

Successful Protein Expression and Purification



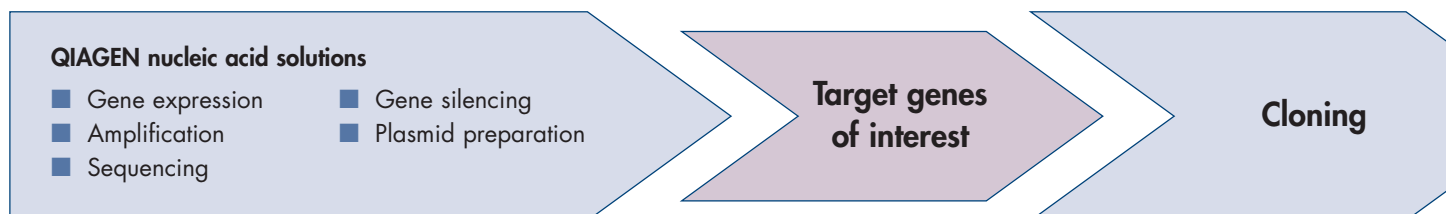
Sample & Assay Technologies

QIAGEN® technologies make proteins available to all

Modern techniques in molecular biology help to provide answers to important scientific questions, such as identification of genes involved in specific cellular processes. Furthermore, these techniques enable target genes to be cloned for expression and detailed analysis of recombinant proteins.

You can produce the amount of protein you need for your downstream applications — which include protein assay, protein binding/interaction studies, antibody generation, structural analysis, expression library screening, screening engineered enzymes, and comprehensive studies of proteins identified in gene expression analysis. And QIAGEN products help you optimize each critical success factor at every step.

High-quality recombinant proteins from your gene of interest



QIAGEN protein solution

Success factors

- QIAgenes — Optimized expression of human genes

Additional products

- Fast access to optimal construct
- Error-free full-length open reading frame
- Enhanced protein yields
- pQE expression vectors*
- Products for PCR and plasmid preparation*

* Find more information at www.qiagen.com.



QIAGEN automated solutions



| | | |
|--|---|--|
| <ul style="list-style-type: none"> ■ EasyXpress® products for cell-free protein expression | <ul style="list-style-type: none"> ■ Ni-NTA matrices for His-tagged proteins | <ul style="list-style-type: none"> ■ Anti-His Antibodies and Antibody Conjugates ■ <i>Strep</i>-tag Antibody |
| <ul style="list-style-type: none"> ■ Fast access to multiple proteins ■ Posttranslational modification ■ Functional expression of membrane proteins | <ul style="list-style-type: none"> ■ High yields and purity ■ Robustness of purification process ■ Protein activity ■ Scalability | <ul style="list-style-type: none"> ■ High sensitivity ■ High specificity ■ Reliable protein interaction assay |
| <ul style="list-style-type: none"> ■ Transfection reagents* | <ul style="list-style-type: none"> ■ <i>Strep</i>-Tactin® matrices for <i>Strep</i>-tagged proteins* | <ul style="list-style-type: none"> ■ HisSorb Strips and Plates* ■ Ni-NTA, <i>Strep</i>-Tactin Magnetic Beads* |

* Find more information at www.qiagen.com.

QIAgenes:* Easy access to high-yield expression of human proteins — in *E. coli*, insect, and mammalian cells

Lack of sufficient amounts of protein often slows down or makes structural and functional analyses of proteins impossible. Expression of optimized genes helps to overcome this obstacle by improving codon usage and avoiding mRNA secondary structure or motifs that interfere with the transcription/translation process (internal ribosomal binding sites, sequence repeats etc.).

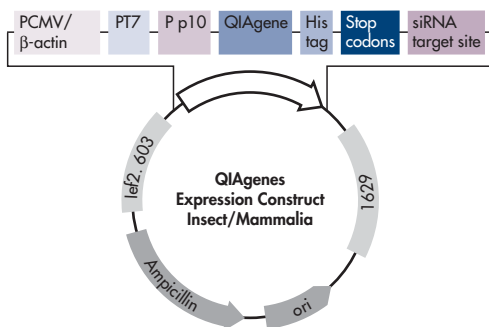


Figure 1. Elements within QIAgenes Expression Constructs Insect/Mammalia.

QIAgenes Expression Kits offer easy access to increased protein expression through:

- Expression-optimized genes — powered by GENEART®
- Genomewide human ORFs ready cloned into expression vectors
- Synthetic cDNA with guaranteed sequence identity
- His tag for Ni-NTA affinity purification
- Easy ordering online through GeneGlobe (www.geneglobe.com)

Using optimized sequences, expression success and expression yields were significantly increased compared to the expression of wildtype sequences (Figure 2).

Proteins expressed with QIAgenes Expression Kits are compatible with:

- Ni-NTA purification technology
- EasyXpress cell-free protein expression kits
- Subcloning of individual domains (Table 1)
- TagZyme™ mediated N-terminal His tag removal (*E. coli* kits)
- C-terminal directed biotinylation for interaction studies (*E. coli* kits)
- siRNA rescue experiments (Insect/Mammalia kits)

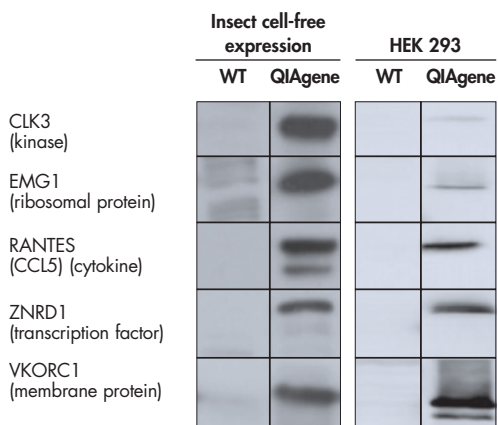


Figure 2. Optimized protein-coding sequences increase yields: Optimized QIAgenes Insect/Mammalia and wild-type (WT) coding sequences of the indicated proteins were cloned to generate QIAgene Expression Constructs and expressed in parallel, either in vitro using the EasyXpress Insect Kit II (insect cell-free expression) or in vivo in HEK 293 cells (HEK 293). Expression levels were visualized after separation of crude lysates by SDS-PAGE and western blotting using Penta-His Antibodies and chemiluminescent detection. **CLK3**: CDC-like kinase 3; **EMG1**: EMG1 nucleolar protein homolog; **RANTES (CCL5)**: chemokine (C-C motif) ligand 5; **ZNRD1**: zinc ribbon domain containing 1; **VKORC1**: vitamin K epoxide reductase complex, subunit 1.

Table 1. pQE vectors for subcloning QIAgenes

| Vector | Promoter | Tag | Expression system |
|--------------------------|-------------------------|---|--|
| pQE-T7 vectors | T7 | C- or N-terminal 10x His tag | <i>E. coli</i> , EasyXpress <i>E. coli</i> Kits |
| pQE-T7-TriSystem vectors | T7, p10, CMV/β-actin | C- or N-terminal 10x His tag or without tag | Insect cells, mammalian, EasyXpress Insect Kits |

For detailed information about QIAgenes, please visit www.qiagen.com/goto/QIAgenes.

* Powered by GENEART — the gene of your choice®

Fast access to multiple proteins — with EasyXpress

Whether you require expression of a target gene identified, for example, in gene expression analysis or expression of a number of different mutations or truncated forms of a gene, the EasyXpress System enables cell-free protein expression, delivering your protein in just a few hours (Figure 3). For proteins with no posttranslational modifications or for which posttranslational modifications are not required, we recommend using the EasyXpress *E. coli* system; the EasyXpress Insect system is the system of choice for expression of eukaryotic proteins with posttranslational modifications and expression of membrane proteins.

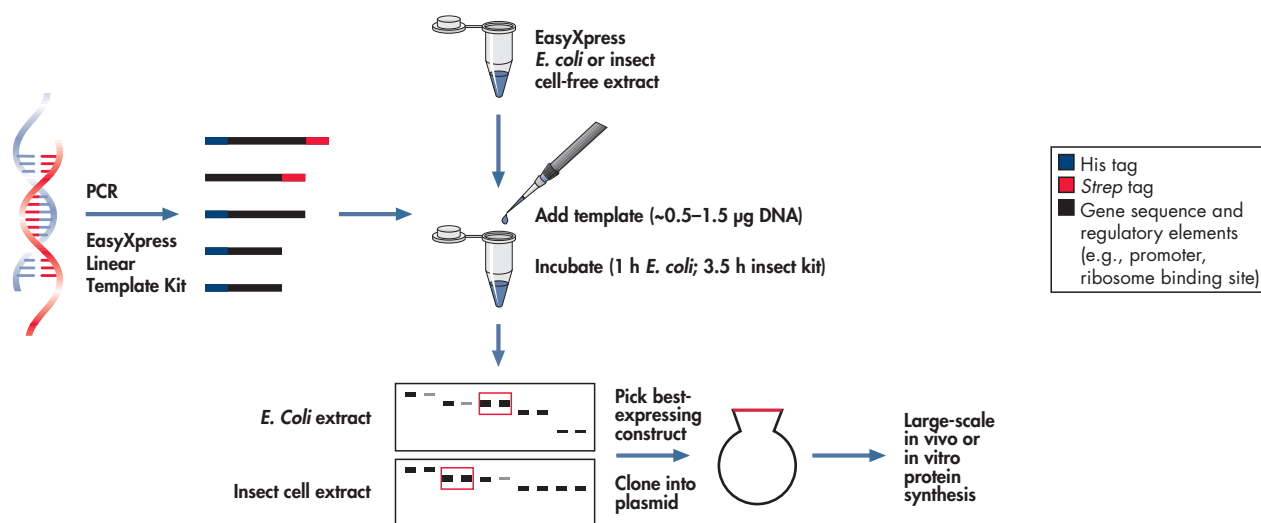


Figure 3. From multiple genes to multiple proteins in a single day using PCR products.

Using the EasyXpress System means:

- No expensive instrumentation for cell-free synthesis
- You can test and produce your QIAgene quickly and easily (Figure 2)
- Easy evaluation of different mutants, truncations, or tag variants (Figure 3)
- An open system allowing addition of various reagents, e.g., co-factors, detergents (see Table 2)
- Up to 1 mg (*E. coli* kits) or 50 µg (Insect Kit II) protein/ml reaction

Table 2. Reagents compatible with EasyXpress *E. coli* cell-free expression reactions*

| Reagent | Effect |
|--|--------------------------------|
| Detergents | Solubilize proteins |
| Redox buffer | Formation of disulfide bridges |
| Hydroxyectoine, D-sorbitol, glycine betaine, L-carnitine, glycerol | Stabilize proteins |
| Metal ions (e.g., Ca ²⁺ , Co ²⁺ , Cu ²⁺ , Fe ²⁺ , Zn ²⁺) | Act as enzyme cofactors |

* For recommended working concentrations please refer to the *EasyXpress Protein Synthesis Handbook*.

Expression of modified eukaryotic and membrane proteins

A broad range of eukaryotic proteins require posttranslational modifications (PTMs) or signal peptide cleavage to display full functional activity. The EasyXpress Insect Kit II, a unique eukaryotic cell-free expression system, enables expression of eukaryotic proteins — including membrane proteins — with PTMs (Table 3).

Figure 4. The indicated *Strep*-tagged protein was synthesized using 500 ng PCR product as expression template in reactions carried out according to manufacturers' instructions. 2.5 μ l aliquots (EasyXpress) or 12.5 μ l (RRL system) of total reaction or supernatant were loaded per lane and visualized using a *Strep*-tag antibody and chemiluminescent detection.

TFIIA $\alpha\beta$: Human transcription factor II, alpha and beta subunits; **MKK3:** Human dual specific mitogen-activated protein kinase 3; **TNF α :** Human tumor necrosis factor alpha. Total (T) and the soluble (S) fraction.

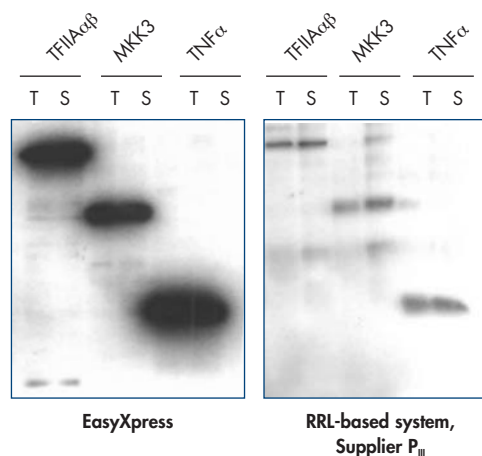


Table 3. Comparison of the EasyXpress Insect Kit II and RRL-based kits

| Criterion | EasyXpress Insect Kit II | RRL-based kit (Supplier P) |
|---------------------------------|--|---|
| Yields | Up to 50 μ g/ml (fully active protein) | 3–6 μ g/ml |
| Posttranslational modifications | Glycosylation, signal peptide cleavage, phosphorylation in homogenous lysates without the need for further additives | Requires additional purchase of Canine Pancreatic Microsomal Membranes |
| Lot-to-lot reproducibility | Good, due to easily controllable production of homogenous insect cell lysates | Poor, due to mixed system (Rabbit reticulocytes and canine microsomes) produced from individual animals |
| Handling | Lysate easy to pipet, all components stay in solution, even on ice | Lysate contains red precipitates (hemoglobin) that make accurate pipetting difficult and may distort photometric assays |

Table 4. Overview of EasyXpress Kits for cell-free protein expression

| Application | Kit | Protein yield | DNA template/promoter | Possible labels |
|--|--|-------------------------------|---|--|
| Construct screening, functional studies | EasyXpress Protein Synthesis Kits | Up to 1 mg/ml reaction | Circular plasmid, PCR product/T7, strong <i>E. coli</i> | Biotin* |
| Construct screening, functional studies, immunization | EasyXpress Protein Synthesis Maxi Kit | Up to 1 mg/ml reaction | Circular plasmid, PCR product/T7, strong <i>E. coli</i> | Biotin* |
| True site-specific labeling for protein–protein interaction studies | EasyXpress Site-Specific Biotin Kit | Up to 150 μ g/ml reaction | Circular plasmid, PCR product with Amber stop codon/T7, strong <i>E. coli</i> | Biotin |
| Labeling for X-ray or NMR studies | EasyXpress Mega Kit, EasyXpress NMR Kits | Up to 10 mg/kit | Circular plasmid/T7, strong <i>E. coli</i> | Selenomethionine, isotopically labeled amino acids |
| Construct screening, functional studies of posttranslationally modified proteins | EasyXpress Insect Kit II | Up to 50 μ g/ml reaction | Linearized plasmid, PCR product/T7, strong <i>E. coli</i> | Biotin* |
| Non-radioactive protein detection | EasyXpress Random Biotin Kit | Up to 1 mg/ml reaction | In combination with EasyXpress <i>E. coli</i> and insect-cell lysates | |
| Generation of PCR products for use as expression templates | EasyXpress Linear Template Kit | | Compatible with EasyXpress <i>E. coli</i> and insect-cell lysates | |

* In combination with the EasyXpress Random Biotin Kit. For more information, visit www.qiagen.com/goto/EasyXpress.

Ni-NTA for the highest purity proteins

Purification of His-tagged proteins with Ni-NTA follows — as usual for QIAGEN purification products — a simple bind-wash-elute procedure (Figure 5). Due to the structure of NTA and its tight binding of nickel ions (Figure 6) proteins can be eluted in high yields with high purity (Figure 7). You can run several cycles without stripping and reloading the resin, which reduces also Ni²⁺ waste. The purification process itself is very robust and allows the use of a broad range of reagents, meaning you can use the specific conditions your proteins need to stay active to purify them (Table 5).

Ni-NTA — the world’s most cited affinity purification resin

- Highest purity of protein after a single purification step (Figure 7)
- Minimal nickel leaching resulting in highly pure proteins and avoiding recharging and toxic waste (Figure 6)
- High yields of up to 50 mg/ml Ni-NTA resin or up to 2 µg/µl magnetic bead suspension (Figure 8)
- Broad reagent compatibility with the only resin that is compatible to 10 mM DTT (Table 5)

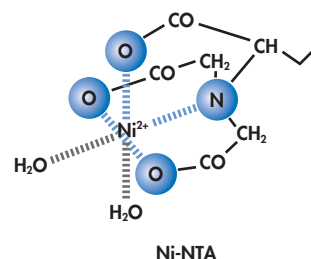


Figure 6. Ni-NTA gives you highly pure proteins. In Ni-NTA resins nickel ions are bound via four coordination sites. Nickel ions bound to IDA-based resins are only bound via three coordination sites and can be lost more easily. Empty charged coordination sites will function like ion-exchange matrices that can bind un-tagged contaminating proteins.

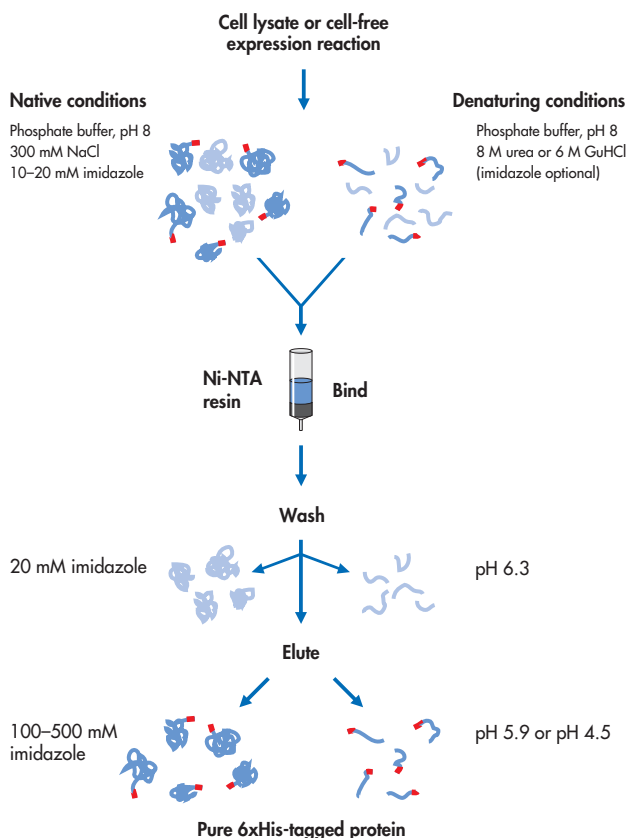


Figure 5. Bind-Wash-Elute purification with Ni-NTA under native (left) and denaturing (right) conditions.

Figure 7. Ni-NTA outperforms a competitor resin for efficient one-step purification of His-tagged proteins. The indicated proteins (arrowed) were expressed in *E. coli* using a QIAgenes Expression Construct. Proteins were purified in parallel from cleared cell lysate using either Ni-NTA Superflow or a nickel resin from supplier G. These two proteins were part of a trial in which 21 of 24 proteins were purified at higher purity on Ni-NTA than on the resin from supplier G. **CL:** Cleared lysate. Each set of three lanes shows from left to right: flow-through, wash, and elution fractions.

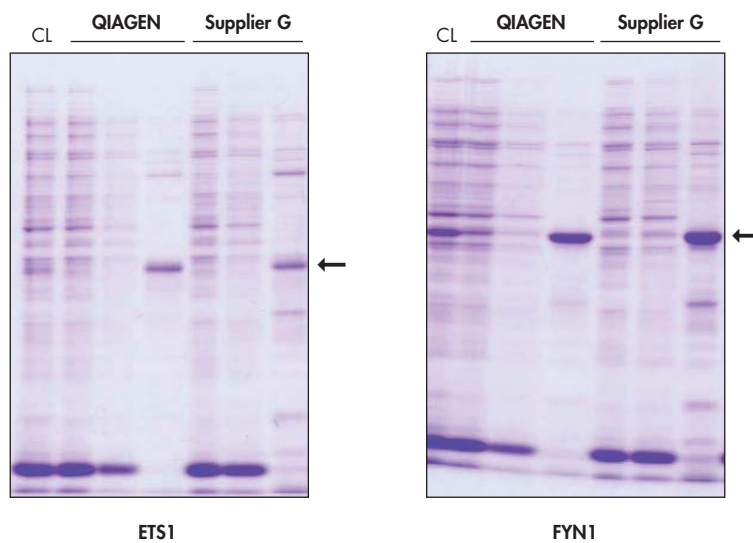


Figure 8. Binding of various His-tagged proteins to Ni-NTA. Binding was performed in batch procedures and proteins quantified using the Bradford method.

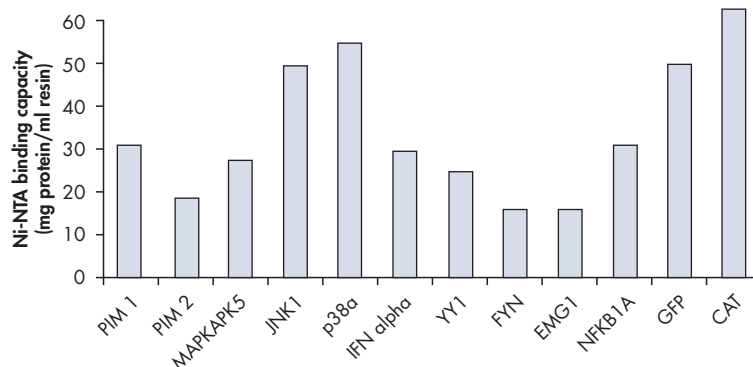


Table 5. Reagents compatible with the 6xHis-Ni-NTA interaction*

| Denaturants | Detergents | Reducing agents | Others | Salts | For long-term storage |
|-------------|-----------------|------------------|------------------------------|------------------------|-----------------------|
| 6 M Gu-HCl | 2% Triton X-100 | 20 mM β-ME | 50% glycerol | 4 M MgCl ₂ | Up to 30% ethanol |
| 8 M Urea | 2% Tween® 20 | 10 mM DTT | 20% ethanol | 5 mM CaCl ₂ | or 100 mM NaOH |
| | 1% CHAPS | 20 mM TCEP | 20 mM imidazole [†] | 2 M NaCl | |
| | 30 mM LDAO | | | | |
| | 20 mM DDM | | | | |
| | 51 mM OG | | | | |

* A more extensive table can be found at www.qiagen.com/goto/Ni-NTAcompatibility.

[†] Can be used at low concentrations (20 mM) to inhibit nonspecific binding and, at higher concentrations (>100 mM), to elute the His-tagged protein from the Ni-NTA matrix.

The faster way to purer proteins with Ni-NTA Superflow™ Cartridges

Fast Protein Liquid Chromatography (FPLC™) is the method of choice for rapidly purifying milligram amounts of protein (e.g., for structural studies). QIAGEN Ni-NTA cartridges — available in 1 ml and 5 ml bed volumes — are compatible with all commonly used FPLC systems (Table 6).



Table 6. Ni-NTA Cartridge specifications

| | 1 ml Ni-NTA Cartridges | 5 ml Ni-NTA Cartridges |
|------------------------------|--|---------------------------------|
| Matrix | Highly cross-linked, 6% agarose | |
| Binding capacity | Up to 50 mg His-tagged protein | Up to 250 mg His-tagged protein |
| Recommended flow rate | 1 ml/min | 5 ml/min |
| Maximum flow rate | 10 ml/min | 40 ml/min |
| Maximum back pressure | 0.5 MPa, 5 bar | |
| Column dimensions (i.d. x h) | 6.7 mm x 28.0 mm | 14.7 mm x 29.8 mm |
| Suitable for | FPLC, ÄKTA™, and Biologic systems, Vision workstation, HPLC instruments, or manual purification using a syringe. | |
| Cartridge connectors | 1/16" (inlet); M 6 (outlet) | |

Ni-NTA Superflow Cartridges include:

- Cartridges containing Ni-NTA — for high yields of high-purity proteins
- Robust construction allowing high maximum flow rates (Table 6)
- Removable screw-top lid for easy maintenance

Table 7. Ni-NTA available in any format

| Product | Protein yield | Applications | Automation option |
|-------------------------------|---|--|--|
| Ni-NTA HisSorb Plates | Up to 0.3 µg/well | Oriented binding of His-tagged proteins | Can be used in combination with standard plate readers |
| Ni-NTA Magnetic Agarose Beads | Up to 2 µg/µl suspension (5%) | Purification from dilute lysates or high-throughput applications | BioSprint® 96, BioRobot® instruments, QIAsymphony® SP |
| Ni-NTA Spin Columns | Up to 300 µg/spin column | Low-throughput expression screening | QIAcube® |
| Ni-NTA Fast Start Kit | Up to 25 mg per prep | Complete purification kit for beginners | |
| Ni-NTA Agarose* | 100 µg – 100 mg | Batch and gravity-flow column purification | |
| Ni-NTA Superflow Columns | Up to 75 mg/column | Automated and gravity-flow column purification | BioRobot instruments |
| Ni-NTA Superflow Cartridges*† | Up to 50 mg (1 ml cartridge) or 250 mg (5 ml cartridge) | Purification using FPLC system or syringe | All commonly-used FPLC instruments |
| Ni-NTA Superflow* | 5 mg – production scale | Batch and FPLC column purification | BioRobot instruments |

* Also available as uncharged NTA products.

† FPLC-compatible cartridges also available for purification of *Strep*-tagged, phospho-, and glycoproteins and serum depletion. Visit www.qiagen.com/goto/cartridges for more information.

For more information about Ni-NTA matrices, visit www.qiagen.com/goto/Ni-NTA.





QIAcube



QIA Symphony SP

Let your instrument do your protein sample preps for you

Whether you want micrograms of protein for expression screening, or milligrams for X-ray crystallography (Table 8), QIAGEN offers instruments and chemistries for highly reproducible sample preparation, so you can use your time for more challenging research.

QIAGEN automated solutions offer:

- Highly reproducible processing and cross-contamination-free pipetting (Figure 10)
- Fully automated solutions on QIAcube and QIA Symphony SP for highest convenience
- Scalable solutions for purifying micrograms to milligrams of pure proteins (Table 8)

Figure 9. Similar yields from automated and manual procedures. The indicated proteins were purified under native conditions from cleared *E. coli* cell lysates in duplicate using Ni-NTA Spin Columns either in an automated procedure on the QIAcube or manually. The average total yields from the duplicate preparations were determined using the Bradford method. **CAT:** Chloramphenicol acetyl transferase; **GFP:** Green fluorescent protein; **HIV-RT:** Human immunodeficiency virus reverse transcriptase; **IL-1b:** Interleukin-1 beta.

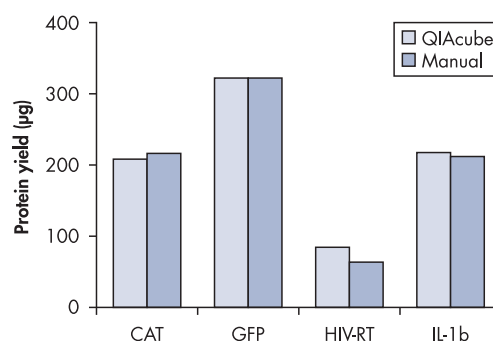


Figure 10. High, reproducible yields of His-tagged GFP from *E. coli* cultures. His-tagged green fluorescent protein (GFP, arrowed) was purified from 5 ml *E. coli* cultures using the QIA Symphony Ni-NTA Denaturing Kit. Pairs of samples show crude lysate and eluate fractions. Average yield was 161 µg per well with a CV of 5.28% (8 out of 24 samples shown).

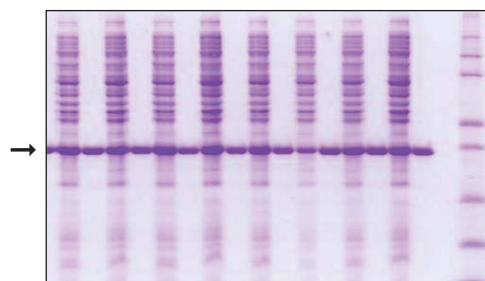


Table 8. QIAGEN automated solutions for protein

| Instrument | Chemistry | Protein yield | Time per run |
|--------------------------------------|--|---|----------------|
| QIAcube* (12 samples) | Ni-NTA Spin Kit | Up to 300 µg His-tagged protein per column | Approx. 30 min |
| | Qproteome® Albumin/IgG Depletion Kit | Up to 0.8 mg protein per column | Approx. 30 min |
| QIA Symphony SP (up to 96 wells) | Ni-NTA Magnetic Beads | Up to 500 µg His-tagged protein per well | 2.5 h |
| BioRobot Protein System (96 samples) | Ni-NTA Magnetic Beads or Strep-Tactin Magnetic Beads | Up to 50 µg His-tagged protein or 10 µg Strep-tagged protein per well | 1 h |
| | Ni-NTA Superflow 96 BioRobot Kit | Up to 10 mg His-tagged protein per well | 2.5 h |
| BioSprint 96 (96 samples) | Ni-NTA Magnetic Beads or Strep-Tactin Magnetic Beads | Up to 400 µg His-tagged protein or 60 µg Strep-tagged protein | 2 h |

* The continuously expanding range of QIAcube protocols has now grown to almost 80 standard protocols. For an up-to-date list of protocols, visit www.qiagen.com/MyQIAcube. To find out more about QIAGEN's automated solutions, visit www.qiagen.com/goto/automation.

Highly sensitive detection of His- and *Strep*-tagged proteins

QIAGEN antibodies enable highly sensitive and specific detection of His- and *Strep*-tagged proteins (Figures 11–13). For various downstream applications such as western blot, dot blot, immunoprecipitation, immunohistochemistry, ELISA, immunolocalization, and FACS (Table 9).

All QIAGEN antibodies are:

- Mouse monoclonal antibodies, free from viruses, mycoplasma, and contaminating immunoglobulins
- Highly specific, displaying no cross-reactivity with host-cell proteins (Figure 11 and 12)
- Available in bulk quantities upon request

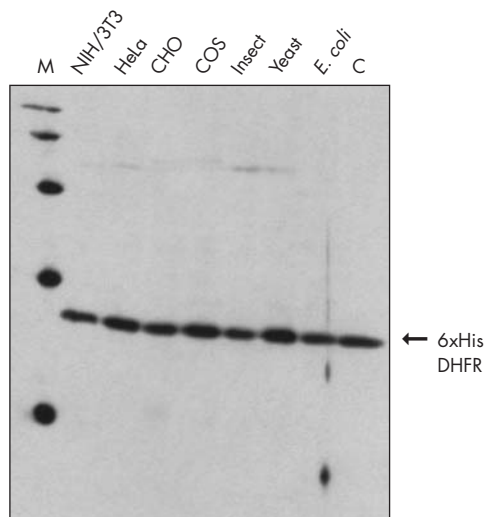


Figure 11. Negligible cross reaction in any expression system. His-tagged DHFR (3 ng) was added to 1 µg of the indicated cell lysate, separated by SDS-PAGE, transferred to membrane by western blotting, and detected in a chemiluminescent procedure using the Penta-His HRP Conjugate. **M:** 6xHis Protein Ladder; **C:** control (purified DHFR, no lysate).

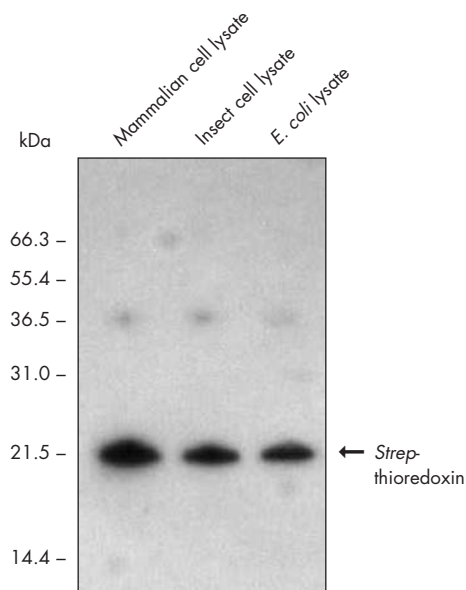


Figure 12. Highly specific detection of *Strep*-tagged proteins in any expression system. Purified *Strep*-tagged thioredoxin (10 ng) was mixed with crude cell lysate from mammalian (NIH-3T3), insect (Sf9), and bacterial (*E. coli*) cells (each 10 µg total protein). After electrophoresis and western transfer, the *Strep*-tagged protein was detected using the *Strep*-tag Antibody and chemiluminescent detection.

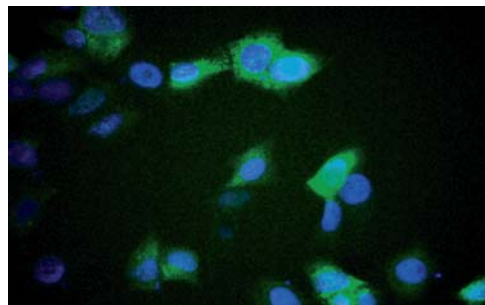
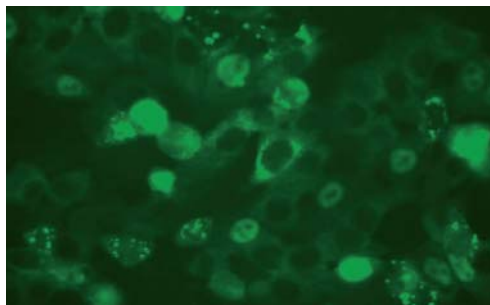


Figure 13. Direct fluorescent localization of proteins. Fluorescence micrograph (400x) of human fibrosarcoma cells transfected with a pQE-TriSystem construct encoding His-tagged decorin. Cells were fixed with 2% paraformaldehyde 24 hours post-transfection and stained using Penta-His Alexa Fluor 488 Conjugate. Fixed cells were stained using Penta-His Alexa Fluor 488 Conjugate and nuclei were counterstained with DAPI (400x).

Table 9. Antibody overview

| | Penta-His Antibodies and HRP Conjugates | Tetra-His Antibodies and HRP Conjugates | RGS-His Antibodies and HRP Conjugates | Penta-His Alexa Fluor Conjugates | Strep-tag Antibody |
|----------------------------------|---|---|---|---|--|
| Epitope | HHHHH | HHHH | RGSHHHH | HHHHH | SAWSHPGFEK |
| Dissociation constant, K_d (M) | $5 \times 10^{-8} - 1 \times 10^{-9}$ | $1 \times 10^{-8} - 5 \times 10^{-8}$ | $1 \times 10^{-8} - 5 \times 10^{-8}$ | $5 \times 10^{-8} - 1 \times 10^{-9}$ | $5 \times 10^{-9} - 1 \times 10^{-10}$ |
| Applications | Western blots, dot blots, ELISA, immunoprecipitation, immunohistochemistry | Western blots, dot blots, ELISA, immunoprecipitation, immunohistochemistry | Western blots, dot blots, ELISA, immunoprecipitation, immunohistochemistry | Immunolocalization, FACS | Western blots, dot blots, ELISA, immunoprecipitation, immunohistochemistry |
| Sensitivity in western blots | 50 pg (chemiluminescent detection); 2 ng (chromogenic detection) | 50 pg (chemiluminescent detection); 2 ng (chromogenic detection) | 50 pg (chemiluminescent detection); 2 ng (chromogenic detection) | n.a. | 1 ng (chemiluminescent detection) |
| Direct detection? | HRP Conjugate enables direct detection. Secondary antibody required for Penta-His Antibody. | HRP Conjugate enables direct detection. Secondary antibody required for Tetra-His Antibody. | HRP Conjugate enables direct detection. Secondary antibody required for RGS-His Antibody. | Detection via fluorescence | Secondary antibody required |

Also available:

Anti-His Antibody Selector Kit — to test for the Anti-His Antibody that best recognizes your protein

6xHis Protein Ladder — a mixture of five His-tagged proteins that serve as size markers and positive controls in detection procedures

Penta-His Biotin Conjugate — for detection using (Strept)avidin reagents

Ni-NTA Conjugates — for direct chromogenic detection of His-tagged proteins in *E. coli* lysates

More information about protein detection and assay can be found at www.qiagen.com/goto/detection.

Ordering Information

| Product | Contents | Cat. no. |
|---|--|----------|
| Cloning | | |
| QIAgenes Expression Kit <i>E. coli</i> | QIAgenes Expression Construct, QIAgenes <i>E. coli</i> Positive Control, Penta-His Antibody, BSA-free, 4 Ni-NTA Spin Columns | Varies |
| QIAgenes Expression Kit Insect/Mammalia | QIAgenes Expression Construct Insect/Mammalia; QIAgenes Insect/Mammalia Positive Control; Penta-His Antibody, BSA-free; Ni-NTA Magnetic Agarose Beads | Varies |
| pQE-T7 Vector 1 | 25 µg; for subcloning QIAgenes (N-terminal 10xHis) | 33013 |
| pQE-T7 Vector 2 | 25 µg; for subcloning QIAgenes (C-terminal 10xHis) | 33023 |
| pQE-T7-TriSystem Vector Set 1 | pQE-T7-TriSystem Vector Set 1 contains: Vector pQE-TriSystem 5, Vector pQE-TriSystem 6, and Vector pQE-TriSystem 7; 25 µg each | 32953 |
| N-Terminus pQE Vector Set | For expressing N-terminally His-tagged proteins in <i>E. coli</i> : 25 µg each; pQE-9, pQE-30, pQE-31, pQE-32, pQE-40 | 32915 |
| C-Terminus pQE Vector Set | For expressing C-terminally His-tagged proteins in <i>E. coli</i> : 25 µg each; pQE-16, pQE-60, pQE-70 | 32903 |
| pQE-TriSystem Vector | For parallel expression of 6xHis-tagged proteins in <i>E. coli</i> , mammalian cells, and baculovirus-infected insect cells: 25 µg pQE-TriSystem Vector DNA | 33903 |
| Expression | | |
| EasyXpress Linear Template Kit Plus (20) | For 20 two-step PCRs: HotStar HiFidelity DNA Polymerase, buffer, RNase-free water, Q-Solution, XE-Solution, positive-control DNA, and optimized PCR primers | 32723 |
| EasyXpress Protein Synthesis Kit (20)* | For 20 x 50 µl reactions: <i>E. coli</i> extract, reaction buffer, RNase-free water, and positive-control DNA | 32502 |
| EasyXpress Protein Synthesis Mega Kit | For 2 x 5 ml reactions: <i>E. coli</i> extract, reaction buffers, amino acid mix w/o methionine, methionine, RNase-free water, gel-filtration columns, and reaction flasks | 32516 |
| EasyXpress NMR Protein Synthesis Kit† | For 2 x 5 ml reactions: <i>E. coli</i> extract, reaction buffers, amino acid mix w/o Arg, Lys, Ser, Thr, Val (supplied as individual amino acids), RNase-free water, gel-filtration columns, and reaction flasks | 32526 |
| EasyXpress Site-Specific Biotin Kit | For 5 x 25 µl reactions: <i>E. coli</i> extract, reaction buffer, RNase-free Water, biotinyl-lysyl tRNA (amber), and positive-control DNA | 32602 |
| EasyXpress Random Biotin Kit | For 60 x 50 µl reactions: 4 x 15 µl EasyXpress Biotinyl-Lysyl tRNA (Phe) | 32612 |
| EasyXpress Insect Kit II | For 20 x 50 µl reactions: <i>Spodoptera frugiperda</i> insect cell extract, reaction buffers, in vitro transcription reaction components, RNase-free water, gel-filtration columns, and positive-control DNA | 32562 |

* Other pack sizes or formats are available; please inquire.

† Kits for labeling other amino acids and for uniform labeling are also available; for more information, visit www.qiagen.com/goto/EasyXpress.



Ordering Information

| Product | Contents | Cat. no. |
|--|---|----------------|
| Purification | | |
| Ni-NTA Magnetic Agarose Beads (2 x 1 ml)* | 2 x 1 ml nickel-charged magnetic agarose beads (5% suspension) | 36111 |
| Ni-NTA Spin Kit (50) | 50 Ni-NTA Spin Columns, Reagents, Buffers, Collection Tubes, 1 µg Control Expression Plasmid | 31314 |
| Ni-NTA Agarose (25 ml)* | 25 ml nickel-charged resin (max. pressure: 2.8 psi) | 30210 |
| Ni-NTA Superflow (25 ml)* | 25 ml nickel-charged resin (max. pressure: 140 psi) | 30410 |
| Ni-NTA Superflow Cartridges (5 x 1 ml)* | 5 cartridges prefilled with 1 ml Ni-NTA Superflow: for automated purification of His-tagged proteins using liquid chromatography systems | 30721 |
| Ni-NTA Superflow Cartridge (1 x 5 ml)* | 1 cartridge prefilled with 5 ml Ni-NTA Superflow: for automated purification of His-tagged proteins using liquid chromatography systems | 30760 |
| NTA Superflow Cartridges (5 x 1 ml)* | 5 cartridges prefilled with 1 ml NTA Superflow: for purification of His-tagged and other metal-binding proteins using liquid chromatography systems | 30821 |
| Ni-NTA Fast Start Kit (6) | For purification and detection of six 6xHis-tagged protein preps: 6 x Fast Start Columns, Penta-His Antibody, Buffers and Reagents | 30600 |
| Ni-NTA Membrane Protein Kit | For 5 detergent screenings and 5 affinity purifications: 7 detergents, buffers, Ni-NTA Superflow, Penta-His Antibody, disposable columns | 30610 |
| <i>Strep</i> -Tactin Magnetic Beads (2 x 1 ml)* | For micro-scale purification of <i>Strep</i> -tagged proteins: 2 x 1 ml <i>Strep</i> -Tactin-charged magnetic agarose beads (10% suspension) | 36311 |
| <i>Strep</i> -Tactin Superflow (2 ml)* | For batch and HPLC purification of <i>Strep</i> -tagged proteins: 2 ml <i>Strep</i> -Tactin-charged Superflow (max. pressure: 140 psi) | 30001 |
| <i>Strep</i> -Tactin Superflow Cartridge (1 ml)* | 1 cartridge prefilled with 1 ml <i>Strep</i> -Tactin Superflow: for automated purification of <i>Strep</i> -tagged proteins using liquid chromatography systems | 30120 |
| PhosphoProtein Purification Cartridge | 5 ml FPLC-compatible cartridge prefilled with PhosphoProtein Purification resin | 37145 |
| Lectin cartridges | ConA, WGA, GNA, LCH, SNA, MAL, AIL, and PNA lectins can be supplied on demand as individual 1ml or 5 ml Cartridges. | Please inquire |

* Other pack sizes or formats are available; please inquire.

† Penta-His Antibodies are also available conjugated to Alexa Fluor 532, 555, and 647.

Ordering Information

| Product | Contents | Cat. no. |
|---|---|----------|
| Detection and Assay | | |
| Anti-His Antibody Selector Kit | RGS-His Antibody, Penta-His Antibody, Tetra-His Antibody, all BSA-free, 3 µg each | 34698 |
| Penta-His Antibody, BSA-free (100 µg) | 100 µg mouse anti-(H)5 (lyophilized, BSA-free, for 1000 ml working solution) | 34660 |
| Penta-His HRP Conjugate Kit | 125 µl Penta-His HRP Conjugate, 5 g Blocking Reagent, 50 ml Blocking Reagent Buffer (10x concentrate) | 34460 |
| Penta-His Alexa Fluor 488 Conjugate† | 125 µl Penta-His Alexa Fluor 488 Conjugate, 200 µg/ml | 35310 |
| <i>Strep</i> -tag Antibody (100 µg) | Mouse monoclonal antibody that recognizes the <i>Strep</i> -tag II epitope; lyophilized, for 1000 ml working solution | 34850 |
| 6xHis Protein Ladder | 6xHis-tagged marker proteins (lyophilized, for 50–100 lanes on western blots) | 34705 |
| Ni-NTA Magnetic Agarose Beads (2 x 1 ml)* | 2 x 1 ml nickel-charged magnetic agarose beads (5% suspension) | 36111 |
| Ni-NTA HisSorb Plates (5) | 5 Ni-NTA-coated, transparent 96-well plates | 35061 |

* Other pack sizes or formats are available; please inquire.

† Penta-His Antibodies are also available conjugated to Alexa Fluor 532, 555, and 647.

Discover the complete range of QIAGEN quality protein products at www.qiagen.com/protein!

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Hoffmann-La Roche owns patents and patent applications pertaining to the application of Ni-NTA resin [Patent series: RAN 4100/63: USP 4,877,830, USP 5,047,513, EP 253 303 B1], and to 6xHis-coding vectors and His-labeled proteins [Patent series: USP 5,284,933, USP 5,130,663, EP 282 042 B1]. All purification of recombinant proteins by Ni NTA chromatography for commercial purposes, and the commercial use of proteins so purified, require a license from Hoffmann-La Roche.

Strep-tag® technology for protein purification and detection is covered by US patent 5,506,121, UK patent 2272698 and French patent 93 13 066; *Strep*-Tactin® is covered by US patent 6,103,493.

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