





Open Existing Project	Enter a DNA sequence, or select from other options, to identify cut sites. Once you submit a sequence, you may choose to customize your digest.	
Name 1 × Name 2 ×	1. Input or choose sequence. Explain	
Name 3 ×	Paste Sequence	
Log in to have unlimited access to your projects.		
✓ Disable cookies Explain		Plain or FASTA/ Max: 300 KBases
	OR Upload File (xx MB max.) Upload File No file selected.	
	OR Enter Gen Bank Browse Gen Bank	<u>S</u>
	OR Select Sequence \$ Select Plasmid Vectors \$ \$ Select Viral a	and Phage
	2. Update preferences. Explain	
	✓ Circular Additional Preferences	
	3. Name project (optional). Explain	

Submit







Open Existing Project	Additional Preferences	×	
Name 1 ×	Enzymes to include:	Methylation sensitivities to include:	
Name 2 ×	■ NEB enzymes (Type II & Type III)	✓ CpG methylation	
Name 3 ×	Time-Saver [™] qualified enzymes	✓ Dam methylation	
Log in to have unlimited	All commercially available specificities	✓ Dcm methylation	
access to your projects.	All specificities	EcoBI methylation	
✓ Disable cookies Ex	All & defined oligonucleotide sequences	✓ EcoKI methylation	
V Bloadle coolings Ex	Only defined oligonucleotide sequences <u>Define Oligos</u>	Minimum ORF length:	Plain or FASTA/ Max: 300 KBases
	In addition to Type II & commercially-available Type III enzymes, also include: Type I & III enzymes Homing endonucleases Nicking enzymes	Genetic code for ORFs: \$\Displays \text{ Select Genetic Code}\$	nk and Phage
	Process this region only: Save Cancel	bp	
	3. Name project (opt	tional). Explain	









Make PDF



Print

Graphical View

Enzyme List

Sequence

ORF Summary

Customize Digest

Results For:

- Linear
- NEB single cutter restriction enzymes
- · Main non-overlapping, min. 100 aa ORFs

Sequence Information:

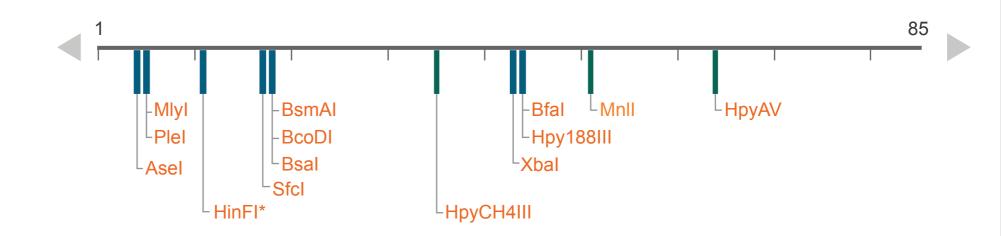
2817 bp GC = 34% AT = 66%

Update Preferences

Start Over

Standard Digest for PURExpr prmr

Use the menu to explore this digest, or to create a custom digest.



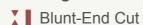
Display







Cleavage Code





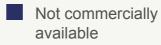


▼ 3′ Extension

Enzyme Code

NEB

Other supplier



Update Colors

Methylation

- * CpG methylation
- # dam/dcm methylation
- () Ambigous site







Make PDF

Graphical View

Enzyme List

Sequence

ORF Summary

Customize Digest

Results For:

- Linear
- NEB single cutter restriction enzymes
- · Main non-overlapping, min. 100 aa ORFs

Sequence Information:

2817 bp GC = 34% AT = 66%

Update Preferences

Start Over

Standard Digest for PURExpr prmr

Cut ▼ Position	Enzyme 🔺	Recognition Sequence	Me Sens.
2	MluCI_	^AATT_	
5	Asel	AT^TA AT	
5	<u>Msel</u>	T^TA_A	
6	<u>FspEI</u>	 CC(N)12^NNNN	
6	Plel_	GAGTCNNNN^N	
6	Mlyl	GAGTC(N)5	
7	<u>FspEI</u>	CC(N)12^NNNN_	
7	<u>MspJI</u>	CNNR(N)9^NNNN_	
12	<u>Hinfl</u>	G^ANT_C	CpG
18	<u>Sfcl</u>	C^TRYA_G	
19	<u>FspEI</u>	CC(N)12^NNNN_	CpG
19	<u>BsmAI</u>	GTCTCN^NNNN_	
19	<u>Bsal</u>	GGTCTCN^NNNN_	
19	<u>BcoDI</u>	GTCTCN^NNNN_	
26	<u>MspJI</u>	CNNR(N)9^NNNN_	
36	HpyCH4III	AC_N^GT	
42	<u>FspEI</u>	CC(N)12^NNNN_	
42	<u>MspJI</u>	CNNR(N)9^NNNN_	
42	<u>MspJI</u>	CNNR(N)9^NNNN_	
44	<u>Xbal</u>	T^CTAG_A	

Display

✓ 0 cutters

✓ 2 cutters

✓ All sites



NEB Restriction Enzyme Activity/Performance Chart







Graphical View

Enzyme List

Sequence

ORF Summary

Customize Digest

Results For:

- Circular
- NEB single cutter restriction enzymes
- Main non-overlapping, min. 100 aa ORFs

Sequence Information:

2817 bp GC = 34% AT = 66%

Update Preferences

Start Over

Standard Digest for LITMUS39

Make PDF

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- 1 GTTAACTACG TCAGGTGGCA CTTTTCGGGG AAATGTGCGC GGAACCCCTA TTTGTTTATT
- 61 TTTCTAAATA CATTCAAATA TGTATCCGCT CATGAGACAA TAACCCTGAT AAATGCTTCA
- 121 ATAATATTGA AAAAGGAAGA GTATGAGTAT TCAACATTTC CGTGTCGCCC TTATTCCCTT
- 181 TTTTGCGGCA TTTTGCCTTC CTGTTTTTGC TCACCCAGAA ACGCTGGTGA AAGTAAAAGA
- 241 TGCTGAAGAT CAGTTGGGTG CACGAGTGGG TTACATCGAA CTGGATCTCA ACAGCGGTAA
- 301 GATCCTTGAG AGTTTTCGCC CCGAAGAACG TTCTCCAATG ATGAGCACTT TTAAAGTTCT
- 361 GCTATGTGGC GCGGTATTAT CCCGTGTTGA CGCCGGGCAA GAGCAACTCG GTCGCCGCAT
- 421 ACACTATTCT CAGAATGACT TGGTTGAGTA CTCACCAGTC ACAGAAAAGC ATCTTACGGA
- 481 TGGCATGACA GTAAGAGAAT TATGCAGTGC TGCCATAACC ATGAGTGATA ACACTGCGGC
- 541 CAACTTACTT CTGACAACGA TCGGAGGACC GAAGGAGCTA ACCGCTTTTT TGCACAACAT
- 601 GGGGGATCAT GTAACTCGCC TTGATCGTTG GGAACCGGAG CTGAATGAAG CCATACCAAA
- 661 CGACGAGCGT GACACCACGA TGCCTGTAGC AATGGCAACA ACGTTGCGCA AACTATTAAC
- 721 TGGCGAACTA CTTACTCTAG CTTCCCGGCA ACAATTAATA GACTGGATGG AGGCGGATAA
- 781 AGTTGCAGGA CCACTTCTGC GCTCGGCCCT TCCGGCTGGC TGGTTTATTG CTGATAAATC
- 841 TGGAGCCGGT GAGCGTGGGT CTCGCGGTAT CATTGCAGCA CTGGGGCCAG ATGGTAAGCC
- 901 CTCCCGTATC GTAGTTATCT ACACGACGGG GAGTCAGGCA ACTATGGATG AACGAAATAG
- 961 ACAGATCGCT GAGATAGGTG CCTCACTGAT TAAGCATTGG TAACTGTCAG ACCAAGTTTA
- 1021 CTCATATATA CTTTAGATTG ATTTACCCCG GTTGATAATC AGAAAAGCCC CAAAAACAGG
- 1081 AAGATTGTAT AAGCAAATAT TTAAATTGTA AACGTTAATA TTTTGTTAAA ATTCGCGTTA
- 1141 AATTTTTGTT AAATCAGCTC ATTTTTTAAC CAATAGGCCG AAATCGGCAA AATCCCTTAT
- 1201 AAATCAAAAG AATAGCCCGA GATAGGGTTG AGTGTTGTTC CAGTTTGGAA CAAGAGTCCA
- 1261 CTATTAAAGA ACGTGGACTC CAACGTCAAA GGGCGAAAAA CCGTCTATCA GGGCGATGGC
- 1321 CCACTACGTG AACCATCACC CAAATCAAGT TTTTTGGGGT CGAGGTGCCG TAAAGCACTA
- 1381 AATCGGAACC CTAAAGGGAG CCCCCGATTT AGAGCTTGAC GGGGAAAGCG AACGTGGCGA
- 1441 GAAAGGAAGG GAAGAAAGCG AAAGGAGCGG GCGCTAGGGC GCTGGCAAGT GTAGCGGTCA
- 1501 CGCTGCGCGT AACCACCACA CCCGCCGCGC TTAATGCGCC GCTACAGGGC GCGTAAAAGG
- 1561 ATCTAGGTGA AGATCCTTTT TGATAATCTC ATGACCAAAA TCCCTTAACG TGAGTTTTCG
- 1621 TTCCACTGAG CGTCAGACCC CGTAGAAAAG ATCAAAGGAT CTTCTTGAGA TCCTTTTTTT
- 1681 CTGCGCGTAA TCTGCTGCTT GCAAACAAAA AAACCACCGC TACCAGCGGT GGTTTGTTTG
- 1741 CCGGATCAAG AGCTACCAAC TCTTTTTCCG AAGGTAACTG GCTTCAGCAG AGCGCAGATA







Graphical View

Enzyme List

Sequence

ORF Summary

Customize Digest

Results For:

- Circular
- NEB single cutter restriction enzymes
- Main non-overlapping, min. 100 aa ORFs

Sequence Information:

2817 bp GC = 34% AT = 66%

Update Preferences

Start Over

Standard Digest for LITMUS39

a.a.

Seq.

286 aa 121 aa Coordinates

143..1003

2424..2789

Protein

*BsrBI

ID

Product

beta-lactamase

alpha fragment

beta-galactosidase

Gene

lacZalpha

bla

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Closest	Flanking	
5´ End	3´ End	Enzymes
<u>Earl</u>	<u>BsaJI</u>	Show

Show

Hpal







	Custom Digest for pU	C19	Make
Graphical View	EcoRI	NEB #R0101	×
Enzyme List	Features:	Methylation:	_
Fragments	 5 minute Time-Saver Buffer: NEBuffer EcoRI Salt: 50 mM NaCI 	! This site overlaps a CpG site: GAATTCg , overlapping sites may impair cleavage if methylated	
Gel	 Main: 100 mM Tris-HCI pH: 7.5 Mg: 10 mM MgCl2 Triton: 0.025% 		
Digested With: EcoRI HindIII Sequence Information: 2686 bp GC = 51% AT = 49%	 • Inton: 0.025% • Rxn temp.: 37 °C • Neoschizomers: None • Isoschizomers: None • Sites in sequence: 1/2 • End produced at 396: 5′ overhang: AATT REBASE page	Enzymes producing compatible ends:	
Jpdate Preferences Start Over	NEB Restriction Enzyme Activity/Performance Chart	Enzymes with no sites in pUC19:	







Create Custom Digest for pUC19

Only enzymes that cut the sequence and match your selection of availability are shown. Non-NEB neoschizomers are not listed, but you can enter it in the search field and its prototype will be selected.

Jump to enzyme(s) or oligo sequence(s)



Select All	Enzyme ▼	Recognition Sequence	Cuts
	<u>AatII</u>	GACGTC	1
	Acc65I	GGTACC	1
	<u>Accl</u>	GTMKAC	1
	<u>Acil</u>	CCGC	34
	<u>AcII</u>	AACGTT	2
	<u>Acul</u>	CTGAAG(N)14NN	2
	<u>AfIIII</u>	ACRYGT	1
	<u>Ahdl</u>	GACNNNNNGTC1	1
	<u>Alul</u>	AGCT	16
	<u>Alwl</u>	GGATCNNNNN	10
	<u>AlwNI</u>	CAGNNNCTG	1
	<u>ApaLI</u>	GTGCAC	3
	<u>ApeKI</u>	GCWGC	12
	<u>Apol</u>	RAATTY	1
	<u>Asel</u>	ATTAAT	3
	<u>Aval</u>	CYCGRG	1
	<u>Avall</u>	GGWCC	2
	<u>BaeGI</u>	GKGCMC	3
	<u>BamHI</u>	GGATCC	1
	<u>Banl</u>	GGYRCC	4

Filter

Enzymes with compatible buffers

Enzymes producing blunt ends

✓ Enzymes producing 5' overhangs

✓ Enzymes producing 3' overhangs

Enzymes with a particular site length

✓ Enzymes cutting N times

Non-palindromic enzymes

Selections:

- ApeKI
- BaeGI
- EcoRI

Digest

Reset







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FEEDBACK ? HELP

Print

Graphical View

Enzyme List

Fragments

Gel

Digested With:

- EcoRI
- HindIII

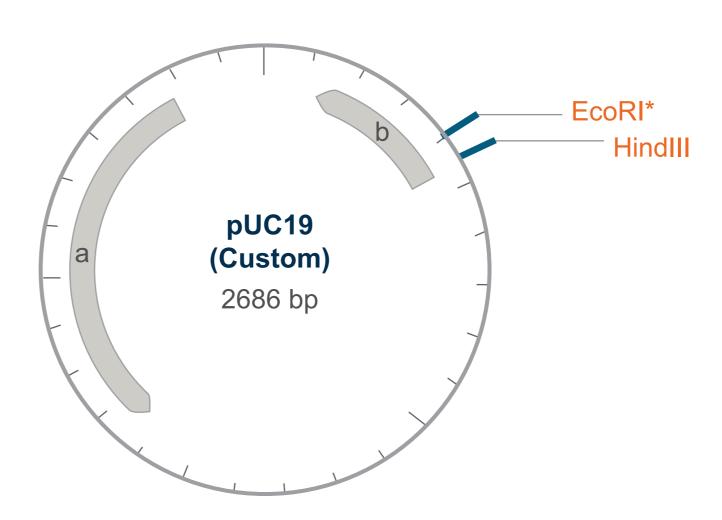
Sequence Information:

2686 bp GC = 51% AT = 49%

Update Preferences

Start Over

Custom Digest for pUC19



Display

Circular

Linear

To zoom into cut sites. use linear display.

Cleavage Code

▼ Blunt-End Cut

Cuts 1 strand

5' Extension

▼ 3′ Extension

Enzyme Code

NEB

Other supplier

Not commercially available

Update Colors

Methylation

- * CpG methylation
- # dam/dcm methylation
- () Ambigous site

ORFs

a: bla

b: lacZalpha







Graphical View

Enzyme List

Fragments

Gel

Digested With:

- EcoRI
- HindIII

Sequence Information:

2686 bp GC = 51% AT = 49%

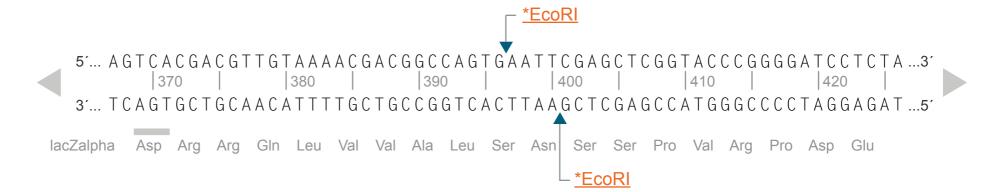
Update Preferences

Start Over

Custom Digest for pUC19

Make PDF

Print



Display

Circular

Linear

To zoom into cut sites, use linear display.

Cleavage Code

I Blunt-End Cut

▼ Cuts 1 strand

5' Extension

▼ 3′ Extension

Enzyme Code

NEB

Other supplier

Not commercially available

Update Colors

Methylation

* CpG methylation

dam/dcm methylation

() Ambigous site

ORFs

a: bla b: lacZalpha







Make PDF



Print

Graphical View

Enzyme List

Fragments

Gel

Digested With:

- EcoRI
- HindIII

Sequence Information:

2686 bp

GC = 51% AT = 49%

Update Preferences

Start Over

Custom Digest for pUC19



#	Enzyme ▼	Recognition Sequence	Cuts	Sites & Flanks	Cut Positions	Blunt/ Overhang
1	<u>EcoRI</u>	G^AATT^C	1	<u>Show</u>	*396/400	5′
2	<u>HindIII</u>	A^AGCT^T	1	Show	447/451	5′

Save as Text

Display



✓ 0 cutters

✓ 2 cutters



✓ All sites



✓ Flanking enzymes



NEB Restriction Enzyme Activity/Performance Chart







Graphical View

Enzyme List

Fragments

Gel

Digested With:

- EcoRI
- HindIII

Sequence Information:

2686 bp GC = 51% AT = 49%

Update Preferences

Start Over

Custom Digest for pUC19

Make PDF

Print

♦ Sort by	
-----------	--

#	Ends	Coordinates	Length (bp)
1	HindIII-EcoRI	448-396	26351
2	EcoRI-HindIII	397-447	51

#	Ends	Coordinates	Length (bp)
1	EcoRI-HindIII	397-447	51
2	HindIII-EcoRI	448-396	2635

Save as Text







Make PDF

Graphical View

Enzyme List

Fragments

Gel

Digested With:

- EcoRI
- HindIII

Sequence Information:

2686 bp

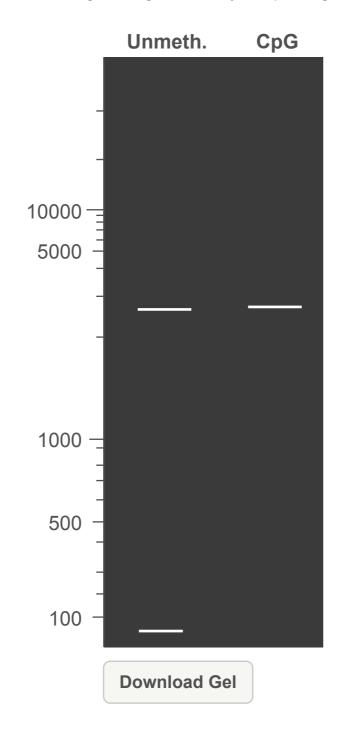
GC = 51% AT = 49%

Update Preferences

Start Over

Custom Digest for pUC19

This virtual gel was generated by interpolating experimental data. See details.



#	Ends	Coordinates	Length (bp)
1	HindIII-EcoRI	448-396	26351
2	EcoRI-HindIII	397-447	51

