycol 4,000			
78. (G6)	0.2 M Magnesium formate dihydrate, 0.1 M Sodium acetate trihydrate pH 4.0, 18% w/v Polyethylene glycol monomethyl ether 5,000			
79. (G7)	2% v/v Polyethylene glycol 400, 0.1 M Imidazole pH 7.0, 24% w/v Polyethylene glycol monomethyl ether 5,000			
80. (G8)	0.2 M Sodium formate, 0.1 M BICINE pH 8.5, 20% w/v Polyethylene glycol monomethyl ether 5,000			
81. (G9)	4% v/v 2-Propanol, 0.1 M BIS-TRIS propane pH 9.0, 20% w/v Polyethylene glycol monomethyl ether 5,000			
82. (G10)	6% v/v Ethylene glycol, 0.1 M Citric acid pH 3.5, 10% w/v Polyethylene glycol 6,000			
83. (G11)	0.15 M Lithium sulfate monohydrate, 0.1 M Citric acid pH 3.5, 18% w/v Polyethylene glycol 6,000			
84. (G12)	10% v/v 2-Propanol, 0.1 M Sodium acetate trihydrate pH 4.0, 22% w/v Polyethylene glycol 6,000			
85. (H1)	0.2 M Sodium chloride, 0.1 M Sodium acetate trihydrate pH 4.0, 22% w/v Polyethylene glycol 8,000			
86. (H2)	20% v/v 2-Propanol, 0.1 M Tris pH 8.0, 5% w/v Polyethylene glycol 8,000			
87. (H3)	10% v/v Polyethylene glycol 200, 0.1 M BIS-TRIS propane pH 9.0, 18% w/v Polyethylene glycol 8,000			
88. (H4)	15% v/v 2-Propanol, 0.1 M Sodium citrate tribasic dihydrate pH 5.0, 10% w/v Polyethylene glycol 10,000			
89. (H5)	0.4 M Sodium malonate pH 6.0, 0.1 M MES monohydrate pH 6.0, 0.5% w/v Polyethylene glycol 10,000			
90. (H6)	0.2 M Potassium sodium tartrate tetrahydrate, 0.1 M BIS-TRIS pH 6.5, 10% w/v Polyethylene glycol 10,000			
91. (H7)	5% v/v (+/-)-2-Methyl-2,4-pentanediol, 0.1 M HEPES pH 7.5, 10% w/v Polyethylene glycol 10,000			
92. (H8)	0.2 M Ammonium acetate, 0.1 M Tris pH 8.0, 16% w/v Polyethylene glycol 10,000			
93. (H9)	5% v/v 2-Propanol, 0.1 M Citric acid pH 3.5, 6% w/v Polyethylene glycol 20,000			
94. (H10)	1.0 M Sodium malonate pH 5.0, 0.1 M Sodium acetate trihydrate pH 4.5, 2% w/v Polyethylene glycol 20,000			
95. (H11)	0.2 M Magnesium chloride hexahydrate, 0.1 M Sodium citrate tribasic dihydrate pH 5.0, 10% w/v Polyethylene glycol 20,000			
96. (H12)	3% w/v Dextran sulfate sodium salt, 0.1 M BICINE pH 8.5, 15% w/v Polyethylene glycol 20,000			