pDRIVE5s-hbAct

A plasmid with a native ubiquitous human beta actin promoter

Catalog # pdrive5s-hbact

For research use only

Version # 09F03-MM

PRODUCT INFORMATION

Content:

- 1 disk of lyophilized GT116 *E. coli* bacteria transformed by a pDRIVE5s plasmid.
- GT116 genotype is: *F-, mcrA, Δ(mrr-hsdRMS-mcrBC), Ø80lacZΔM15, ΔlacX74, recA1, endA1 Δdcm ΔsbcC-sbcD.*
- 4 pouches of *E. coli* Fast-Media® Zeo (2 TB and 2 Agar)

Shipping and storage:

- Products are shipped at room temperature.
- Transformed bacteria should be stored at -20˚C. Bacteria are stable up to one year when properly stored.
- Store *E. coli* Fast-Media® Zeo at room temperature. Fast-Media® pouches are stable 18 months when stored properly.

Quality control:

- Plasmid construct has been confirmed by restriction analysis and sequencing.
- Bacteria have been lyophilized, and their viability upon resuspension has been verified.

GENERAL PRODUCT USE

pDRIVE is an expression plasmid containing a native or composite promoter of interest. **pDRIVE** may be used to:

- Subclone a promoter of interest into another vector. Unique restriction sites are present at each end of the promoter allowing convenient excision. The 5' sites are *Sda* I, and *Spe* I. *Sda* I is compatible with *Nsi* I and *Pst* I. *Spe* I is compatible with *Avr* II, *Nhe* I and *Xba* I. The 3' restriction site is *Nco* I which includes the ATG start codon, and is compatible with *BspH* I and *BspL*U11 I.

- Compare the activity of different promoters in transient transfection experiments. Each pDRIVE promoter drives the expression of the SEAP reporter gene which allows for testing of the promoter's activity in transient transfection experiments. Furthermore, the SEAP gene is flanked by unique restriction sites (*Nco* I and *Nhe* I) for easy replacement with a different gene of interest.

PROMOTER CHARACTERISTICS

Human beta actin promoter

Complete Promoter size: 2387 bp

Beta-actin is a highly conserved protein ubiquitously expressed in all eukaryotic cells as a component of the cytoskeleton and as a mediator of internal cell motility. A 1.2-kb fragment of the β-actin 5' flanking region is sufficient for efficient transcription[1]. This 1.2-kb fragment contains a TATA box, a CCAAT box and two highly conserved elements. The region between +765 and +783 downstream from the cap site has enhancer activity and acts as a serum-response element in quiescent cells[2]. This 19-bp element contains a CC(A/T)6GG sequence which is found in the promoter region of many cytoskeletal and muscle-specific actin genes. The human β-actin promoter was shown to drive high levels of expression of a transgene in vitro[3]. InvivoGen provides a larger fragment that includes the 5'UTR of the β-actin gene.

1. Sugiyama H. et al. 1988. Strong transcriptional promoter in the 5' upstream region of the human beta-actin gene. Gene. 65(1):135-9. **2. Orita S. et al., 1989.** Identification of a site that mediates transcriptional response of the human beta-actin gene to serum factors. Gene.75(1):13-9. **3. Muller SR. et al. 1990.** Efficient transfection and expression of heterologous genes in PC12 cells. DNA Cell Biol 9(3):221-9.

PLASMID FEATURES

- **SEAP gene** encodes an engineered secreted embryonic alkaline phosphatase. The levels of SEAP in the culture medium of transfected cells expressing the reporter gene can be assayed with chromogenic or luminescent methods
- **SV40 pAn:** The Simian Virus 40 late polyadenylation signal enables efficient cleavage and polyadenylation reactions resulting in high levels of steady-state mRNA.
- **pMB1 Ori** is a minimal *E. coli* origin of replication with the same activity as the longer Ori.
- **EM2K** is a bacterial promoter that enables the constitutive expression of the antibiotic resistance gene in *E. coli.*
- ***Zeo*** gene confers zeocin resistance therefore allowing the selection of transformed *E. coli* carrying a pDRIVE plasmid.

Note: *Stable transfection of clones cannot be performed due to the absence of an eukaryotic promoter upstream of the Sh ble gene.*

METHODS

Growth of pDRIVE-transformed bacteria:

Use sterile conditions to do the following:

1- Resuspend the lyophilized *E. coli* by adding 1 ml of LB medium in the tube containing the disk. Let sit for 5 minutes. Mix gently by inverting the tube several times.

2- Streak bacteria taken from this suspension on a zeocin LB agar plate prepared with the *E. coli* Fast-Media® Zeo agar provided (see below).

3- Place the plate in an incubator at 37˚C overnight.

4- Isolate a single colony and grow the bacteria in TB supplemented with zeocin using the Fast-Media® Zeo liquid provided (see below).

5- Extract the pDRIVE plasmid DNA using the method of your choice.

Selection of bacteria with *E. coli* Fast-Media Zeo:

E. coli Fast-Media® Zeo is a **new, fast and convenient** way to prepare liquid and solid media for bacterial culture by using only a microwave. *E. coli* Fast-Media® Zeo is a TB (liquid) or LB (solid) based medium with zeocin, and contains stabilizers.

E. coli Fast-Media® Zeo can be ordered separately (catalog code # fas-zn-l, fas-zn-s).

Method:

1- Pour the contents of a pouch into a clean borosilicate glass bottle or flask.

2- Add 200 ml of distilled water to the flask

3- Heat in a microwave on MEDIUM power setting (about 400Watts), until bubbles start appearing (approximately 3 minutes). **Do not heat a closed container. Do not autoclave Fast-Media®.**

4- Swirl gently to mix the preparation. **Be careful, the bottle and media are hot, use heatproof pads or gloves and care when handling.**

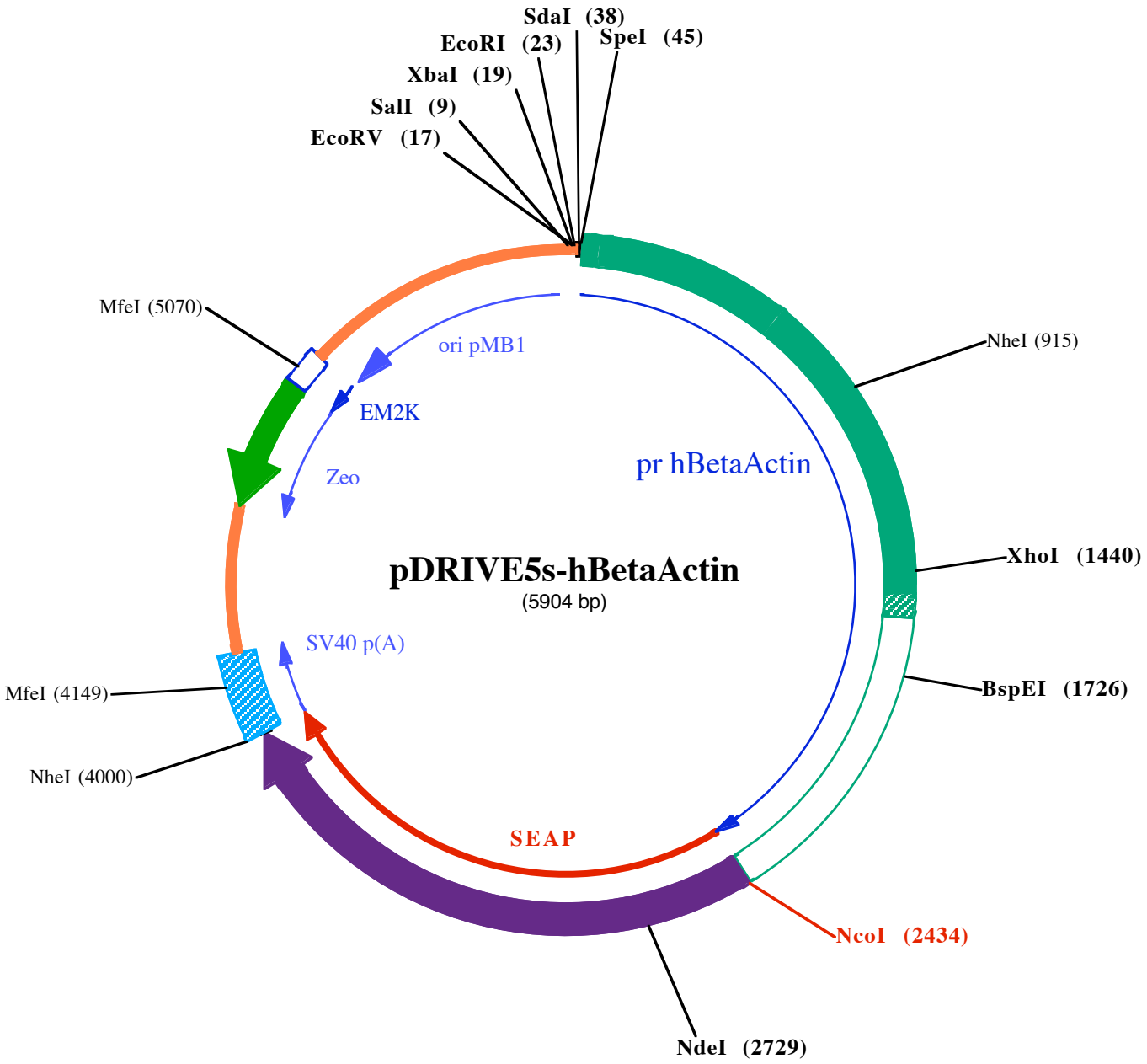
5- Reheat the media for 30 seconds and gently swirl again. Repeat as necessary to completely dissolve the powder into solution. But be careful to avoid overboiling and volume loss.

6- Let agar medium cool to 45˚C before pouring plates. Let liquid media cool to 37˚C before seeding bacteria.

Note: *Do not reheat solidified Fast-Media® as the antibiotic will be permanently destroyed by the procedure.*

TECHNICAL SUPPORT
Toll free (US): 888-457-5873
Outside US: (+1) 858-457-5873
Europe: +33 562-71-69-39
E-mail: info@invivogen.com
Website: www.invivogen.com

InvivoGen
3950 Sorrento Valley Blvd. Suite A
San Diego, CA 92121 - USA


SdaI (38)
SpeI (45)
EcoRI (23)
XbaI (19)
SalI (9)
EcoRV (17)
MfeI (5070)
ori pMB1
EM2K
Zeo
pr hBetaActin
NheI (915)
XhoI (1440)
pDRIVE5s-hBetaActin
(5904 bp)
SV40 p(A)
MfeI (4149)
BspEI (1726)
NheI (4000)
SEAP
NcoI (2434)
NdeI (2729)

InvivoGen

EcoRI (23)
EcoRV (17)
SalI (9) XbaI (19) SdaI (38) SpeI (45)

1 GCGGCCGCGTCGACGATATCTAGAATTCGGATCCTGCAGGGCCCACTAGTTCCATGTCCTTATATGGACTCATCTTTGCCTATTGCGACACACACTCAAT

101 GAACACCTACTACGCGCTGCAAAGAGCCCCGCAGGCCTGAGGTGCCCCCACCTCACCACTCTTCCTATTTTTGTGTAAAAATCCAGCTTCTTGTCACCAC

201 CTCCAAGGAGGGGGAGGAGGAGGAAGGCAGGTTCCTCTAGGCTGAGCCGAATGCCCCTCTGTGGTCCCACGCCACTGATCGCTGCATGCCCACCACCTGG

301 GTACACACAGTCTGTGATTCCCGGAGCAGAACGGACCCTGCCCACCCGGTCTTGTGTGCTACTCAGTGGACAGACCCAAGGCAAGAAAGGGTGACAAGGA

401 CAGGGTCTTCCCAGGCTGGCTTTGAGTTCCTAGCACCGCCCCGCCCCCAATCCTCTGTGGCACATGGAGTCTTGGTCCCCAGAGTCCCCCAGCGGCCTCC

501 AGATGGTCTGGGAGGGCAGTTCAGCTGTGGCTGCGCATAGCAGACATACAACGGACGGTGGGCCCAGACCCAGGCTGTGTAGACCCAGCCCCCCCGCCCC

601 GCAGTGCCTAGGTCACCCACTAACGCCCCAGGCCTGGTCTTGGCTGGGCGTGACTGTTACCCTCAAAAGCAGGCAGCTCCAGGGTAAAAGGTGCCCTGCC

701 CTGTAGAGCCCACCTTCCTTCCCAGGGCTGCGGCTGGGTAGGTTTGTAGCCTTCATCACGGGCCACCTCCAGCCACTGGACCGCTGGCCCCTGCCCTGTC

801 CTGGGGAGTGTGGTCCTGCGACTTCTAAGTGGCCGCAAGCCACCTGACTCCCCCAACACCACACTCTACCTCTCAAGCCCAGGTCTCTCCCTAGTGACCC

NheI (915)
901 ACCCAGCACATTTAGCTAGCTGAGCCCCACAGCCAGAGGTCCTCAGGCCCTGCTTTCAGGGCAGTTGCTCTGAAGTCGGCAAGGGGGAGTGACTGCCTGG

1001 CCACTCCATGCCCTCCAAGAGCTCCTTCTGCAGGAGCGTACAGAACCCAGGGCCCTGGCACCCGTGCAGACCCTGGCCCACCCCACCTGGGCGCTCAGTG

1101 CCCAAGAGATGTCCACACCTAGGATGTCCCGCGGTGGGTGGGGGGCCCGAGAGACGGGCAGGCCGGGGGCAGGCCTGGCCATGCGGGGCCGAACCGGGCA

1201 CTGCCCAGCGTGGGGCGCGGGGGCCACGGCGCGCGCCCCCAGCCCCCGGGCCCAGCACCCCAAGGCGGCCAACGCCAAAACTCTCCCTCCTCCTCTTCCT

1301 CAATCTCGCTCTCGCTCTTTTTTTTTTTCGCAAAAGGAGGGGAGAGGGGGTAAAAAAATGCTGCACTGTGCGGCGAAGCCGGTGAGTGAGCGGCGCGGGG

XhoI (1440)
1401 CCAATCAGCGTGCGCCGTTCCGAAAGTTGCCTTTTATGGCTCGAGCGGCCGCGGCGGCGCCCTATAAAACCCAGCGGCGCGACGCGCCACCACCGCCGAG

1501 ACCGCGTCCGCCCCGCGAGCACAGAGCCTCGCCTTTGCCGATCCGCCGCCCGTCCACACCCGCCGCCAGgtaagcccggccagccgaccggggcaggcgg

1601 ctcacggcccggccgcaggcggccgcggccccttcgcccgtgcagagccgccgtctgggccgcagcgggggcgcatgggggggaaccggaccgccgtg

BspEI (1726)
1701 gggggcgcgggagaagcccctgggcctccggagatgggggacaccccacgccagttcggaggcgcgaggccgcgctcgggaggcgcgctccgggggtgcc

1801 gctctcggggcgggggcaaccggcggggtctttgtctgagccgggctcttgccaatggggatcgcagggtgggcgcggcggagcccccgccaggcccggt

1901 gggggctggggcgccattgcgcgtgcgcgctggtcctttgggcgctaactgcgtgcgcgctgggaattggcgctaattgcgcgtgcgcgctgggactcaa

2001 ggcgctaactgcgcgtgcgttctggggcccggggtgccgcggcctgggctggggcgaaggcgggctcggccggaaggggtggggtcgccgcggctcccgg

2101 gcgcttgcgcgcacttcctgcccgagccgctggccgcccgagggtgtggccgctgcgtgcgcgcgcgccgacccggcgctgtttgaaccgggcggaggcg

2201 gggctggcgcccggttgggagggggttggggcctggcttcctgccgcgcgccgcggggacgcctccgaccagtgtttgccttttatggtaataacgcggc

2301 cggcccggcttcctttgtccccaatctgggcgcgcgccggcgcccctggcggcctaaggactcggcgcgccggaagtggccagggcgggggcgacctcg

NcoI (2434)
2401 gctcacagcgcgcccggctattctcgcagCTCACCATGGTTCTGGGGCCCTGCATGCTGCTGCTGCTGCTGCTGCTGGGCCTGAGGCTACAGCTCTCCCT
1 M V L G P C M L L L L L L L G L R L Q L S L

2501 GGGCATCATCCCAGTTGAGGAGGAGAACCCGGACTTCTGGAACCGCGAGGCAGCCGAGGCCCTGGGTGCCGCCAAGAAGCTGCAGCCTGCACAGACAGCC
22 G I I P V E E E N P D F W N R E A A E A L G A A K K L Q P A Q T A

2601 GCCAAGAACCTCATCATCTTCCTGGGCGATGGGATGGGGGTGTCTACGGTGACAGCTGCCAGGATCCTAAAAGGGCAGAAGAAGGACAAACTGGGGCCTG
56 A K N L I I F L G D G M G V S T V T A A R I L K G Q K K D K L G P

NdeI (2729)
2701 AGATACCCCTGGCTATGGACCGCTTCCCATATGTGGCTCTGTCCAAGACATACAATGTAGACAAACATGTGCCAGACAGTGGAGCCACAGCCACGGCCTA
89 E I P L A M D R F P Y V A L S K T Y N V D K H V P D S G A T A T A Y

2801 CCTGTGCGGGGTCAAGGGCAACTTCCAGACCATTGGCTTGAGTGCAGCCGCCCGCTTTAACCAGTGCAACACGACACGCGGCAACGAGGTCATCTCCGTG
122 L C G V K G N F Q T I G L S A A A R F N Q C N T T R G N E V I S V

2901 ATGAATCGGGCCAAGAAAGCAGGGAAGTCAGTGGGAGTGGTAACCACCACACGAGTGCAGCACGCCTCGCCAGCCGGCACCTACGCCCACACGGTGAACC
156 M N R A K K A G K S V G V V T T T R V Q H A S P A G T Y A H T V N

3001 GCAACTGGTACTCGGACGCCGACGTGCCTGCCTCGGCCCGCCAGGAGGGGTGCCAGGACATCGCTACGCAGCTCATCTCCAACATGGACATTGATGTGAT
189 R N W Y S D A D V P A S A R Q E G C Q D I A T Q L I S N M D I D V I

3101 CCTGGGTGGAGGCCGAAAGTACATGTTTCGCATGGGAACCCCAGACCCTGAGTACCCAGATGACTACAGCCAAGGTGGGACCAGGCTGGACGGGAAGAAT
222 L G G G R K Y M F R M G T P D P E Y P D D Y S Q G G T R L D G K N

3201 CTGGTGCAGGAATGGCTGGCGAAGCGCCAGGGTGCCCGGTATGTGTGGAACCGCACTGAGCTCATGCAGGCTTCCCTGGACCCGTCTGTGACCCATCTCA
256 L V Q E W L A K R Q G A R Y V W N R T E L M Q A S L D P S V T H L

3301 TGGGTCTCTTTGAGCCTGGAGACATGAAATACGAGATCCACCGAGACTCCACACTGGACCCCTCCCTGATGGAGATGACAGAGGCTGCCCTGCGCCTGCT
289 M G L F E P G D M K Y E I H R D S T L D P S L M E M T E A A L R L L

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3401 GAGCAGGAACCCCCGCGGCTTCTTCCTCTTCGTGGAGGGTGGTCGCATCGACCACGGTCATCACGAAAGCAGGGCTTACCGGGCACTGACTGAGACGATC
 322  S  R  N  P  R  G  F  F  L  F  V  E  G  G  R  I  D  H  G  H  H  E  S  R  A  Y  R  A  L  T  E  T  I
3501 ATGTTCGACGACGCCATTGAGAGGGCGGGCCAGCTCACCAGCGAGGAGGACACGCTGAGCCTCGTCACTGCCGACCACTCCCACGTCTTCTCCTTCGGAG
 356  M  F  D  D  A  I  E  R  A  G  Q  L  T  S  E  E  D  T  L  S  L  V  T  A  D  H  S  H  V  F  S  F  G
3601 GCTACCCCCTGCGAGGGAGCTCCATCTTCGGGCTGGCCCCTGGCAAGGCCCGGGACAGGAAGGCCTACACGGTCCTCCTATACGGAAACGGTCCAGGCTA
 389  G  Y  P  L  R  G  S  S  I  F  G  L  A  P  G  K  A  R  D  R  K  A  Y  T  V  L  L  Y  G  N  G  P  G  Y
3701 TGTGCTCAAGGACGGCGCCCGGCCGGATGTTACCGAGAGCGAGAGCGGGAGCCCCGAGTATCGGCAGCAGTCAGCAGTGCCCCTGGACGAAGAGACCCAC
 422  V  L  K  D  G  A  R  P  D  V  T  E  S  E  S  G  S  P  E  Y  R  Q  Q  S  A  V  P  L  D  E  E  T  H
3801 GCAGGCGAGGACGTGGCGGTGTTCGCGCGCGGCCCGCAGGCGCACCTGGTTCACGGCGTGCAGGAGCAGACCTTCATAGCGCACGTCATGGCCTTCGCCG
 456  A  G  E  D  V  A  V  F  A  R  G  P  Q  A  H  L  V  H  G  V  Q  E  Q  T  F  I  A  H  V  M  A  F  A
                                                                                                    NheI
3901 CCTGCCTGGAGCCCTACACCGCCTGCGACCTGGCGCCCCCCGCCGGCACCACCGACGCCGCGCACCCGGGGCGGTCCCGGTCCAAGCGTCTGGATTGAAG
 489  A  C  L  E  P  Y  T  A  C  D  L  A  P  P  A  G  T  T  D  A  A  H  P  G  R  S  R  S  K  R  L  D  •
4001 CTAGCTGGCCAGACATGATAAGATACATTGATGAGTTTGGACAAACCACAACTAGAATGCAGTGAAAAAAATGCTTTATTTGTGAAATTTGTGATGCTAT

                                                MfeI (4149)
4101 TGCTTTATTTGTAACCATTATAAGCTGCAATAAACAAGTTAACAACAACAATTGCATTCATTTTATGTTTCAGGTTCAGGGGGAGGTGTGGGAGGTTTTT

4201 TAAAGCAAGTAAAACCTCTACAAATGTGGTATGGAATTAATTCTAAAATACAGCATAGCAAAACTTTAACCTCCAAATCAAGCCTCTACTTGAATCCTTT

4301 TCTGAGGGATGAATAAGGCATAGGCATCAGGGGCTGTTGCCAATGTGCATTAGCTGTTTGCAGCCTCACCTTCTTTCATGGAGTTTAAGATATAGTGTAT
4401 TTTCCCAAGGTTTGAACTAGCTCTTCATTTCTTTATGTTTTAAATGCACTGACCTCCCACATTCCCTTTTTAGTAAAATATTCAGAAATAATTTAAATAC
4501 ATCATTGCAATGAAAATAAATGTTTTTTATTAGGCAGAATCCAGATGCTCAAGGCCCTTCATAATATCCCCCAGTTTAGTAGTTGGACTTAGGGAACAAA
4601 GGAACCTTTAATAGAAATTGGACAGCAAGAAAGCGAGCTTCTAGCTTATCCTCAGTCCTGCTCCTCTGCCACAAAGTGCACGCAGTTGCCGGCCGGGTCG
                                                    125  •  D  Q  E  E  A  V  F  H  V  C  N  G  A  P  D
4701 CGCAGGGCGAACTCCCGCCCCCACGGCTGCTCGCCGATCTCGGTCATGGCCGGCCCGGAGGCGTCCCGGAAGTTCGTGGACACGACCTCCGACCACTCGG
 108  R  L  A  F  E  R  G  W  P  Q  E  G  I  E  T  M  A  P  G  S  A  D  R  F  N  T  S  V  V  E  S  W  E  A
4801 CGTACAGCTCGTCCAGGCCGCGCACCCACACCCAGGCCAGGGTGTTGTCCGGCACCACCTGGTCCTGGACCGCGCTGATGAACAGGGTCACGTCGTCCCG
  75  Y  L  E  D  L  G  R  V  W  V  W  A  L  T  N  D  P  V  V  Q  D  Q  V  A  S  I  F  L  T  V  D  D  R
4901 GACCACACCGGCGAAGTCGTCCTCCACGAAGTCCCGGGAGAACCCGAGCCGGTCGGTCCAGAACTCGACCGCTCCGGCGACGTCGCGCGCGGTGAGCACC
  42  V  V  G  A  F  D  D  E  V  F  D  R  S  F  G  L  R  D  T  W  F  E  V  A  G  A  V  D  R  A  T  L  V
                                                                                MfeI (5070)
5001 GGAACGGCACTGGTCAACTTGGCCATGATGGCTCCTCCTGTCAGGAGAGGAAAGAGAAGAAGGTTAGTACAATTGCTATAGTGAGTTGTATTATACTATG
   8  P  V  A  S  T  L  K  A  M
5101 CAGATATACTATGCCAATGATTAATTGTCAAACTAGGGCTGCAGGTTAATTAAGAACATGTGAGCAAAAGGCCAGCAAAAGGCCAGGAACCGTAAAAAGG

5201 CCGCGTTGCTGGCGTTTTTCCATAGGCTCCGCCCCCCTGACGAGCATCACAAAAATCGACGCTCAAGTCAGAGGTGGCGAAACCCGACAGGACTATAAAG

5301 ATACCAGGCGTTTCCCCCTGGAAGCTCCCTCGTGCGCTCTCCTGTTCCGACCCTGCCGCTTACCGGATACCTGTCCGCCTTTCTCCCTTCGGGAAGCGTG

5401 GCGCTTTCTCATAGCTCACGCTGTAGGTATCTCAGTTCGGTGTAGGTCGTTCGCTCCAAGCTGGGCTGTGTGCACGAACCCCCCGTTCAGCCCGACCGCT

5501 GCGCCTTATCCGGTAACTATCGTCTTGAGTCCAACCCGGTAAGACACGACTTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGAGGTA

5601 TGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCCTAACTACGGCTACACTAGAAGAACAGTATTTGGTATCTGCGCTCTGCTGAAGCCAGTTACCTTC

5701 GGAAAAAGAGTTGGTAGCTCTTGATCCGGCAAACAAACCACCGCTGGTAGCGGTGGTTTTTTTGTTTGCAAGCAGCAGATTACGCGCAGAAAAAAAGGAT

5801 CTCAAGAAGATCCTTTGATCTTTTCTACGGGGTCTGACGCTCAGTGGAACGAAAACTCACGTTAAGGGATTTTGGTCATGGCTAGTTAATTAACATTTAA

5901 ATCA
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Fast-Media®

Microwaveable media for selection and propagation of *E. coli* transformants

Catalog # fas-xx-l, fas-xx-s, fas-xx-xgal

For research use only

Version # 05C28-SV

PRODUCT INFORMATION

Contents:

E. coli **Fast-Media®** are prepared as individual sealed pouches containing the necessary amount of powder for preparation of 200 ml of selective liquid or agar medium.

30 pouches are supplied for each order of TB or Agar and 20 pouches are supplied for each order of XGal Agar.

Storage and stability:

Fast-Media® are shipped at room temperature, and must be stored in a dry and cool place. They are stable for at least one year at room temperature.

When properly prepared, **Fast-Media®** plates or TB are stable several weeks at 4˚C, and remain sterile and selective**.**

Quality control:

The high quality and performance of each formulation has been tested with some widely used and proprietary *E. coli* K12 derived strains*. These include DH5α, Top10, MC1061, XL1 blue, JM 109, TB1, GT100, GT110.

The adequate plasmids carrying the appropriate *E. coli* resistance genes are used as positive control.

**E. coli* recipient strains carrying the Tn5 transposon are resistant to Kanamycin and Zeocin™.

GENERAL PRODUCT USE

E. coli **Fast-Media®** are microwaveable ready-to-use solid or liquid media, supplied with a selective antibiotic, and chromogenic substrates (for five references), therefore designed for the growth or selection of *E. coli* transformant colonies, as well as detection of blue/white colonies.

- Fast-Media® Agar formulation is LB based agar medium supplemented with selective antibiotic, it's used for selection of resistant *E. coli* colonies after transformation by vectors carrying a selection resistance gene.

- Fast-Media® X-Gal formulation is a LB based agar medium supplemented with selective antibiotic, X-Gal and IPTG. It's used for detection of blue/white resistant colonies after transformation by a vector carrying *LacZ* gene.

- Fast-Media® TB formulation is a Terrific Broth based liquid medium supplemented with selective antibiotic. It's used for high cell density culture of transformed bacteria, and extraction of high quantity and quality of required plasmid.

FAST-MEDIA® FEATURES

E. coli **Fast-Media®** offer researchers a quick and convenient way to prepare 200 ml of liquid culture medium, or 8-10 agar plates in about five minutes USING A MICROWAVE INSTEAD OF AN AUTOCLAVE

E. coli **Fast-Media®** are available with a large variety of prokaryotic selective agents including Ampicillin, Blasticidin S, Hygromycin B, Kanamycin, Puromycin and Zeocin™ (see table below).**Fast-Media®** is also available with no selective agent (Base) that can be prepared with or without antibiotics.

	Agar	X-Gal	TB
Base	√		√
Ampicillin	√	√	√
Blasticidin	√	√	√
Hygromycin	√	√	√
Kanamycin	√	√	√
Puromycin	√		√
Zeocin	√	√	√

SPECIAL HANDLING

Caution should be exercised during handling of Fast-Media® due to potential allergenic properties of antibiotics. Wear protective gloves, do not breath the dust**.**

METHOD

For customer convenience, procedure is directly printed on each pouch.

1- Pour the pouch contents into a clean borosilicate glass bottle or flask.
2- Add 200 ml of distilled or deionized water.
3- Heat in a microwave oven on MEDIUM power setting (about 450W) until bubbles start to appear (about 3 minutes).
Do not heat in a closed container.
4- Swirl gently to mix the preparation and re-heat for 30 seconds. Swirl gently again.
5- Repeat step 4 if necessary until the medium is completely dissolved. Do not overboil.
6- Allow the medium to cool to 50-55 °C, use directly for liquid medium, or pour plates for solid medium.

Caution: *Any solution heated in a microwave oven may become superheated and suddenly boil when moved or touched. Handle with extreme care. Wear heat-proof gloves.*

Note*: Do not repeat this above procedure once the medium is prepared because the antibiotic will be adversely affected.*

For preparation of Fast-Media Base.

- Follow the instructions above and when media has cooled to 50-55 ˚C add the antibiotic at the appropriate concentration for selection of *E. coli*.

TECHNICAL SUPPORT
Toll free (US): 888-457-5873
Outside US: (+1) 858-457-5873
Europe: +33 562-71-69-39
E-mail: info@invivogen.com
Website: www.invivogen.com

InvivoGen
3950 Sorrento Valley Blvd. Suite A
San Diego, CA 92121 - USA