

# Glutathione S-transferase (GST) Gene Fusion System

The Glutathione S-transferase (GST) Gene Fusion System from GE Healthcare is a versatile system for the expression, purification, and detection of GST-tagged proteins produced in *E. coli*. The system consists of three major components: pGEX plasmid expression vectors, products for GST purification, and a variety of GST detection products. A series of site-specific proteases for cleavage of the GST tag complements the system. The GST affinity tag permits a mild purification process that does not affect a protein's native structure and function.

## GST Gene Fusion System benefits include:

- All pGEX vectors offer a *tac* promoter for chemically inducible, high-level expression
- Mild, nondenaturing buffer compositions for isolation of active proteins
- Affinity chromatography products based on Glutathione Sepharose™ media for one-step purification of samples from low microgram to gram scale
- Convenient prepacked formats suitable for single samples or parallel screening of multiple cloning constructs
- PreScission™ Protease, Thrombin, or Factor Xa recognition sites on pGEX vectors for cleaving the target protein from the fusion product
- Easy detection of fusion protein using Anti-GST Antibody

GST occurs naturally as an M<sub>r</sub> 26 000 protein, which can be expressed in *E. coli* with full enzymatic activity. The crystal structure of recombinant *Schistosoma japonicum* GST from pGEX vectors has been determined and matches that of the native protein. The pGEX plasmid vectors are designed for inducible, high-level intracellular expression of genes or gene fragments as fusions with *S. japonicum* GST.



**Fig 1.** Glutathione Sepharose High Performance, Glutathione Sepharose 4 Fast Flow, and Glutathione Sepharose 4B are all available as bulk media and in prepacked columns and multiwell plates for purification of GST-tagged proteins.

Recombinant proteins are easily purified from, for example *E. coli* cell lysates by affinity chromatography using Glutathione Sepharose media (Fig 1), either with prepacked columns or by batch purification.

Cleavage of the target protein from GST is achieved using a site-specific protease, which possesses a recognition sequence located immediately upstream from the multiple cloning site on the pGEX vectors. Cleavage of the GST tag is performed on-column as a part of the purification protocol or off-line after purification. Recombinant proteins can be detected using an immunoassay provided in the GST Detection Module, Western blotting with Anti-GST Antibody, or by a colorimetric assay.

## pGEX vectors

GST-tagged proteins are constructed by inserting a gene or gene fragment into the multiple cloning site of one of the pGEX vectors. The vectors provide all three translational reading frames beginning with the EcoRI restriction site (see Table 1).



The pGEX vectors are designed for inducible, high-level intracellular expression of genes or gene fragments. Expression in *E. coli* yields tagged proteins with the GST moiety at the amino terminus and the protein of interest at the carboxyl terminus. Thirteen pGEX vectors are available (see Fig 2); all of them have a *tac* promoter for chemically inducible, high-level expression and an internal *laq1<sup>a</sup>* gene for use in any *E. coli* host.

**Table 1.** Protease cleavage sites of pGEX vectors

Vector	Cleavage enzyme
pGEX-6P-1, pGEX-6P-2, pGEX-6P-3	PreScission Protease
pGEX-4T-1, pGEX-4T-2, pGEX-4T-3	Thrombin
pGEX-5X-1, pGEX-5X-2, pGEX-5X-3	Factor Xa
pGEX-2TK	Thrombin

Nine of the vectors have an expanded multiple cloning site (MCS) that contains six restriction sites. The expanded MCS facilitates the unidirectional cloning of cDNA inserts obtained from libraries constructed using many available lambda vectors. pGEX-6P-1, pGEX-6P-2, and pGEX-6P-3 each encode the recognition sequence for site-specific cleavage by PreScission Protease between the GST domain and the multiple cloning site. pGEX-4T-1, pGEX-4T-2, and pGEX-4T-3 are derived from pGEX-2TK and contain a Thrombin recognition site. pGEX-5X-1, pGEX-5X-2, and pGEX-5X-3 are derivatives of pGEX-3X and possess a Factor Xa recognition site (see Table 1).

pGEX-2TK is uniquely designed to allow the detection of expressed proteins by directly labeling the fusion products *in vitro*. This vector contains the recognition sequence for the catalytic subunit of cAMP-dependent protein kinase obtained from heart muscle. The protein kinase site is located between the GST domain and the MCS. Expressed proteins can be directly labeled using protein kinase and [ $\gamma$ - $^{32}$ P]ATP and readily detected using standard radiometric or autoradiographic techniques. pGEX-2TK is a derivative of pGEX-2T; its fusion proteins can be cleaved with Thrombin. Collectively, the pGEX vectors provide all three translational reading frames beginning with the EcoRI restriction site. pGEX-1 $\lambda$ T, pGEX-6P-1, pGEX-4T-1, and pGEX-5X-1 can directly accept and express cDNA inserts isolated from  $\lambda$  gt11 libraries.

To complement the pGEX vectors, GST Vector Primers for Sequencing are available for immediate use in sequencing double-stranded DNA inserted into the pGEX vectors.

A wide variety of *E. coli* host strains can be used for cloning and expression with the pGEX vectors. A lyophilized protease-deficient *E. coli* host strain for optimal expression of recombinant protein, *E. coli* BL21, is available separately.

## GST-tagged protein purification and screening

GST-tagged proteins are easily purified from, for example bacterial lysates by affinity chromatography using glutathione immobilized to a matrix. The high specificity between GST and glutathione ensures that high purity is obtained in a single step. Elution is performed under mild, nondenaturing conditions so that protein antigenicity and function is preserved. A variety of affinity chromatography products are available from GE Healthcare. Glutathione Sepharose media (Table 2) are available in several formats ranging from prefilled MultiTrap™ 96-well filter plates to prepacked, SpinTrap™, GraviTrap™, HiTrap™, and HiPrep™



**Fig 2.** Map of the glutathione S-transferase fusion vectors showing the reading frames and main features. All thirteen vectors have stop codons in all three frames downstream from the multiple cloning site (not depicted in this map).

columns or Lab packs (media packs in sizes from 25 ml to 500 ml). The different formats provide options of purification of sample from low microgram scale to gram quantities.

**Table 2.** Characteristics of Glutathione Sepharose media

Characteristics	Glutathione Sepharose High Performance	Glutathione Sepharose 4 Fast Flow	Glutathione Sepharose 4B
Matrix	Highly cross-linked, 6% agarose	Highly cross-linked, 4% agarose	4% agarose
Average particle size	34 $\mu\text{m}$	90 $\mu\text{m}$	90 $\mu\text{m}$
Binding capacity <sup>1</sup>	> 10 mg	> 10 mg	> 10 mg
Recommended flow rate <sup>2</sup>	< 150 cm/h	50–300 cm/h	< 75 cm/h

<sup>1</sup> Binding of recombinant glutathione S-transferase/ml medium. The binding of GST-tagged proteins depends on size, conformation, and concentration of the protein in the sample loaded. Binding of GST to glutathione is also flow-dependent, and lower flow rates often increase the binding capacity. This is important during sample loading, Protein characteristics, pH, and temperature, but also the medium used can affect the binding capacity.

<sup>2</sup> H<sub>2</sub>O at room temperature.

The choice of equipment will depend on the specific purification. Elution is performed in a benchtop centrifuge when using microspin columns, manually using gravity-flow columns, or by step-gradient elution using a peristaltic pump or syringe in combination with prepacked GSTrap columns. GST MultiTrap 96-well plates are designed for high-throughput screening of small volumes and can purify up to approximately 500  $\mu\text{g}$  of GST-tagged protein per well. GST SpinTrap columns can purify up to 500  $\mu\text{g}$  per column. For purification of larger quantities, prepacked columns such as GST GraviTrap, GSTrap, and GSTPrep 16/10 provided excellent formats.

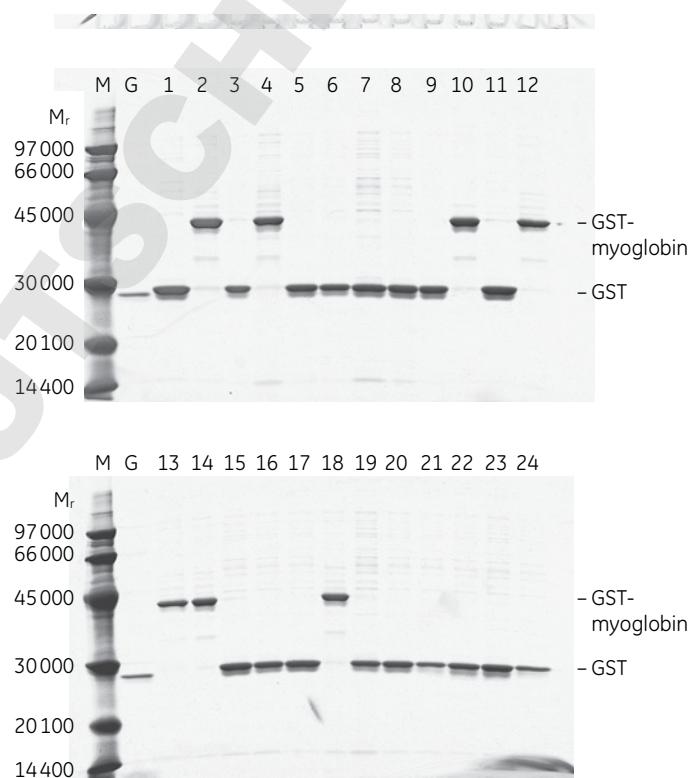
### GST SpinTrap columns

GST SpinTrap columns are excellent for screening of expression levels, and purification conditions prior to scaling up. The columns are designed for use in a microcentrifuge (Fig 3). Each column contains 50  $\mu\text{l}$  of Glutathione Sepharose 4B, enough for purifying up to 500  $\mu\text{g}$  of recombinant GST. The columns are pre-equilibrated in 1x phosphate buffered saline (PBS) with 0.05% Kathon™ (an antibacterial preservative). Each package contains 50 columns.



**Fig 3.** GST SpinTrap columns are designed for efficient, small-scale purification of GST-tagged proteins using a microcentrifuge.

*E. coli* transformants containing cDNA expressing a GST-tagged human myoglobin were randomly selected, expressed, and purified using GST SpinTrap columns. A human myoglobin cDNA was ligated to linearized pGEX-5X-1 and used to transform in *E. coli* BL21 cells. Twenty-four randomly selected colonies were used to inoculate 3 ml cultures, which were grown overnight. Expression was induced with isopropyl  $\beta$ -D-thiogalactopyranoside (IPTG) for 2 h. Lysates were prepared from 1.5 ml aliquots of each culture by a freeze-thaw procedure and applied to GST SpinTrap columns. Aliquots of each reduced glutathione eluate were applied on an SDS gel for analysis by SDS-PAGE (Fig 4). The results showed that 7 of the 24 transformants expressed the GST-tagged myoglobin.



**Fig 4.** SDS-PAGE analysis of eluates from a screening of 24 randomly selected *E. coli* transformants containing cDNA expressing GST-tagged human myoglobin. M = LMW-SDS Marker Kit. Lanes 1–24 contain products eluted from the GST SpinTrap columns using reduced glutathione.

### GST GraviTrap columns

GST GraviTrap provides convenient, disposable columns prepacked with 2 ml of Glutathione Sepharose 4B, sufficient for purification of up to 20 mg of GST-tagged protein. Each package contains 10 prepacked columns manufactured from biocompatible polypropylene (Fig 5). Special frits protect the medium from running dry during purification. GST GraviTrap columns are delivered in a package that converts conveniently into a column stand (Workmate). The plastic tray in the product package can be used to collect liquid waste. When handling volumes above 10 ml, connecting Labmate™ PD-10 Buffer Reservoir to the column increases the loading capacity to approx. 35 ml.



**Fig 5.** GST GraviTrap together with Workmate (column stand) for increased convenience (left) and GST Bulk Kit (right) are two options available for gravity-flow purification of GST-tagged proteins.

### GST Bulk Kit

GST Bulk Kit contains a 10 ml bulk pack of Glutathione Sepharose 4B and five disposable columns. With this kit, GST-tagged proteins can be purified using either column chromatography or a batch method. GST Bulk Kit contains sufficient reagents for purification of up to 50 mg of GST-tagged protein.

### GST MultiTrap 96-well filter plates

GST MultiTrap 96-well filter plates are available in two options: GST MultiTrap FF and GST MultiTrap 4B, prepacked with Glutathione Sepharose 4 Fast Flow and Glutathione Sepharose 4B, respectively. Both products provide highly reproducible, high-throughput screening and rapid, small-scale purification of GST-tagged proteins from unclarified or clarified samples (Fig 6). Typical applications include expression screening of different constructs, screening for solubility of proteins, and optimization of the conditions for small-scale parallel purification. Purification of up to 500 µg of GST-tagged proteins/well directly from unclarified cell lysate is achieved using GST MultiTrap, which shortens handling time and minimizes degradation of sensitive target proteins. The 96-well plate format gives great flexibility, both when working with automated robotic systems and manually using centrifugation or vacuum. Consistent well-to-well and plate-to-plate performance ensures high reproducibility.

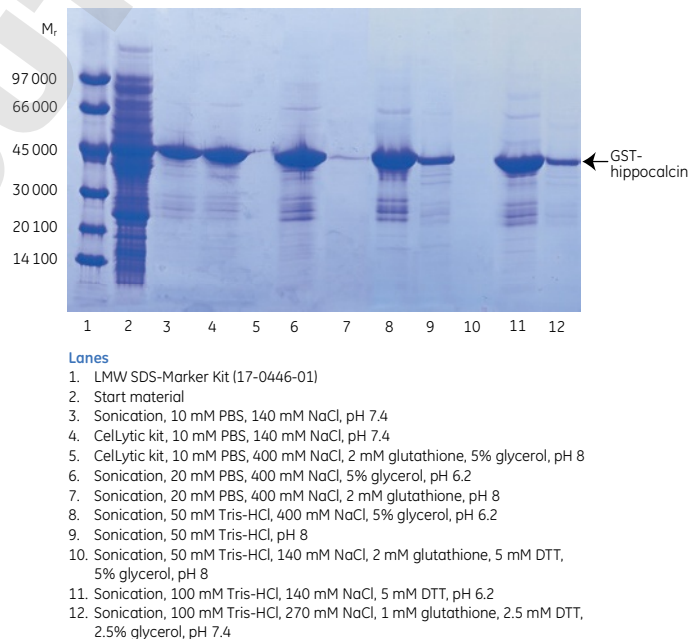


**Fig 6.** GST MultiTrap 96-well filter plates are available in two options; GST MultiTrap FF and GST MultiTrap 4B.

A buffer-screening study for determination of optimal buffer conditions for purification of GST-hippocalcin using GST MultiTrap FF was designed. The parameters tested were buffer, pH, sodium chloride, glycerol, DTT, and glutathione. A comparison between sonication and use of a commercial cell lysis kit was also performed. Factorial design (design-of-experiments) and statistical analysis were performed using MODDE™ software (Umetrics). The different buffer conditions and sample preparation methods were applied randomly on the filter plate.

The presence of glutathione in sample, binding buffer, or wash buffer decreased the yield of purified GST-hippocalcin significantly, while the buffer had no effect on yield. Low pH improved yield while pH and additives such as DTT and glycerol, or sodium chloride, did not affect purity significantly (Fig 7).

The screening results showed that buffer conditions for purifying GST-hippocalcin with highest yield and purity were between 10 and 20 mM sodium phosphate, 140 to 400 mM NaCl, pH 6.2 to 7.4. Sample preparation could be performed with both a commercial cell lysis kit and sonication without significantly affecting the purification result.



**Fig 7.** SDS-PAGE (reducing conditions, ExcelGel™ SDS Gradient 8–18; Coomassie™ staining) of collected fractions of eluted GST-hippocalcin from some of the GST MultiTrap FF filter plate wells.

### GST Buffer Kit

GST Buffer Kit contains stock solutions of binding and elution buffers for purification of GST-tagged proteins. The kit eliminates time-consuming buffer preparation and promotes fast, reproducible, and convenient purification work. Sufficient reagents are supplied to purify up to 20 mg of GST-tagged protein. GST Buffer Kit contains 10× PBS, reduced glutathione, dilution buffer, and an instruction booklet.

## Protein purification scale-up

Purification protocols can easily be scaled-up; Glutathione Sepharose 4 Fast Flow, Glutathione Sepharose 4B, and Glutathione Sepharose High Performance are all available in larger prepacked columns, and as bulk media. Addition of a second gel filtration step is recommended to polish the target protein and remove possible aggregates.

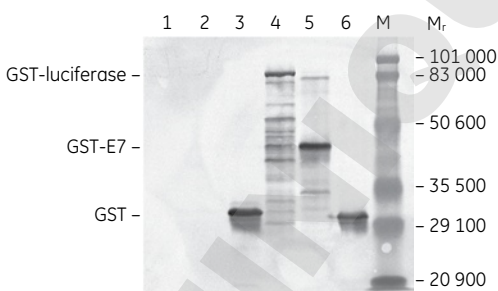
GSTrap affinity columns are 1 ml and 5 ml prepacked HiTrap columns for purification of GST-tagged proteins in high milligram quantities. GSTrap columns are available packed with all three different Glutathione Sepharose, media—GSTrap FF, GSTrap 4B, and GSTrap HP. Sample application, washing, and elution can be performed using a syringe with a supplied connector, a peristaltic pump, or a liquid chromatography system such as ÄKTA™ design.

GSTPrep FF 16/10 is a 20 ml HiPrep column prepacked with Glutathione Sepharose 4 Fast Flow, and can be used for purification of milligram to gram quantities of GST-tagged protein.

## Detection of GST

### Anti-GST Antibody

Anti-GST Antibody is a polyclonal antibody purified from the sera of goats for highly sensitive and specific detection of recombinant GST-tagged proteins. The strength of a polyclonal antibody is that it can recognize different GST epitopes, so that GST-tagged proteins are detected even if some binding sites are masked due to protein folding. The Anti-GST Antibody is extensively cross-adsorbed to remove antibodies that bind to *E. coli* proteins, and affinity purified using Glutathione Sepharose chromatography media. Anti-GST Antibody is supplied unconjugated for use with any enzyme-conjugated anti-goat antibody, and is recommended for use in Western blots and dot blots (Fig 8).



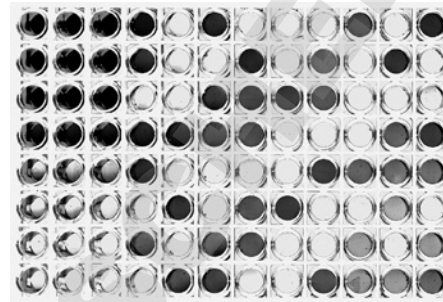
#### Lanes

- 1-2. Sonicate of *E. coli* TG1 and KL45 cells, respectively
3. Sonicate of induced pGEX-5X-1 containing cells
4. Sonicate of induced pGEX-5X-luciferase containing cells (expressing GST-luciferase recombinant protein)
5. Sonicate of induced pGEX-4T-E7 containing cells (expressing GST-E7 recombinant protein)
6. Purified GST
- M. Prestained molecular weight marker

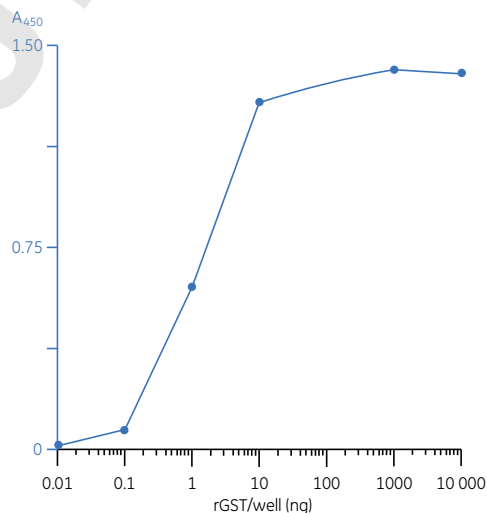
**Fig 8.** Western blot of *E. coli* lysates containing GST-tagged proteins. For detection, Anti-GST Antibody, anti-goat IgG alkaline phosphatase conjugate, and 1-chloro-2-4-dinitrobenzene (CDNB)/nitro-blue tetrazolium chloride (NBT) enzyme substrate were used.

## GST 96-Well Detection Module

GST 96-Well Detection Module permits rapid, sensitive determination of GST fusion proteins in a variety of samples. Clarified lysates or intermediate purification fractions can be applied directly into the wells of GST 96-Well Detection Plates. GST-tagged proteins are captured on Anti-GST Antibody immobilized on the walls of each well. Captured GST-tagged proteins are detected with HRP/Anti-GST Conjugate provided in the module (Fig 9). Standard curves for quantitation of GST-tagged proteins can be made with recombinant GST (rGST), which is included as a control (Fig 10). The product contains five microplates and reagents.



**Fig 9.** Screening of bacterial lysates for GST fusion protein expression using GST 96-Well Detection Module. Cultures of randomly selected *E. coli* colonies resulting from a pGEX-6P-1/luciferase gene cloning experiment.



**Fig 10.** Sensitive detection of recombinant GST using the GST 96-Well Detection Module. The indicated amounts of rGST protein were applied directly to the wells of a GST 96-Well Capture Plate. After binding and washing, the wells were treated with a HRP/Anti-GST Conjugate, and detection was performed adding 3, 3', 5, 5'-tetramethylbenzidine (TMB) substrate (A<sub>450</sub>).

## GST Detection Module

GST Detection Module enables sensitive detection of GST-tagged proteins and contains components for detection using either a biochemical assay where glutathione and 1-chloro-2-4-dinitrobenzene (CDNB) serve as substrates for GST to yield a yellow product detectable at 340 nm, or an immunoassay. GST Detection Module contains components sufficient for 50 detection reactions using both assays, and an instruction booklet.

## Removal of GST tag by enzymatic cleavage

Removal of the GST tag can be performed before functional or structural studies of the target protein. The amount of protease, temperature, and length of incubation required for complete digestion varies according to the nature of the target protein.

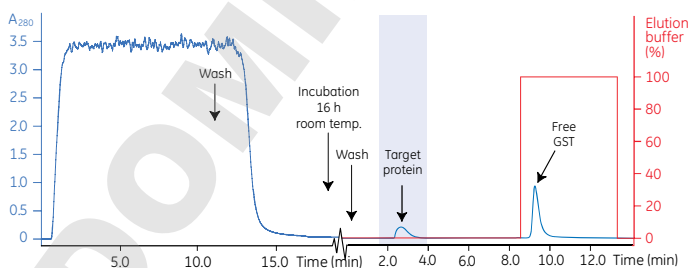
Tagged proteins containing a PreScission Protease, Thrombin, or Factor Xa recognition site can be cleaved either while bound to Glutathione Sepharose chromatography media or in solution after elution. When the GST protein is bound to the column, cleavage releases the target protein, which is eluted with binding buffer while the GST moiety remains bound to the medium. On-column cleavage is generally recommended as the method of choice since many potential contaminants can be washed out and the target protein eluted with a higher level of purity. Cleavage after elution is suggested if optimization of cleavage conditions is necessary.

Removal of serine proteases such as Thrombin, Factor Xa, and PreScission Protease from a protein or peptide preparation is performed using Benzamidine Sepharose 4 Fast Flow, which is available in bulk packs. For convenience, Benzamidine Sepharose 4 Fast Flow is also available in prepacked HiTrap Benzamidine FF 1 ml and 5 ml columns.

### Thrombin

Thrombin enables site-specific cleavage of fusion proteins with an accessible Thrombin recognition sequence and can be used to digest GST-tagged proteins prepared from pGEX vectors containing the recognition sequence for Thrombin (pGEX-1λT, pGEX-2T, pGEX-2TK, pGEX-4T-1, pGEX-4T-2, and pGEX-4T-3). One unit of enzyme cleaves  $\geq 90\%$  of 100  $\mu\text{g}$  of a test GST-tagged protein when incubated in 1  $\times$  PBS at 22°C for 16 h (Fig 11).

**Column:** GSTrap FF 1 ml  
**Sample:** 10 ml of clarified cytoplasmic extract from *E. coli* expressing a GST-tagged protein  
**Binding buffer:** PBS, pH 7.3  
**Elution buffer:** 50 mM Tris-HCl, 10 mM reduced glutathione, pH 8.0  
**Flow rate:** 1 ml/min  
**System:** ÄKTAexplorer™ 10



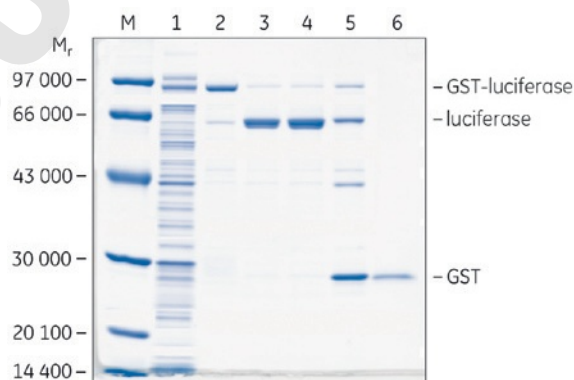
**Fig 11.** On-column Cleavage of the GST affinity tag with Thrombin in conjunction with purification.

### Factor Xa

Factor Xa purified from bovine plasma cleaves proteins specifically after the tetrapeptide Ile-Glu-Gly-Arg and can be used to digest GST-tagged proteins prepared from pGEX vectors containing this sequence (pGEX-3X, pGEX-5X-1, pGEX-5X-2, and pGEX-5X-3). One unit cleaves  $\geq 90\%$  of 100  $\mu\text{g}$  of a test GST-tagged protein when incubated in 1 mM  $\text{CaCl}_2$ , 100 mM NaCl, and 50 mM Tris-HCl (pH 8.0) at 22°C for 16 h.

### PreScission Protease

PreScission Protease is a genetically engineered fusion protein consisting of human rhinovirus 3C protease and GST (Fig 12). This protease was specifically designed to facilitate removal of the protease by allowing simultaneous protease immobilization and cleavage of GST-tagged proteins produced from the pGEX-6P vectors (pGEX-6P-1, pGEX-6P-2, and pGEX-6P-3). PreScission Protease specifically cleaves between the Gln and Gly residues of the recognition sequence of LeuGluValLeuPheGlnGlyPro. The protease has maximal activity at 4°C, and cleavage can thus be performed at low temperatures, improving the stability of the target protein. One unit will cleave  $\geq 90\%$  of 100  $\mu\text{g}$  of a test GST-tagged protein in 50 mM Tris-HCl, 150 mM sodium chloride, 1 mM EDTA, 1 mM DTT, pH 7.0 at 5°C for 16 h.



#### Lanes

M = LMW SDS-Marker Kit (17-0446-01)

1. Sonicate of *E. coli* BL21 cells containing a pGEX-6P-1 plasmid that codes for GST-luciferase recombinant protein
2. Eluate following purification of sonicate on Glutathione Sepharose and elution with buffer containing 10 mM of reduced glutathione
3. Flowthrough following PreScission Protease digestion (4 h, 5°C, 80 units/ml medium bed) of GST-luciferase recombinant protein bound to Glutathione Sepharose
4. Flowthrough following PreScission Protease digestion (16 h, 5°C, 80 units/ml medium bed) of GST-luciferase recombinant protein bound to Glutathione Sepharose
5. Eluate following PreScission Protease digest of GST-luciferase recombinant protein bound to Glutathione Sepharose and elution with buffer containing 10 mM of reduced glutathione
6. Purified GST tag

**Fig 12.** Expression of a tagged GST-luciferase recombinant protein (GST-luciferase) in pGEX-6P-1 and digestion by PreScission Protease while bound to Glutathione Sepharose.

## Ordering information

Product	Quantity	Code no.	Product	Quantity	Code no.
<b>pGEX vectors</b>			<b>Scale-up purification products</b>		
pGEX-4T-1	25 µg	28-9545-49	GSTrap HP columns	5 × 1 ml*	17-5281-01
pGEX-4T-2	25 µg	28-9545-50		1 × 5 ml	17-5282-01
pGEX-4T-3	25 µg	28-9545-52		5 × 5 ml*	17-5282-02
pGEX-5X-1	25 µg	28-9545-53	Glutathione Sepharose High Performance	25 ml	17-5279-01
pGEX-5X-2	25 µg	28-9545-54		100 ml	17-5279-02
pGEX-5X-3	25 µg	28-9545-55	GSTrap FF columns	2 × 1 ml	17-5130-02
pGEX-2TK	25 µg	28-9546-46		5 × 1 ml*	17-5130-01
pGEX-6P-1	25 µg	28-9546-48		1 × 5 ml	17-5131-01
pGEX-6P-2	25 µg	28-9546-50		5 × 5 ml*	17-5131-02
pGEX-6P-3	25 µg	28-9546-51	GSTPrep FF 16/10 column	1 × 20 ml	28-9365-50
pGEX-2T	25 µg	28-9546-53	Glutathione Sepharose 4 Fast Flow	25 ml	17-5132-01
pGEX-3X	25 µg	28-9546-54		100 ml	17-5132-02
pGEX -1λT EcoRI/BAP	5 µg	28-9546-56		500 ml	17-5132-03
<b>GST vector primers for sequencing</b>			GSTrap 4B columns	5 × 1 ml*	28-4017-45
pGEX 5' Sequencing Primer 5'-d[GGGCTGGCAAGCCACGT TTGGTG]-3'	0.05 A <sub>260</sub> unit	27-1410-01		1 × 5 ml	28-4017-47
pGEX 3' Sequencing Primer 5'-d[CCGGGAGCTGCATGTGTC AGAGG]-3'	0.05 A <sub>260</sub> unit	27-1411-01		5 × 5 ml*	28-4017-48
<i>E. coli</i> BL21	1 vial	27-1542-01	Glutathione Sepharose 4B	10 ml	17-0756-01
<b>Small-scale purification products</b>				300 ml	
GST SpinTrap	50 columns	28-9523-59		100 ml	
GST Bulk Kit	1 kit	27-4570-01	Glutathione Sepharose 4B (prepacked disposable column)	2 × 2 ml	17-0757-01
GST GraviTrap	10 columns	28-9523-60	* 100-pack size available by special order.		
GST MultiTrap FF	4 × 96-well filter plates	28-4055-01	<b>Related products</b>		
GST MultiTrap 4B	4 × 96-well filter plates	28-4055-00	Collection plate, 500 µl V-bottom (for use with multiwell plates)	5 × 96-well plates	28-4039-43
GST Buffer Kit	1 kit	28-9523-61	Labmate PD-10 Buffer Reservoir	10	18-3216-03
<b>Detection</b>			<b>Related literature</b>		
Anti-GST Antibody	0.5 ml, 50 detections	27-4577-01	<b>Code no.</b>		
GST Detection Module	50 detections	27-4590-01	Glutathione Sepharose – Total solutions for preparation of GST-tagged proteins, Selection guide	28-9168-33	
GST 96-Well Detection Module	5 plates	27-4592-01	Recombinant Protein Purification Handbook, Principles and Methods,	18-1142-75	
Anti-GST HRP Conjugate	75 µl	RPN1236	GST Gene Fusion System Handbook	18-1157-58	
ECL GST Western Blotting Detection Kit	1 kit	RPN1237	Glutathione Sepharose High Performance, GSTrap HP, Data file	18-1174-32	
<b>Cleavage</b>			Glutathione Sepharose 4 Fast Flow, GSTPrep FF 16/10, GSTrap FF, Data file	18-1174-85	
Thrombin	500 units	27-0846-01	GSTrap 4B columns, Data file	28-4048-14	
Factor Xa	400 units	27-0849-01	Benzamidine Sepharose 4 Fast Flow (high sub)/HiTrap Benzamidine FF (high sub), Data file	18-1139-38	
PreScission Protease	500 units	27-0843-01	Pure simplicity for tagged proteins, Brochure	28-9353-64	
HiTrap Benzamidine FF (high sub)	2 × 1 ml	17-5143-02	Prepacked chromatography columns for ÄKTA design systems, Selection guide	28-9317-78	
	5 × 1 ml	17-5143-01			
	1 × 5 ml	17-5144-01			
Benzamidine Sepharose 4 Fast Flow (high sub)	25 ml	17-5123-10			

For contact information for your local office,  
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