ATCC

Product Sheet

lambdaMGU2 *C. elegans* cDNA library (ATCC® 77366™)

Please read this FIRST

Storage Temp.
Store unopened frozen vial at -80°C or lower. Vapor phase liquid nitrogen is preferred for long term storage.

Biosafety Level
1

Intended Use

This product is intended for research use only. It is not intended for any animal or human therapeutic or diagnostic use.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: lambdaMGU2 *C. elegans* cDNA library (ATCC® 77366™)

Shipping Information

Frozen bacteria-free phage lysate

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

Or contact your local distributor

Description

Designation: lambdaMGU2 *C. elegans* cDNA library
Distribution Host: *Escherichia coli* q358 (ATCC® 47018™) (for propagation)
Escherichia coli 1046 [pCRE1] (ATCC® 77368™) (for conversion to phagemid)

Propagation

Thaw contents of the vial in a 37°C water bath with gentle agitation until no ice crystals remain. Library can be diluted and plated following standard protocols. Recommended growth media is LB. Recommended growth temperature is 37°C.

Starting and Amplifying ATCC Bacteriophage Lambda Clones and Vectors:

1. Prepare fresh plating bacteria. Grow *E. coli* host strain overnight or at least to A_{600} = 0.4 in medium containing 0.2% maltose (to give higher titers).
2. Spin down cells in a low speed centrifuge. Resuspend in 0.4 volumes 10 mM $MgSO_4$ or SM buffer. Store at 4°C until ready to use. These cells are good for up to 2 weeks if stored at 4°C.
3. Open freeze dried vial containing the phage according to instructions.Asceptically add 0.3 to 0.4 mL of liquid medium to the freeze-dried pellet and mix well.
4. Pipette 100 µL of the host suspension to a sterile test tube. Add 3 mL. of warm (50°C) LB lambda top agar (see below) containing 0.2% maltose and mix gently. Pour onto plates. Allow the plates to solidify.
5. Spot a loopful or two of the phage suspension on the lawn of the freshly poured bacteria.
6. Incubate overnight at 37°C. Fresh plates give larger plaques.
7. Cut plaques out of agar and add them to 0.5 mL of 10 mM $MgSO_4$ or SM buffer and store at 4^0C overnight.
8. Add 100 µL of the overnight phage dilution to 100 µL prepared plating bacteria and mix gently. Incubate in a 37°C water bath for 20 minutes to allow phage to adsorb.
9. Add 3 mL. LB lambda top agar containing 0.2% maltose and mix gently. Pour onto plates. Incubate overnight at 37°C.
10. Invert open plate over a chloroform-saturated adsorbent paper for 10 minutes.
11. Add 7.5 mL of 10 mM $MgSO_4$ or SM buffer to the plate and allow it to stand at room temp for 1 hour or in 4°C overnight.
12. Collect and save the liquid on the plate. This should be a high titer lysate. Add a few drops of chloroform if its going to be stored for more than a few days.

LB Lambda top agar medium:
NaCl, 5 g
Tryptone, 10 g
Yeast extract, 5 g
Distilled water to 1 L

Sterilize at 121°C, 15 minutes. Cool to approximately 50°C and add the following sterile solutions.

1M $CaCl_2$, 5 mL
$MgSO_4$ H_2O to a final concentration of 0.2% w/v
50% maltose, 5 mL

Vector Information

Library information:
Genome: *Caenorhabditis elegans*
Strain: N2
Type of insert: cDNA
Vector: lambdaMGU2
Insert size range (kb): 2-3 kb average
Vector ends: Modification: BamHI
Number of independent recombinants: 1.0 x 10^7
Titer: 5 x 10^7 pfu/mL

Vector information:
Vector: lambdaMGU2
Vector type: phage
Intact vector size (kb): 41.7

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Frozen bacteria-free phage lysate

Construction: lambda2690, pMGU
Features:
Insert detection: Spi+
Marker: ampR
Replicon: lambda, pMB1, m13

References

References and other information relating to this product are available online at www.atcc.org.

Notes

To prepare phagemid from lambdaMGU2, grow recombinants on a RecA- host expressing the Cre protein such as *E. coli* 1046 [pCRE1] (ATCC77368) and select for ampicillin resistance. The pMGU product is 4.185 kb. Efficiency of phagemid recovery is approximately 20%. Plasmid pCRE1 may be a low level contaminant, but is easily distinguished from pMGU DNA. Inserts can be amplified using the following primers flanking the BamHI cloning site : upstream 5' -AAGAGGCAGAACTGGCAG-3' and downstream 5' -ATCGATGCATAGCGATTC-3'. The order of the major features in the cloning region of the lambda vector is : lambda J - SmaI - SalI - loxP - EcoRI - M13 ori - ampR - pMB1 ori - hindIII - 3'gam/BamHI/5'gam - XhoI - loxP - SalI - lambda N.
- Gene 120 : 135-141, 1992.
Single stranded DNA may be recovered from phagemid constructs using M13KO7 helper phages.
- Gene 120 : 135-141, 1992

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

ATCC Warranty

The viability of ATCC® products is warranted for 30 days from the date of shipment, and is valid only if the product is stored and cultured according to the information included on this product information sheet. ATCC lists the media formulation that has been found to be effective for this strain. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this strain. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.

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This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of strains on deposit, ATCC is not liable for damages arising from the misidentification or misrepresentation of cultures.

Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at www.atcc.org

Additional information on this culture is available on the ATCC web site at www.atcc.org.

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