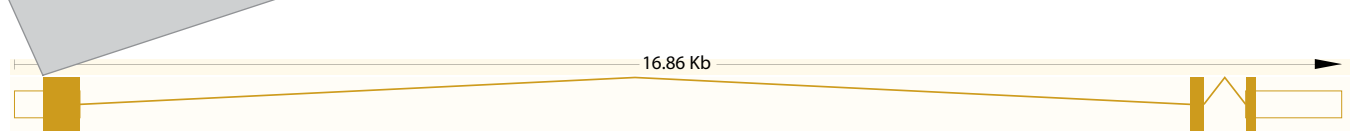


Meox1-nucTagRFPT-ires-CE Construct Overview

Created 5 April 2012
Updated 17 May 2011

Gene Overview

nucTagRFPT-ires-CE



Meox1-001 ENSMUST0000057054

Design comments

There is a single reported transcript for the Meox1 locus (Ensembl). A nuclear-localized TagRFP-T was linked to a CreERT2 fragment with the EMCV IRES sequence to create a bicistronic reporter cassette. (See Bochkov and Palmenberg (2006) BioTechniques 41 (3):283-8.) There is a putative splice variant, however this does not include the first exon where the reporter was placed.

Target site in cDNA

cDNA for Meox1-001

Transcript length: 2,228bps Translation length: 253 residues

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1  CGGGTCCAGCCCTCTGCAGGCACCTCAATCACTCAGGGTCCCGGGAAGGGCTGTGCAGTA
61  TGGGTGGTCCAAAGTAGGACAGTCAAAATGTTTCAGCATGGTAGGAAACAACCTCCGTGCC
121  CGATAGTCAGCGTGTGTCTGGGGCAGGAAGGCAGACGTGAGCCCTAGACAGGTGTGGACA
181  CGCACATGTGTGTCTCCGGCCACGTGTTTGTGGAATTTGAGGCAAAATTTTTGTTTTGGT
241  TCCTGGGGTAAAGTTTCCATTCAACATTTTCCCTACTGTTTAATTTTTTTTTTAATTTT
301  AAATTACAAAACCTCTGACTAGAAAAAGCCCAATACCTTTTGAAGGACTGGGGCAGGCAGT
361  GGACAGCAGATGGATCCAGTGGCCCAACAGCTGTGTGAGGAACCCCGAGCCCGAGCCCT
421  GTCTGGGGTGCCTTCGAAACCCCACTCAGAAGATAGCAGCCCTCAGGGCTGTCCCAT
481  TATCCCCAACCCCGTTTTCTTCCACCAAAAATCAGACTTCCAGCGCACAGCAGCATAC
541  CCGACTTCTCTGCTTCTGCTGGCTGGCAGCCACCCACACAGCTGCCCCGGACTGAGCGA
601  ATCTTCAACGAGCAGCATCTCCCTTCCACAGACCCCTGACTGGCACTTCCCTATCTCA
661  GAAGCCGGGCAGAGGCTCAACCTAGGCCACGCTGGGAGCCAGGAGATGGGAGCCGGC
721  AGCCCCGGCCTGGTGGATGGCACAGCAGGATTTGGGGAGGATTCATGGTACTTGGGAGC
781  ATCGCCAATGAGACGGAGAAGAAATCATCCAGAAGGAAAAAGAGAGGTGAGACACCCAG
841  GAGAACGGAGGAGGGAAGCCAGAAGGCAGCAGCAAGCCCGAAGGAGAGGACAGCCCTTC
901  ACCAAGGAGCAGCTACGGGAGCTGGAGGCAGAGTTTGCCACCACTACCTGACCCGG
961  CTCGGGAGATATGAGATTGCAGTCAACTGGACCTTCTGAGCGGCAGGTCAAAGTCTGG
1021  TTCAGAACCGGAGGATGAAGTGGAAACGTGTGAAGGGGGTTCAGCCTGTGTCGCCACAG
1081  GAGCAGGACCGAGAGGATGGGACTCTGCAGCTTCTCCAAAGTTCAGAGTGAGTGTCCCA
1141  AAGACCAAAACCAAGAAAGACTGAAGAAACCCCTCTCCAGTTCCACACCCCGGTTCCCA
1201  CCCTCTCACATCTCTGGACCCACCCAGGGCAGCCTGCACACACTTAAGGTGTGAAGTTG
1261  CCGATATGTGGGAGCCCTGAATTTCCCAAGAAGGCTTAGCTCAGCGTCTTACATCTCAG
1321  CAGCCCTTCCCAGGCCCTTCAACTTCCACTCTCTCTGAACTCCAAGGACTGGGAAA
1381  CTATAGAAAACAGACCTGGGGCCCTTTTCTGGGTCTCTTGGCTACTACCACACTAG
1441  CATCCCCCTTACATCAGGGTCTCTCTGGGTCTGGCCCATCTGGCCTATGAGAGATC
1501  CATCTCTGGTACCAGCTGGTACTTGTAAAAGCAAACTCTCTCCAGATGTTACACCCG
1561  TTGAGTTGAAGGTAGGAAGTGGCTCCTGTGTCTAAGGACGGAGGATAGCACAAGAGC
1621  TGATGGATGAGGACTGAGATACGTGACCTTGAAGTGAAGCTTCTCAGGCTCAGCGT
1681  CTTGTGTTCTCAAGGACTTGACTGCAGATAAGGACAGGAAAAACAACCATCTCTCTCC
1741  ATGGGATGCATTTGGACCTATCTCTGCGTGTTCGGGAAAAAGCTTTGTGGGAGACCTCCC
1801  AGGTTACACACATGCGCAGCTCAGATCTCAGACCAACTTCTGAGGATGCTCTCTGTGAG
1861  GACTGGTGGGAAAAACAGCCTCAGGCAACAGTCTCTTGGAAAGACCCCTGTGTGCCTCA
1921  GTATTAGATGGTGGATCTCCAGATCTGCTGATGTGCAAGGAGGGTTCAGCAAGTAT
1981  TTGAATTTCTTGCATGGAGATCTGATTTGTGAGTGTTTAAAAATAACCCAGTCTCCCTTCC
2041  CCACCCATCAAGACAGAAGCTGTGGAAAATGATTTGTCAAATGAGATGCCAAGTTAGAGC
2101  ATGTATCAGTTTTCCCTACAGCATATTTTCATATGTTTTTTTTTCTTAAGATTACATCA
2161  GCTAATTTGCAAGGTCAATTCACTTTGTGAAGAAAACCTCAGAGAAATAAATCAACAAA
2221  AAAATGCA
    
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Reporter Cassette

The nucTagRFPT-ires-CE reporter was inserted at the consensus start ATG of the Meox1 coding region. The Neo/Kan component is used for selection in bacteria and removed with transient expression of Flp-recombinase prior to microinjection.



pBS-nucTagRFPT-i-CE

5621 bp (molecule 8498 bp)

Meox1-nucTagRFPT-ires-CE Target Site Details

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Endogenous Targeting Site (includes homology arms)

Exon 1 p55-2
agcatCGGGT CCAGCCCTCT GCAGGCACTC ATTCACCTAG GGTCCCCGGG AAGGGCTTGT CAGTATGGGT GGGTCCAAGG TAGGACAGTC AAAATGTTCA
tcgtaGCCCC GGTCCGGAGA CTCCCGTAG TAAGTGAGTC CCAGGGGCC TTCCCGAACA GTCATACCCA CCCAGGTTCC ATCCTGTGAG TTTTACAAGT

Exon 1
GCATGGTAGG AAACAACCTC GTGCCGATA GTCAGCGTGT GTCGGGGGCA GGAAGGCAGA CTGAAGCCT AGACAGGTGT GGACACGCAC ATGTGTGTTT
CGTACCATCC TTTGTTGAGG CACGGGCTAT CAGTCGCACA CAGCCCCCT CTTCCGTCT GCACTTCGGA TCTGTCCACA CCTGTGCGTG TACACACAAG

Exon 1
CTGCCACGT GTTGTGGAA TTTGAGGCAA AATTTTGTG TTGGTTCCTG GGGTAAAGTT TCCATTCAAC ATTTTCCTCT ACTGTTTAAT TTTTNTTTTA
GACCGGTGCA CAAACACCTT AAATCCGTT TTAATAACAA AACCAAGGAC CCCATTTCAG AGTAAGTTG TAAAGGAGA TGACAAATTA AAAAAAAT

Exon 1
ATTTTAAAT ACAAACTCT GACTAGAAA AGCGCAATAC CTTTGAAAG ACTGGGGCAG GCAGTGGACA GCAGATGGAT CCAGTGGCCA ACAGCTGTGT
TAAATTTAA TGTTTGGAGA CTGATCTTTT TCGCGTTATG GAAACTTTC TGACCCCGTC GTGCACCTGT CGTCTACCTA GGTACCCGGT TGTCGCACACA

Exon 1
GAGGAACCC CAGCCCCAG CCCCTGTCTG GGGCTGCCTT CGAAACCCCT ACTCAGAAGA TAGCAGCGCC TCAGGGCTGT CCCATTATCC CCCAACCCCG
CTCCTTGGGG GTCGGGGGTC GGGGACAGAC CCCGACGAA GCTTTGGGG TGAGTCTTCT ATCGTCGCGG AGTCCCGACA GGGTAATAGG GGGTGGGGG

Targeted Site - 5'

Meox1 mRNA (partial)
TTCTGGCCA CGTGTGAGT GAATTTGAGG CAAAATTTT GTTTTGGTTC CTGGGGTAAA GTTTCATTC AACATTTTC TCTACTGTTT AATTTTNTT
AAGGACCGGT GCACAAACAC CTTAACTCC GTTTTAAAA CAAAACCAAG GACCCCATTT CAAGGTAAG TTGTAAAAGG AGATGACAAA TTAATAAAAA

Meox1 mRNA (partial) Kozak nucTagRFPT-T
TTAATTTTAA ATTACAAAAC TCTGACTAGA AAAAGCGCAA TACCTTGAA AGGACTGGGG CAGGCAGTGG ACAGCAGGCC ACCATGGTGC ACGTGGATCC
AATTAATAAT TAATGTTTGG AGACTGATCT TTTTCGCGTT ATGAAAACCT TCCTGACCCC GTCCTGCACC TGTCGTCGG TGTGACCACG TGCACCTAGG

nucTagRFPT-T
AAAAAAGAAG AGAAAGGTAG ATCCAAAAA GAAGAGAAA GTAGATCCAA AAAAGAAGAG AAAGGTACAC GTGAGCATGG TGTCTAAGGG CGAAGAGCTG
TTTTTCTTTC TCTTCCATC TAGGTTTTT CTCTCTTTC CATCTAGGTT TTTCTTCTC TTCCATGTT CACTCGTACC ACAGATTCCT GCTTCTCGAC

Targeted Site - 3'

BGH pA
GGGGGTGGGG TGGGCAGGA CAGCAAGGGG GAGGATTGGG AAGACAATAG CAGGCATGCT GGGGATGCGG TGGGCTCTAT GGATCCAGT GGCCAACAGC
CCCCACCCC ACCCGTCTCT GTCGTTCCCT CTCTAACCCT TTCTGTTATC GTCCGTACGA CCCCTACGCC ACCCGAGATA CCTAGTCA CCGTTGTCG
TGTGTGAGGA ACCCCAGCC CCCAGCCCTT GTCTGGGCT GCCTTCGAAA CCCCCTCA GAAGTAGCA GCGCCTCAGG GCTGTCCAT TATCCCCAA
ACACTCCT TGGGGTGGG GGGTCGGGGA CAGACCCGA CGGAAGCTTT GGGGTGAGT CTCTATCGT CGCGGAGTCC CGACAGGTA ATAGGGGGTT
CCCCGTTTC CTCCACAAA AAATCAGACT TCCAGCGAC AGCAGCATA CCGACTTCT CTGCTTCCTG CTGGCAGCC ACCCCACACA GCCTGCCCC
GGGCAAAAG GAAGTGGTT TTAGTCTGA AGGTCGCTG TCGTCGTATG GGGCTGAAGA GACGAAGGAC GGACCGTCG TGGGTGTGT CCGACGGGGC
GACTGAGCGA ATCTTCAACG AGCAGCATCC TGCCTTCCCA CAGACCCCTG ACTGGCACTT CCCTATCTCA GAAGCCGGGC AGAGGCTCAA CCTAGGCCA
CTGACTCGCT TAGAAGTTGC TCGTCGTAGG ACGGAAGGTT GTCTGGGAC TGACCGTAA GGGATAGAGT CTTCGGCCCG TCTCCAGTT GGATCCGGT
pGT33
GCTGGGAGCG CCAGGGAGAT GGGAGCCGGC AGCCCCGGCC TGGTGGATGG CACAGCAGGA TTGGGGGAGG ATTGCATGTT ACTTGGGACG ATCGCCAATG
CGACCTCGC GGTCCCTCTA CCCTCGGCC TCGGGGCCGG ACCACTTACC GTGTCGTCTT AACCCCTCC TAACGTACCA TGAACCTGC TAGCGGTTAC
AGACGGAGAA GAAATCATCC AGAAGAAAA AAGAGAGGTC AGGTAGTGA TGGAGGAA
TCTGCCTCT CTTTAGTAG TCTTCTTTT TTCTCTCCAG TCCATCCACT ACCTCCTT
p33-2

Meox1-nucTagRFPT-ires-CE BAC Transgene

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BAC clone RP23-5O22 was targeted by recombineering. The genomic context of the nucTagRFP-ires-CE reporter is shown below. The BAC and the target gene are highlighted in yellow.

