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Overnight Express™ Autoinduction Systems

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Can Merck Chemicals make my protein expression process easier?

Yes! With Overnight Express™ Autoinduction Systems, the protein expression process begins without having to manually add induction agent.
We've done it for you, so that you don't have to!!

That's what's in it for you. Merck Chemicals

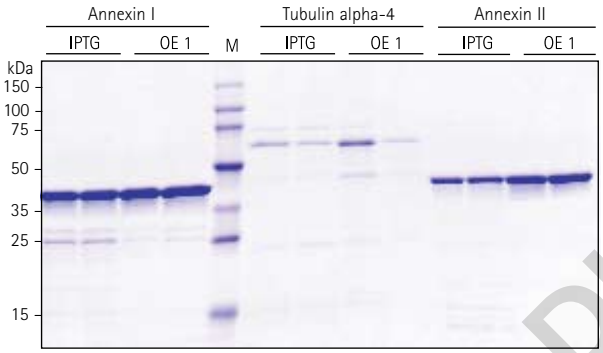
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The Overnight Express™ Autoinduction Systems enable regulated protein expression in *E. coli*, without monitoring the culture or adding inducer during cell growth. The simplified protocol offers great convenience, allowing you to focus on your research while it does its job.



Overnight Express™ Autoinduction Systems

With Overnight Express™ Autoinduction Systems, a period of cell growth is followed by spontaneous induction of protein expression – without monitoring cell density and without conventional induction with IPTG. The method is based on media components that are metabolized differentially to promote growth to high density and automatically induce protein expression from lac promoters.



Lanes	Sample volume
Annexin I	4 µl
Tubulin alpha-4	4 µl
Annexin II	IPTG, 8 µl; OE1, 4.5 µl

Lanes	Sample
IPTG	IPTG induction
OE1	Overnight Express System 1 autoinduction
M	Perfect Protein™ Markers, 15-150 kDa

Analysis of eukaryotic target proteins purified from cultures induced with Overnight Express System 1 versus IPTG
pET recombinants encoding the indicated His•Tag® fusion proteins were transformed into BL21(DE3). Overnight Express System 1 induction was accomplished by inoculating 5 ml medium in 10-ml × 24-well plates with a single colony and incubating overnight (approximately 16 h) at 30°C with shaking at 250 rpm. IPTG induction was accomplished by inoculating 5 ml medium in 10-ml × 24-well plates with a single colony and incubating at 16°C with shaking at 250 rpm to an average OD600 of 1.0 followed by addition of IPTG to 1 mM final concentration and incubating an additional 16 h prior to harvest. Target proteins were extracted and purified using the RoboPop Ni-NTA His•Bind® Purification Kit. The indicated sample volumes were analyzed by SDS-PAGE (10–20% gradient gel) and Coomassie blue staining.



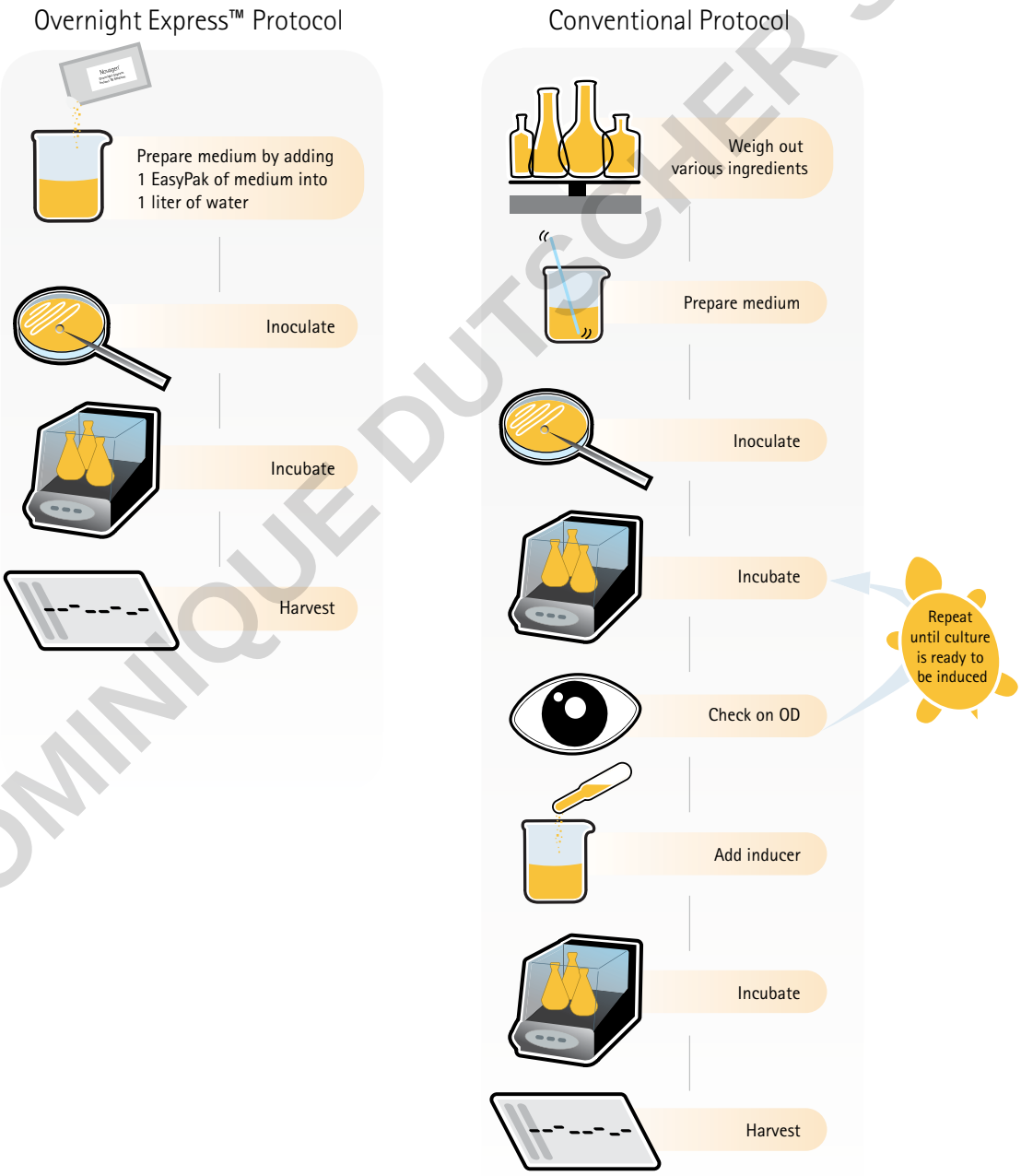
Overnight Express™ systems... for a good night's sleep

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Overnight Express™ Autoinduction Systems

We've done it for you, so that you don't have to!

The Overnight Express Autoinduction System is extremely convenient for routine expression of proteins in multiple cultures and is ideal for high-throughput parallel analysis of protein expression, solubility, and purification from multiple expression clones



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Overnight Express™ Autoinduction Medium Selection Guide

Please reference the selection guide below for the medium for your specific application.

Overnight Express™ Instant LB Medium 6

Medium Complete autoinduction media in granulated Luria-Bertani formulation

Application Most routine recombinant protein expression applications

Overnight Express™ Instant TB Medium 6

Medium Complete autoinduction media in granulated Terrific Broth formulation

Application Most routine recombinant protein expression applications

Overnight Express™ Autoinduction System 1 7

Medium Autoinduction media to be added to glucose-free medium (eg. 2X YT, SOC, LB and TB)

Application Most routine recombinant protein expression applications

Overnight Express™ Autoinduction System 2 9

Medium Chemically defined autoinduction media

Application Compatible with selenomethionyl (Se-Met) labeling of proteins for x-ray crystallography

Overnight Express™ NMR Medium - Optimization 11

Medium Chemically defined autoinduction media

Application Determine optimal culture conditions for high-level protein expression before isotopic protein labeling. It can also be used for ^{15}N protein labeling when user provides ^{15}N-ammonium chloride.

Overnight Express™ NMR Medium - ^{15}N 11

Medium Chemically defined autoinduction media

Application High level incorporation of ^{15}N for initial NMR analysis, to assess suitability for structure determination

Overnight Express™ NMR Medium - ^{15}N, ^{13}C 11

Medium Chemically defined autoinduction media

Application High level incorporation of ^{15}N and ^{13}C for backbone and side-chain assignments and for restraint measurements in structure determination

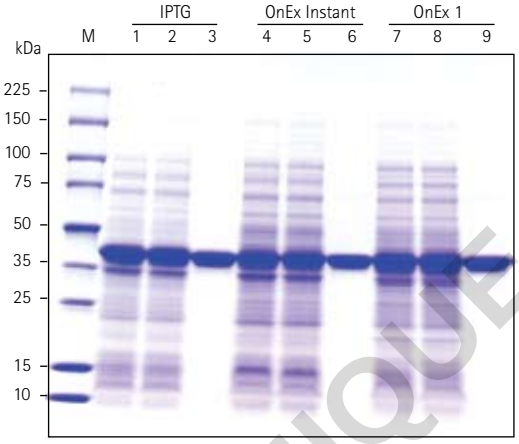
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Overnight Express™ Instant LB and TB Medium

The complete solution for high level protein expression without the need to monitor cell growth

Autoinduction media	▶	Simplifies protocol by eliminating the monitoring and induction steps
Rich culture media	▶	High cell densities and protein expression level
Granulated format	▶	Rapid and uniform dissolution in water Minimize the chance of inhaling hazardous powdered ingredients
Singe use EasyPak packaging	▶	Minimal sample handling, significantly reduce the chance of contamination Convenient, no need to spend extra time on weighting out different ingredients

Granulated Instant LB Medium and TB Medium formulations are combinations of Overnight Express™ System 1 with either Luria-Bertani Broth (LB) or Terrific Broth (TB). These extremely convenient, rich-culture autoinduction media provide high-level protein production in pET and other IPTG-inducible bacterial expression systems.



Lane	Sample
M	Perfect Protein™ Markers, 10-225 kDa
1, 4, 7	pET - Lipocortin 1, TCP
2, 5, 8	pET - Lipocortin 1, Soluble protein
3, 6, 9	pET - Lipocortin 1, Ni-NTA His•Bind® purified protein (3 µg)

15-L fermentations were grown in TB and induced with IPTG or autoinduced during growth in Overnight Express Instant TB Medium (Cat. No. 71491) or during growth in TB medium supplemented with Overnight Express Autoinduction System 1 (Cat. No. 71300). Equivalent cell masses of total cellular protein (TCP) and soluble protein fractions from cellular extracts and purified protein were analyzed by SDS-PAGE (10-20% gradient gel) and stained with Coomassie blue.

Overnight Express™ Instant LB and Instant TB Medium are ideal for routine expression of proteins in multiple cultures and for high-throughput parallel analysis of protein expression, solubility and purification from multiple expression clones.

Two packaging formats are available. The EasyPak aluminum foil pouch contains enough granulated medium to prepare 1 L culture. Just add the EasyPak contents to 1 L sterile water, supplement with 10 ml glycerol, and bring it to a boil in a microwave oven for 2 minutes.

Ordering Information

Product	Size	Cat. No.	Price
Overnight Express™ Instant LB Medium	1 EasyPak (1 x 45 g)	71757-3	€49
	5 EasyPak (5 x 45 g)	71757-4	€200
	1 kg	71757-5	€493
	10 kg	Bulk.	Enquire.

Product	Size	Cat. No.	Price
Overnight Express™ Instant TB Medium	1 EasyPak (1 x 60 g)	71491-3	€49
	5 EasyPak (5 x 60 g)	71491-4	€200
	1 kg	71491-5	€493
	10 kg	Bulk.	Enquire.

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Overnight Express™ Autoinduction System 1

Self induced protein expression system compatible to your specific culture medium

Autoinduction media	Simplifies protocol by eliminating the monitoring and induction steps Ideal for protein production in pET expression system or other IPTG-inducible bacterial system
Sterile stock solutions	Minimal sample handling, significantly reduce the chance of contamination Convenient, no need to spend extra time on weighting out different ingredients
Compatible to any conventional glucose-free bacterial growth medium	Just add it into your existing culture medium; offers you with simplicity and flexibility

The unique Overnight Express™ Autoinduction Medium provides high-level protein production in pET and other IPTG-inducible bacterial expression systems. The Overnight Express™ Autoinduction System 1 kit contains three concentrated sterile solutions:

OnEx™ Solution 1 A blend of carbon sources optimized for tightly regulated uninduced growth to high cell density, followed by high-level induction

OnEx™ Solution 2 A concentrated buffer and nitrogen-source bend that mediates metabolic acid production and provides additional nitrogen for increased protein synthesis

OnEx™ Solution 3 Provides magnesium for maximal cell density.

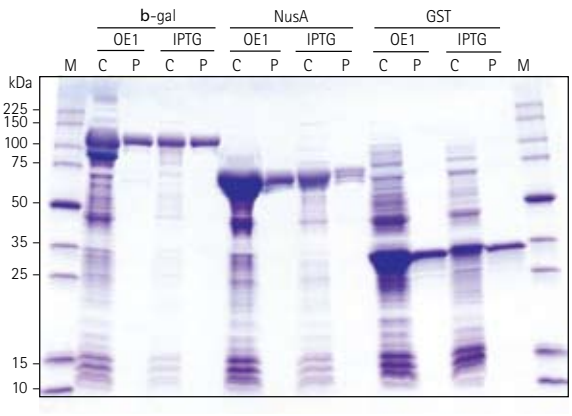


Save time!
We've done the prep work
for you, so that
you don't have to!

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Overnight Express™ Autoinduction System 1 (continued)

Cell mass and target protein yield are often increased several-fold compared to conventional protocols using induction with IPTG.



Lane(s)	Sample volume
M	Perfect Protein™ Markers, 10-225 kDa
OE1	Overnight Express autoinduction
IPTG	IPTG induction
C	Crude protein extract (equal volumes loaded)
P	Purified target protein (equal protein mass loaded)

Expression and purification of target proteins from cultures induced with Overnight Express System 1 (OE1) versus IPTG
pET recombinants encoding ß-gal, NusA, and GST His•Tag® fusion proteins were transformed into BL21(DE3). Protein expression was induced in parallel cultures either by Overnight Express System 1 or 1 mM IPTG. Cells were harvested by centrifugation and protein was extracted with BugBuster® HT Protein Extraction Reagent plus rLysozyme® Solution. After removing a sample, the extract was used for robotic affinity purification using Ni-NTA His•Bind® Resin. Samples of the crude protein extract (7 ml) and of the purified target protein (2 ml) were analyzed by SDS-PAGE (10-20% gel) and Coomassie blue staining.

Ordering Information

Product	Size	Components	Cat. No.	Price
Overnight Express™ Autoinduction System 1	1 Liter Kit	OnEx™ Solution 1: 1 x 20 ml OnEx™ Solution 2: 1 x 50 ml OnEx™ Solution 3: 1 x 1 ml	71300-3	€79
	5 Liter Kit	OnEx™ Solution 1: 5 x 20 ml OnEx™ Solution 2: 2 x 125 ml OnEx™ Solution 3: 5 x 1 ml	71300-4	€315

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Overnight Express™ Autoinduction System 2

Complete, chemically defined medium to selenomethionine (Se-Met) label proteins for crystallography

Chemically defined	Consistent product performance, minimal lot-to-lot variability Ideal for Se-Met labeling of proteins for crystallography
Autoinduction media	Simplifies protocol by eliminating the monitoring and induction steps Ideal for protein production in pET expression system or other IPTG-inducible bacterial system
Sterile stock solutions	Minimal sample handling, significantly reduce the change of contamination Convenient, no need to spend extra time on weighting out different ingredients

The Overnight Express™ Autoinduction System 2 features the capability to label proteins with selenomethionine (Se-Met) for downstream crystallization and x-ray diffraction studies. All components is chemically defined, resulting in consistent product performance by minimizing lot-to-lot variability. The Overnight Express™ Autoinduction System 2 kit contains six concentrated sterile solutions:

OnEx™ Solution 1 A blend of carbon sources optimized for tightly regulated uninduced growth to high cell density, followed by high-level induction

OnEx™ Solution 2 A concentrated buffer and nitrogen-source bend that mediates metabolic acid production and provides additional nitrogen for increased protein synthesis

OnEx™ Solution 3 Provides magnesium for maximal cell density.

OnEx™ Solution 4 Provides trace metals to minimize growth limitations associated with mineral deficiencies and satisfies the metal requirements of metal-containing target proteins, even at high expression levels

OnEx™ Solution 5 A mixture of amino acids lacking methionine

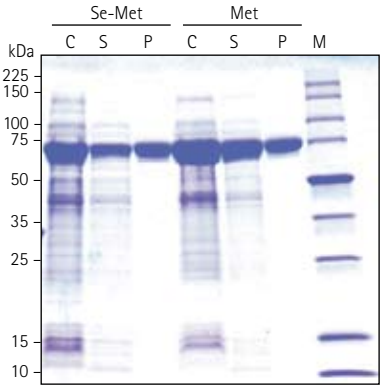
OnEx™ Solution 6 A separate methionine solution. Sufficient methionine (Met) is provided to support growth of the Met auxotroph B834 while providing the ability to reduct the level of unlabeled Met for slenomethionine incorporation by Met auxotrphs

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Overnight Express™
systems...
for a good
night's sleep

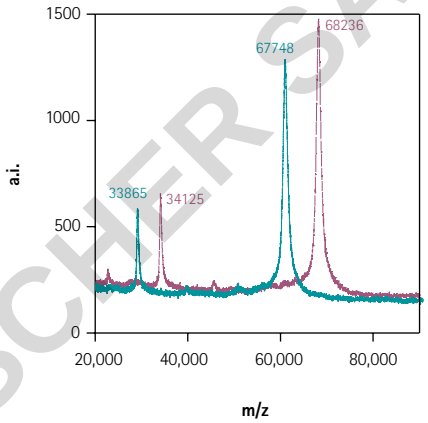
Overnight Express™ Autoinduction System 2 (continued)

The Overnight Express Autoinduction System 2 is the perfect solution for routine expression of proteins in multiple cultures and is ideal for high-throughput parallel analysis of protein expression, solubility screening, and purification from multiple expression clones



Analysis of crude, soluble, and purified proteins from cultures grown in Overnight Express Autoinduction System 2 medium

pET-44b(+) plasmids expressing a His•Tag®/NusA fusion protein were transformed into B834(DE3) cells and grown for 16 h at 37°C in Overnight Express System 2 medium containing Se-Met (125 µg/ml) or Met (OnEx Solution 6). Cultures were centrifuged and cells were resuspended in Ni-NTA Bind Buffer containing AEBSF Hydrochloride, Benzamidine Hydrochloride, and Lysonase™ Bioprocessing Reagent. The suspension was sonicated and centrifuged at 12,000 x *g* for 10 min.Soluble cell extracts were processed by Ni-NTA His•Bind® chromatography. Two micrograms of purified protein (P), 5 µl of soluble extract (S), and a sample of crude extract (C; standardized to harvest OD_{600}) were analyzed by SDS-PAGE (10–20% gradient gel) and Coomassie blue staining. The predicted molecular mass for the pET-44b(+) protein is 67.8 kDa. Lane M: Perfect Protein™ Markers, 10–225 kDa.



Mass spectra

Mass spectroscopy analysis shows Se-Met incorporation into target proteins with Overnight Express System 2 autoinduction as described in figure at left. (Spectra provided by the Mass Spectrometry/Bioanalytical Facility at the University of Wisconsin Biotechnology Center.)

Ordering Information

Product	Size	Components	Cat. No.	Price
Overnight Express™ Autoinduction System 2	1 Liter Kit	OnEx™ Solution 1: 1 x 20 ml OnEx™ Solution 2: 1 x 50 ml OnEx™ Solution 3: 1 x 1 ml OnEx™ Solution 4: 1 x 1 ml OnEx™ Solution 5: 1 x 20 ml OxEx™ Solution 6: 1 x 20 ml	71366-3	€141
	5 Liter Kit	OnEx™ Solution 1: 5 x 20 ml OnEx™ Solution 2: 2 x 125 ml OnEx™ Solution 3: 5 x 1 ml OnEx™ Solution 4: 5 x 1 ml OnEx™ Solution 5: 1 x 100 ml OnEx™ Solution 6: 1 x 100 ml	71366-4	€561

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Overnight Express™ Autoinduction NMR Media

Complete, chemically defined medium to ^{15}N- and ^{13}C- label proteins for NMR spectroscopy

Chemically defined	Consistent product performance, minimal lot-to-lot variability
Stable isotope-labeled nutrients	Reliable and efficient isotopic labeling of proteins. Ideal for ^{15}N- and ^{13}C- labeling of proteins for NMR analysis
Autoinduction media	Simplifies protocol by eliminating the monitoring and induction steps. Ideal for protein productin in pET expression system or other IPTG-inducible bacterial systems
Sterile stock solutions	Minimal sample handling, significantly reduce the change of contamination. Convenient, no need to spend extra time on weighing out different ingredients

The Overnight Express™ Autoinduction NMR Media offer convenient, high-yield expression and efficient isotopic labeling of recombinant proteins for NMR analysis. Each kit includes reagents to make non-inducing, chemically defined starter medium. Starter medium can be used for staged growth without expression, prior to labeling, and for stable storage of bacterial strains containing expression plasmids.

Overnight Express™ Autoinduction NMR Medium – Optimization Intended Purpose: To optimize cell growth and protein expression in minimal medium, before preparing isotopically labeled target proteins for NMR spectroscopy

Overnight Express™ Autoinduction NMR Medium – ^{15}N Intended Purpose: To produce [U-^{15}N] labeled proteins to assess suitability for further analysis

Overnight Express™ Autoinduction NMR Medium – ^{15}N, ^{13}C Intended Purpose: To produce [U-^{15}N, U-^{13}C] labeled proteins to determine structure when supplemented with 13C-labled glycerol (available separately, Cat. No. CLM 1510-EMD)

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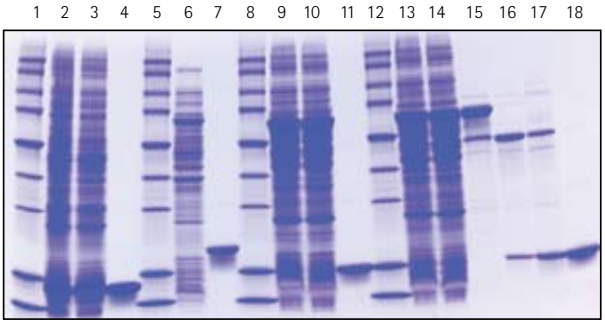
Overnight Express Autoinduction NMR Media Components

Solution	Description	NMR Medium-Optimization ml/L	NMR Medium-^{15}N ml/L	NMR Medium-^{15}N, ^{13}C* ml/L	Non-inducing starter medium ml/200 ml
OnEx™ NMR Solution 1	Autoinducing carbon source; single label	20	20		
OnEx™ Dual-NMR Solution 1	Autoinducing carbon source; dual label			20	
OnEx™ Solution 2	Buffer salts with nitrogen				10
OnEx™ NMR Solution 2	Buffer salts without nitrogen	50	50	50	
OnEx™ Solution 3	Magnesium	2	2	2	0.2
OnEx™ Solution 4	Trace metals	0.2	0.2	0.2	0.04
OnEx™ NMR Solution 5	Amino acids				4
OnEx™ NMR Solution 6	Carbon source (non-autoinducing)				5
†1.0 M ammonium chloride (unla-beled)	Nitrogen source for optimization media	50			
†1.0 M ^{15}N-ammonium chloride	^{15}N source		50	50	
†2.5% (w/v) ^{13}C-glucose*	^{13}C source*			20	

*For high level incorporation of ^{13}C, suitable for NMR structural determination, Overnight Express Autoinduction NMR Medium-^{15}N, ^{13}C must be supplemented with 13C3 Glycerol, available separately (Cat. No. CLM 1510-EMD). 13C3 Glycerol is manufactured by Cambridge Isotope Laboratories, Inc.
†Solution is prepared using the solid component included in the medium kit.

Overnight Express™ Autoinduction NMR Media (continued)

SDS-PAGE analysis of expression and purification samples of proteins produced using Overnight Express Autoinduction NMR Media



Lane	Sample	Lane	Sample
1	Perfect Protein™ Markers 10-225 kDa (PPM)	10	At1g01470 - (S)
2	At5g22580 - total cellular protein (TCP)	11	At1g01470 - (P) (3 mg)
3	At5g22580 - soluble protein (S)	12	PPM
4	At5g22580 - purified protein (P) (3 mg)	13	At1g01470 - (TCP)
5	PPM	14	At1g01470 - (S)
6	At5g01610 (S)	15	At1g01470 - Ni-NTA His•Bind® elution
7	At5g01610 (P) (3 mg)	16	At1g01470 - after protease cleavage
8	PPM	17	At1g01470 - subtractive Ni-NTA His•Bind flowthrough
9	At1g01470 - (TCP)	18	At1g01470 - gel filtration flowthrough

For additional information on this experiment, including isotopic incorporation and NMR data, please see the *inNovations* 26 article at **www.merck4biosciences.com/innovations26OnEx**

Ordering Information

Product	Size	Components	Cat. No.	Price
Overnight Express™ Autoinduction NMR Medium – Optimization	1 Liter Kit	OnEx™ NMR Solution 1: 1 x 20 mL OnEx™ Solution 2: 1 x 10 mL OnEx™ NMR Solution 2: 1 x 50 mL OnEx™ Solution 3: 3 x 1 mL OnEx™ Solution 4: 1 x 1 mL OnEx™ NMR Solution 5: 1 x 4 mL OnEx™ NMR Solution 6: 1 x 5 mL Ammonium Chloride: 2.7 g	71760-3	€98
Overnight Express™ Autoinduction NMR Medium – ^{15}N	1 Liter Kit	OnEx™ NMR Solution 1: 1 x 20 mL OnEx™ Solution 2: 1 x 10 mL OnEx™ NMR Solution 2: 1 x 50 mL OnEx™ Solution 3: 3 x 1 mL OnEx™ Solution 4: 1 x 1 mL OnEx™ NMR Solution 5: 1 x 4 mL OnEx™ NMR Solution 6: 1 x 5 mL Ammonium Chloride: 2.7 g	71759-3	€211
Overnight Express™ Autoinduction NMR Medium – ^{15}N	5 Liter Kit	OnEx™ NMR Solution 1: 1 x 100 mL OnEx™ Solution 2: 1 x 50 mL OnEx™ NMR Solution 2: 2 x 125 mL OnEx™ Solution 3: 1 x 11 mL OnEx™ Solution 4: 2 x 1 mL OnEx™ NMR Solution 5: 1 x 20 mL OnEx™ NMR Solution 6: 1 x 25 mL [^{15}N]Ammonium Chloride: 5 x 2.7 g	71759-4	€843
Overnight Express™ Autoinduction NMR Medium – ^{15}N, ^{13}C	1 Liter Kit	OnEx™ NMR Solution 1: 1 x 100 mL OnEx™ Solution 2: 1 x 50 mL OnEx™ NMR Solution 2: 2 x 125 mL OnEx™ Solution 3: 1 x 11 mL OnEx™ Solution 4: 2 x 1 mL OnEx™ NMR Solution 5: 1 x 20 mL OnEx™ NMR Solution 6: 1 x 25 mL [^{15}N]Ammonium Chloride: 5 x 2.7 g	71789-3	€270
13C3 Glycerol	5 g		CLM 1510-EMD	€1482

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Additional guidelines

Glycerol stock preparation

When growing cultures to prepare glycerol stocks that will be used to inoculate Overnight Express cultures, we recommend the addition of 0.5% glucose to a glucose-free medium (e.g., TB, LB broth, or 2X YT) to maintain plasmid stability. Grow the cells to an OD_{600} of 0.6–0.8 and add 0.1 vol of sterile 80% glycerol. Mix well and store at –70°C.

Aeration: Efficient growth to saturation and utilization of carbon sources provided in Overnight Express medium requires vigorous agitation and proper aeration. Optimized culture volume:vessel dimension ratio is required for proper aeration.

Temperature and length of incubation

It is important to grow the cells to stationary phase when using the Overnight Express Systems. Stationary phase is usually reached as quickly as 8–10 hours, if the cultures are incubated at 37°C. When lower incubation temperatures are used, saturation may only be reached by incubation for 24 hours or more. Continued incubation for several hours after stationary phase appears to have no deleterious effects.

To export the target protein using the signal sequence leaders present in a number of pET vectors or to improve the yield of soluble protein, growth, and induction at 25°C or 30°C may be optimal.

Bacterial strains: Because lactose is used for induction, expression hosts should produce functional **lac** permease (encoded by the *lacY* gene) and β-galactosidase (encoded by the *lacZ* gene) for consistent results in both complex and defined media. *lacY* mutant strains will not efficiently transport lactose for induction and *lacZ* mutants will not convert a portion of the transported lactose into the allolactose inducer. Elevated levels of target gene expression in *lacY* and *lacZ* mutant strains may occur as cells approach stationary phase in some complex media, including Overnight Express medium. However, this induction may vary depending upon medium composition, cell growth stage, and nutrient availability, all of which affect pH and the levels of cyclic AMP and acetate (Grossman et al., 1998).

If using a plasmid with a T7lac promoter for expression, a host strain that does not contain a pLysS plasmid is recommended [e.g., BL21(DE3)]. The combination of the T7 lysozyme expressed by the pLysS plasmid and the lac repressor encoded by pET vectors carrying the T7lac promoter results in significantly reduced levels of protein expression when using the Overnight Express Autoinduction Systems. When the “plain” T7 promoter is used, the low level of lysozyme provided by pLysS has little effect on expression of target proteins.

Expression vectors

Overnight Express Autoinduction Systems are compatible with pET bacterial expression vectors and other IPTG-inducible bacterial expression systems.

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Related Products

Benzonase® Nuclease
Benzonase® Nuclease is a genetically engineered endonuclease from *Serratia marcescens*. It degrades all forms of DNA and RNA (single-stranded, double-stranded, linear, and circular) while having no proteolytic activity. It is effective over a wide range of conditions and possesses an exceptionally high specific activity. The enzyme completely digests nucleic acids to 5´-monophosphate terminated oligonucleotides 2 to 5 bases in length (below the hybridization limit), which is ideal for removal of nucleic acids from recombinant proteins, enabling compliance with FDA guidelines for nucleic acid contamination. The ability of Benzonase® Nuclease to rapidly hydrolyze nucleic acids makes the enzyme an excellent choice for viscosity reduction, to reduce processing time and to increase yields of protein. For example, the enzyme is compatible with BugBuster® and PopCulture® Protein Extraction Reagents and can be added along with these reagents to eliminate viscosity by removing nucleic acids from *E. coli* extracts.

BugBuster® 10x Protein Extraction Reagent
BugBuster® 10X is a concentrated formulation of the proprietary detergents employed in BugBuster® Protein Extraction Reagent without the addition of salts or buffer components. This concentrated formula provides a flexible alternative to the ready-to-use standard 1X reagent, allowing user-defined dilution and addition of buffer components. BugBuster 10X has all of the bioprocessing benefits of standard BugBuster® Protein Extraction Reagent plus the freedom to control pH, reagent concentration, and buffer additives necessary for maximum extraction and activity of your target protein.

PopCulture® Reagent
PopCulture Reagent is a buffered mixture of concentrated detergents formulated to extract proteins from *E. coli* cells directly in their culture medium. Using this method, cell culture, protein extraction and purification performed in the original culture tube or multiwell plate. PopCulture reagent perforates the *E. coli* cell wall without denaturing soluble protein and protects protein from the pH extremes produced in high density culture media. Recombinant proteins can be assayed directly or purified by adding an affinity matrix, washing the matrix-target protein complex to remove culture medium and cellular contaminants and eluting the purified protein from the matrix. Purification of fusion proteins from total culture extracts has been demonstrated using both IMAC and GST affinity chromatography methods. *This product is covered by EP Patent 1,432,822 owned by EMD Chemicals Inc. or its Affiliates.*

RoboPop™ Ni-NTA His•Bind® Purification Kit
The RoboPop™ Ni-NTA His•Bind® Purification Kit is designed for filtration-based 96-well format purification of soluble His•Tag® fusion proteins directly from *E. coli* cultures without harvesting cells. The kit features PopCulture® Reagent, rLysozyme™ Solution, and Benzonase® Nuclease for centrifuge-free cell lysis and extract preparation in one step. The combination of PopCulture extraction, Ni-NTA His•Bind Resin, and a 2-ml filter plate allows high-throughput processing of up to 5 ml of *E. coli* culture per well. Whereas the magnetic-based His•Mag™ kit purifies up to 125 µg target protein per 1 ml culture, the filtration-based kit purifies up to 1 mg His•Tag fusion protein per 5 ml culture. Bacterial culture, cell lysis, and resin binding steps are carried out in standard 24-well plates (not supplied), which accommodate a maximum volume of 5 ml per well. The reaction slurry is then transferred to a 96-well Filter Plate (included) and the washing and elution steps are carried out on a vacuum manifold. The Filter Plate is compatible with standard filter manifolds for manual sample processing, and the entire purification has been validated for robotic sample processing with the Genesis® Freedom™ Workstation from Tecan and the MultiPROBE® II liquid handling work station from PerkinElmer Life Sciences. A 96-well Collection Plate (1-ml wells) with an air-tight aluminum foil sealer is provided for storage of the purified proteins.

Antibiotics
Antibiotics are natural substances secreted by microorganisms that are toxic to other microorganisms, but are generally non-toxic toward higher organisms. We offer a wide range of antibiotics for your protein production needs.

Ampicillin, Sodium Salt
Ampicillin is a ß-lactam antibiotic that kills growing cells by interfering with the terminal reaction in bacterial wall synthesis. Active against Gram-negative bacteria. Inactivated by lactamases. Suitable for use in research that uses ampicillin-resistant plasmids.

Chloramphenicol
Chloramphenicol is a synthetic bacteriostatic antibiotic that inhibits translation of RNA by binding to the 50S ribosomal subunit, blocking the peptidyltransferase reaction of ribosomes. Chloramphenicol resistance is encoded by the resistance gene cat, whose product, chloramphenicol acetyltransferase, inactivates the antibiotic via acetylation. Typically used at 30-50 µg/ml for selection of chloramphenicol-resistant *E. coli*.

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Kanamycin Sulfate, *Streptomyces kanamyceticus* sp.
Kanamycin Sulfate is an aminoglycoside antibiotic effective against Gram-positive and Gram-negative bacteria. It inhibits protein biosynthesis by acting on the 30S ribosome, causing misreading of the genetic code. In mammals, kanamycin may cause renal damage and is ototoxic.

Rifampicin
Antibiotic that specifically inhibits DNA-dependent RNA polymerase in bacteria by forming an inactive complex. Does not affect mammalian RNA polymerase. Inhibits transcription by preventing the initial transcription complex from entering the elongation mode.

Spectinomycin, Dihydrochloride, Pentahydrate, *Streptomyces* sp.
Spectinomycin is a broad-spectrum aminoglycoside antibiotic containing two glucose moieties. It is effective against Gram-positive and Gram-negative bacteria. It inhibits initiation, elongation, and termination of protein synthesis in prokaryotes and induces misreading of the genetic code. Footprint studies indicate that spectinomycin exerts regional effects on ribosomal structure.

Streptomycin Sulfate, *Streptomyces* sp.
Streptomycin Sulfate is an antibiotic effective against Gram-positive and Gram-negative bacteria. It inhibits initiation, elongation, and termination of protein synthesis in prokaryotes, induces misreading of the genetic code. It is often used in culture media to control growth of microorganisms.

Tetracycline, Hydrochloride
Tetracycline, Hydrochloride blocks protein synthesis by inhibiting aminoacyl tRNA binding to the A-site of ribosomes. It induces a cold shock-response and enhances P450 expression in bacteria.

Ordering Information

Product	Size	Cat. No.	Price
Benzonase® Nuclease, Purity > 99%	10 KU	70664-3	€182
Benzonase® Nuclease HC, Purity > 99%	25 KU	71206-3	€376
BugBuster® Master Mix	100 ml	71456-3	€184
	500 ml	71456-4	€580
BugBuster® Protein Extraction Reagent	100 ml	70584-3	€80
	500 ml	70584-4	€320
BugBuster® 10x Protein Extraction Reagent	10 ml	70921-3	€69
	50 ml	70921-4	€304
	100 ml	70921-5	€591
PopCulture® Reagent	15 ml	71092-3	€45
	75 ml	71092-4	€200
	250 ml	71092-5	€604
RoboPop™ Ni-NTA His•Bind® Purification Kit	1 Kit	71188-3	€794
Ampicillin, Sodium Salt	5 gm	171254-5GM	€47
	25 gm	171254-25GM	€161
Chloramphenicol	25 gm	220551-25GM	€43
	100 gm	220551-100GM	€163
Kanamycin Sulfate, *Streptomyces kanamyceticus* sp	5 gm	420311-5GM	€59
	25 gm	420311-25GM	€220
Rifampicin	1 gm	557303-1GM	€64
	5 gm	557303-5GM	€2471
Spectinomycin, Dihydrochloride, Pentahydrate, *Streptomyces* sp.	100 gm	567570-100GM	€53
Streptomycin Sulfate, *Streptomyces* sp.	100 gm	5711-100GM	€53
Tetracycline, Hydrochloride	10 gm	58346-10GM	€31
	25 gm	58346-25GM	€42
	50 gm	58346-50GM	€79

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Finland	00800 1166 8811	00800 1166 8811	00800 1166 8822
France	0800 699 620	0800 126 461	0800 348 630
Germany	0800 6931 000	0800 100 3496	0800 6236 100
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Spain	00800 1166 8811	00800 1166 8811	00800 1166 8822
Sweden	020 796 110	020 796 110	00800 1166 8822
Switzerland	0800 561 470	0800 561 470	00800 1166 8822

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Orders:	**customer.service@merckbio.eu**
Technical Support:	**techservice@merckbio.eu**
Bulk Enquiries:	**bulk@merckbio.eu**

Other Local Merck Offices

Bulgaria
Merck EAD
Tel: +359 2 807 5174
Fax: +359 2 807 5101
E-mail: merck-aquachim@bgnet.bg

Czech Republic
Merck spol. s r.o.
Tel: +420 272 084 272
Fax: +420 272 084 303
E-mail: chemie@merck.cz
Web: www.merck.cz

Hungary
Merck Kft.
Tel: +36 1 463 8100
Fax: +36 1 463 8101
E-mail: kemia@merck.hu
Web: www.merck.hu

Romania
Merck Romania S.R.L.
Tel: +40 21 319 8850
Fax: +40 21 319 8848
E-mail: info.merck@merck.ro
Web: www.merck.ro

Slovenia
Merck d.o.o.
Tel: +386 1 560 3800
Fax: +386 1 560 3830
E-mail: info.chem@merck.si
Web: www.merck.si

Croatia
Merck d.o.o.
Tel: +385 1 4864 111
Fax: +385 1 4864 191
E-mail: kemija@merck.t-com.hr

Greece
Merck S.A.
Tel: +302 10 9885 300, 349, 350
Fax: +302 10 9885 400
E-mail: merck.chem.gr@merck.de

Poland
Merck Sp. z o.o.
Tel: +48 22 53 59 700
Fax: +48 22 53 59 703
E-mail: merck@merck.pl
Web: www.merck.pl

Slovakia
Merck spol. s r.o.
Tel: +421 2 49267 111
Fax: +421 2 49267 777
E-mail: merck@merck.sk
Web: www.merck.sk

For any other country not listed:	**Orders:** +44 (0)115 943 0840	**Technical Support:** +44 (0)115 943 0840	**Fax:** +44 (0)115 943 0951

Merck Chemicals Ltd.
Boulevard Industrial Park,
Padge Road, Beeston
Nottingham NG9 2JR
United Kingdom



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