pSELECT-zeo-LacZ
A LacZ Reporter Gene System Selectable with Zeocin™
Catalog # psetz-lacz
For research use only
Version # 11C08-MM

PRODUCT INFORMATION

Content:
- 20 µg of pSELECT-zeo-LacZ plasmid provided as lyophilized DNA
- 4 pouches of E. coli Fast-Media® Zeo (2 TB and 2 Agar)

Storage and Stability:
Product is shipped at room temperature. Lyophilized DNA should be resuspended upon receipt and stored at -20°C. Lyophilized DNA is stable 3 months at -20°C. Resuspended DNA is stable more than one year at -20°C. Store E. coli Fast-Media® Zeo at room temperature. Fast-Media® pouches are stable 18 months when stored properly. Fast-Media® is a TB (liquid) or LB (solid) based medium that already contains the selective antibiotic.

Quality control:
Plasmid construct has been confirmed by restriction analysis and sequencing. Plasmid DNA was purified by ion exchange chromatography and lyophilized.

GENERAL PRODUCT USE
pSELECT plasmids are specifically designed for strong and constitutive expression of a gene of interest in a wide variety of cell lines. They allow the selection of stable transfrectants and offer a variety of selectable markers. pSELECT plasmids contain two expression cassettes: the first drives the expression of the gene of interest and the second drives the expression of a gene of interest in a wide variety of cell lines. They allow the selection of stable transfecants and offer a variety of selectable markers.

pSELECT-LacZ plasmids can be used as control vectors for cloning of an open reading frame, as the LacZ gene is flanked by two unique restriction sites: Nco I at the 5’ end that encompasses the Start codon, and Nhe I at the 3’ end.

pSELECT-LacZ can serve as a gene reporter system for the study of eukaryotic gene expression and regulation. The E. coli lacZ gene encoding β-galactosidase is the classical histochemical reporter gene. β-Galactosidase catalyzes the hydrolysis of X-Gal producing a blue precipitate that can be easily visualized under a microscope. InvivoGen provides two LacZ staining kits for simple and convenient visual detection of LacZ expression within cells or tissues.

- LacZ Cell Staining Kit (sold separately, see RELATED PRODUCTS) allows you to determine the percentage of transfected cells expressing the lacZ gene. The assay can be completed in 30 minutes, and the blue precipitate can take from 30 minutes to overnight to appear.

- LacZ Tissue Staining Kit (sold separately, see RELATED PRODUCTS) allows the detection of transduced cells expressing the lacZ gene within fresh or frozen tissues. The staining of the tissues can be observed in 5 to 24 hours.

PLASMID FEATURES

First expression cassette
- **hEF1-HTLV prom** is a composite promoter comprising the Elongation Factor-1a (EF-1a) core promoter and the R segment and part of the U5 sequence (R-U5’) of the Human T-Cell Leukemia Virus (HTLV) Type 1 Long Terminal Repeat. The EF-1a promoter exhibits a strong activity and yields long lasting expression of a transgene in vivo. The R-U5’ has been coupled to the EF-1a core promoter to enhance stability of RNA.

- **LacZΔCpG gene**: a humanized and CpG-free allele of the LacZ gene. This CpG-free gene is ten times more active than the wild-type gene in mammalian cells. It can be used for in vitro or in vivo applications.

- **SV40 pAn**: The Simian Virus 40 late polyadenylation signal enables efficient cleavage and polyadenylation reactions resulting in high levels of steady-state mRNA.
- **ori**: a minimal E. coli origin of replication to limit vector size, but with the same activity as the longer Ori.

Second expression cassette
- **CMV enh/prom**: The human cytomegalovirus immediate-early gene 1 promoter/enhancer was originally isolated from the Towne strain and was found to be stronger than any other viral promoters.
- **EM7** is a bacterial promoter that enables the constitutive expression of the antibiotic resistance gene in E. coli.
- **Zeo**: Resistance to Zeocin™ is conferred by the Sh ble gene from Streptococcichrobium Hindustanus. The Sh ble gene is driven by the CMV enhancer/promoter in tandem with the bacterial EM7 promoter allowing selection in both mammalian cells and E. coli.
- **ßGlo pAn**: The human beta-globin 3' UTR and polyadenylation sequence allows efficient arrest of the transgene transcription.

METHODS

Plasmid resuspension:
Quickly spin the tube containing the lyophilized plasmid to pellet the DNA. To obtain a plasmid solution at 1 µg/µl, resuspend the DNA in 20 µl of sterile H2O. Store resuspended plasmid at -20°C.

References:

RELATED PRODUCTS

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<th>Product</th>
<th>Catalog Code</th>
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<tr>
<td>Fast-Media® Zeo TB (20 pouches)</td>
<td>fas-zn-l</td>
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<tr>
<td>Fast-Media® Zeo Agar (20 pouches)</td>
<td>fas-zn-s</td>
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<td>LacZ Cell Staining Kit (100 reactions)</td>
<td>rep-lz-c</td>
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<td>LacZ Tissue Staining Kit (100 ml)</td>
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TECHNICAL SUPPORT
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InvivoGen
3950 Sorrento Valley Blvd. Suite 100
San Diego, CA 92121 - USA
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 # 10G07-MM

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 2 years at room temperature.

When properly prepared, Fast-Media® plates or broths are stable for 4 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, GT115, GT116.

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 is 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 is 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.

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<tr>
<th></th>
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SPECIAL HANDLING

Caution should be exercised during handling of Fast-Media® due to potential allergic 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- Mix thoroughly by swirling the glass bottle or flask.
4- 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.**
5- Swirl gently to mix the preparation and re-heat for 30 seconds. Swirl gently again.
6- Repeat step 4 if necessary until the medium is completely dissolved. Do not overboil.
7- 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 supplemented 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

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