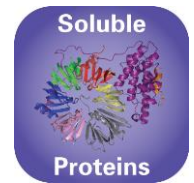
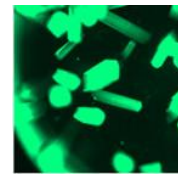


Molecular  
Dimensions



## PACT *premier*<sup>™</sup> HT-96 Green Screen

## MD1-52

PACT *premier* is a pH, Anion, Cation crystallization trial devised to test pH within a PEG/Ion screen environment with fluorescent dye added for superb UV performance.

MD1-52 contains 96 x 1 mL reagents in a deep-well block.

### Features of PACT *premier*<sup>™</sup> green screen:

- Optimized sparse matrix screen with non-covalent fluorescent dye added.
- Increased crystal contrast- observe protein crystals <30 $\mu$ M and also those lacking intrinsic fluorescence
- A modern, comprehensive PEG/ion screen ready-to-use with fluorescent dye added for superb UV performance.
- This 96-well screen is really 3 screens in one:
  - 24-well pH/PEG screen
  - 24-well cation/PEG screen
  - 48-well anion/PEG screen

### Rationale for a new PEG/Ion screen

The first step in crystallization is often to reach for a commercially available “sparse matrix” kit, and hope that one of the conditions produces something that looks harvestable, or optimizable. If no obvious leads come out of the screen, it is hard to learn anything from the negative (precipitate and clear) results.

There are a few screens that try to test crystallization space in a more rational manner – for example, the Clear Strategy<sup>™</sup> Screen and The Solubility Tool Kit.

Traditional PEG/Ion screens provide a logical test of seven cations and eleven anions using PEG 3350 as the precipitation agent. However, the user has no control over pH and hence cannot determine the effect of one cation or anion over another.

For this reason the PEG/ION/pH (PACT) screen has been developed to systematically test the effect of pH, anions and cations, using PEG as the precipitant. This screen has been implemented very successfully at the Netherlands Cancer Institute (NKI), and at the Oxford Protein Production Facility (OPPF).

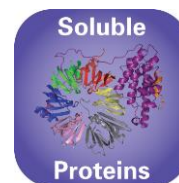
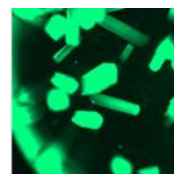
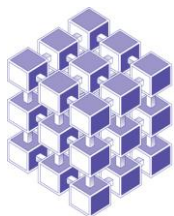
### PACT *premier*<sup>™</sup>

#### pH/PEG screen

This consists of four broad range buffer systems (1) versus PEG 1500. These buffers allow one to scan the pH range from 4 to 9, without changing the chemistry of the system, so effectively isolating the effect of pH from the effect of the buffer that causes the change in pH.



Protein crystals grown successfully with PACT *premier*<sup>™</sup>.



## **PACT premier™**

### **Cation/PEG screen**

This is made up of six cations (all with chloride counter ions) that are combined with PEG 6000 at four different pHs: Acetate pH 5.0, MES pH 6.0, HEPES pH 7.0 and Tris pH 8.0. The cations tested are Na<sup>+</sup>, NH<sub>4</sub><sup>+</sup>, Li<sup>+</sup>, Mg<sup>2+</sup>, Ca<sup>2+</sup> and Zn<sup>2+</sup>. The zinc ion is tested at lower concentration than the other cations in the screen (0.01 M vs. 0.2 M)

### **Anion/PEG screen**

This is made up of 12 anions, with either sodium or potassium counter ions, which are tested at 0.2 M against PEG 3350. The anions include fluoride, bromine, iodide, thiocyanate, nitrate, formate, acetate, sulfate, tartrate, phosphate, citrate and malonate. The phosphate solution is tested at a concentration of 0.02 M. Chloride is not included here as it is the counter ion in the cation screen. Three sets of reagents are tested at pH 6.5, 7.5, and 8.5 with the Bis-Tris propane buffer system whilst one set of reagents is tested without buffering.

### **References:**

(1)Newman *et al* (2005). Towards rationalization of crystallization screening for small- to medium-sized academic laboratories: the PACT/JCSG+ strategy. *Acta Cryst.* **D61**, 1426-1431.

### **Formulation Notes**

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

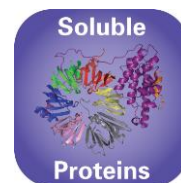
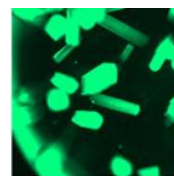
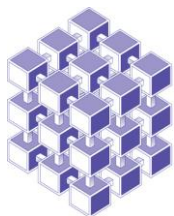
Final pH may vary from that specified on the datasheet. Molecular Dimensions will be happy to discuss the precise formulation of individual reagents.

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

Enquiries regarding **PACT premier™** 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](http://www.moleculardimensions.com)

PACT premier™, when used together with JCSG-plus™ as a primary screening strategy, is an extremely powerful and successful combination (i.e. a combination of a modern sparse matrix approach and an information yielding systematic trial).



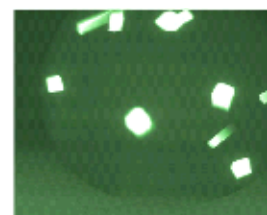
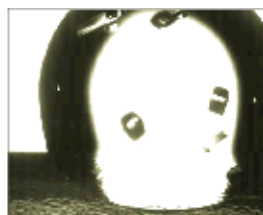
### Rationale behind Green Screens

In recent years UV fluorescence imaging has become an established method for protein/salt crystal discrimination. Whilst the majority of proteins contain at least one tryptophan residue there remain a number that fluoresce weakly or not at all, yielding a false negative.

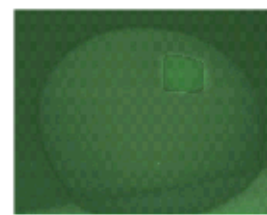
**Green Screens** contain a non-covalent fluorescent dye (1) which conveys fluorescence on most proteins when illuminated with UV light. This not only increases the signal-to-noise ratio (important for very small crystals), but also allows the identification of protein crystals lacking intrinsic fluorescence that would otherwise remain ambiguous. Green screens are available in three of our most successful screens, **PACT premier™**, **JCSG-plus™** and **MemGold™**. Use these screens together with UV transparent plates (MD11-00U-100) and ClearVue sheets (MD6-01S) for optimum UV performance.

### Reference:

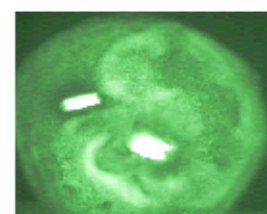
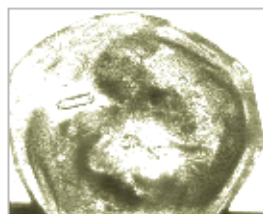
Groves et al (2007). A method for the general identification of protein crystals in crystallization experiments using a non-covalent fluorescent dye. *Acta Cryst.* **D63**, 526-535.



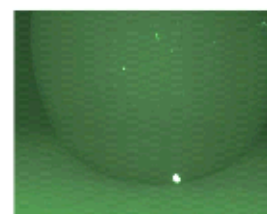
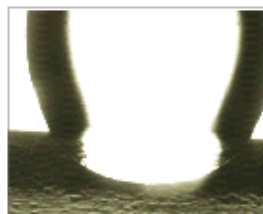
Protein



Salt



Protein

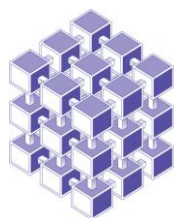


Microcrystal Protein

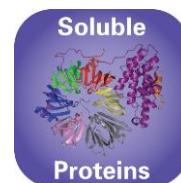
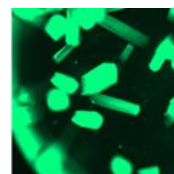
Images kindly provided by Matthew Groves, EMBL Outstation- Hamburg.

### Features of the Green Screens:

- **Non-covalent** – binds in channels and not observed to affect crystallization or diffraction quality in any of the proteins tested\*.
- **Increased crystal contrast** – observe protein crystals <30  $\mu$ M and also those lacking significant intrinsic fluorescence.
- **Improved signal-to-noise** – on average quantum yield ratio of fluorescence is increased from 0.2 to 0.7.
- **Easy-to-use** – available in three proven to be successful screens - PACT premier™, JCSG-plus™ and MemGold™.
- **Standard format** – for all automated systems – 1 mL HT-96 block (10 mL kits available on request).



Molecular  
Dimensions



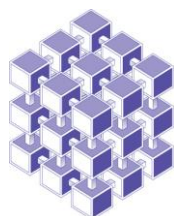
**PACT premier™ HT-96 Green Screen**

**Conditions A1 – D12**

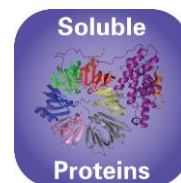
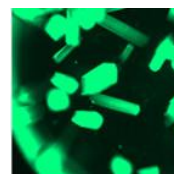
**MD1-52**

Well #	Conc.	Salt	Conc.	Buffer	pH	Conc.	Precipitant	Dye concentration
A1			0.1 M	SPG	4.0	25 % w/v	PEG 1500	100µM 1,8-ANS
A2			0.1 M	SPG	5.0	25 % w/v	PEG 1500	100µM 1,8-ANS
A3			0.1 M	SPG	6.0	25 % w/v	PEG 1500	100µM 1,8-ANS
A4			0.1 M	SPG	7.0	25 % w/v	PEG 1500	100µM 1,8-ANS
A5			0.1 M	SPG	8.0	25 % w/v	PEG 1500	100µM 1,8-ANS
A6			0.1 M	SPG	9.0	25 % w/v	PEG 1500	100µM 1,8-ANS
A7	0.2 M	Sodium chloride	0.1 M	Sodium acetate	5.0	20 % w/v	PEG 6000	100µM 1,8-ANS
A8	0.2 M	Ammonium chloride	0.1 M	Sodium acetate	5.0	20 % w/v	PEG 6000	100µM 1,8-ANS
A9	0.2 M	Lithium chloride	0.1 M	Sodium acetate	5.0	20 % w/v	PEG 6000	100µM 1,8-ANS
A10	0.2 M	Magnesium chloride hexahydrate	0.1 M	Sodium acetate	5.0	20 % w/v	PEG 6000	100µM 1,8-ANS
A11	0.2 M	Calcium chloride dihydrate	0.1 M	Sodium acetate	5.0	20 % w/v	PEG 6000	100µM 1,8-ANS
A12	0.01 M	Zinc chloride	0.1 M	Sodium acetate	5.0	20 % w/v	PEG 6000	100µM 1,8-ANS
B1			0.1 M	MIB	4.0	25 % w/v	PEG 1500	100µM 1,8-ANS
B2			0.1 M	MIB	5.0	25 % w/v	PEG 1500	100µM 1,8-ANS
B3			0.1 M	MIB	6.0	25 % w/v	PEG 1500	100µM 1,8-ANS
B4			0.1 M	MIB	7.0	25 % w/v	PEG 1500	100µM 1,8-ANS
B5			0.1 M	MIB	8.0	25 % w/v	PEG 1500	100µM 1,8-ANS
B6			0.1 M	MIB	9.0	25 % w/v	PEG 1500	100µM 1,8-ANS
B7	0.2 M	Sodium chloride	0.1 M	MES	6.0	20 % w/v	PEG 6000	100µM 1,8-ANS
B8	0.2 M	Ammonium chloride	0.1 M	MES	6.0	20 % w/v	PEG 6000	100µM 1,8-ANS
B9	0.2 M	Lithium chloride	0.1 M	MES	6.0	20 % w/v	PEG 6000	100µM 1,8-ANS
B10	0.2 M	Magnesium chloride hexahydrate	0.1 M	MES	6.0	20 % w/v	PEG 6000	100µM 1,8-ANS
B11	0.2 M	Calcium chloride dihydrate	0.1 M	MES	6.0	20 % w/v	PEG 6000	100µM 1,8-ANS
B12	0.01 M	Zinc chloride	0.1 M	MES	6.0	20 % w/v	PEG 6000	100µM 1,8-ANS
C1			0.1 M	PCTP	4.0	25 % w/v	PEG 1500	100µM 1,8-ANS
C2			0.1 M	PCTP	5.0	25 % w/v	PEG 1500	100µM 1,8-ANS
C3			0.1 M	PCTP	6.0	25 % w/v	PEG 1500	100µM 1,8-ANS
C4			0.1 M	PCTP	7.0	25 % w/v	PEG 1500	100µM 1,8-ANS
C5			0.1 M	PCTP	8.0	25 % w/v	PEG 1500	100µM 1,8-ANS
C6			0.1 M	PCTP	9.0	25 % w/v	PEG 1500	100µM 1,8-ANS
C7	0.2 M	Sodium chloride	0.1 M	HEPES	7.0	20 % w/v	PEG 6000	100µM 1,8-ANS
C8	0.2 M	Ammonium chloride	0.1 M	HEPES	7.0	20 % w/v	PEG 6000	100µM 1,8-ANS
C9	0.2 M	Lithium chloride	0.1 M	HEPES	7.0	20 % w/v	PEG 6000	100µM 1,8-ANS
C10	0.2 M	Magnesium chloride hexahydrate	0.1 M	HEPES	7.0	20 % w/v	PEG 6000	100µM 1,8-ANS
C11	0.2 M	Calcium chloride dihydrate	0.1 M	HEPES	7.0	20 % w/v	PEG 6000	100µM 1,8-ANS
C12	0.01 M	Zinc chloride	0.1 M	HEPES	7.0	20 % w/v	PEG 6000	100µM 1,8-ANS
D1			0.1 M	MMT	4.0	25 % w/v	PEG 1500	100µM 1,8-ANS
D2			0.1 M	MMT	5.0	25 % w/v	PEG 1500	100µM 1,8-ANS
D3			0.1 M	MMT	6.0	25 % w/v	PEG 1500	100µM 1,8-ANS
D4			0.1 M	MMT	7.0	25 % w/v	PEG 1500	100µM 1,8-ANS
D5			0.1 M	MMT	8.0	25 % w/v	PEG 1500	100µM 1,8-ANS
D6			0.1 M	MMT	9.0	25 % w/v	PEG 1500	100µM 1,8-ANS
D7	0.2 M	Sodium chloride	0.1 M	Tris	8.0	20 % w/v	PEG 6000	100µM 1,8-ANS
D8	0.2 M	Ammonium chloride	0.1 M	Tris	8.0	20 % w/v	PEG 6000	100µM 1,8-ANS
D9	0.2 M	Lithium chloride	0.1 M	Tris	8.0	20 % w/v	PEG 6000	100µM 1,8-ANS
D10	0.2 M	Magnesium chloride hexahydrate	0.1 M	Tris	8.0	20 % w/v	PEG 6000	100µM 1,8-ANS
D11	0.2 M	Calcium chloride dihydrate	0.1 M	Tris	8.0	20 % w/v	PEG 6000	100µM 1,8-ANS
D12	0.002 M	Zinc chloride	0.1 M	Tris	8.0	20 % w/v	PEG 6000	100µM 1,8-ANS





Molecular  
Dimensions

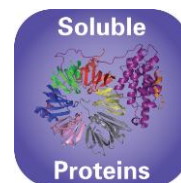
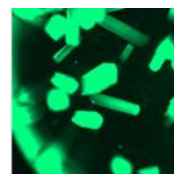
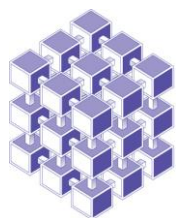


**PACT premier™ HT-96 Green Screen**

**Conditions E1 – H12**

**MD1-52**

Well #	Conc.	Salt	Conc.	Buffer	pH	Conc.	Precipitant	Dye concentration
E1	0.2 M	Sodium fluoride				20 % w/v	PEG 3350	100µM 1,8-ANS
E2	0.2 M	Sodium bromide				20 % w/v	PEG 3350	100µM 1,8-ANS
E3	0.2 M	Sodium iodide				20 % w/v	PEG 3350	100µM 1,8-ANS
E4	0.2 M	Potassium thiocyanate				20 % w/v	PEG 3350	100µM 1,8-ANS
E5	0.2 M	Sodium nitrate				20 % w/v	PEG 3350	100µM 1,8-ANS
E6	0.2 M	Sodium formate				20 % w/v	PEG 3350	100µM 1,8-ANS
E7	0.2 M	Sodium acetate trihydrate				20 % w/v	PEG 3350	100µM 1,8-ANS
E8	0.2 M	Sodium sulfate				20 % w/v	PEG 3350	100µM 1,8-ANS
E9	0.2 M	Potassium sodium tartrate tetrahydrate				20 % w/v	PEG 3350	100µM 1,8-ANS
E10	0.02 M	Sodium/potassium phosphate				20 % w/v	PEG 3350	100µM 1,8-ANS
E11	0.2 M	Sodium citrate tribasic dihydrate				20 % w/v	PEG 3350	100µM 1,8-ANS
E12	0.2 M	Sodium malonate dibasic monohydrate				20 % w/v	PEG 3350	100µM 1,8-ANS
F1	0.2 M	Sodium fluoride	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F2	0.2 M	Sodium bromide	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F3	0.2 M	Sodium iodide	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F4	0.2 M	Potassium thiocyanate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F5	0.2 M	Sodium nitrate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F6	0.2 M	Sodium formate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F7	0.2 M	Sodium acetate trihydrate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F8	0.2 M	Sodium sulfate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F9	0.2 M	Potassium sodium tartrate tetrahydrate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F10	0.02 M	Sodium/potassium phosphate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F11	0.2 M	Sodium citrate tribasic dihydrate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
F12	0.2 M	Sodium malonate dibasic monohydrate	0.1 M	Bis-Tris propane	6.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G1	0.2 M	Sodium fluoride	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G2	0.2 M	Sodium bromide	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G3	0.2 M	Sodium iodide	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G4	0.2 M	Potassium thiocyanate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G5	0.2 M	Sodium nitrate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G6	0.2 M	Sodium formate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G7	0.2 M	Sodium acetate trihydrate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G8	0.2 M	Sodium sulfate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G9	0.2 M	Potassium sodium tartrate tetrahydrate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G10	0.02 M	Sodium/potassium phosphate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G11	0.2 M	Sodium citrate tribasic dihydrate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
G12	0.2 M	Sodium malonate dibasic monohydrate	0.1 M	Bis-Tris propane	7.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H1	0.2 M	Sodium fluoride	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H2	0.2 M	Sodium bromide	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H3	0.2 M	Sodium iodide	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H4	0.2 M	Potassium thiocyanate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H5	0.2 M	Sodium nitrate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H6	0.2 M	Sodium formate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H7	0.2 M	Sodium acetate trihydrate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H8	0.2 M	Sodium sulfate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H9	0.2 M	Potassium sodium tartrate tetrahydrate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H10	0.02 M	Sodium/potassium phosphate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H11	0.2 M	Sodium citrate tribasic dihydrate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS
H12	0.2 M	Sodium malonate dibasic monohydrate	0.1 M	Bis-Tris propane	8.5	20 % w/v	PEG 3350	100µM 1,8-ANS



**Abbreviations:**

**HEPES**; N-(2-hydroxyethyl)-piperazine-N'-2-ethanesulfonic acid, **MES**; 2-(N-morpholino)ethanesulfonic acid, **PEG**; Polyethylene glycol, **Tris**; 2-Amino-2-(hydroxymethyl)propane-1,3-diol, **SPG buffer**; Succinic Acid, sodium phosphate monobasic monohydrate, Glycine, **MIB buffer**; Sodium malonate dibasic monohydrate, Imidazole, Boric acid, **PCTP buffer**; Sodium propionate, Sodium cacodylate trihydrate, Bis-Tris propane, **MMT buffer**; DL-Malic acid, MES monohydrate, Tris.

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



**Re-Ordering details:**

Catalogue Description	Pack size	Catalogue Code
PACT <i>premier</i> <sup>™</sup>	96 x 10 mL	MD1-29
PACT <i>premier</i> <sup>™</sup> HT-96	96 x 1 mL	MD1-36
PACT <i>premier</i> <sup>™</sup> FX-96	96 x 100 µL	MD1-36-FX
<b>Eco Screens</b>		
PACT <i>premier</i> <sup>™</sup> Eco Screen	96 x 10 mL	MD1-29-ECO
PACT <i>premier</i> <sup>™</sup> HT-96 Eco Screen	96 x 1 mL	MD1-36-ECO
<b>Green Screens</b> (contain fluorescent green dye - ideal for UV)		
PACT <i>premier</i> <sup>™</sup> Green Screen	96 x 10 mL	MD1-55
PACT <i>premier</i> <sup>™</sup> HT-96 Green Screen	96 x 1 mL	MD1-52
<b>Combo Packs</b>		
Super2 Combo Value Pack (JCSG- <i>plus</i> <sup>™</sup> + PACT <i>premier</i> <sup>™</sup> )	2 x 96 x 10 mL	MD1-75
Super2 Combo HT-96 Value Pack (JCSG- <i>plus</i> <sup>™</sup> HT-96 + PACT <i>premier</i> <sup>™</sup> HT-96)	2 x 96 x 10 mL	MD1-75-HT
<b>Single Reagents</b>		
PACT <i>premier</i> <sup>™</sup> single reagents	100 mL	MDSR-29-tube number
PACT <i>premier</i> <sup>™</sup> HT-96 single reagents	100 mL	MDSR-36-well number

For PACT *premier*<sup>™</sup> stock solutions please visit the Optimization section on our website.